ORIGIN & OUTLOOK

5th International Sponge Symposium 1998

QUEENSLAND MUSEUM

Book of ABSTRACTS

WORKSHOPS ON *'SYSTEMA PORIFERA'* & *'PALAEONTOLOGICAL SPONGE BUILD-UPS'*, 27-28 June 1998

Saturday 27 June 1998

Registration desk open (9.00am-5.00pm)

9.00-10.45: Concurrent workshop sessions

1. '*Systema Porifera*' (Chairman: Dr John Hooper) (Venue: Museum Theatre, Level 2)

Aims: *'To summarise, and revise where necessary, the supraspecific classification of the Phylum Porifera, based on re-evaluation of type material and recent biological evidence. The resultant classification, to be published as a book, will be a synthesis of contemporary revisions, morphological systematics and non-morphological evidence. Wherever possible systematics will be interpreted in a phylogenetic framework, although for some groups this may not yet possible. Illustrations will be provided to the major characters used to define genera, families and higher taxa, and keys to these taxa will be presented'*. It is expected that this completed project will provide a previously inaccessible window onto the taxonomy of this difficult phylum. The book will be divided into chapters based on families or logical family groups, plus one or more overview chapter, to be written by 33 authors. Its scheduled date of publication by Plenum (New York) is circa mid 2000.

This (3rd) '*Systema*' workshop aims to facilitate the completion of all chapters, discuss the contents and format of the overview chapter(s), and try to resolve some of the problems remaining (or agree on the best strategies for presenting the unresolvable issues). The workshop will commence with a status report on the project by the editors. Those authors attending the 5th ISS will be then asked to provide status reports and estimates of time to submission of their respective chapter(s). These presentations will be informal. Authors' presentations and subsequent discussions will be conducted along the lines of a forum. Authors may have **up to 15 minutes each** to speak on their respective groups and the status and problems of these groups. A brief written summary or progress report should be provided to the editors at the close of the workshop. The workshop will be open to delegates of the 5th ISS, who are welcome to participate in the discussions.

2. '*Palaeontological sponge build-ups*' (Chairman: Prof Joachim Reitner) (Venue: Student Room 1, Level 1)

Aims: This workshop will focus on the questions of origins of the Porifera and the metazoa, and their subsequent evolution as interpreted using geobiological approaches; the role and preservation potential of Porifera in build-ups throughout the earth's history (e.g. "mudmounds", reefs); and the future and directions of sponge paleontology.

❒ **10.45-11.15: morning tea**
11.15-1.00: workshops continue

❒ **1.00-2.00: lunch (provided)**
2.00-3.30: workshops continue

❒ **3.30-4.00: afternoon tea**
4.00-5.30: workshops continue

❒ **Evening Free**

Sunday 28 June 1998

Registration desk open (9.00am-6.00pm)

9.00-10.30: concurrent workshops continue

❒ **10.30-11.00: morning tea**
11.00-12.00: workshops conclude

1.00-6.00: Delegate registration, posters assembled, etc.

- ❒ **6.00pm-8.00pm: Official Welcome and Reception, Queensland Museum Foyer, Level 2**

5th INTERNATIONAL SPONGE SYMPOSIUM, *'ORIGIN & OUTLOOK'*

29 June – 3 July 1998

Monday 29 June 1998 (VENUE: MUSEUM THEATRE, LEVEL 2)

Registration desk open from 8.00am

- **The Astra Pharmaceuticals (Australia) Keynote & Plenary Session**

9.00-9.05: **Opening remarks (Convener)**

9.05-9.35: **The Ian Potter Foundation Keynote Address** – *'Origin & Outlook'* – **Prof Claude Lévi** (Ian Potter Foundation Visiting Scholar) (Muséum National d'Histoire Naturelle, Paris, France)

9.35-10.05: *Plenary Address:'The Past'*: **Dr Françoise Debrenne** (Muséum National d'Histoire Naturelle, Paris, France)

10.05-10.35: *Plenary Address: 'The Present'*: **Prof Patricia Bergquist** (University of Auckland, New Zealand)

10.35-11.05: *Plenary Address: 'The Future'*: **Dr Jean Vacelet** (Station Marine d'Endoume, Marseille, France)

❒ **11.05-11.35: morning tea**

11.35-11.50: Group photograph (in theatre)

- **Session 1: Palaeontology & Palaeoecology (Chairman: Prof Joachim Reitner, University of Göttingen, Germany)**

11.50-12.10: Distinctive Middle Cambrian sponge-calcimicrobe reefs in Iran. **Dr Peter Kruse** (NT Geological Survey, Department of Mines and Energy, Darwin, Australia)

12.10-12.30: Remarks on the paleoecology and reef building potential of Late Jurassic siliceous sponges. **Mr Manfred Krautter** (Institut für Geologie und Paläontologie, Universität Stuttgart, Germany)

12.30-12.50: Taphonomy and Preservation Potential of Sponge Tissue. (Reitner, Neuweiler, Schumann-Kindel & Thiel) **Prof Joachim Reitner** (Institut und Museum für Geologie und Paläontologie, Universität Göttingen, Germany)

12.50-1.10: An overview of Stromatoporoid-dominated Middle Devonian reef complexes in north Queensland. **Dr Alex Cook** (Queensland Museum, Brisbane, Australia)

1.10-1.30: The phylogenetic history of sponges in Palaeozic time. **Dr Dorte Mehl** (Institut für Paläontologie, Freie Universität Berlin, Germany)

❒ **1.30-2.30: lunch**

2.30-2.50: Meso-Cenozoic history of the siliceous sponges with rigid skeleton. **Dr Andrzej Pisera** (Institute of Paleobiology, Polish Academy of Sciences, Warszawa, Poland)

2.50-3.10: [Title to be announced]. **Dr Theo Engeser** (Institut und Museum für Geologie und Paläontologie, Universität Göttingen, Germany)

- **Session 2A: Systematics (I), Morphometrics & Phylogeny (Chairman: Dr Sven Zea, INVEMAR, Universidad Nacional de Colombia, Colombia)**

3.10-3.30: Origin and early fossil record of Sponges-A geobiological approach. (Reitner, Schumann-Kindel & Thiel). **Prof Joachim Reitner** (Institut und Museum für Geologie und Paläontologie, Universität Göttingen, Germany)

3.30-3.50: New hexactinellids from the Mendocino Ridge off Oregon, USA. **Dr Henry Reiswig** (Redpath Museum, McGill University, Montreal, Canada)

3.50-4.10: Phylogenetics of the euretids (Euretidae; Hexactinellida). **Mr Ben Wheeler** (Biological Sciences, Redpath Museum, McGill University, Montreal, Canada)

❒ **4.10-4.40: afternoon tea**

4.40-5.00: The freshwater sponges from a Neotropical sand dune area. (Volkmer-Ribeiro, Correia, Brenha & Mendonça) **Prof Cecilia Volkmer-Ribeiro** (Museu de Ciências Naturais, Fundaçao Zoobotânica do Rio Grande do Sul, Porto Alegre, Brazil)

5.00-5.20: Sponges of the Low Isles, Great Barrier Reef: an important scientific site, or a case of mistaken identity? (Hooper, List, Kennedy, Cook & Valentine) **Dr John Hooper** (Queensland Museum, Brisbane, Australia)

5.20-5.40: *Isodictya* Bowerbank: Poecilosclerid or Haplosclerid (Samaai, Kelly-Borges & Gibbons) **Mr Toufiek Samaai** (Zoology Department, University of the Western Cape, Bell Ville, South Africa)

5.40-6.00: The demosponge fauna of the Houtman Abrolhos Islands. **Dr Jane Fromont** (Western Australian Museum, Perth, Australia)

6.00-6.20: Do we need subgenera in Haplosclerida? Convenience versus phylogenetic reliability. **Dr Ruth Desqueyroux-Faundez** (Muséum d'Histoire Naturelle, Geneva, Switzerland)

6.20-6.40: Sponge distribution off northern Jamaica down to 107m. **Dr Helmut Lehnert** (Institut und Museum für Geologie und Paläontologie (IMGP), Universität Göttingen, Germany)

6.40-7.00: Proposal of a phylogenetic classification of the mycalids with anisochelae, and comments on the status of *Naviculina* Gray, 1867. **Dr Eduardo Hajdu** (Museu Nacional, Departamento de Invertebrados, Universidade Federal do Rio de Janeiro, Brazil)

❒ **Evening free**

Tuesday 30 June 1998

Registration desk open from 8.00am

- **Session 2B: Systematics (II), Chemotaxonomy & Biogeography (Chairman: Dr Shirley Pomponi, Harbor Branch Oceanographic Institution, Inc., USA).**

8.30-8.50: Pushing the Boundaries - a new genus and species of Dictyoceratida. (Sorokin, Bergquist & Karuso) **Ms Shirley Sorokin** (Museum of Tropical Queensland, Townsville, Australia)

8.50-9.10: Biogeography and taxonomy of the reef cave dwelling coralline demosponge *Astrosclera willeyana* throughout the Indo-Pacific. (Woerheide & Reitner) **Dr Gert Woerheide** (Institut und Museum für Geologie und Paläontologie, Universität Göttingen, Germany; currently Queensland Museum, Brisbane, Australia)

9.10-9.30: Biodiversity, species composition and distribution of marine sponges in northeastern Australia. (Hooper, Kennedy, List-Armitage, Cook & Quinn) **Dr John Hooper** (Queensland Museum, Brisbane, Australia)

- **Session 3: Marine Natural Products, Biochemistry & Biosynthesis (Chairman: Assoc. Prof Mary Garson, University of Queensland, Brisbane, Australia)**

9.30-9.50: Production of bioactive metabolites by symbiotic microorganisms in marine sponges. (Faulkner, Bewley, Salomon, Schmidt & Unson) **Prof. John Faulkner** (Scripps Institution of Oceanography, University of California, San Diego, USA)

9.50-10.10: Cyanide and thiocyanate-based biosynthesis in tropical marine sponges (Simpson & Garson) **Dr Mary Garson** (Department of Chemistry, University of Queensland, Brisbane, Australia)

10.10-10.30: Theonellapeptolides from the deep-water New Zealand sponge *Lamellomorpha strongylata*. **Dr Murray Munro** (Chemistry Department, University of Canterbury, Christchurch, New Zealand)

❒ **10.30-11.00: morning tea**

11.00-11.20: [Title to be announced] **Prof Ron Quinn** (Queensland Pharmaceutical Research Institute, Griffith University, Brisbane, Australia)

11.20-11.40: Drug Farming in the 21st Century. (Duckworth, Battershill, Schiel & Bergquist) **Dr Chris Battershill** (National Institute of Water and Atmospheric Research Ltd, Oceanographic (NZOI), Wellington, New Zealand)

11.40-12.00: Chemical and antimalarial investigations of some *Cymbastela* species. (Koenig & Wright) **Dr Tony Wright** (Institute for Pharmaceutical Biology, Technical University of Braunschweig, Germany)

12.00-12.20: Antimicrobial activity of Caribbean sponge extracts (Newbold, Pawlik, Jensen & Fenical) **Dr Rochelle Newbold** (Biological Sciences, University of North Carolina at Wilimington, North Carolina, USA)

❒ **12.20-1.20: lunch**

- **Session 4: Chemical Ecology (Chairman: Dr Jane Fromont, Western Australian Museum, Perth, Australia)**

1.20-1.40: Discovery and sustainable supply of marine natural products as drugs, industrial compounds and agrochemicals: chemical ecology, genetics, aquaculture and cell culture. (Battershill, Page, Duckworth, Miller, Bergquist, Blunt, Munro, Northcote, Newman & Pomponi) **Dr Chris Battershill** (National Institute of Water and Atmospheric Research Ltd, Oceanographic (NZOI), Wellington, New Zealand)

1.40-2.00: The ecological role of cytotoxic alkaloids: *Haliclona* n.sp., an unusual sponge/dinoflagellate association (Clark, Field, Charan, Flowers, McCaffrey, Garson & Webb) **Dr Mary Garson** (Department of Chemistry, University of Queensland, Brisbane, Australia)

2.00-2.20: An assessment of 'space wars' as a determining factor in the production of novel bioactive indoles by *Ircinia* sp. (Evans-Illidge, Bourne & Wolff) **Ms Libby Evans-Illidge** (Australian Institute of Marine Science, Townsville, Australia)

2.20-2.40: Production of bioactive furanosesterterpene tetronic acids as possible internal chemical defense mechanism in the sponge *Ircinia felix* (Porifera: Demospongiae) (Zea, Parra, Martinez & Duque) **Dr Sven Zea** (INVEMAR, Universidad Nacional de Colombia, Santa Marta, Colombia)

2.40-3.00: The release of allelochemicals by three tropical sponges (Demospongiae) and their toxic effects on coral substrate competitors. (Nishiyama & Bakus) **Mr Greg Nishiyama** (Department of Biological Sciences, University of Southern California, Los Angeles, USA)

3.00-3.20: Intraspecimen variation of secondary metabilites in *Oceanapia* sp. and effects towards generalist and specialist predators. (Schupp, Eder, Paul & Proksch) **Mr Peter Schupp** (Lehrstuhl für Pharmazeutische Biologie, University of Würzburg, Germany)

3.20-3.40: *Negombata magnifica* - a magnificent (chemical) pet. **Dr Micha Ilan** (George S. Wise Faculty of Life Sciences, Department of Zoology, Tel-Aviv University, Israel)

❒ **3.40-4.10: afternoon tea**

4.10-4.30: Serotonin in Porifera? Evidence from developing *Tedania ignis*, the Caribbean fire sponge (Demospongiae) (Weyrer, Rützler & Rieger) **Dr Klaus Rützler** (Invertebrate Zoology, National Museum of Natural History Smithsonian Institution, Washington DC, USA)

4.30-4.50: Temperate sponge assemblages exposed to grazing: the role of refugia and chemical defence. **Dr Andy Davis** (Australian Flora and Fauna Research Centre, Department of Biological Sciences, University of Wollongong, Australia)

4.50-5.10: Predation on Caribbean sponges. The importance of chemical defenses. **Dr Joe Pawlik** (Biological Sciences and Centre for Marine Science Research, University of North Carolina at Wilmington, North Carolina, USA)

5.10-5.30: Sponge Distribution and Lake Chemistry among northern Wisconsin Lakes: Minna Jewell's Survey Revisited (Colby, Frost & Fischer) **Ms Alison Colby** (Center for Limnology, University of Wisconsin – Madison, Wisconsin, USA)

5.30-5.50: The relationship of silicate levels to the shallow water distribution of hexactinellids in British Columbia. **Dr Bill Austin** (Marine Ecology Station, Cowichan Bay, British Columbia, Canada)

- **'The Commonwealth Bank of Australia' Poster Session (Chairman: Dr Sally Leys, Queensland Museum, Brisbane, Australia)**

6.00-7.00: Authors are requested to stand by their posters from 6-7pm. **(Light refreshments will be provided). See list of poster titles at end of this document.**

- ❒ **7.30pm-10.30pm: US National Science Foundation Sponsored Workshop on 'Regional research opportunities and career development'. Venue: *Synergy III* Riverboat dinner cruise. Boarding at 7.30pm for 7.45 departure, returning 10.30pm from Southbank Charter Terminal (the closest to Victoria Bridge on Southbank). All delegates are welcome, but cost is <u>not</u> included in the registration fees (cost is $46, including meal and some drinks)**

Wednesday 1 July 1998

Registration desk open from 8.00am

- **Session 5: Molecular Phylogeny (Chairman: Dr Nicole Boury-Esnault, Marine d'Endoume, Marseille, France)**

8.30-8.50: Phylogenetic relationships within the Phylum Porifera utilising the 18s rRNA gene (Adams, McInerney & Kelly-Borges) **Ms Christi Adams** (Molecular Biology Unit, The Natural History Museum, London, UK)

8.50-9.10: Approach to the phylogeny of Axinellidae (Porifera: Demospongiae) using morphological and molecular data. (Alvarez, Crisp, Driver, Hooper & Van Soest) **Dr Belinda Alvarez de Glasby** (National Institute of Water and Atmospheric Research Ltd., Oceanographic (NZOI), Wellington, New Zealand)

9.10-9.30: Good congruence between morphology and molecular phylogeny of Hadromerida, or how to bother sponge taxonomists (Chombard & Boury-Esnault). **Ms Catherine Chombard** (Service de Systematique Moleculaire, Muséum National d'Histoire Naturelle, Paris, France)

9.30-9.50: Morphology and molecules in lithistid taxonomy: new solutions for old problems.(Kelly-Borges, McCormack & McInerney) **Dr Michelle Kelly-Borges** (Dept. of Landscape & Plant Sciences, UNITEC Institute of Technology, Auckland, New Zealand)

9.50-10.10: Phylogeny of Lithistid sponges (McInerney & Kelly-Borges) **Dr James McInerney** (Department of Zoology, The Natural History Museum, London, UK)

❒ **10.10-11.00: morning tea and preparation for excursion**

- ❒ **11.00am–until approximately 7.30pm: Half-day rainforest excursion and lunch at "O'Reilly's", Lamington National Park (about 2 hours bus trip southwest of Brisbane)**

❒ **Evening free**

Thursday 2 July 1998

Registration desk open from 8.00am

- **Session 6: Molecular Biology & Genetics (Chairman: Dr Bernie Degnan, University of Queensland, Brisbane, Australia)**

8.30-8.50: Patterns of inter and intraspecific genetic divergence in marine sponges (Solé-Cava & Boury-Esnault) **Dr Nicole Boury-Esnault** (Station Marine d'Endoume, Marseille, France)

8.50-9.10: Homeobox genes expressed in the adult and reaggregating sponge. (Larroux & Degnan) **Ms Claire Larroux** (Department of Zoology, University of Queensland, Brisbane, Australia)

9.10-9.30: Homeobox containing genes in freshwater sponges (Richelle-Maurer & Van de Vyver) **Dr Evelyn Richelle-Maurer** (Physiologie Cellulaire et Genetique des Levures, Université Libre de Bruxelles, Belgium)

9.30-9.50: Random amplified polymorphic DNA (RAPD) analysis can reveal micro- and macroevolutionary patterns in Porifera (Lôbo-Hajdu, Salgado, Rosário, Hajdu, Muricy & Albano) **Dr Gisele Lôbo-Hajdu** (Departmento de Bioquimica, Instituto de Biologia, Universidade Estadual do Rio de Janeiro, Brazil)

- **Session 7A: Ecology (I), Population Dynamics, Spatial Distribution, Disturbance & Bioerosion (Chairman: Dr Roland Pitcher, CSIRO Marine Research, Brisbane, Australia)**

9.50-10.10: Coral reef community patchiness and sponge distribution off Mactan Island, Cebu, Phillippines (Bakus & Nishiyama) **Prof Jerry Bakus** (Department of Biological Sciences, University of Southern California, Los Angeles, USA)

10.10-10.30: Population dynamics of a sponge/macroalgal symbiosis: Possible causes for a patchy distribution at One Tree Reef. **Dr Donelle Trautman** (Department of Science (Biological Sciences), University of Sydney, Australia)

❒ 10.30-11.00: morning tea

11.00-11.20: Limits on the bathymetric distribution of keratose sponges: a field test in deep-water (Maldonado & Young) **Dr Manuel Maldonado** (Department of Aquatic Ecology, Centro d' Estudios Avancados de Blanes (CSIS), Blanes, Spain)

11.20-11.40: The role of early life-history stages in determining adult spatial patterns of encrusting sponges. (Uriz, Maldonado, Turon & Martí) **Dr Maria Uriz** (Centro d'Estudios Avancados de Blanes, C.E.A.B. (CSIS), Blanes, Spain)

11.40-12.00: Research on the natural dynamics of some structurally dominant tropical sponges and other sessile fauna. (Pitcher, Wassenberg, Smith, Cappo, Hooper & Doherty) **Dr Roland Pitcher** (CSIRO Division of Marine Research, Brisbane, Australia)

12.00-12.20: Sponge assemblages as bioindicators of anthropogenic disturbance (Roberts, Davis & Cummins) **Mr Danny Roberts** (Strategic Planning Department, Wyong Shire Council, Wyong, Australia)

12.20-12.40: Recovery and growth of the Giant Barrel Sponge (*Xestospongia muta*) following physical injury from a vessel grounding in the Florida Keys. **Mr George Schmahl** (Florida Keys National Marine Sanctuary, Key West, Florida, USA)

❒ 12.40-2.00: lunch

2.00-2.20: The replacement of natural hard substrata by artificial substrata: its effects on sponges and ascidians. **Mr Nathan Knott** (Special Research Centre on Ecological Impacts of Coastal Cities, University of Sydney, Australia)

2.20-2.40: An experimental approach to the ecological significance of movement in sponges (Maldonado & Uriz) **Dr Manuel Maldonado** (Department of Aquatic Ecology, Centro d' Estudios Avancados de Blanes (CSIS), Blanes, Spain)

2.40-3.00: Resource partitioning by Caribbean coral reef sponges: Is there enough food for everyone? **Dr Adele Pile** (Department of Larval Ecology, Harbor Branch Oceanographic Institution Inc, Fort Pierce, Florida, USA)

3.00-3.20: The infectiveness of a bioeroding clionid sponge (Schönberg & Wilkinson) **Ms Christine Schönberg** (Australian Institute of Marine Science, Townsville, Australia)

3.20-3.40: *Cliona lampa* and disturbance on the coral reefs of Castle Harbour, Bermuda (McKenna & Ritter) **Dr Sheila McKenna** (Marine Laboratory, University of Guam, USA)

3.40-4.00: Bioerosion and bioconstruction by the symbiotic sponge *Cliona nigricans* (Porifera, Demospongiae) (Calcinai, Cerrano, Bavestrello & Sarà) **Prof Michele Sarà** (Istituto di Zoologia dell'Universita, Genova, Italy)

❒ 4.00-4.30: afternoon tea

4.30-4.50: The planktonic armoured propagules of the excavating sponge *Alectona* (Porifera, Demospongiae) are larvae: evidence from *Alectona wallichii* and *A. mesatlantica* n.sp. **Dr Jean Vacelet** (Centre d'Oceanologie de Marseille, Station Marine d'Endoume, Univ. d'Aix-Marseille 2, France)

4.50-5.10: Reproduction of some demosponges in a temperate Australian shallow water habitat. **Dr Jane Fromont** (Western Australian Museum, Perth, Australia)

- **Session 7B: Ecology (II), Mutualism, Predation & Reproductive Biology (Chairman: Prof Roberto Pronzato, Istituto di Zoologia dell'Universita Genova, Italy)**

5.10-5.30: A sponge that cheats on diffuse mutualism among other sponge species. **Dr Janie Wulff** (Biology Department, Middlebury College, Middlebury, Vermont, USA)

5.30-5.50: Relationship between sponges and a taxon of obligatory inquilines: the Siliquariid molluscs. (Pansini, Cattaneo-Vietti & Schiaparelli) **Dr Maurizio Pansini** (Istituto di Zoologie dell'Universita, Genova, Italy)

5.50-6.10: Ecological and evolutionary significance of morphological variation in the tropical sponge *Anthosigmella varians*: niche partitioning and predator-induced phenotypic plasticity. **Dr Malcolm Hill** (Department of Biology, Fairfield University, Connecticut, USA)

6.10-6.30: Ecological adaptions of a freshwater sponge association in a large river (Porifera, Spongillidae). **Dr Jochen Gugel** (Department of Zoology, Tel Aviv University, Israel)

6.30-6.50: Do Caribbean sponges have physical defenses (Chanas & Pawlik) **Mr Brian Chanas** (Laboratory of Pharmacology and Chemistry, National Institute of Environmental Health Sciences, North Carolina, USA)

6.50-7.10: Polly want a sponge?: Field examination of spongivory by Caribbean parrotfishes in reef and mangrove habitats (Dunlap & Pawlik) **Mr Matthew Dunlap** (Department of Biological Sciences, University of North Carolina-Wilmington, North Carolina, USA)

- **❒ 7.30pm-11.30pm: Symposium Dinner, "Io Ti Amo", South Bank**

Friday 3 July 1998

Registration desk open from 8.30am

- **Session 8: Cell Biology, Physiology & Anatomy (Chairman: Prof Gisele Van de Vyver, Université Libre de Bruxelles, Belgium)**

9.00-9.20: Long term survey of the calcification rate in Jamaican coralline sponges. (Willenz & Hartman) **Dr Phillipe Willenz** (Department des Invertebres, Institut Royal des Sciences Naturelles de Belgique, Bruxelles, Belgium)

9.20-9.40: Characterization of calcium-binding matrix proteins from distinct coralline demosponges. (Bergbauer, Lange, Szewzyk & Reitner) **Dr Matthias Bergbauer** (Fachgebiet Oekologie der Mikroorganismen, Technische Universitaat Berlin, Germany)

9.40-10.00: Annual cycle of foreign matter incorporated by *Chondrosia reniformis* (Porifera, Demospongiae): the role of the water movement (Cerrano, Bavestrello, Benatti, Cattaneo-Vietti, Giovine & Sarà) **Dr Carlo Cerrano** (Istituto di Zoologia dell' Universita, Genova, Italy)

❒ 10.00-10.30: morning tea

10.30-10.50: Sponge Cell Adhesion: an Evolutionary Ancestor of Histocompatibility Systems? (Fernandez & Burger) **Dr Xavier Fernandez-Busquets** (Friedrich Miescher-Institut, Basel, Switzerland)

10.50-11.10: Regulatory Mechanisms of Immune Cells in Sponges. **Dr Tom Humphreys** (Kewalo Marine Laboratory, University of Hawaii, Honolulu, USA)

11.10-11.30: Propagated electrical impulses in a sponge. (Leys & Mackie) **Dr Sally Leys** (Department of Biology, University of Victoria, British Columbia, Canada; currently Queensland Museum, Brisbane, Australia)

11.30-11.50: Time-lapse studies of sponge motility and anatomical rearrangements. **Dr Calhoun Bond** (Department of Biology, Greensboro College, North Carolina, USA)

❒ 11.50-1.00: lunch

- **Session 9: Microbial Symbioses & Nutrition (Chairman: Dr Henry Reiswig, Redpath Museum, McGill University, Montreal, Canada)**

1.00-1.20: Photosynthesis and respiration of the cyanobacterium-containing sponge, *Dysidea herbacea* (Hinde, Borowitzka & Pironet) **Dr Mike Borowitzka** (School of Biological and Environmental Sciences, Murdoch University, Perth, Australia)

1.20-1.40: Microbial symbionts of Great Barrier Reef sponges (Burja, Webster, Murphy & Hill) **Dr Russell Hill** (Australian Institute of Marine Science, Townsville, Australia)

1.40-2.00: Perspectives on sponge-cyanobacterial symbioses (Diaz & Ward) **Dr Christina Diaz** (Marine Sciences, University of California Santa Cruz, California, USA)

2.00-2.20: Membrane-bounded nuclear bodies in a diverse range of microbial symbionts of Great Barrier Reef sponges (Fuerst, Webb, Garson, Hardy & Reiswig) **Dr John Fuerst** (Department of Microbiology, University of Queensland, Brisbane, Australia)

2.20-2.40: Evidence of transfer of photosynthate from a red algal macrophyte to its symbiotic sponge (Grant, Hinde & Borowitzka) **Dr Adrienne Grant** (School of Biological Sciences, University of Sydney, Australia)

2.40-3.00: Nitrogen fixation in symbiotic marine sponges: ecological significance and difficulties in detection. (Wilkinson, Summons, Roksandic & Evans) **Dr Clive Wilkinson** (Australian Institute of Marine Science, Townsville, Australia)

3.00-3.20: Nitrogen Flux in a Sponge - Macroalgal Symbiosis (Davy & Hinde) **Dr Rosalind Hinde** (School of Biological Sciences, University of Sydney, Australia)

❒ **3.20-3.50: afternoon tea**

3.50-4.10: Effect of photosynthetic activity in endosymbiotic zoochlorellae on gemmule germination of a freshwater sponge, *Radiospongilla cerebellata.* (Satoh, Fujimoto, Yamamoto & Kamishima) **Prof Yoshihisa Kamishima** (Department of Biology, Faculty Education, Okayama University, Japan)

4.10-4.30: Wide phylogenetic spectrum of heterotrophic microbes associated with the sponge *Discodermia dissoluta* characterized by microbiological and molecular techniques (Lopez, McCarthy, Janda, Willoughby & Pomponi) **Dr Joe Lopez** (Division of Biomedical Marine Research, Harbor Branch Oceanographic Institution, Inc., Fort Pierce, Florida, USA)

4.30-4.50: Optimisation of the growth medium for an in vitro sponge culture. (Osinga, Tramper & Wijffels) **Dr Ronald Osinga** (Department of Food Sciences and Technology, Food and Bioprocess Engineering Group, Wageningen Agricultural University, The Netherlands)

4.50-5.10: Carnivorous sponge [The Movie !] **Dr Jean Vacelet** (Station Marine d'Endoume, Marseille, France)

❒ **5.10-6.00: "Where to Next ?" Closing remarks by convenor, and subsequent adjournment for refreshments.**

POSTER TITLES (* indicates no abstract received)

1. The growth of the branching sponge, *Axinella australiensis* **Dr Edward Abraham***; 2. Dynamics of sponge/coral interactions **Dr Lisanne Aerts***; 3. [Title to be announced] **Mr Michael Assmann***; 4. Sponges on Multimedia - Demonstration **Prof. Jerry Bakus**; 5. Sponges, indicators of marine environmental health. (Battershill & Abraham) **Dr Chris Battershill**; 6. The Feeding Biology of *Polymastia croceus.* (Bell, Bergquist & Battershill) **Mr Andrew Bell**; 7. Pharmacological screening **Dr Roberto Berlink***; 8. Chemistry and biological activities of substances isolated from *Amphimendon viridis* **Dr Roberto Berlink***; 9. [Title to be announced] **Dr Tony Carroll***; 10. Molecular phylogeny of demosponges **Ms Catherine Chombard***; 10. Remarks about the status of the genus *Myxilla* (Porifera, Poecilosclerida) in the Galician coast (NW of Iberian Peninsula) (Cristobo, Rios & Urgorri) (communicated via Dr Nicole Boury-Esnault) **Dr Javier Cristobo**; 11. Trace element and stable isotope profiles from the coralline sponge (*Astrosclera willeyana*) (Fallon, McCulloch & Hooper) **Mr Stewart Fallon**; 12. Historical occurence of sponge populations in Connecticut lakes: analysis of spicules in dated sediment cores (going back 100+ years) **Dr Paul Fell***; 13. Fijian Sponges **Mr Stanley Flavel***; 14. Spongivory by the Brazilian starfish *Echinaster brasiliensis* (Guerrazzi, Hajdu, Morgado Amaral & Duarte) **Dr Eduardo Hajdu**; 15. Cellular localization of jaspamide **Ms Mary Kay Harper***; 16. Sponges of the Heron, Wistari and Sykes Reefs, Capricorn-Bunker Group, Great Barrier Reef (Hooper, Cook, Kennedy & List) **Mr Stephen Cook**; 17. Molecular phylogeny of freshwater sponges (families Lubomirskiidae and Spongillidae) and their relationships with sea sponges. (Itskovich, Belikov, Efremova & Masuda) **Dr Valeria Itskovich**; 18. [Title to be announced] **Dr Janka Jarchow***; 19. [Title to be announced] **Dr Clifford Jones***; 20. Experimental optimisation of growth rate and morphology of the cultured bath sponge *Coscinoderma mathewsi* Lendenfeld in Pohnpei, Micronesia. (Kelly-Borges, Harkins & Croft) **Dr Michelle Kelly-Borges**; 21. [Title to be announced] **Dr Young Kim***; 22. Convergence in the Time-Space Continuum: A Predator-Prey Interaction (Knowlton & Highsmith) **Ms Ann Knowlton**; 23. [Title to be announced] **Mr Kyung Jin Lee***; 24. Antimicrobial activity of sponges from southern Brazil. Atlantic coast (Lerner, Schapoval, Mothes, Possuelo, Costa, Rech, Farias, Mans & Henriques) **Ms Clea Lerner**; 25. A new dendroceratid sponge with reticulate skeleton (Maldonado & Uriz) **Dr Manuel Maldonado**; 26. A new genus of freshwater sponges from the Lake Tana (Manconi, Cubeddu & Pronzato) **Dr Renata Manconi**; 27. Spatial and temporal variation of the natural toxicity in sponges of a Mediterranean cave: is there a trend ? (Martí, Uriz, Turon & Ballesteros) **Dr Maria Uriz**; 28. Study on the distribution of Baikalian sponges (Masuda, Iskovich, Veinberg & Efremova) **Dr Yoshiki Masuda**; 29. An ultrastructural study on the contractile pinacocyte of a freshwater sponge. (Matsuno, Kuroda & Masuda) **Prof. Akira Matsuno**; 30. The phylogenetic position of the sponge *Spongosorites suberitoides* determined by analysis of 28S rRNA gene sequence. (McCormack, McInerney & Kelly-Borges) **Dr Grace McCormack**; 31 [Title to be announced] **Prof. Ted Molinski***; 32. The genus *Erylus* Gray, 1867: a revision of Brazilian species with description of a new species (Astrophorida, Geodiidae) (Mothes, Lerner & Da Silva) **Dr Beatriz Mothes**; 33. [Title to be announced] **Dr Biswapati Mukherjee***; 34. An evaluation of biochemical, morphological and cytological data sets for the phylogeny of the Homosclerophorids (Porifera, Demospongiae) **Prof. Guilherme Muricy**; 35. A taxonomic key to the marine sponges of North Carolina **Mr Scott Nichols**; 36. Sponges of the Andaman Sea (Pattanayak & Hooper) **Mr Joygopal Pattanayak***; 37. *Setidium obtectum* Schmidt, 1879 rediscovered **Dr Andrzej Pisera**; 38. The impact of trawling on some tropical sponges and other sessile fauna. (Pitcher, Burridge, Wassenberg & Smith) **Dr Roland Pitcher**; 39. Sponge farming in the Mediterranean Sea: new perspectives (Pronzato, Bavestrello, Cerrano, Magnino, Manconi, Pantelis, Sarà & Sidri) **Prof. Roberto Pronzato**; 40. Sponge external shape as a taxonomic character: the case of *Spongia officinalis* and *Spongia agaricina* (Pronzato, Sidri, Dorcier, Cappello & Manconi) **Prof. Roberto Pronzato**; 41. Chemical defenses of the Caribbean sponges *Aplysina fulva* and *A. insularis* (Puyana, Pawlik & Fenical) **Ms Monica Puyana**; 42. New approaches to the biomineralization processes of calcified skeletons in coralline demosponges. (Reitner, Bergbauer, Wörheide, Lange, Thiel, Eisenhauer, Reimer & Fliege) **Prof. Joachim Reitner**; 43. "Mud Mound" Structures and Coralline Sponges from the Osprey Reef (Queensland Plateau, Coral Sea, Australia) (Reitner, Wörheide & Hooper) **Prof. Joachim Reitner**; 44. New colonial *Vaceletia*-type sphinctozoan from the Pacific. (Reitner, Wörheide & Hooper) **Prof. Joachim Reitner**; 45. A strange suberitid demosponge from a marine high alkaline crater lake (Satonda Island, Indonesia) (Reitner, Wörheide, Hooper, Arp, Reimer & Kempe) **Prof. Joachim Reitner**; 46. Carbon Isotope Time Series of Coralline Sponges from the Coral Sea, Philippines and Caribbean. (Boehm, Eisenhauer, Joachimski, Lehnert, Dullo, Woerheide & Reitner) **Prof. Joachim Reitner**; 47. Carbon Isotope History of Caribbean Surface Waters Revealed by Coralline Sponges. (Boehm, Eisenhauer, Joachimski, Lehnert, Dullo & Reitner) **Prof. Joachim Reitner**; 48. [Title to be announced] **Dr. Evelyn Richelle-Maurer***;49. The sponges of Paraiso nearshore fringe reef Cozumel, Mexico **Mr Jason Ritter**; 50. Freshwater sponges (Porifera: Spongillidae) of the Guanacaste Conservation Area, Costa Rica: A preliminary survey **Mr Scott Roush**; 51. Taxonomic Evaluation of jasplakinolide-containing sponges of the family Coppatidae (Sanders & Diaz) **Ms Miranda Sanders**; 52. Wanted: the names of common bioerodiing sponges of the central Great Barrier Reef (5 poster panels) **Ms Christine Schönberg**; 53. Microbial influenced pyritization of marine sponges (communicated by Dr Matthias Bergbauer) **Ms Gabriela Schumann-Kindel**; 54. *Chondrosia reniformis*: hitherto unknown bacteria (Schumann-Kindel, Bergbauer, Manz, Reitner & Szewzyk) (communicated by Dr Matthias Bergbauer) **Ms Gabriela Schumann-Kindel**; 55. Relationship of sand and fibre in the Psammocinian horny sponge **Prof. Chung-Ja Sim**; 56. [Title to be announced] **Ms Kathleen Smith***; 57. Genetic confirmation of the specific status of two sponges of the genus *Cinachyrella* in the Southwest Atlantic (Lazoski, Peixinho, Russo & Solé-Cava) **Dr Antonio Solé-Cava** (communicated by **Dr Nicole Boury-Esnault**); 58. Photosynthesis and respiration by the symbiotic association between a coral reef sponge and its macroalgal symbiont **Dr Donelle Trautman**; 59. Non-feeding periods in the Poecilosclerid sponge *Crambe crambe*: an ultrastructural study. (Turon & Uriz) **Dr Maria Uriz**; 60. [Title to be announced] **Ms Clare Valentine***; 61. [Title to be announced] **Ms Clare Valentine***; 62. CD-ROM Guide to the North East Atlantic Sponges (Van Soest, Picton & Morrow) **Dr Rob Van Soest***; 63. Biology of sponge natural products **Dr Rob Van Soest**; 64. The peculiarities of sponge spicule composition in Holocene - Pleistocene sediments of the underwater Akademichesky Ridge of Lake Baikal (Weinberg, Eckert, Efremova & Masuda) **Ms Elena Weinberg**; 65. The Cross-shelf distribution of South West Sulawesi Open Reef Sponges **Ms Nicole de Voogd***; 66. Computer demonstration of Platypus: the Zoological Catalogue of Australia, Porifera **Dr Alice Wells***; 67. Biocalcification in the Indo-Pacific coralline demosponge *Astrosclera willeyana* Lister - the role of basopinacoderm (Woerheide & Reitner) **Dr Gert Woerheide**; 68. Climatic changes of the last 450 years recorded in the skeleton of the coralline demosponge *Astrosclera willeyana* (Woerheide & Reitner) **Dr Gert Woerheide**; 69. Three new scalarane based sesterterpenes from the tropical marine sponge *Carteriospongia calciformis.* (Jahn, Koenig & Wright) **Dr. Tony Wright**; 70. Rapid change and stasis in a coral reef community **Dr Janie Wulff**; 71. New finds of early Cambrian spicular sponges of extra-ordinary preservation (Siberia) **Dr A. Yu Zhuravlev***.

5TH ISS. LIST OF REGISTERED PARTICIPANTS

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ABSTRACTS OF THE 5TH ISS

PHYLOGENETIC RELATIONSHIPS WITHIN THE PHYLUM PORIFERA UTILISING THE 18S rRNA GENE

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Sponges are classically considered to be a monophyletic group, however it has been suggested that they may be paraphyletic or polyphyletic. Recent molecular data utilising 18S rRNA sequences support the sponges as a paraphyletic group comprised of the Porifera and Ctenophora. The 18S rRNA database was expanded with newly acquired near full-length sequences (1300 bps) for two hexactinellids, four demosponges and one calcareous sponge. Phylogenetic relationships were re-examined within the Porifera at the class level as well as between Porifera, Ctenophores and Choanoflagellates at the phylum level.

Key words: *Hexactinellida, Demospongiae, Calcarea, phylogeny, rRNA sequences*

APPROACH TO THE PHYLOGENY OF AXINELLIDAE (PORIFERA: DEMOSPONGIAE) USING MORPHOLOGICAL AND MOLECULAR DATA

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A set of 27 species of marine sponges of the Axinellidae and related families was selected with the aim of testing the monophyly of Axinellidae and investigating their phylogenetic relationships using cladistic methods. Partial 28S rDNA sequences, including the D3 domain, and traditional morphological characters were used independently to construct phylogenetic trees. Alignment of the sequences using the appropriate model of secondary structure of the RNA was compared to that produced by the ClustalW. The alignment using secondary structure constraints produced a better estimate of the phylogeny and was demonstrated to be an effective and objective method.

The results from the analyses of the molecular and morphological data sets are not fully congruent; the morphological data suggest that Axinellidae is monophyletic, however the molecular data suggest that it is not monophyletic. In both cases the sampled members of the family are closely related to those of Halichondriidae and Dictyonellidae. Tests of heterogeneity (reciprocal T-PTP and partition homogeneity test) shown that the data partitions are heterogeneous, which could be due to sampling errors (in either data set) or differences in the underlying phylogenies, and therefore data were not combined in a single analysis.

Key words: *Axinellidae, secondary structure, D3 domain, 28S ribosomal DNA, phylogeny*

PECULIARITIES OF FERTILIZATION PROCESS IN THE SPONGE *LEUCOSOLENIA COMPLICATA* MONTAGU (CALCISPONGIAE, CALCARONEA) FROM THE BARENTS SEA

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The fertilization process in the Barents-Sea sponge *Leucosolenia complicata* Mont. (Calcispongia, Calcaronea) was studied on the ultrastructural level and light microscopy level with histochemical methods being applied (tests for the total content of proteins, lipids, mucopolysaccharides, nucleic acids were carried out). As with all other Calcispongiae, fertilisation is conducted with the special carrier cell. At present, carrier-cell fertilisation is found in a series of species, mainly in sycon- and leucon-structured Calcaronea. *L. complicata* has an anatomical organisation of the ascon type and specific fertilisation processes in *L. complicata* might be due to its sperm's unique organisation.

The mature spermatozoon has neither acrosome nor flagellum, but is just a spherical cell 4.8 µm in diameter, occupied mainly with the nucleus (d=4.6 µm. In *L. complicata* spermiogenesis takes place in choanocytes where the protein capsule around the sperm nucleus is synthesised. During the massive sperm release, any cell from the nurse cells complex, can seize a sperm and transform into a sperm-carrying cell *in situ*. The transformation of a seized sperm into the spermiocyst is accompanied with rapid isolation of the sperm's nucleus from its protein capsule. These processes are correlated with protein-dying tests and might be a result of either protein accumulation or structural transformation of the proteins that comprise the capsule and nuclear chromatin. The spermiocyst formation goes along with the hypertrophic changes of the carrier cell, i.e. its diameter increases from 8.8 µm to 19.4 µm and the nurse-cells increase in their size (from 8.8 µm to 11 µm in diameter), evident from transmission electron micrographs.

The fertilisation process begins with the protein capsule penetrating the oocyte and gradually resolving in its ooplasm. The extra swelling of the sperm nucleus within the carrier cell coincides with this process. The sperm nucleus penetrates into the oocyte's cytoplasm and the maturation divisions in the egg proceed. They take place in the egg's animal part, turned towards the choanoderm. Both meiotic divisions from metaphase1 to telophase2 follow. The chromosomes' arrangement within the metaphasal plates during both maturation divisions appear to be most characteristic of the *L. complicata* oocytes' meiosis: the chromosomes arrange themselves annulus-like (ring-like). During either the first or the second maturation divisions the annulus-like metaphasal plates turn 90 degrees round and only then the chromosomes begin to move towards the maturation spindle poles. The polar bodies are separated under the choanoderm. The completion of the oocyte's maturation divisions coincide with the beginning of the sperm-nucleus's transformation into the male pronucleus. Therefore the processes of male and female pronuclei formation is concurrent.

The sperm nucleus begins transforming into the male pronucleus with its own nuclear membrane destruction, male chromatin swells and loosens, and the building of the pronucleus membrane follows. After the maturation divisions are completed, the chromosomes are condensed into a tight spherical chromatin mass, which then gradually loosens and is transformed into a chromatin net. The female-pronucleus membrane is then formed. Definitive pronuclei are similar in their size and show up as large (d=22µm) bubble-like nuclei filled with finely-granulated chromatin net. In *L. complicata*, pronuclei do not fuse together. As a rule, two groups of chromosomes are formed in the zygote, they unite into one and arrange into a metaphasal plate of the first cleavage division.

Key words: *Calcispongiae, fertilisation, spermatozoon, carrier cell, spermiocyst, oocyte, meiosis, zygote*

THE RELATIONSHIP OF SILICATE LEVELS TO THE SHALLOW WATER DISTRIBUTION OF HEXACTINELLIDS IN BRITISH COLUMBIA

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The boot sponge, *Rhabdocalyptus dawsoni*, occurs at depths as shallow as 10 m in the Strait of Georgia, British Columbia. The cloud sponge, *Aphrocallistes vastus*, typically occurs in slightly deeper water but has been found as shallow as 5m in Johnstone Strait, British Columbia. These species also form bioherms several meters thick in some localities. Initial surveys of the literature indicate that shallow water silicate levels in regions of British Columbia are high compared to levels in other shallow marine waters. Areas of the Antarctic are an exception and hexactinellids also occur here in shallow water. Hexactinellids are absent in habitats which might be expected to support populations, such as Norwegian fjords, but where silicate levels are low. The recently described shallow water occurrence of hexactinellids in the Mediterranean may be an exception. Additional input from participants at the Symposium may help support or refute a relationship between shallow water occurrence of hexactinellid populations and high levels of silicates.

Key words: *hexactinellid, silicates, British Columbia, shallow water*

CORAL REEF COMMUNITY PATCHINESS AND SPONGE DISTRIBUTION OFF MACTAN ISLAND, CEBU, PHILIPPINES

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Coral reef community structure was studied during 1994-95 at Mactan Island, off Cebu City, Cebu, Philippines. Three line intercepts were surveyed from depths of 7 to 32 m and straight line distances of 55-68 m. Live coral represented 29-42% (=36%) of categories intercepted and sponges 1-5% (=3%), representing the two most abundant groups of benthic organisms. All remaining benthic taxa together comprised only an average of 1% of the intercept distance. The number of sponges along the intercepts ranged from 4 to 19 (=12). Sponge approximate density from line intercept data ranged from 1/20 m^2 to 1/250 m^2 (=1/40 m^2) and they were typically large specimens. A transition frequency matrix was calculated for all line intercepts and a test for a Markov chain was conducted. The most frequently encountered sequence was coral rubble followed by live coral (11% sequence frequency). Live coral followed by sponges was 5% and sponges followed by live coral was 5%. In a similar study of the benthos with five line intercepts at depths of 7-12 m, the most frequently encountered sequence was live coral to sponge and sponge to live coral (36%). Sponge to sponge transitions represented 4%. None of the sequences were significant at P = 0.05, that is, the succession of substratum types were independent of each other. Reasons given for this independence include: 1) high species richness resulting in a high degree of patchiness of species; 2) high intensity fish predation on plankton and benthos; 3) high intensity predation on larvae and spores by hard corals, octocorals and millepores, among others; and 3) allelochemical defenses in competition for space.

Key words: *Philippines, coral reef, community ecology, patchiness, predation, allelochemicals*

SPONGES ON MULTIMEDIA - DEMONSTRATION

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The world's first multimedia CD-ROM (for PC's and NTSC monitor systems) was peer-reviewed then completed during Fall 1996. It covers all aspects of sponge biology, attempting to make the arcane subject informative yet humorous to young university students interested in invertebrate zoology or marine biology.

Key words: *multimedia, CD-ROM*

SPONGES, INDICATORS OF MARINE ENVIRONMENTAL HEALTH

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There is an urgent need for marine ecosystem indicators to facilitate management aimed at either ameliorating impacts or guiding sustainable utilisation of marine resources. We propose that qualitative and quantitative examination of marine benthic communities will provide robust indication of responses to short and long term environmental conditions, and further suggest that information exists which permits the creation of a hierarchy of indicators for establishing ecosystem health in a regional context. These are in the form of identifiable marine community assemblages, together with biomass and growth indices determined from morphological parameters associated with the characterising species for each assemblage. Examples are provided to demonstrate the sensitivity of such indicators by focusing on sponge characterised communities. The composition of assemblages and population statistics of key species reflect ecosystem disturbances following catastrophic sediment deposition following cyclones, and in response to more recent and relatively short-term impacts. The latter include responses to sediment disruption from trawling and sand mining, and responses to water quality change during algal bloom events.

Marine environmental indicators are likely to take the form of well-defined ecotypes described by characterising species presence. These species have known ranges of tolerance to environmental variables such as light, current, food supply, turbidity, BOD, and sediment regime. They are by their very nature, relevant at a regional level and will be set in the context of a biogeographic classification for any coast or shelf. They can be further refined by interrogation of models relating population structure of key species to biological and physical attributes of the environment.

Key words: *growth, morphology, indicators, environmental health, marine resources, benthic communities*

THE FEEDING BIOLOGY OF *POLYMASTIA CROCEUS*

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Polymastia croceus is a yellow encrusting marine sponge endemic to New Zealand's coastal waters. It has recently attracted interest due to its production of proteinaceous secondary metabolites that have

potential for use in anti-cancer and possibly anti-HIV pharmaceuticals. Studies have been initiated to determine the best options to supply biomass for metabolite extraction, in anticipation of drug industry demand should active compounds succeed in clinical trials. This species also represents a suitable model for experimentation as findings will have relevance to other species that may become targets for production in the future. In-sea aquaculture of *P. croceus* has been found to be very successful as a method of inexpensive production of biomass for extraction of target metabolites. Aquarium based aquaculture is however more desirable, but advance in this area is dependant on understanding the feeding biology of the sponge. Investigations on *P. croceus* feeding demonstrate that the sponge can process large quantities of water. This suggests that food may also be processed in large quantities and thus could become a limiting factor for *P. croceus* distribution and growth in the wild and in any culture facility. Investigation of the food types and rates of consumption were thus undertaken.

Previous studies into sponge feeding and pumping in general have shown that water can be processed at very high rates and the main component of their diet is plankton less than 10 µm. The work undertaken in this study concentrated on the ultraplankton component of the diet. Single beam flow cytometry was used to quantify diet with a method similar to that used by Pile (1996). It was found that *P. croceus* had high retention efficiencies (up to 94%) of ultraplankton, which was similar to previous findings. However, in contrast to previous work it was found that cyanobacteria and picoeukaryotes were the species retained most efficiently. High rates of water transport were also found (averaging 0.85 l/min) suggesting that ecologically significant depletion of ultraplankton could be taking place. Spatial variation in the retention efficiencies was also found suggesting selectivity in feeding.

Key words: *sponge feeding, sponge pumping, ultraplankton, flow cytometry, metabolite extraction*

CHARACTERIZATION OF CALCIUM-BINDING MATRIX PROTEINS FROM DISTINCT CORALLINE DEMOSPONGES

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Calcified sponges played an important role as reef building organisms during different geological time periods. Living relatives of this group investigated here, *Spirastrella (Acanthochaetetes) wellsi, Astrosclera willeyana* and *Vaceletia n. sp.*, can be found in cryptic niches of indopacific coral reefs. The first known relatives of some of these sponges are known since the upper permian.The mode of biomineralization of the examined species seems to be extremely conservative, since they are phylogenetically very old and exhibit merely minor alterations in their calcareous skeletons. Each of the three species exhibits a unique type of basal skeleton with its own specific modifications of carbonate crystals. Each species was shown to have a specific array of calcium-binding macromolecules enclosed within its intraskeletal matrix. The proteins are separated by SDS polyacrylamide gel electrophoresis. A single protein was detected in *S. wellsi*, two proteins in *A. willeyana*, and four proteins in *Vaceletia n. sp.*. All proteins were characterized by their molecular weight and isoelectric point. The soluble matrix constituents of each species were tested for their potential to decrease precipitation of calcium and strontium carbonate, respectively, in a saturated solution. The findings strongly suggest that these soluble proteins function as the template for skeletal formation and are responsible for determining the particular type of calcium carbonate polymorphs.

Key words: biomineralization, organic matrix, calcium-binding proteins, calcite, aragonite

CARBON ISOTOPE TIME SERIES OF CORALLINE SPONGES FROM THE CORAL SEA, PHILIPPINES AND CARIBBEAN.

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Live coralline sponges [*Ceratoporella nicholsoni, Astrosclera willeyana, Spirastrella (Acanthochaetetes) wellsi*] were collected from reef caves and deeper reef slopes of the Caribbean, the Visaya Sea (Philippines) and the Coral Sea (Great Barrier Reef). The specimens were dated by either radiocarbon or uranium-thorium methods. Ages were found to range from 200 to 600 years. We tested the reproducibility of d13C values measured on the aragonite of *Ceratoporella nicholsoni* by investigating variations along single layers of a well-laminated specimen. We also compared values measured on the outermost layers of several specimens. The reproducibility for d13C is excellent in most cases. Only few samples show depletion by up to 0.2 permil. Two parallel transects through a specimen of *Astrosclera willeyana* also display excellent reproducibility of d13C values. All specimens show the well-known industrial decline in d13C values starting ca. in 1850 A.D (e.g. Druffel and Benavides 1986, Nature 321, 58-61). In comparing the magnitude of this decline measured in our samples and in d13C of atmospheric CO2 (Francey et al., Tellus, in press), we can estimate the local degree of isotopic equilibration between atmosphere and sea-water. We find values range from 40% of the atmospheric change at the Great Barrier Reef and in the Philippines to 65% in Jamaica. We compared, for each site, the preindustrial d13C from total CO2 (DIC) of the surface water, calculated from our sponge records, with published phosphate concentrations. The values agree with a high input of nutrient-rich subsurface water at the Philippine site and at the Great Barrier Reef. At the Great Barrier Reef local upwelling at the reef front has been reported. However, the measured d13C values are much lower than expected for average phosphate concentrations. Either the upwelling is much more intense than assumed, or the Astrosclera record is affected by secondary processes and/or a vital/kinetic effect.

Key words: *coralline sponges, Philippines, Great Barrier Reef, Caribbean*

CARBON ISOTOPE HISTORY OF CARIBBEAN SURFACE WATERS REVEALED BY CORALLINE SPONGES.

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Live coralline sponges of the species *Ceratoporella nicholsoni* were collected from caves of north Jamaican reefs (20 m depth) and from the deeper slope of Pedro Bank (125 m depth). These sponges build a very dense aragonitic basal skeleton in apparent isotopic equilibrium with ambient water. Uranium-thorium dating of four specimens resulted in ages of 450 to 600 years. Within that timeframe, the sponge skeletons provide a continuous carbon isotope record, which starts at the end of the medieval warm period (1400 A.D.) and covers the "Little Ice Age" (about 1550 to 1850 A.D.), as well as the industrial period (since ca. 1850 A.D.). With a sample resolution of 0.7 mm and growth rates of 0.2 to 0.4 mm/year the temporal resolution is about 2 to 4 years. The carbon isotope records show an excellent linear correlation with the atmospheric pCO2 history, as recently reconstructed from Antarctic ice cores (Etheridge et al. 1996, JGR 101, 4115-4128). We find no significant difference between the preindustrial and the industrial regression slopes (-0.013 permil/ppm) which agrees with a common

mechanism for the observed surface water carbon isotope variations, i.e. addition/removal of isotopically "light" organic carbon to/from the atmosphere-surface ocean-biosphere system. The "Little Ice Age" is characterized by a slight increase of d13C values (+0.1 permil), peaking around 1700 A.D. During the same period, pCO2 was about 6 ppm lower than during the medieval warm period. Both can be explained by an increase in the terrestrial organic carbon reservoirs or in oceanic productivity. The Pedro Bank specimen, collected from the uppermost thermocline, shows only a dampened d13C increase during the Little Ice Age and a slightly subdued industrial d13C decline. This is expected because of the greater influence of deep-water at this depth. A comparison of the observed variation of marine d13C values and d13C of atmospheric CO2 included in Antarctic ice (Francey et al., Tellus, in press) allows one to constrain the maximum global average cooling of the ocean surface layer during the Little Ice Age to ca. -0.7 K (possible range 0 to -2 K). Further comparison to the simultaneous pCO2 decrease of 6 ppm suggests an even smaller cooling. Alternatively, an enhanced oceanic export productivity could partly explain the observations.

Key words: *carbon isotope history, coralline sponges*

TIME-LAPSE STUDIES OF SPONGE MOTILITY AND ANATOMICAL REARRANGEMENTS

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Sponges have a general reputation as sessile and static animals, but this view has been contradicted by time-lapse microscope studies of live intact sponges belonging to several taxa (2 freshwater and 5 marine genera). These studies have demonstrated that adult sponges form leading margins made of crawling cells (pinacocytes and mesohyl cells), and that these crawling margins appear capable of generating shape changes and locomotion of the entire sponge. These together with tracing studies have shown that sponges can move up to 160µm/hr (4mm per day). Observed sponges also display continuous cell movements and anatomical rearrangements in their marginal regions. These rearrangements produce slow continuous changes in the spicule skeleton and in the canal systems. Both whole-sponge motility and the internal rearrangements appear to be strongly affected by factors such as substratum adhesiveness, grooves, internal tensile forces, and water flow patterns. These ongoing changes may be an important source for plasticity in a sponge's life history.

Key words: *anatomy, cells, crawling, locomotion motility, anatomical rearrangement, time-lapse*

PHOTOSYNTHESIS AND RESPIRATION OF THE CYANOBACTERIUM-CONTAINING SPONGE, *DYSIDEA HERBACEA*

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Marine sponges containing cyanobacterial endosymbionts are common in tropical waters, and the dictyoceratid sponge, *Dysidea herbacea*, is one of the most abundant sponges in the shallow lagoon at One Tree Reef, Great Barrier Reef. This sponge contains large numbers of the filamentous cyanobacterium, *Oscillatoria spongeliae*. The *O. spongeliae* trichomes are located free in the sponge mesohyl, although they are often in contact with archaeocytes. The high biomass of the cyanobacteria is illustrated by the chlorophyll *a* content of the association, which is about 335 $\mu g.mL^{-1}$ sponge volume, or 180.3 $\mu g.g^{-1}$ sponge wet weight. These values are much higher than for any other sponges so far studied.

Photosynthetic and dark respiration rates were measured using an oxygen electrode in summer and winter at ambient lagoon temperatures and at saturating irradiances. The compensation point for

photosynthetic O_2 production is reached at about 30-50 µmol photons.m^{-2}.sec^{-1} and photosynthesis saturates at about 300 µmol photons.m^{-2}.sec^{-1}. No seasonal differences in the photosynthetic and respiration rates could be detected indicating that the sponge adapts to changing environmental conditions. The *D. herbacea/O. spongeliae* association, does however respond to changes in temperature, with a Q_{10} for photosynthesis of about 5. Photosynthesis and respiration rates are also sensitive to the O_2 concentration in the seawater. The implications of these results for the ecology of this symbiotic association will be discussed.

Key words: *dictyoceratid sponge, cyanobacterium, symbiosis, photosynthesis, respiration, temperature*

MICROBIAL SYMBIONTS OF GREAT BARRIER REEF SPONGES

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Microbial symbioses with Great Barrier Reef (GBR) invertebrates, and in particular sponge species, are of biotechnological interest since microbes associated with invertebrates may produce important bioactive compounds previously attributed to the invertebrates. Several potentially useful compounds have been found from sponge species sampled from the GBR and two of these sponges are being studied in detail. *Rhopaloeides odorabile* is a Dictyoceratid sponge common throughout the GBR that produces novel norsesterterpenes (rhopaloic acids) which exhibit potent cytotoxic activities and "Very White Fan" (VWF), a previously undescribed Demospongiae, contains the compound fanolide which retards the growth of several tumour cell lines. As a first step to investigate whether bacteria are implicated in producing any of these compounds, bacteria isolated from *R. odorabile*, VWF, and the surrounding ambient seawater were characterised using morphological, biochemical, and molecular techniques. In the case of *R. odorabile*, a single bacterium was found to dominate the culturable bacterial community associated with the sponge but was absent from ambient seawater samples. This bacterium, designated NW001, was predominant in all individual sponges sampled (N=40) from different regions of the GBR, generally at more than an order of magnitude greater than the second most common bacterium (NW002). The bacterial community associated with *R. odorabile* appears to be highly stable. In the case of VWF, the culturable bacterial community was more diverse and showed greater variation between individuals. This community generally comprised eight predominant bacteria, rarely isolated from water samples and constituting ca. 70% of the total culturable bacteria. The remaining 30% of culturable bacteria were highly variable and generally were also found in the water column samples, indicating that they are transient sponge-associated bacteria rather than symbionts.

Extensive biochemical testing was performed on all isolates to give data for cluster analyses to identify the major groups of bacteria present. Most isolates were Gram-negative (*R. odorabile* 91%, VWF 93%) and oxidative positive (*R. odorabile* 87%, VWF 50%), with 48% of isolates from VWF and 4% of those from *R. odorabile* being pigment producers (either diffusible or cell-associated). The predominant isolates from each sponge were characterised at the molecular level by PCR amplification and sequencing of 16S ribosomal RNA gene fragments. Full sequence analysis (>1,400 bp) from NW001 and two of the predominant isolates from VWF, designated AB001 and AB002, indicated that these isolates are members of the α-Proteobacteria, related to intracellular mammalian pathogens and plant symbionts. NW001, AB001, and AB002 are related to each other but only distantly related to previously characterised α-Proteobacteria, indicating that they may be members of a new genus of sponge symbionts. Partial sequence data from other VWF isolates indicate that this assemblage includes members of the genera *Thermatoga*, *Vibrio* (including *Vibrio alginolyticus*), *Bacillus*, and *Arthrobacter*. In addition, one isolate is a member of the *Cytophaga/Flexibacter/Bacteroides* group, closely related to *Microbacterium* spp. and one isolate is most closely related to an uncultured microbe, str. SCB111, a distant member of the Vibrionaceae. Several cyanobacteria have recently been isolated from both sponges and will be characterised by using the same molecular approach. This study has resulted in an array of well-characterised microbes for natural products screening, in particular for important compounds known to be produced by the sponges.

Key words: *symbiont, Dictyoceratida, bacteria characterisation, cell culture, biochemical testing, PCR, 16S ribosomal RNA,* Rhopaloeides odorabile, *α-Proteobacteria*

ANNUAL CYCLE OF FOREIGN MATTER INCORPORATED BY *CHONDROSIA RENIFORMIS* (PORIFERA, DEMOSPONGIAE): THE ROLE OF THE WATER MOVEMENT

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Many demosponges strengthen their skeleton by incorporating various allochtonous matter such as sand grains, sponge spicules and other inorganic particles. In spongin fibres of horny sponges this uptake seems to be definitive: the particles increase in number with the sponge growth. *Chondrosia reniformis* is characterised by a collagenous skeleton which lacks endogenous spicules and by an ectosome which is reinforced by foreign matter, collected from the water column thanks to a continuous production of a thin mucous film on the sponge surface. *C. reniformis* shows a high capability of particle selection, collecting mainly exogenous siliceous spicules, silicates and quartz particles and avoiding carbonates. Moreover, *C. reniformis* shows an evident turnover of the incorporated foreign matter, probably linked to its unusual ability to dissolve quartz. The possible relationships between the annual cycle of the foreign matter present inside the sponge body, the local sedimentation and the sea conditions along the rocky cliffs of Portofino Promontory (Ligurian Sea, Italy) were evaluated monthly at two sites (Punta del Faro and Paraggi Bay), characterised by different sedimentary conditions.

At Punta del Faro, where the cliff ends at a depth of 55m, specimens of *C. reniformis* were sampled monthly by SCUBA diving from March 1994 to June 1995 at depths of 3m, 12m and 25m. At these last two depths, two sediment traps were placed to collect the fraction of sediments available to the sponges. At Paraggi Bay, where the cliff ends at 25m depth, sponges were collected at a 3m and 15m depth, and a sediment trap was placed in this last station. At both localities, a superficial (3m depth) sediment trap was also placed, but strong wave action prevented a sufficient continuity in data collection.

A quantitative comparison among the annual trends of the sediment engulfed by the sponges (sand grains and opaline spicules), the sediments collected by traps, and the sea condition evidences a strong influence of this last parameter on the uptake process in function of depth.

Moreover, there is an inverse relationship between the size of the engulfed particles and sea conditions. During calm periods, the sponge can also take up larger particles, whereas during rough periods, the sponge's sticky surface is unable to do so. In addition, resuspension processes play an important role near the sea floor where only fine particles became available. Thus, it is possible to assume that the rough sea can, firstly, limit the uptake of the particles and, secondly, increase their availability by resuspension processes.

Interestingly, the large amount of quartz grains continuously incorporated and dissolved by *Chondrosia*, suggests a possible local role played by this sponge in the silica balance in the shallow coastal waters. Our data estimated that the *C. reniformis* population of the Portofino Promontory makes about $2x10^6$ g of dissolved silica available each year.

Key words: *uptake, foreign matter, silica, sedimentation, sea condition, horny sponges, Mediterranean Sea*

DO CARIBBEAN SPONGES HAVE PHYSICAL DEFENSES ?

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Sponges are conspicuous members of the Caribbean marine ecosystem, but are preyed upon by a very select group of consumers called spongivores. Like other sessile reef invertebrates such as ascidians and octocorals, sponges possess a variety of novel secondary metabolites and as well as mineral and organic skeletal components. Several studies have shown that sponges possess chemical defenses that inhibit feeding by browsing generalist fish, but no study to date has demonstrated that sponge skeletal components deter predation. Sponges are soft-bodied and seem to lack an obvious physical defense, such as a mineralized shell. However, the tissues of most sponges often contain a collagen-like substance called spongin and sharp siliceous spicules in high concentrations. Spicules serve as important structural components by increasing tissue rigidity and could potentially act as a defense by irritating the mouth parts and the digestive system of predators. Calcified structures, similar in size to spicules, from octocorals and algae have been shown to reduce feeding by fish and invertebrates. Surprisingly, field and laboratory aquarium assays of sponge spicules employing predatory reef fish did not support a defensive function. Consumption by reef fish was reduced only when spicules were assayed using foods of low nutritional quality. In assessing the chemical defenses of Caribbean sponges, 31% of the species we studied possessed organic extracts palatable to reef fish. Interestingly, many of these undefended sponge species are abundant and consumed only by spongivores. Sponges lacking a chemical defense may be protected from generalist predators by having tissues of low nutritional value. Protein, carbohydrate, lipid, ash, and caloric content of 71 Caribbean sponge species were measured to investigate the relationship between chemical defense and nutritional value. Except for lipid content, no significant differences in nutritional quality were found between chemically defended and undefended species. Sponges lacking a chemical defense may rely on tactics other than a physical or "nutritional" defense, such as faster growth rates, to avoid predation by generalist consumers.

Key words: *chemical defenses, physical defenses, spicules, silica, nutritional quality, predatory-prey interactions, Caribbean reef ecosystems.*

GOOD CONGRUENCE BETWEEN MORPHOLOGY AND MOLECULAR PHYLOGENY OF HADROMERIDA, OR HOW TO BOTHER SPONGE TAXONOMISTS

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Within Demospongiae, the order Hadromerida is well defined and there is a strong consensus among systematicians about its composition and validity. This order is characterised by the presence of tylostyles radially arranged at least in the periphery, and by microscleres, when present, of the aster type. All Hadromerida are oviparous and the choanocytes have a periflagellar sleeve. Ten families are without any doubt attributed to Hadromerida, six of which with microscleres of the aster type and four of which without microscleres.

The first work on molecular phylogeny of Porifera was made on the Hadromerida (Kelly-Borges, Bergquist & Bergquist, 1991). The molecule used was the 18S rRNA, which appeared to be not sufficiently informative to resolve the phylogeny at that taxonomic level.

In this work we have used the 5' end of the 28S rRNA (about 1000 bp) to explore the internal phylogeny of this order. 15 species belonging to 12 genera and 8 families were sequenced. Five outgroup species were sequenced belonging to Axinellida, Tetractinellida, and Halichondrida. Parsimony and Neighbor-Joining analyses have been done. Trees were rooted by using Tetractinellida (*Cinachyrella* and *Discodermia*) as a monophyletic outgroup. Both analyses (Parsimony and Neighbor-Joining) show that the Hadromerida are composed of four monophyletic taxa. Taxon 1 is composed of 6 species belonging to the Spirastrellidae, Acanthochaetetidae, Clionidae, and Placospongiidae. All these families have microscleres of the spiraster-type. Taxon 2 is composed by 5 species of Timeidae and Tethyidae. These two families have microscleres of the euaster-type. Taxon 3 is composed of only one species *Polymastia mamillaris* belonging to the family Polymastiidae, which has no microsclere of aster type. The validity of this taxon has to be checked with other genera belonging to the Polymastiidae family. Taxon 4 is composed of three Suberitidae and an external species *Halichondria panicea*, which belongs to the family Halichondriidae (order Halichondrida). Neither the Suberitidae nor the Halichondriidae have microscleres of the aster type. The monophyly of each of these four taxa is well supported with high bootstrap proportions. The monophyly of the four taxa together is also well supported but the relationships between them cannot be ascertained.

The monophylies of taxa 1 and 2 are congruent with morphology, both taxa corresponding to the hadromerid families with spirasters and with euasters, respectively. An important and unexpected problem of classification appeared with taxon 4. The result obtained with our sequence of *Halichondria panicea* was confirmed with a shorter sequence of *Hymeniacidon heliophila* available in GenBank. When the sequence of *Hymeniacidon* is included, taxon 4 remains monophyletic and strongly supported by BP. From the morphological and cytological point of view there is no synapomorphy between the two groups. The Halichondrida are defined mostly by negative characters. However, we observed a fine morpho-molecular synapomorphy for taxon 4. This is the loss of a small loop of 15 bp in the secondary structure of the D2 domain, which is probably the result of only one deletion event. From the chemical point of view, there is another synapomorphy: a large amount of stanols have been described both in the Suberitidae and the Halichondrida.

The best hypothesis seems to reallocate Halichondriidae to the Hadromerida. The order Hadromerida remains monophyletic. With the exception of this reallocation the classification obtained with 28S rRNA is perfectly congruent with the existing classification. All the families are monophyletic. We propose a subordinal classification : Spirastrellina, Timeina, Polymastiina and Suberitina.

Key words: *Demospongiae, molecular phylogeny, 28S rRNA, Hadromerida, Halichondrida, monophyly*

SPONGE DISTRIBUTION AND LAKE CHEMISTRY AMONG NORTHERN WISCONSIN LAKES: MINNA JEWELL'S SURVEY REVISITED

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An extensive survey of the regional distribution of freshwater sponges, and of the factors that control their occurrence, was conducted by Minna Jewell in Northern Wisconsin, USA during the 1930s. We returned to 18 of her original 102 study lakes in 1996-97 to determine the 60-year stability of the sponge distribution patterns that she reported. Comparisons of Jewell's data and our recent survey suggest that the distribution of *Spongilla lacustris* has largely remained stable over time. This species was originally observed in 76 of the Jewell study lakes, and we found no change in the distribution of *S. lacustris* in 13 of the 18 lakes revisited. Additionally, we applied a discriminant model to the historical data set, to test how effectively four chemical variables (pH, colour, conductivity, and SiO_2) could predict the distribution of *S. lacustris*. Based on these four variables, however, this model failed to predict current distribution patterns in 17 additional lakes sampled in 1996, indicating that other environmental variables are likely influencing *S. lacustris* distribution in northern Wisconsin lakes.

Key words: *ecology, freshwater sponges, distribution patterns, chemical variables, Wisconsin*

AN OVERVIEW OF STROMATOPOROID DOMINATED MIDDLE DEVONIAN REEF COMPLEXES IN NORTH QUEENSLAND

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Middle Devonian stromatoporoid buildups are known from the Burdekin Subprovince and the Broken River Province in the Townsville hinterland, north Queensland.

Recent studies have placed these buildups within a reliable stratigraphic and sedimentologic framework. Buildups within the Burdekin Subprovince developed in a restricted near to proximal shore setting in a partially enclosed basinal setting. Those buildups within the Broken River province developed upon a more open marine shelf.

Major Burdekin stromatoporoid-coral buildups were of two types: low relief extensive biostromes and associated stromatoporoid pavements, and a biohermal system of one to two metres relief from the sea floor. Additional buildups of note are small patch reefs developed within nearshore siliciclastic muddy lagoons adjacent to granitic headlands. In a number of such metre scale buildups within dominantly siliciclastic settings, assemblages of stromatoporoids and corals show repetitive growth interruption surfaces suggesting episodic stress and killing events.

Storm disturbance during development the biostromal pavements was high and an important sedimentologic factor for the "reef" growth. Minor sponge s.s. buildups are known from the uppermost Burdekin Formation, but have not been studied.

In the Broken River Province, Givetian buildups are more extensive and can be traced on the hundreds of metre scale, these have received little detailed sedimentologic study, but are of similar style to biostromal pavements from the neighbouring Burdekin Basin. Minor biohermal occurrences are found within the Papilio Mudstone, and formed on a muddy shelf, and include both stromatoporoid and sponge s.s. buildups.

Stromatoporoid taxonomy has revealed the presence of eight stromatoporoid communities in the Burdekin Basin, comprising 35 taxa. Dominant stromatoporoids were dendroids *Amphipora, Stachyodes and Trupetostroma,* frame building. *Trupetostoma Pseudotrupetostroma, Hermatostroma, Actinostroma* and *Ferestromatopora. Coenostroma, Clathrocoilona,* and *Stromatopora* were accessory to reef growth. In the Broken River detailed taxonomic work has only been partially completed. Significant overlap exists at generic level with the two adjacent provinces, but species level differences are strong suggesting distinct partitioning of open marine versus embayment faunas. This phenomenon is reflected in other faunal elements (gastropods, rugose corals).

Key words: *stromatoporoid, biostromes*

SPONGES FROM HERON ISLAND, CAPRICORN-BUNKER GROUP, GREAT BARRIER REEF

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The Capricorn–Bunker Group consists of 20 reefs lying in the southern section of the Great Barrier Reef (23°26'S, 151°55'E). To the north lie 14 reefs (Capricorn Group), 8 of which have vegetated cays; whereas in the south there are 6 reefs, 3 with vegetated cays (Bunker Group). The Queensland Museum

recently surveyed the sponge faunas of both groups of reefs, although the present study is restricted to the reefs around Heron Island (Heron, Wistari and Sykes Reefs), depicting some of the more common or prominent (or photogenic) species.

A total of 216 species of sponges were collected from these three reefs, comprising 2 classes, 14 orders and 43 families. Collections made intertidally (on exposed reef flats and rock pools) were less diverse than those made subtidally (off the edge of the reef flat): 69 species in 43 genera, versus 155 species in 81 genera, resepectively. Surprisingly, only 8 species occur on both the reef flat and subtidal habitats, of which 3 belong to the Calcarea. To date 71 of these 216 species have not been found elsewhere ('apparent endemics'), either on the Great Barrier Reef or the Queensland coast.

Key words: *Heron Island, Great Barrier Reef, intertidal, subtidal, sponge fauna.*

REMARKS ABOUT THE STATUS OF THE GENUS *MYXILLA* (PORIFERA, POECILOSCLERIDA) IN THE GALICIAN COAST (NW OF IBERIAN PENINSULA)

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The genus *Myxilla* Schmidt, 1862 is represented on the Iberian Peninsula by six species, five of which have been collected off the coast of Galicia, and are studied in this paper: *M. incrustans, M. iotrochotina, M. macrosigma,* and *M. fimbriata*; the sixth, (*M. tarifensis.*), has been recently described in the Strait of Gibraltar (Carballo & García-Gomez, 1996).

The material under study comprises 188 specimens taken from 72 stations all along the coast of Galicia between 1979 and 1991. The intertidal specimens were collected during the neap tides occurring every month, while the subtidal samples were obtained from hard substrates by Scuba diving and by means of a horizontally operated rectangular naturalist benthic dredge (Holme & McIntyre, 1984) in soft bottoms.

Based on these data this paper presents a complete illustrated description of the their habitus, skeletal arrangement and spicules in the form of underwater photographs, figures and SEM photography. Also included are autoecology, distribution and a biometric study of the spicules in addition to a morphological-systematic discussion in which the details of the specimens collected in comparison to those from other latitudes are described.

Lastly an identification key is provided for the species of the genus *Myxilla* in the NE Atlantic.

Key words: Myxilla, *taxonomy, habitat, Galicia, NE Atlantic*

TEMPERATE SPONGE ASSEMBLAGES EXPOSED TO GRAZING: THE ROLE OF REFUGIA AND CHEMICAL DEFENSE.

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In the shallow (<15m) subtidal zone of southeastern Australia a 'barrens' habitat is maintained by the diadematoid sea urchin *Centrostephanus rodgersii.* High densities of this urchin remove all algae and encrusting invertebrates with the exception of grazer-resistant coralline algal crusts. Nevertheless, sponges and other sessile invertebrates occur in this habitat, usually on vertical surfaces, and cover around 2% of the substratum. I sought to determine how these invertebrates maintain space in the face

of intense grazing pressure by the urchins. Sessile invertebrate cover was positively correlated with the density of the large barnacle *Austrobalanus imperator* while the intensity of urchin grazing, as determined by scrape marks on plastic disks, was negatively correlated with barnacle density. Manipulation of barnacle density at two sites, involving the addition and removal of barnacles, indicated that invertebrate recruitment and the cover of sponges was highest among high densities of *Austrobalanus* (>220 barnacles m^{-2}). These experiments also revealed that well established sponges do not require barnacles to maintain space. Urchin feeding trials with sponge extracts indicate that a number of common sponges in this assemblage possess chemical antifeedants. These data suggest that sponges maintain space in this system in two ways: (i) they only recruit successfully to refugia from urchin grazing provided by high densities of the barnacle *Austrobalanus imperator* and (ii) they dissuade urchins from grazing by using secondary metabolites. Identifying the natural products responsible for the feeding-deterrent properties is the focus of current work.

Key words: *antifeedant activity, experimental ecology, invertebrate recruitment, refugia from grazing, sponge secondary metabolites*

DO WE NEED SUBGENERA IN HAPLOSCLERIDA? CONVENIENCE VERSUS PHYLOGENETIC RELIABILITY

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There are some genera such as *Callyspongia, Haliclona, Mycale, Myxilla*, etc., which include a large number of species. Within these genera there are currently no phylogenetically meaningful groupings due to the absence of stable character combinations present in other groups of sponges. From a practical point of view, this makes these genera difficult to manage. For improving this situation, authors have created subdivisions for species that are difficult to place, and it has been proposed to introduce subgenera. How do we decide when and by which method we have to erect a new subgenus? We should obtain the realisation and rationalisation of the number of nominal haplosclerid/petrosid genera, versus the number which are actually "valid" (= "phylogenetically meaningful"). We need to understand why there exists such a great difference between "nominal and actual" genera and why most taxonomists do not accept artificial genera.

We should decide between the use of subgenera that are "convenient", easily manageable and consequently useful for species identification, but not strictly "phylogenetic", or subgenera where the characters have been tested from a phylogenetic point of view and which form monophyletic groupings. It is suggested here to aim for the phylogenetic solution where we will obtain a classification which is homogenous, testable, consistent, objective and just as useful for species identification as an artificial convenient classification.

Key words: *Haplosclerida, phylogenetics, generic subdivisions, cladistics*

PERSPECTIVES ON SPONGE-CYANOBACTERIAL SYMBIOSES

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Insights on the evolution of sponge-cyanobacterial symbioses are drawn from biogeographic and molecular data. The taxonomic and geographic distribution of sponge-cyanobacteria associations is analysed after surveying their occurrence at eight localities in the Eastern and Western Tropical Pacific, and the Caribbean. Three methods - fluorescent microscopy, thin layer chromatography and transmission electron microscopy - were used to infer the existence of endosymbiotic cyanobacteria.

Thirty-eight species, representing 17 families and 11 orders of Demospongiae, and one family and order of Calcarea, are added to the list of sponges involved in these associations. This number represents an increase of more than 50% over previously known occurrences of this type of metazoan-microbial association. However this increase of species numbers represents only an addition of twelve genera and two families to the taxonomic distribution of these associations. Species from 26 of the 72 recognised Demospongiae families, and 3 of the 17 recognised Calcarea families are found to harbour cyanobacterial endosymbionts. These data suggest a rather restricted taxonomic range for sponge-cyanobacterial assemblages, and invites a search for evolutionary trends among the families involved. The genera with highest number of species harbouring cyanobacteria are: Aplysina (10 spp), Xestospongia (7 spp), Dysidea (5 spp), and Theonella (5 spp). Although the updated list of sponge-cyanobacterial assemblages shows a few biogeographic trends, the understanding of the evolution of these associations requires the study of more extensive geographic areas.

The use of 16S rDNA analysis to understand the phylogenetic relationships of endosymbiotic cyanobacteria is discussed. Genetic analyses promise to shed light on the understanding of the evolution and specificity of these associations. 16S rDNA gene analyses carried out so far suggest that sponge-cyanobacterial assemblages comprise diverse and complex evolutionary histories, some of which might share evolutionary pathways with other important marine symbiotic assemblages involving cyanobacteria.

Key words: *cyanobacteria, endosymbioses, biogeography, evolutionary trends, 16S ribosomal genes*

DRUG FARMING IN THE 21ST CENTURY

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Commercial farming of sponges for drug production has much potential. For this potential to be realised, however, requires research into some fundamental questions. These include, assessment of environmental conditions that promote maximum sponge growth and survivorship, while allowing stable or enhanced biosynthesis of target metabolites. Additionally, farming methods and structures that optimise these sponge and metabolite characteristics, must be examined. Aquaculture techniques must be cheap, easy and practicable to use for large-scale production, and ideally flexible for a variety of sponge, and indeed other invertebrate, species. This paper describes the essence of sponge aquaculture, providing case studies of *Polymastia croceus, Latrunculia brevis* and *Raspailia agminata*. All three sponges elicit bioactive metabolites that have chemotherapeutic potential.

Ecological observations suggest that all sponges exhibit seasonal fluctuations in growth rates and metabolite chemistry. There were also latitude/site/depth-related variations in metabolite biosynthesis. To examine where best to carry out aquaculture for these species, a number of hypotheses relating to growth and target metabolite biosynthesis were tested.

To determine what environmental conditions are important we cloned and seeded *P. croceus* and *L. brevis* in each season to differing exposures and depths. *P. croceus* had the most promising results with some explants doubling their weight in two months. Best growth and survivorship occurred in spring, and in all seasons at exposed locations. The optimal depth depended on season of cloning and deployment. In comparison, *L. brevis* had greatest growth and survivorship in winter, although at the sheltered and moderately exposed locations, growth and survivorship was generally good. *L. brevis* could be farmed at all depths within the range examined.

In developing a commercial farming method, we experimented with four general techniques. Explants of *P. croceus, L. brevis* and *R. agminata* were (1) contained inside something; (2) attached to rope/cord; (3) had rope/cord passed through them; and (4) had rope/cord passed around them. For all three sponges we found that explants contained inside mesh had the most promising growth and survivorship. From these experiments we have devised a commercial farming structure.

Key words: *commercial farming, environmental conditions, seasonal conditions, propagation techniques*

DISCOVERY AND SUSTAINABLE SUPPLY OF MARINE NATURAL PRODUCTS AS DRUGS, INDUSTRIAL COMPOUNDS AND AGROCHEMICALS: CHEMICAL ECOLOGY, GENETICS, AQUACULTURE AND CELL CULTURE.

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Using chemical ecological clues, it is now possible to target habitats and eco-taxonomic groups of marine organisms to increase the likelihood of discovery of species which elicit natural compounds with chemotherapeutic or industrial application. Using the same clues, combined with Geographic Information System interrogation of the benthic geomorphology and oceanography associated with target species, it is possible to identify locations allowing recollection of species of interest. The information gained from both primary collections and focused recollections, provides the basis for hypothesis driven experiments examining sustainable supply options for extracted target metabolites where synthesis is not practicable.

We describe recent results from an integrated multi-disciplinary programme designed to develop sustainable production options for a variety of marine natural products that have interesting biological activities. Three species of sponge from the genera *Lissodendoryx, Mycale* and *Latrunculia*, produce novel metabolites with anti-tumour activity. The natural abundance of each would not support a production industry based on wild harvest should their metabolites be required for drug production. Each has been successfully cultured in-sea demonstrating very good to excellent growth parameters. Each can be cultured with maintenance of target metabolite biosynthesis. In addressing the question of how to optimally produce target compounds, it has been necessary to examine a number of key biological issues pertaining to each species. These include genetic identity of populations supplying seed material, correlates with variable target metabolite biosynthesis in natural populations, origin of target metabolite biosynthesis (symbiont or sponge), and the efficacy of artificial production techniques (sea or land aquaculture or cell culture).

We conclude that the guess-work can now be taken out of artificial culture of sponges with a view to produce desirable natural products. It is possible to select for a high yielding culture stock and provide techniques to enhance biosynthesis or target metabolites.

Key words: *marine natural products, aquaculture genetics, cell culture*

POLLY WANT A SPONGE? : FIELD EXAMINATION OF SPONGIVORY BY CARIBBEAN PARROTFISHES IN REEF AND MANGROVE HABITATS

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Caribbean sponge species such as *Xestospongia muta* frequently display linear grazing scars that appear to have been made by parrotfishes, yet there are few scientific reports of parrotfish spongivory. We used a video camera to monitor 40 specimens of *X. muta* for a minimum of 0.5 hr/sponge to determine the frequency of parrotfish bites on this species. Ten hours of taping captured 45 bites on normally coloured sponges, and 527 bites on four bleached sponges. Also, the guts from parrotfishes collected in mangrove and reef habitats were digested in nitric acid and analysed for spicule content. Parrotfishes collected in the mangroves (*Sparisoma aurofrenatum, Scarus croicensis*, and *Sc. taeniopterus*) had a significantly greater mass of spicules in their guts than did parrotfishes collected on the reef (*Sp. aurofrenatum, Sp. viride, Sp. chrysopterum, Sc. vetula, Sc. coelestinus*, and *Sc. taeniopterus*). Up to 148mg of spicules were present in the guts of mangrove parrotfishes. The spicules of *Geodia gibberosa*, a sponge that is common in the mangroves but rare in exposed locations on the reef, were abundant in the gut samples. Our results suggest that some sponge species are palatable not only to specialist predators such as sea turtles and angelfishes, but also to species that are not usually recognised as sponge predators.

Key words: *spongivory, parrotfishes*, Xestospongia muta, Geodia gibberosa, Sparisoma spp., Scarus spp., *spicules, ecology, predation*

DEVELOPMENT OF *HALISARCA DUJARDINI* JOHNSTON 1842 (PORIFERA, CERACTINOMORPHA, HALISARCIDA) FROM EGG TO FREE LARVA

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Embryonic development in the sexual viviparous sponge *Halisarca dujardini* from the White Sea (Arctic) shallow water was studied. Complete, equal, asynchronal cleavage is characterised with variability of analogous developmental stages and the lack of the strictly determined cleavage spindles position. The cytoplasm is filled with numerous yolky granules with heterogenic contents. At the 16-24 cell-stage a small cavity is formed. Blastomeres and the embryo polarity are not expressed. Large nuclei containing pronucleolar bodies are situated at the central parts of the cells. From the 16-24 cell- stage, true nucleolus formation starts. The polarisation of blastomeres is expressed by the distal movement of nuclei and changes in cell form. Cleavage furrow planes obtain the similar radial pattern forming roundish stereoblastula 130-170μm in diameter with the small cavity restricted with long wedge-shaped cells.

The internal layer of the larva is formed at the 100-130 cell-stage owing to the individual cells' apolar migration out of the blastula walls. At the same time flagella are formed on the cells' apical surfaces, yolk granules being concentrated basally. Internal cells proliferate actively, differentiating into nucleolated amoebocytes, granular cells, collencytes and choanoblasts.

The larvae are of parenchymella type and are covered with flagella. Flagellated cells are less numerous at the posterior pole. Flagellated epithelial cells are wedge-shaped. At their apical parts they contain a drop-like nucleus with nucleolus and a flagellum embedded into a pocket-like cytoplasmic invagination. The basal 2/3 of the cell volume is filled with numerous yolk granules. Flagellated cells are connected at their apical end by outgrowths of the plasma membrane embedded into similar invaginations of the neighbouring membrane. Posterior flagellated cells are trapeziform or rectangular, and contain numerous yolk granules. The nuclei are roundish, with large nucleoli. There is one spherical cavity inside the larva, which is formed by a layer of cylindrical cells that have flagella and microvilli inside the cavity. Their piriform nuclei contain nucleoli, and there are yolk granules in the cytoplasm. There are no specialized cell contacts between blastomeres and larval cells. The spiral symbiotic bacteria are present in the central part of the larva and in intercellular spaces.

Some peculiarities of *H. dujardini* embryogenesis are unique among Ceractinomorpha and are a matter of principle for comparative embryological studies of Porifera. They are: 1) total equal asynchronic cleavage; 2) equal, apolar coeloblastula with a small cavity; 3) unexpressed polarity of blastomeres; 4)

subsequent of the same type radial cleavage leading to the cell polarisation and stereoblastula formation; 5) formation of an internal cell mass larva in the larva by multipolar cell ingression at the 100-130 cell-stage; 6) development of special choanoblasts. All the features mentioned can serve as additional arguments for separation of the Halisarcida as an order (Bergquist, 1996).

Key words: Halisarca dujardini, *embryology, cleavage, larva, cells, ultrastructure*

AN ASSESSMENT OF 'SPACE WARS' AS A DETERMINING FACTOR IN THE PRODUCTION OF NOVEL BIOACTIVE INDOLES BY *IRCINIA SP.*

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Ircinia sp from Salamander Reef has yielded a plethora of bioactive indoles including mono, di and non brominated variants. We have isolated and identified seven of these compounds and at least forty more await attention. Metabolite composition varies widely within and between individual sponges, and the question arises as to the cause of this variability.

Ircinia sp is a common sponge at Salamander Reef, an isolated inshore rocky reef near Townsville where competition for space is fierce. All viable hard substrate is covered by sessile biota which perpetually interact with neighbouring rivals, hence the possible role of 'space wars' as a determining factor in metabolite variability in *Ircinia sp* was investigated.

Tissue biopsies were taken from different interaction circumstances within sponge individuals, and metabolite profiles were compared. HPLC chromatograms were used, along with UV spectra, to identify gross patterns in the relative contributions of numerous novel indoles. Identification of the major components in the HPLC profile was accomplished by negative ion electrospray mass spectrometry of collected fractions.

Two predominant populations of metabolites were identified on the basis of the relative importance of a close group of conjugated brominated indoles and their putative precursor. Diminished precursor and increased product occurred in tissue from interaction sites or fleshy projections. However this trend was not absolute and the results suggest that factors determining metabolite composition are complex. Hypotheses for further investigation are presented.

Key words: *chemical ecology, indoles, competition, metabolites*

TRACE ELEMENT AND STABLE ISOTOPE PROFILES FROM THE CORALLINE SPONGE (*ASTROSCLERA WILLEYANA*)

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Techniques developed for laser-ablation-ICP-MS analysis of corals have now been utilised for the analysis of trace elements in the coralline sponge *Astrosclera willeyana.* In scleractinian corals the elements B, Mg, Sr, Ba and U show seasonal variations consistent with environmental parameters, predominantly sea surface temperature and variations in upwelling. We report here a preliminary investigation to determine whether elemental distributions in sclerosponges will provide meaningful proxy information about past oceanographic conditions.

Samples from Taveuni, Fiji, Ruby Reef, GBR and Truk, Caroline Islands have been analysed at a sampling resolution of ~40 µm. With current techniques and data reduction methods, sampling at this resolution produces too much variation to show any elemental correlations. When samples are filtered to ~100µm resolution, longer-term (annual to several year) patterns appear, which are consistent between the B/Ca, Mg/Ca, Sr/Ca and Ba/Ca cycles. This suggests a common incorporation mechanism between these four elements. If this variation is temperature related, the method of incorporation is markedly different than corals. The boron, magnesium and barium concentrations in sclerosponges are 2-5 times lower than in corals, with concentrations of ~20 ppm, ~200 ppm and ~4 ppm, respectively. The strontium and uranium concentrations are 1-2.5 times higher than in corals with concentrations of ~9000 ppm and ~7 ppm respectively. We will also present preliminary stable isotope data ($\delta^{18}O$ and $\delta^{13}C$) to compare with the trace element profiles.

Key words: Astrosclera, *Sr/Ca, Mg/Ca, Ba/Ca, laser ablation ICP-MS, $\delta^{18}O$, $\delta^{13}C$, environmental parameters*

PRODUCTION OF BIOACTIVE METABOLITES BY SYMBIOTIC MICROORGANISMS IN MARINE SPONGES

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Marine invertebrates are constantly in contact with microorganisms that can live on or within their tissues but only those microorganisms that maintain a constant association with their host can be considered symbionts. Furthermore, it is generally accepted that the relationship between a microbial symbiont and a host organism is one from which both derive some benefit. We are particularly interested in the role of symbiotic microorganisms in the production of bioactive marine natural products that are isolated from marine sponges and tunicates.

In the past, it has been proposed that many bioactive metabolites from marine invertebrates were produced by symbiotic microbes. The rationale for these proposals was usually that the compounds resembled metabolites from cultured microorganisms or that the same compound was isolated from invertebrates of different phyla. We have now demonstrated that arguments based on "chemical logic" are not always correct and that every instance where chemical production by a symbiont has been proposed should be carefully investigated.

This presentation will highlight methods used to identify the cellular location of bioactive metabolites in marine invertebrates. Some metabolites have been located in bacterial and cyanobacterial cells, while others are clearly found in the invertebrate cells. We have previously reported that chlorinated and brominated metabolites from the sponge *Dysidea herbacea* were localized in cyanobacterial cells and that in the sponge *Theonella swinhoei*, the bicyclic peptide theopalauamide was located in filamentous eubacteria while swinholide was found in a fraction containing a mixture of unicellular bacteria. The most recent research from our laboratory will be presented.

Key words: *symbionts, bacteria, cyanobacteria, bioactive compounds*

SPONGE CELL ADHESION: AN EVOLUTIONARY ANCESTOR OF HISTOCOMPATIBILITY SYSTEMS?

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Sponges have been traditionally used as models to study cell adhesion because their rather loose and porous extracellular matrix allows a mild cell dissociation and the recovery of intercellular components in virtually native state. Species-specific cell recognition and adhesion in sponges is mediated by extracellular proteoglycan-like complexes termed aggregation factors (AFs), still not identified in higher animals. Polyvalent glycosaminoglycan interactions are involved in the species-specificity, representing one of the few known examples of a regulatory role for carbohydrates.

A surprising characteristic of sponges, considering their low phylogenetic position, is that they possess an exquisitely sophisticated histocompatibility system. Any grafting between two different sponge individuals is almost invariably incompatible in the many species investigated, exhibiting a variety of transitive qualitatively and quantitatively different responses, which can only be explained by the existence of a highly polymorphic gene system regulating sponge allogeneic reactions. The development of variable-region molecules is thought to have been a crucial event in the evolution of primordial vertebrate immune systems, followed by gene rearrangement to provide more diversity. Early in the evolution of the immune system, then, a gene must have duplicated to allow such diversity to arise. Unfortunately, there is an absolute lack of protein sequence information concerning the molecules involved in invertebrate histoincompatibility reactions. Recently, we deduced from cDNA the sequence of the aggregation factor core protein from the red beard sponge, *Microciona prolifera*, and Southern blot analysis suggested the existence of several related genes.

We have screened individual sponge cDNA libraries, identifying multiple related forms for the AF core protein (MAFp3). Northern blots show the presence in several human tissues of transcripts strongly binding a MAFp3-specific probe. We have studied tissue histocompatibility within a sponge population, finding 100% correlation between rejection behaviour and the individual-specific restriction fragment length polymorphism pattern using AF-related probes. PCR amplifications with specific primers showed that at least some of the MAFp3 forms are allelic and distribute in the population used. A pronounced polymorphism is also observed when analysing purified AF in polyacrylamide gels. Protease digestion of the polymorphic glycosaminoglycan-containing bands indicates that glycans are also responsible for the variability. The data presented reveal a high polymorphism of aggregation factor components which matches the elevated sponge alloincompatibility, suggesting an involvement of the cell adhesion system in sponge allogeneic reactions. Our present work will be discussed in the context of the evolution of histocompatibility systems and their possible divergence from primitive cell-cell interaction molecules.

Key words: *graft rejection, proteoglycans, invertebrate immunity, aggregation factors, cell adhesion, porifera genes, cDNA, histocompatibility*

THE DEMOSPONGE FAUNA OF THE HOUTMAN ABROLHOS ISLANDS

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The Houtman Abrolhos Islands lie off the West Coast of Australia within a biogeographic zone that has overlapping temperate and tropical components, and a proportion of endemic species. The islands are situated in the path of the warm, southward flowing Leeuwin current. Studies on marine biota of these islands has found a dominant tropical component to the fauna. The marine sponges of the Houtman Abrolhos islands are poorly studied, therefore a field program was established to comprehensively collect sponges and document the species that occur there, and to determine if this biota was principally tropical or temperate in origin. Seventy-six demosponge species are reported from the two localities examined in this study. Twenty-eight of the species are known to science, thirteen are identified to species but require confirmation by comparison with type material, and thirty five species are probably new. Three genera are reported for the first time from Australia. This study brings the total number of demosponge species that have been documented as occurring at the Houtman Abrolhos to one hundred and eight. Preliminary assessment of the tropical and temperate components of the sponges are presented, and related to comparable studies on other components of the marine biota of these islands.

Key words: *Demosponges, Houtman Abrolhos Islands, Western Australia, biogeography*

REPRODUCTION OF SOME DEMOSPONGES IN A TEMPERATE AUSTRALIAN SHALLOW WATER HABITAT

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Species of *Tethya, Chondrilla, Mycale* and *Echinodictyum* have been monitored for two years at South Mole, Fremantle (32°04'S:115°45'E) to determine onset of reproductive activity, sex phenotype, and reproductive mode of the various species. Most reproductive activity occurs in the summer months November to January. The majority of the species are contemporaneous hermaphrodites and both ovipary and vivipary have been found. Details of the reproductive development of these species is reported and discussed in relation to environmental variables such as temperature.

Key words: *Demosponges, Fremantle, Western Australia, reproduction, environmental variables*

MEMBRANE-BOUND NUCLEAR BODIES IN A DIVERSE RANGE OF MICROBIAL SYMBIONTS OF GREAT BARRIER REEF SPONGES

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Thin sections of chemically fixed tissue of several sponge species collected from Heron Island on the Great Barrier Reef, including *Jaspis stellifera, Pseudoceratina crassa,* and *Axinyssa* sp., were examined to investigate the cell organisation of the bacteria-like microbial symbionts present. Such symbionts have been observed in these sponges to occur as a diverse range of morphotypes based on cell shape and cell wall type. A variety of different symbiont morphotypes were found to possess a membrane-bound nucleoid, a feature not expected in prokaryotes. These had been previously observed by us in one symbiont morphotype in the Micronesian sponges *Stromatospongia micronesica* and *Astrosclera willeyana.* Several distinct microbial morphotypes containing membrane-bound nuclear bodies were observed in Great Barrier Reef sponges, only one of which resembled the type which we have previously observed in Micronesian sponges. In all these forms, the fibrillar nucleoid was surrounded by a single bilayer membrane, in most morphotypes defining a compartment also containing electron-dense particles resembling ribosomes or other nucleoplasmic material; such material was sometimes less dense and sometimes more dense than the cytoplasmic particulate material. Cell wall structure of the morphotypes broadly included both Gram-positive and Gram-negative, outer membrane-bound types, as well as cell walls with a clear subunit S-layer type structure resembling that of known Archaea including crenarcheotes and other less clearly classifiable types. Cytoplasmic membranes can be clearly seen in some cases as distinct from nuclear body membranes, excluding plasmolysis as an explanation for membrane-boundedness of nuclear bodies. The phylogenetic relationships of these microbes may be diverse if reflecting wall type, but at least some appear to be most likely to represent members of the Domain Archaea, perhaps resembling the crenarcheote *Cenarchaeum symbiosum* described from North American *Axinella* sp.

Key words: *Bacteria, Archaea, nucleoids, membrane-bound, symbionts, electron microscopy, ultrastructure*

CYANIDE AND THIOCYANATE-BASED BIOSYNTHESIS IN TROPICAL MARINE SPONGES

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Inorganic cyanide is a biosynthetic precursor to isonitrile metabolites found in marine sponges, and also to the co-occurring isothiocyanates. Thiocyanate is now also an established precursor to isonitriles and isothiocyanates[2]. The sponge *Axinyssa* n.sp. incorporates both sodium [^{14}C] cyanide and sodium [^{14}C] thiocyanate into 2-thiocyanatoneopupukeanane as well as the 9-isothiocyanatopupukeanane, however not into 9-isocyanopupkeanane. The specificity of incorporation into the thiocyanate carbon was confirmed by chemical degradation.

Stylotella aurantium incorporates sodium [^{14}C] cyanide and sodium [^{14}C] thiocyanate into the rare dichloroimine functionality present in stylotellanes A and B, as well as into farnesyl isothiocyanate. The specificity of incorporation into the dichloroimine carbon atom was confirmed by chemical degradation.

This work represents the first detailed study of the biosynthetic origin of organic thiocyanates and dichloroimines, and extends the range of functionality known to be biosynthesised from cyanide and thiocyanate. Our results raise the interesting question of the interconversion of inorganic cyanide and thiocyanate and/or the interconversion of the resulting organic metabolites in marine sponges. Advanced precursor experiments, which are in progress, may shed further light on these conversions.

Key words: *biosynthesis, cyanide, terpenes, thiocyanate,* Axinyssa n.sp., Stylotella aurantium

THE ECOLOGICAL ROLE OF CYTOTOXIC ALKALOIDS: *HALICLONA* N. SP., AN UNUSUAL SPONGE/DINOFLAGELLATE ASSOCIATION

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Light microscopy and electron microscopy studies of the tropical marine sponge *Haliclona* n. sp. (Order: Haplosclerida; Family: Haliclonidae) from Heron Island, Great Barrier Reef, have revealed that this sponge is characterised by the presence of a dinoflagellate symbiont and by nematocysts. The dinoflagellates are morphologically similar to *Symbiodinium microadriaticum*, the common intracellular zooxanthellar symbiont of corals. The major sponge cell types found in *Haliclona* n. sp. are spongocytes, choanocytes and archaeocytes; groups of dinoflagellates are enclosed within large vacuoles in the archaeocytes. The sponge grows on coral substrates, from which it may acquire the nematocysts, and shows features such as mucus production which are typical of excavating sponges. The cytotoxic alkaloids haliclonacyclamines A and B associated with *Haliclona* n. sp. are shown by Percoll density gradient fractionation to be localised within the sponge cells rather than the algal symbionts. The ability to synthesise bioactive compounds such as the haliclonacyclamines may help *Haliclona* n. sp. preserve its remarkable ecological niche. An investigation into the effect of the haliclonacyclamines has indicated that these compounds cause coral tissue necrosis at concentrations of 5 ppm within 24 hours.

Key words: *alkaloids, haliclonacyclaminesl,* Acropora, *dinoflagellates,* Symbiodinium microadriaticum, *Percoll density gradient fractionation, secondary metabolites,* Haliclona

EVIDENCE OF TRANSFER OF PHOTOSYNTHATE FROM A RED ALGAL MACROPHYTE TO ITS SYMBIOTIC SPONGE

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Symbiotic cyanobacteria are quite common in coral reef sponges providing much of the sponge's supply of carbon. There are also several sponge species with macroalgal symbionts. In these sponges, the role of the algae is unknown. One of these symbioses is that of the sponge, *Haliclona cymiformis* (Haplosclerida) and the red alga, *Ceratodictyon spongiosum* (Rhodymeniales) which is common in the shallow tropical waters of fringing reefs of the Indo-Pacific region. The sponge tissue comprises about one third of the dry weight of the association and grows over the external surface of the alga and between the algal branchlets. In the field, the alga is dark green to purple with thick branches of tightly anastomosed (fused) branchlets. However, in culture, the branchlets are red and thin and do not fuse. Neither symbiont has been found growing separately in nature suggesting that the symbiosis is obligate. The physiological basis of this well integrated association is not yet known.

The sponge obtains nutrients from the water column in the form of dissolved and particulate organic matter at rates that are similar to those of free-living sponges (Trautman, 1997). We have found that some photosynthate is transferred from the alga to the sponge, in a time-dependent manner. After 1 h incubation in the light with $Na_2{}^{14}CO_3$ the amount of photosynthetically fixed carbon transferred to the sponge (range 22.77- 48.3 nmol carbon/mg dry wt. of sponge) represents 0.6-1.28% of the total carbon fixed by the alga during this period. When the fixed carbon in the sponge tissue is extracted using methanol/chloroform/water (24/10/4 v/v/v), to give an aqueous-soluble fraction (low molecular weight metabolites) and a chloroform-soluble fraction (lipids, sterols, chlorophyll etc.) followed by extraction in 2 M KOH (high molecular weight metabolites such as proteins, polynucleotides, polysaccharides) 75-88% of the ^{14}C -labelled carbon is found in the aqueous fraction, about 11-20% in the KOH-soluble fraction, 2-3% in the chloroform-soluble fraction and <3% in KOH-insoluble material.

When the aqueous-soluble fraction is further fractionated by ion exchange chromatography into neutral (sugars), basic (amino acids), acidic (organic acids) and phosphate ester fractions, most of the fixed carbon is found in the basic (47%) and neutral (38%) fractions. Some fixed carbon is found in organic acids (14%) with very little in phosphate esters (<2%). Our data suggest that while the alga may supply the sponge with some essential nutrients, the major source of organic carbon is the particulate and dissolved organic matter in the ambient seawater. It may be that the primary role of the algal symbiont is structural rather than nutritional.

Key words: *symbiosis, red alga, carbon metabolism, photosynthate, translocation*

ECOLOGICAL ADAPTIONS OF A FRESHWATER SPONGE ASSOCIATION IN A LARGE RIVER (PORIFERA, SPONGILLIDAE)

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In the years 1993-1995 the species composition and autecology of freshwater sponges (Porifera, Spongillidae) was investigated in the Rhine between Karlsruhe and Bonn (Germany). The species *Ephydatia fluviatilis, Ephydatia muelleri, Trochospongilla horrida, Spongilla lacustris, Eunaplus fragilis* and *Eunapius carteri* were found. *E. fluviatilis* was classified as r-strategist due to its high ability to colonise new habitats. All other species laid their emphasis in successful establishment in more stable habitats and should therefore be classified as K-strategists among freshwater sponges. Only in *E. fluviatilis* the production of larvae was an integral part of the life cycle, all other species put their

main efforts in producing gemmules. It is therefore discussed, whether the dominance of asexual reproduction vs. sexual reproduction is a feature of running-water habitats.

Key words: *Spongillidae, life cycle, adaptions, central Europe, river, running water*

PROPOSAL OF A PHYLOGENETIC CLASSIFICATION OF THE MYCALIDS WITH ANISOCHELAE, AND COMMENTS ON THE STATUS OF *NAVICULINA* GRAY, 1867.

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The phylogenetic relationships for the mycalids with anisochelae are revised. Several likely monophyletic assemblages, currently assigned subgeneric rank or lower are included, totalling 12 groups, with special reference to Gray's *Naviculina*. The type species of the latter assemblage, viz. *N. clifioni* (from SW Australia) is revised, and its recent allocation to the vicinity of *Arenochalina mirabilis* contested, being suggested a more likely affinity to *Aegogropila*, as currently understood. Its main anisochelae are here dubbed naviculichelae. A preliminary revision of over 230 published names for *Mycale* and allied taxa with anisochelae has been undertaken looking for *N. clifioni's* kinship, yielding four likely candidates, viz. *M. cleistochela* (from Madagascar and Indonesia), *M. diastrophochela* (from the Vema Seamount, SE Atlantic), *M. obscura* (from Indonesia and all over Australia), and *M. peculiaris* (from Papua New Guinea). A phylogeny is proposed for the mycalids with anisochelae, though not fully resolved, and alternative phylogenetic classification schemes are advanced, with a discussion on the pros and cons of each one.

Acknowledgement of financial support: CNPq, FAPERJ, FAPESP, FUJB-UFRJ.

Key words: *phylogenetic classification*, Mycale, Naviculina, *phylogeny, Linnaean hierarchy*

SPONGIVORY BY THE BRAZILIAN STARFISH *ECHINASTER BRASILIENSIS*

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The feeding ecology of *Echinaster brasiliensis* has been studied on a temporal gradient (January 1995 - September 1996, 11 observation periods), along a shallow-water transect parallel to the coastline (1.5-6 m depth, 2000 m^2) at Ponta do Baleeiro (23°49.727'S - 45°25.364'W), São Sebastião Channel (São Sebastião, SP, Brazil). In total, 3025 starfish were observed, 44% of which were feeding (1337/3025). Of these, 42% (557/1337) were feeding on sponges, a significantly higher proportion than the real availability of sponges in terms of area coverage by organisms. Of the 33 sponge species recognised, the most wanted prey was *Mycale* sp.n., representing 40% (221/557) of the total number of observed spongivory events. Other common sponge prey items were *Amphimedon* sp., *Haliclona* sp.n., *Mycale angulosa, Mycale microsigmatosa* and *Tedania ignis*, with ca. 5% of the spongivory events each. Semiquantitative arbitrary estimations point toward these species' high abundance in the study area. Therefore, we cannot discard the possibility of a direct link between the sponge's abundance and apparent starfish preferences. Of the 33 sponge species eaten, at least 61% (20/33) belong to genera from which species were found (literature) to possess toxins, thus raising the question: 'What are these toxins good for?' The conspicuous habit of *Echinaster brasiliensis* suggests that it may be unpalatable to many potential predators, perhaps through the use of sequestered toxins of dietary origin. The temporal gradient studied did not reveal clear patterns, thus suggesting that inter-annual climatic oscillations may play an important role in shaping the starfish's feeding ecology.

Acknowledgement of financial support: CNPq, FAPERJ, FAPESP, FUJB-UFRJ.

Key words: *spongivory, southwestern Atlantic, Echinaster, Mycale, feeding ecology, chemical ecology*

ECOLOGICAL AND EVOLUTIONARY SIGNIFICANCE OF MORPHOLOGICAL VARIATION IN THE TROPICAL SPONGE *ANTHOSIGMELLA VARIANS*: NICHE PARTITIONING AND PREDATOR-INDUCED PHENOTYPIC PLASTICITY

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Anthosigmella varians (Duchassaing & Michelotti) is a common Caribbean demosponge for which two ecotypes have been described. Encrusting *A. varians* forma *incrustans* (hereafter referred to as *incrustans*) is conspicuous on fore and back reefs while branching *A. varians* forma *varians* (hereafter *varians*) is found in shallow, lagoonal areas. Wave energy has been hypothesised to be responsible for this distribution since *varians* is presumed to be unable to handle strong currents and the periodic heavy wave action that open reefs receive. In the Florida Keys, USA, *varians* is found close to shore on both bay and ocean sides of many islands while *incrustans* is found on reefs 5 miles offshore. During this study, a third morph (identified as *A. varians* forma *rigida*) was encountered on Floridian reefs in sympatry with *incrustans*. The *rigida* form has lobate branches similar to *varians* but a much stiffer skeleton due to a thick spicule and collagen rich cortex; *rigida* individuals were often found less than a meter from *incrustans* individuals.

I conducted a series of field experiments with each morph to examine ecological influences on phenotypic variation in *A. varians*. For experiments involving *incrustans*, caging of reef populations (i.e., removal of predator effects) had no effect on morphology. In addition, *incrustans* individuals transplanted into the low flow and predator free environment of the Florida Bay did not modify growth form. Experiments involving *varians*, however, demonstrated the importance of predators in the distribution and morphology of this sponge. Transplanted *varians* individuals that were not caged were immediately consumed by angelfish on the fore reef. Survivorship was high for caged individuals (~91%) and low (45%) for uncaged replicates. At the end of the experiment, surviving uncaged *varians* individuals lost all pre-transplantation height and were only present within calcareous blocks (since *A. varians* is a boring sponge). Artificial predation experiments involving *varians* resulted in increases in spicule concentration compared to unmolested controls. Injured areas were noticeably white and could be clearly demarcated from non-injured tissue. The *rigida* morph was discovered late in the study, and short term caging and transplant experiments did not provide clear evidence about whether wave energy or predation were responsible for the observed stiffness in this morph. However, both *varians* and *rigida* began to encrust when they were placed in low sediment habitats.

In addition to the field experiments, I performed a genetic analysis on the different morphotypes using allozyme electrophoresis. Fixed differences were found at two loci between encrusting (i.e., *incrustans*) and branching (i.e., *varians* and *rigida*) forms, but no differences were observed between either *varians* or *rigida*. Thus, it appears that *varians* and *rigida* are reproductively isolated from *incrustans* individuals. The evolutionary factors responsible for the divergence are unknown, however, sedimentation may be a factor restricting *incrustans* to the reef. It is not clear whether a branching morphology allowed *A. varians* to exploit high sediment environments, but data presented here indicate that predators restrict *varians* individuals to shallow, lagoonal habitats. The branching form may not have been able to colonise open reef environments until the evolution of a predator-induced defense in the form of increased cortex thickness (e.g., collagen production). In any case, my data indicates that predation and sedimentation are primary factors responsible for the adaptive morphological variation observed in *A. varians*.

Key words: *phenotypic plasticity, adaptation, speciation, predator-induced defense niche diversification, allozyme electrophoresis, reciprocal transplant, caging experiments*

NITROGEN FLUX IN A SPONGE - MACROALGAL SYMBIOSIS

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Nutrient cycling between corals and their zooxanthellal symbionts is important to the conservation of limiting nutrients, such as nitrogen, in oligotrophic reef waters. The tropical reef sponge *Haliclona cymiformis* forms an intercellular symbiosis with the red macroalga *Ceratodictyon spongiosum*, but it is unknown whether this association also promotes the cycling and conservation of essential nutrients. We therefore determined the potential importance of ammonium excreted by the sponge to the nitrogen-status and growth of the macroalga, using specimens collected from One Tree Island Lagoon on the Great Barrier Reef.

The association possessed the capacity to take up ammonium, nitrate and perhaps nitrite from the ambient seawater in the light. However these nutrients were commonly present at concentrations of less than 3 μM in the water at One Tree Island and so this seawater was probably only a minor source of nitrogen for the macroalga. In contrast, when the association was pre-incubated in darkness for 24 hours and its dark ammonium excretion rate measured, ammonium levels in the surrounding seawater (1 litre) increased from 0.25±0.1 μmol/g dry weight to 2.2±0.6 μmol/g dry weight over a 6-hour period; shorter dark pre-incubations resulted in lesser rates of ammonium excretion. It therefore appears that sponge waste is a major source of inorganic nitrogen for *C. spongiosum*, and this will be illustrated by means of a preliminary nitrogen budget. However, the enhancement of dark carbon fixation by ammonium (20 μM NH_4Cl), which increases as algae become more nitrogen limited, suggested that *C. spongiosum* was still nitrogen-limited in One Tree Island Lagoon. The ammonium enhancement ratio of freshly-collected *C. spongiosum* was 1.4 ± 0.06, which compared to a ratio of 1.5 ± 0.07 for cultured *C. spongiosum* when deprived of inorganic nitrogen for one week; the ratio ranged from 1.1 ± 0.1 for cultured *C. spongiosum* supplemented with a regular source of nitrogen (100 μM NH_4Cl) to 2.1 ± 0.6 for cultured *C. spongiosum* deprived of nitrogen for 6 weeks.

We therefore propose that the situation in the *Haliclona-Ceratodictyon* symbiosis is analogous to that in corals and other zooxanthellate invertebrates, with the animal partner being an important source of nitrogen for the alga. Furthermore, when combined with evidence for the translocation of nitrogenous compounds from the macroalga to the sponge (Grant *et al.*, in prep), it is evident that the cycling and conservation of nitrogen within the symbiosis may be an important factor in the success of this association in nutrient-poor habitats.

Key words: *sponge-macroalgal symbiosis, nitrogen flux, nitrogen conservation, nitrogen budget*

SPONGES OF THE LOW ISLES, GREAT BARRIER REEF: AN IMPORTANT SCIENTIFIC SITE, OR A CASE OF MISTAKEN IDENTITY ?

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Much of our early, reliable scientific knowledge on marine taxonomy, biological and other processes of coral reefs in general, and the Great Barrier Reef (GBR) in particular, comes from the 1928-29 GBR

Expedition based on the Low Isles. 106 species of sponges were collected from northern reefs of the GBR Expedition and described by Burton (1934), 36 from the Low Isles.

Burton concluded that the sponge fauna contained: "species characteristic of the Indo-Pacific" (38% of his species); many "common also to the coasts of Australia" (17%) "with a mixing of the Australian and Malayan sponge-faunas"; substantial cosmopolitanism (12%) with species "also found in the West Indies, Azores and Mediterranean"; and only few indigenous species (14% unique to the Low Isles, 19% exclusive to N. Australia).

Re-examination of BMNH voucher and type material found 30% of species of these species were misidentified, mainly concerning the so-called 'widely distributed' taxa. Recent collections from the Low Isles by the Queensland Museum discovered 109 species, and together with the revised Burton collection indicate a sponge fauna of 134 species (in 63 genera and 35 families). Surprisingly only 12 species (9% of the Low Isles fauna) were common to both the Burton and our collections.

Taxonomic comparisons with other provinces show several major trends for Low Isles sponges: 1) The fauna contains a generalist element comprising 'typical GBR species', found on virtually all reefs surveyed so far (23% of Low Isles species). 2) The fauna also contains an indigenous component of species unique to the northern GBR (48% of Low Isles species), with 32% of these not yet recorded from anywhere else, and another 16% known only from both the Low Isles and Lizard Island (200km to the north). 3) Affinities with coastal faunas are low, contrary to Burton's hypothesis, with only 13% of Low Isles species also found on adjacent coastal regions. 4) Affinities with oceanic coral reef species are also low, with only 10% of Low Isles species found on the Coral Sea seamounts. 5) The concept of an 'east Australian coast' sponge fauna is not supported, contrary to both Lendenfeld (1888, 1889) and Burton, with only 10% of Low Isles species extending southwards into more temperate Queensland waters, and only 2% extending further into southern New South Wales. 6) The concept of 'cosmopolitan' species is unsubstantiated.

Keywords: *Low Isles, Great Barrier Reef, biodiversity, biogeography, taxonomy.*

BIODIVERSITY, SPECIES COMPOSITION AND DISTRIBUTION OF MARINE SPONGES IN NORTHEASTERN AUSTRALIA.

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Biodiversity, species composition and biogeographic relationships were compared between 18 regional populations of marine sponges along the NE. Australian coastline (extending from the Sydney region to the eastern Gulf of Carpentaria), based exclusively on samples of living populations. Much of the older literature concerning Australian sponge taxonomy is too unreliable to be used effectively as a tool to determine conspecificity and explore faunistic relationships (Hooper & Wiedenmayer, 1994), and consequently this literature was ignored completely in our analyses. Levels of biodiversity varied considerably between many regions, related in part to the size and diversity of habitats present in particular regions, but also to differences in collection effort. Several regions with apparently low sponge diversity (e.g. 3 seamounts in the Coral Sea) were clearly biased by differential collection efforts, whereas in other regions these biodiversity data appear to be more realistic indicators of species richness. Faunas of the Gulf of Carpentaria and Turtle Islands were more intensively sampled but had relatively low sponge diversity, whereas those of the Swain Reefs, Capricorn-Bunker Group, Lizard Island and Moreton Bay regions had much higher species diversity with equivalent (and sometimes lower) collection effort. Five of the seven relatively highly diverse regions lay in the south (Swain Reefs, Capricorn-Bunker Group, Moreton Bay, Sydney-Illawarra, Sunshine Coast), with only two northern regions showing comparable diversity (Lizard Island, Low Isles), contrary to latitudinal trends in diversity found in some other marine phyla. Statistically these trends do not appear to be artifacts of

sampling effort but reflect true differences in provincial diversity. The number of unique ('apparently endemic') species within each of the 18 regions had a median value of about 33%, although this value varied considerably between particular regional faunas. 'Endemism' was seen to be largely a function of their biogeographic isolation or proximity to other regional faunas, and to ecological factors such as the possession of unique habitat types. Regions with highest levels of relative 'endemism' were Sydney-Illawarra (the most southern region; 81% of species), Wreck Reef (the most isolated oceanic region; 46%), and the Gulf of Carpentaria (differing substantially from all other regions in its habitat composition; 45%). Consistent discovery of about 33% of 'new species' from each reef system we surveyed suggests that the possible sponge biodiversity in NE. Australia greatly exceeds previous estimates of about 1500 species. Within NE. Australia (ignoring the southern NSW outgroup), 5 provincial faunas are presently recognised, grouped heirarchically based on parsimony analysis, each showing greater similarities in species composition within-regions, and fewer similarities between-regions: 1) Tweed River (Byron Bay to the Gold Coast); 2) SE. QLD (Moreton Bay to Hervey Bay; 3) GBR (Capricorn-Bunker Group to the Cockburn Is); 4) Far N. coastal reef and islands, extending into the Gulf of Carpentaria (although the latter is probably a valid province in itself); and 5) Coral Sea (although not yet substantially surveyed). The concept of a homogeneous 'east Australian coastal' fauna is rejected (cf. Lendenfeld, 1888, 1889), and the possibility that both the GBR and Coral Sea regions comprise more than single provinces (cf. Burton, 1934) requires further investigation.

Keywords: Biodiversity, biogeography, faunistics, eastern Australia, Great Barrier Reef, Coral Sea.

REGULATORY MECHANISMS OF IMMUNE CELLS IN SPONGES

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Gray cells, large granular wandering cells present throughout the tissues of many species of sponges, have been identified as immunocytes in two species of sponges, *Microciona prolifera* and *Callyspongia diffusa*. When the tissues of two different sponge individuals are apposed, the gray cells accumulate at the boundary of contact at the time of tissue rejection. I have suggested that these cells may be viewed as the most primordial examples of evolutionary predecessors of the well-known vertebrate lymphocytes. This comparison implies that gray cells share features of vertebrate lymphocytes and I have examined this idea with studies on two prominent aspects of activation of T and B cells. The primary signalling event upon activation of a lymphocyte by recognition of an appropriate immune target is the synthesis and release of cytokines that alert and coordinate the activity of other lymphocytes in the surrounding tissue and throughout the body. In addition the activation of lymphocytes involves internal second messenger pathways converging on the transcription factor, NFkB, that are inhibited by Cyclosporin A, a drug often used medically to prevent rejection in human transplants. Using Boyden Chamber assays, the assays originally used to identify vertebrate immune system cytokines, I have succeeded in establishing in *M. prolifera* that contact with foreign tissue stimulates the release of cytokines activating the migration of gray cells toward the contacting tissue. Similarly, doses of Cyclosporin A commonly used to inhibit the activation of vertebrate T cells, suppresses histoincompatibility in *M. prolifera* and allows the healing together of tissue from two individual sponges that would normally undergo tissue rejection. These results provide further evidence that the foundations of the cellular immune system of animals were already established in the sponges and that study of gray cells will provide insight into the course of evolution of animal immunity.

Key words: *immunology, immunocytes, gray cells*

NEGOMBATA MAGNIFICA - A MAGNIFICENT (CHEMICAL) PET

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Negombata magnifica (Latrunculia) is a Red Sea sponge known to produce the toxin latrunculin (Lat). Since synthesis of this compound is economically non-viable, we evaluated various ways of producing it, while determining its natural mechanism of production and ecological relevance. We examined the possibility of: 1. identifying the cells which produce and harbour latrunculin; 2. establishing cell cultures; 3. forming an underwater sponge "garden"; and 4. taking advantage of the sponge's own reproduction and new larval settlement. Early in the study it became evident that *Negombata magnifica* might actually comprise two closely related species of *Negombata*, one of them an undescribed new species. The work reported here refers to the original *Negombata magnifica*.

1) The location of Lat B, was studied using specific rabbit anti-Lat B antibodies. Rabbits were immunised with a conjugate of Lat B with Keyhole Limpet Hemocyanin (KLH) and the antibodies were affinity purified over a Lat B-Sepharose column. Thick and thin sections of the sponge were analysed by immunohistochemical and immuno-gold techniques using light and transmission electron microscopy, respectively. Latrunculin B was prominently labelled in the sponge ectosome, especially in the dense cell layer beneath the cortex. Immuno-gold localisation within the sponge revealed that Lat B resides in the sponge cells and not in its prokaryotic symbionts. The labelling density of gold particles in the archeocytes and choanocytes was significantly higher than that of the other sponge cell types (special cells and skeleton associated cells). The antibodies labelled primarily archeocytes and choanocytes, membrane-limited inclusions which are perhaps Lat B secretory and/or storage vesicles. The concentration of Lat B in the sponge periphery correlates with the defensive role of the toxin, since encounters with epibionts, predators and competitive neighbours take place through the ectosome. It may, therefore, be useful to isolate these cells for culture. 2) Primary cell cultures were established from adults and embryos. Mechanical dissociation of inner parts (without the external layer) proved to be superior (less contamination and more cell types) to other techniques. Primary cultures from embryos lasted significantly longer (up to 280 days) and cells survived a freezing phase. Cell lines, however, have not yet been established. 3) Initial steps were taken toward establishing an *in situ* "garden" of *N. magnifica* from sponge fragments. Although growth rate of sponge fragments was superior to that of natural sponges in their vicinity, fragment survival over a year proved to depend on sponge handling, water depth and environmental conditions (currents, sedimentation etc.). 4) *Negombata magnifica* had a peak in sexual reproduction during the summer. Sexually produced, naturally released, larvae were settled on plates and their growth and development were followed for up to 4 months.

Key words: *latrunculin, natural product, localization, antibodies, sponge reproduction, immunohistochemical and immuno-gold techniques, cell culture*

MOLECULAR PHYLOGENY OF FRESHWATER SPONGES (FAMILIES LUBOMIRSKIIDAE AND SPONGILLIDAE) AND THEIR RELATIONSHIPS WITH SEA SPONGES.

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Two families belonging to phylum Porifera are represented in Lake Baikal:cosmopolitan Spongillidae and endemic Lubomirskiidae. The endemic family of Baikal sponges, Lubomirskiidae, is not well known. Previous results of morphological and embryological methods indicate that Lubomirskiidae and Spongillidae are closely related, but the relationship is unclear. Indeed, there little agreement on the

origin of freshwater sponges on the whole. These groups are considered to be polyphyletic. We used molecular methods with the aim to make clear the relationships within the family Lubomirskiidae and to determine the closest relatives of the Spongillidae and Lubomirskiidae among sea sponges. We had obtained partial sequences of 18S rRNA for some sponges species belonging to the families Lubomirskiidae, Spongillidae and Halichohdriidae and compared them with available sequences of 18S rRNA of other Porifera. As sponges harbour a large number of symbionts, sponge specific primers were designed and synthesized.

We have obtained partial sequences (630 bases) of 18S rRNA of 6 species of sponges. These sequences have high homology with each other. *H. japonica* was found to be significantly different, while only a single substitution was observed in Lubomirskiidae and Spongillidae. Hence, we observed that 18S rRNA has an insufficient sequence variability for statistical confirmation of relationships between closely related species of freshwater sponges, but it is sufficient to attempt to establish their ancestors among sea sponges.

Parsymony and neighbour-joining analyses give trees with similar topology. Bootstrap values show that all clusters are statistically significant. Topology of the tree indicates that Ceractinomorpha and Demospongiae form two different monophyletic groups. The results conform with morphological systematics and with the conclusion obtained by analyses of 28S rRNA (B.Lafay et.al.). According to our tree, Ceractinomorpha is molyphyletic, *L.abietina* and *S.lacustres* are placed in the same clade, confirming that Lubomirskiidae are likely to be closely related. The marine groups closely related belong to the families Halichondriidae and Axinellidae, as they emerged as sister groups.

Key words: *Lake Baikal, Spongillidae, Lubomirdkiidae, 18S rRNA, Phylogeny, Freshwater sponges*

NEW DATA ABOUT MORPHOLOGY AND FEEDING PATTERNS OF BARENTZ SEA *HALICHONDRIA PANICEA* (PALLAS)

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Visual observations in the marine aquaria and transmission electron microscopy studies on the larvae of the intertidal sponge *Halichondria panicea* demonstrated individual variations in external and internal morphology, behaviour and type of metamorphosis. Parenchymulae of this species were found to possess the ability to actively feed by endocytosis (phago- and pinocytosys). The larvae crawled over the substrate and cast numerous unicellular organisms (bacteria and flagellates from 2 to 4μm in size) onto the body surface by a flagellum. During this, the apical parts of the flagellated cells formed large lobopodia that served for catching and ingesting food particles. I monitored the consequent patterns of contact of the flagellates with the surface of lobopodia, their entrapment, submersion, the formation and transport of the digestive phagosomes into the basal parts of the surface cells. Each surface locomotory cell was capable of catching and ingesting food. No morphological and/or functional differences between the surface cells were found. Nevertheless, singular flagellated cells packed with the phagosomes submerged inside the larva. Here these cells could be easily distinguished by the presence of a flagellum and the typical shape of the nucleus. Later on, the submerged flagellated cells withdrew the flagellum and acquired an amoeboid shape. Final digestion of the caught organisms occurred only inside the larva. It was suggested that endosymbionts found in the surface and inner cells of the larvae served as an additional food source for the larvae. Presence of the numerous pinocytosis vacuoles in the apical parts of the flagellated cells suggested that the sponge larvae are also able to absorb dissolved low-molecular matter.

To conclude, parenchymula of *H. panicea* could be recognised as a living embodiment (a living model) of the hypothetical phagocytella of Mechnikov in which the differentiation of the body layers into kinoblast and phagocytoblast is only primordial, purely functional and still reversible.

Key words: *intertidal, larva, feeding, digestion, endocytosis, digestive phagosomes*

REGENERATION ABILITIES OF THE SPONGILLID LARVAE

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Free-swimming larvae of *Spongilla lacustris* were cut into two halves with a razor blade under a binocular microscope. Two samples of the larvae were used. In the first sample, larvae were cut in the tangential plane (two equal halves with a similar set of cells and structures). In the second sample, larvae were cut in the transverse plane (two unequal parts). The 'anterior' fragment contained a large cavity lined with pinacocytes, the halved amount of the surface flagellated cells, and underlying collencytes. The 'posterior' part of the larva had a halved amount of the surface flagellated cells and all of the internal structures typical for the fully developed spongillid larvae. Each half of the larva was maintained in a separate Petri dish with the celloidine-covered bottom in well-aerated river water.

Visual observations and transmission electron microscopy yielded the following preliminary results. The halves of the larvae closed the edges of the wound immediately after dissection while continuing to move. However, trajectory, velocity and direction of the movements differed in different types of experimentally cut larvae. This was directly related to the presence or absence and the development of the larval cavity. Thirty minutes following the dissection, the tangentially split halves of the larvae looked normal (movement, attachment and metamorphosis generally similar), but half the size of the control larvae. In 2-3 days after the settlement these half-larvae metamorphosed into normally functioning small sponges. Developmental capabilities of the transverse halved larvae were different. The anterior half-larvae soon closed the cut edges, acquired a shape of a hollow sphere and swam easily and rapidly in the water. They maintained activity for two or more days, attached, formed pinacoderm and few flagellated chambers, and the sponges died. The posterior halves recovered the integrity of the flagellar cover in an hour following the dissection. They acquired the shape of spheres tightly packed with cells, covered with slightly elongated flagellated cells, and swam heavily and slowly near the bottom, with a maximum free life of 18 hours. After the settlement and attachment they formed pupae covered with pinacoderm and within 2 days developed into normal sponges.

Transmission electron microscopy showed the surface flagellated cells played an important role. These cells provided restoration of the surface cell cover, however, their role greatly differed in development of the 'anterior' and 'posterior' half-larvae. In the 'anterior' halves, the flagellated cells migrated inside; some were ingested by underlying collencytes (phagocytosis); some transformed into choanocytes giving rise to few flagellated chambers. During development of the 'posterior' half- larvae, some surface flagellated cells transformed into the pinacocyte-like cells *in situ* and still retained flagella for a long time. The leading role in the transformation of the flagellated cells belonged to the centrioles (both flagellated and flagellum-less) and to the root structures of flagellum connected with the centrioles.

Collencytes played the important role in the attachment and development of the settled half-larvae. These cells actively migrated to the surface of the settled larva, phagocyted the cells damaged during dissection, secreted a large amount of collagen and contributed to the flattening of the half-larvae and their attachment to the substrate. The post- settlement fate of flagellated surface cells of the half-larvae was partially dependent on the amount and the activity of collencytes. The next major morphogenetic role belonged to archaeocytes, the main source for the formation of choanoblasts, spiculocytes, collencytes, pinacocytes and other cells. The archaeocytes mitotically divided several times, losing their storage inclusions, and thus gave rise to several differentiated cell lineages. Probably, the lack of the necessary amount of the cells is responsible for the developmental retardation of the settled 'anterior' half-larvae.

Key words: *Spongillidae, larva, metamorphosis, development, ontogeny, transmission electron microscopy*

THREE NEW SCALARANE BASED SESTERTERPENES FROM THE TROPICAL MARINE SPONGE *CARTERIOSPONGIA CALCIFORMIS*

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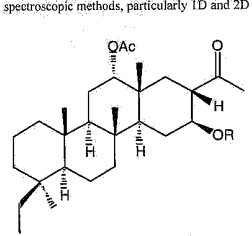
From the dichloromethane extract of the tropical marine sponge *Carteriospongia calciformis* (Carter, 1885), collected from The Great Barrier Reef Australia, three new (**1**,**2** and **9**), and seven known (**3**-**8** and **10**) scalarane based sesterterpenes were isolated. All structures were secured by employing spectroscopic methods, particularly 1D and 2D NMR, and accurate mass measurement.



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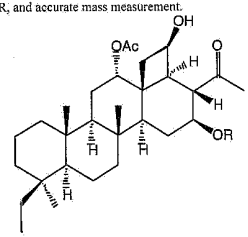

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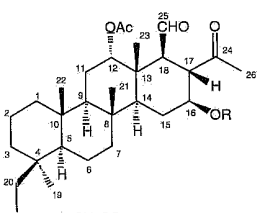
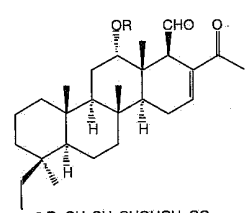


3 R=CH_3CO-
4 R=$CH_3CHOHCH_2CO$-
5 R=$CH_3CH_2CHOHCH_2CO$-



6 R=$CH_3CH_2CHOHCH_2CO$-
7 R=$CH_3CH(OCOCH_3)CH_2CO$-
8 R=$CH_3CH_2CH(OCOCH_3)CH_2CO$-
9 R=$CH_3CH(OCOCH_2CH_3)CH_2CO$-
10 R=$CH_3CH_2CH(OCOCH_2CH_3)CH_2CO$-

Key words: *sesterterpenes*, Carteriospongia calciformis, *spectroscopic analysis, novel compounds*

EFFECTS OF PHOTOSYNTHETIC ACTIVITY IN ENDOSYMBIOTIC ZOOCHLORELLAE ON GEMMULE GERMINATION OF A FRESHWATER SPONGE, *RADIOSPONGILLA CEREBELLATA*.

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Many of freshwater sponges thrive in oligotrophic clear waters in cooperation with photosynthetic endosymbionts, such as zoochlorellae in their mesenchymal cells. They also withstand unfavourable winter season in dormant forms as gemmules. Annandale sponge *Radiospongilla cerebellata* is a green freshwater demospongiae with zoochlorellae in their archaeocytes and flourishes only in warmer season in southern district of Japan. Gemmules of the Annandale sponge also contain zoochlorellae in thesocytes and germinate only under illumination even if all other conditions are properly provided. Although the light sufficient to induce the germination was very low in intensity and extremely short in illuminating period, photosynthesis seems to be essential for the germination, because a photosynthetic inhibitor, atrazine, strongly inhibited the germination under optimal condition.

Since gemmules of the Annandale sponge contain a rich storage of nutrients in the thesocytes, photosynthetic nutrients, produced during the incubating period under very low intensity and short length of illumination, seem to have little effect on the induction of the gemmule germination. We undertook to observe the effects of other factors on gemmule germination, that is, gaseous components such as oxygen evolved and carbon dioxide consumed by photosynthesis. To accomplish gas experiments, we devised a glass slide with a hollow chamber of 3 cm^3 sealed with a glass plate. In gas experiments, gemmules were placed in the chamber with M-medium (previously boiled to eliminate dissolved gaseous components). The chamber was tightly sealed with a glass plate and the desired amount of gas was introduced to the medium as a bubble under the glass plate. Gemmules were illuminated at 3000 lx through the glass plate by ordinal fluorescent tubes for 10hrs daily and kept at 24°C for 8 days. When gemmules were incubated in degassed medium that was tightly sealed and isolated from the atmospheric gases, no gemmules were germinated. When an air bubble was introduced to the incubating chamber by one tenth or more the volume of the incubation medium 100% germination was achieved.

Of the major elements of air, only oxygen induced germination efficiently, and brought full germination at much less quantity than the air (about one hundredth of the incubating medium in volume). On the other hand, nitrogen and carbon dioxide, showed no effects on the gemmule germination under the optimal condition. On the contrary, carbon dioxide showed strong inhibition of gemmule germination in the oxygenated or aerated media. These results show that gemmules of the Annandale sponge are induced to germinate by cytoplasmic oxygen concentration under the favourable condition, but can be substantially suppressed by carbon dioxide. Thus, it is regarded that light applied to the gemmules would initially promote photosynthesis in the symbiotic chlorellae in the thesocytes, which would absorb cytoplasmic carbon dioxide and block the initiation and/or progression of gemmule germination, and evolve oxygen that promotes gemmules to germinate. To confirm this assumption, we have tried to induce germination in darkness with fully oxygenated and carbon dioxide free media. However, no gemmules have germinated in total darkness, in spite of other optimal conditions.

The failure of germination in darkness can be understood as follows: when optimal temperature and oxygen were provided to the gemmule cells, (which had been dormant under the cold temperature), they seemed to rouse and begin to respire, and generate carbon dioxide. The carbon dioxide could not be eliminated from the cytoplasm readily, due to the lack of photosynthesis under darkness, so its accumulation in the thesocytes appears to inhibit germination of the gemmules.

Key words: *freshwater sponge, gemmule, germination, symbionts, photosynthesis, O2, CO2*

EXPERIMENTAL OPTIMISATION OF GROWTH RATE AND MORPHOLOGY OF THE CULTURED BATH SPONGE *COSCINODERMA MATHEWSI* LENDENFELD IN POHNPEI, MICRONESIA

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It is now generally agreed that the cultivation of natural bath-sponges is an important alternative to the harvesting of wild sponge stocks. However, consistency of quality, sustainability of production and a reliable supply of farmed sponges to markets, has been a major barrier to the commercial success of previous cultivation initiatives. Although several sponge species of commercial quality can be found throughout Micronesia and the Central west Pacific, the people of Pohnpei Island are the only Pacific Islanders known to have harvested bath-sponges for traditional domestic use. Recently an increasing perception by local people that this highly valued marine resource was threatened by over harvest has lead to the development of simple and inexpensive cultivation techniques. This has now become the focus of a major research effort to develop an environmentally and economically sustainable cottage industry for locals that requires low labour and material investments. Cultured bath-sponges will be released into the European markets in 1999.

At present, research emphasis is focused on optimisation of the growth rate to commercial size, and the final shape of the cultured sponge. Towards this end 3 long-term experiments are being carried out to determine 1) the nature of growth-rate variability in wild sponges and the explant cuttings that are derived from them, and 2) the role of the position of excision of the explant from the donor sponge on growth rate, and 3) the biological basis of morphology variability in the wild and cultured sponge.

Experiment 1 - The first experiment attempted to determine whether significant differences existed in the growth rates of cuttings that had previously shown high (mean >120%) and low (mean <60%) rates of annual increase. Of particular interest was the high growth rate displayed by some cuttings and whether this could be maintained through subsequent generations. All "high growth" sponges retained their high growth rate, and all "low growth" rate sponges retained low-growth rate. Thus, high growth rate sponges can be reliably used to seed new farms for optimal grow-out rates and productivity.

Experiment 2 - Extensive field work by 2 of the authors indicated the presence of four morphotypes of *Coscinoderma mathewsi*, only two of which are optimal for the domestic market. Early cultivation attempts did not control for morphology and so many sponges were not acceptable commercially. Early cultivation revealed that cuttings with pinacoderm intact, were more likely to survive, and thus should be planted in preference to cuttings which lacked pinacoderm (eg. interior parts of large sponges). A second experiment was designed to examine the effects of position within the seed-stock sponge from which the cuttings were derived (inside, outside, upper, middle, lower) and the morphology of the seed-stock sponge (vasiform, digitate, ball-shaped, and ring-shaped) on the growth rates of the cutting and its final morphology.

Of the four parameters tested within the second experiment (variability of sponge, seed-stock morphotype, cutting level within the sponge, excision position of cutting), only *position of excision* showed significant differences. Growth rates of inside and outside cuttings were significantly different within several weeks and remained so for the duration of the experiment (24 months). Results on the effect of the "starting seed-stock" morphology on the cultured sponge morphology is not available until harvest of the experiment in 1999.

A third long-term experiment is presently ongoing - to try to alter the final morphology through altering the physical environment in which the cuttings are grown.

Key words: *sponge mariculture,* Coscinoderma mathewsi, *growth rate, morphology, optimisation, Pohnpei, Micronesia, commercial farming*

MORPHOLOGY AND MOLECULES IN LITHISTID TAXONOMY: NEW SOLUTIONS FOR OLD PROBLEMS.

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Most lithistid sponges lack an adequate range of taxonomic characters for differentiation, and in most genera these characters are extremely plastic. As a consequence of this, the generation of morphological hypotheses comparison with molecular phylogenies is nearly impossible due to the absence of reliable synapomorphies. Historically, lithistids have been grouped together in a single order on the basis of common possession of an interlocking siliceous skeleton. Recent morphological and palaeontological data indicate, however, that lithistid sponges are polyphyletic; several genera possess skeletal characters that suggest affinity with non-lithistid demosponges. We have found that in many cases these characters are probably non-homologous and misleading.

Ongoing research on the phylogeny of lithistid sponges has revealed some interesting "anomalies" of identification. In this talk I will highlight three examples within the lithistid families Theonellidae and Corallistidae where 28S rDNA analyses indicate unexpected phylogenetic affinities; the nearest relatives of de Laubenfel's (1954) "*Plakinalopha" mirabilis* are *Theonella* spp., *Theonella atlantica* is more closely related to *Corallistes* spp than *Theonella* spp, and *Theonella tubulata* van Soest is more closely related to *Macandrewia azorica* in the Corallistidae.

What is to be done in this situation? To what extent can molecular hypotheses be accepted over morphological hypotheses or vice versa? We have found that rather than having to "accept" one over the other, which often goes against *instinctual phylogeny*, molecular data requires us to re-examine these problems by reciprocal illumination, through the generation of higher quality morphological research and the examination of characters that are often, not at first, obvious. With this group of lithistid sponges, triaene rhabd and clade morphology, microsclere ornamentation, and the patterns of desma zygoses, and shaft ornamentation become crucially important in the differentiation of taxa.

Thus, for this particular group of organisms we have found that morphological hypotheses between closely related taxa are often strongly informative and can lend crucial evidence for the acceptance of certain molecular phylogenies over others. Molecular data can clearly indicate relationships between organisms where morphological data had previously failed, and molecular data often requires us to re-examine morphological characters from new perspectives, leading the discovery of new taxonomic discriminators.

Key words: *Phylogeny, 28S rDNA, morphology, congruence, lithistid, Theonellidae, Corallistidae*

THE REPLACEMENT OF NATURAL HARD SUBSTRATA BY ARTIFICIAL SUBSTRATA: ITS EFFECTS ON SPONGES AND ASCIDIANS.

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Subtidal reefs around coastal cities such as Sydney are composed of a variety of natural and artificial substrata. Commonly these are natural rocky reefs, breakwalls, seawalls and pier pilings. These types of hard substrata differ in their structure. Most natural hard substrata consist of horizontal surfaces; most surfaces on artificial hard substrata are vertical. Therefore, replacing natural hard substrata with artificial hard substrata is likely to change the surface of substrata from predominantly horizontal to mostly vertical. To understand and predict the potential effects of these changes on the assemblage of sponges and ascidians it is important to determine their distribution on horizontal and vertical surfaces.

The few ecological studies on the distribution of algae and invertebrates on horizontal and vertical surfaces have reported that there are more sponges and ascidians on vertical than on horizontal surfaces. It has not been tested whether these patterns exist in the temperate waters around Sydney. Furthermore, of the studies that have examined the effects of horizontal and vertical surfaces on the distribution of sponges and ascidians, none has experimentally tested the factors that cause these distributions.

Here, I present results of my tests of the hypothesis that sponges and ascidians are more abundant on vertical than horizontal surfaces in the shallow subtidal zone around Sydney. I will also discuss future manipulative experiments to determine which factors are important in creating these distributions.

Key words: *Ascidiacea, distribution, hard substrata, shallow subtidal, habitat*

CONVERGENCE IN THE TIME-SPACE CONTINUUM: A PREDATOR-PREY INTERACTION

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Community structure is influenced by many biotic and abiotic factors. Predation is a key structuring mechanism for some marine communities. Prey abundances may fluctuate with strength of predator recruitment and persistence, except in cases where some of the prey population has a refuge in space or time from predation. Consistent, moderate predation levels on a predicably available prey resource should lead to stable community structure with relatively small fluctuations in predator and prey population densities. Conversely, prey species lacking a refuge from predation are subject to major population fluctuations commensurate with strength of predator recruitment and abundance.

The sponge *Halichondria panicea* is patchily distributed in the rocky intertidal on the south shore of Kachemak Bay, southcentral Alaska, and in certain locations is the spatial dominant. At one site approximately 55m in horizontal length, *H. panicea* has dominated the mid-intertidal for at least 10 years, with low densities of potential molluscan predators such as *Archidoris montereyensis, Katherina tunicata*, and *Diadora aspera* present. Percent cover estimates of primary space occupiers at the site were collected from 10 0.25m^2 permanent quadrats established in August 1994. *H. panicea* averaged 53.4% +/-9.9% cover through August 1996. Other major cover categories were algae, 14.6% +/-6.4%, and open rock, 26.1% +/-10.2%. Visits to the site in early spring of 1997 revealed that the sponge colonies overwintered with few indications of major mortality events. No percent cover data were collected at that time.

Total numbers of the nudibranch *Archidoris montereyensis*, which is a specialist predator on *H. panicea*, present at the site were recorded and ranged from 12 to 42 from 1994-1996. In the spring of 1997, strong recruitment resulted in an average population of 151 *A. montereyensis* on site from May to July. Percent cover of *H. panicea* declined from visual estimates of 40% in May to 15% in July. By August 1997, when the 10 permanent quadrats and 10 haphazardly placed quadrats were measured, essentially no sponge could be found at the study site. After July, the abundance of nudibranchs declined to 32 individuals commensurate with sponge reduction. By September, only one small sponge colony and 7 predatory nudibranchs were present at the site. Even though *H. panicea* is abundant in the region and potential recruits should be numerous, as of April 1998, the site once dominated by *H. panicea* is predominantly open rock with some recruitment of annual macroalgae occurring. The predator-prey relationship of *A. montereyensis* and *H. panicea* is an example of a chase through space and time with convergence resulting in extreme population fluctuations and an unstable community.

Key words: *predation, nudibranch, intertidal, predator/prey interaction, community structure, Alaska, recruitment,*

REMARKS ON THE PALEOECOLOGY AND REEF BUILDING POTENTIAL OF LATE JURASSIC SILICEOUS SPONGES

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In the early Late Jurassic (Oxfordian) siliceous sponges developed extensively. They formed a discontinous siliceous sponge reef belt extending over more than 7000 km from New Foundland, Iberia, France, Switzerland, Germany, Poland, Romania to the Caucasian Mountains.

Siliceous sponges are no systematic unit but belong to the different taxonomic groups Hexactinellida and the polyphyletic lithistid demosponges. Due to their different organisation and biology, the ecological demands of the different siliceous sponges groups differ remarkably. The two major groups must be carefully distinguished for paleoenvironmental interpretations.

In general, lithistid demosponges are active filter feeding organisms. They feed on nannoplancton mainly bacteria. The bathymetric distribution of demosponges corresponds to a great extent with the bathymetric distribution of bacteria. The fairly high preservation potential of rigid demosponges is explained by a high amount of mesohyl-dwelling bacteria, causing rapid calcification after death.

Osmotrophy is an important feeding strategy of Hexactinellida. Dissolved organic matter is enriched in deeper water low-energy settings, causing the majority of Hexactinellida to dwell in such habitats. As the mesohyl of Hexactinellida consists of very thin collagenous material, there is hardly any room to harbour bacteria. This easily explains why microbially induced post-mortem calcification of the sponge by microbial autmicrites occurs at a much lower rate so that fossilisation potential is much lower in comparison with rigid demosponges.

The taxonomic composition of fossil siliceous sponge populations is mainly controlled by sedimentation rate, nutrition and hydrodynamics. The dominance of major taxa is strongly influenced by bathymetry, due to changes of hydrodynamics and nutrition along a bathymetrical gradient. However, the quality of substrates, water energy or extreme oligotrophy may strongly modulate bathymetric distribution.

Key words: *Late Jurassic, Hexactinellida, lithistid demosponges, paleoecology, calcification, fossilisation*

DISTINCTIVE MIDDLE CAMBRIAN SPONGE-CALCIMICROBE REEFS IN IRAN

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Following the virtual demise of archaeocyaths in the Toyonian stage and consequent collapse of the Early Cambrian archaeocyath-calcimicrobe reef consortium, Middle and Late Cambrian reefs remained generally devoid of metazoan input, being almost entirely microbial. The few exceptions in this interval generally include some minor contribution by spiculate sponges. One such spiculate sponge-calcimicrobe reef system in the Middle Cambrian of northern Iran is distinctive in that the spiculate sponges constitute a major component of the reef framework.

The reefs are in units 2 and 3 of the Mila Formation in the eastern Elburz (Alborz) Mountains. The Mila Formation consists of five units, together ranging in age from Middle Cambrian to Early Ordovician. Trilobites permit correlation of unit 2 and the reef-bearing lower unit 3 with the late Middle Cambrian, and upper unit 3 with the Chinese Kushanian (terminal Middle to earliest Late Cambrian) and

Changshanian (Late Cambrian) stages. The reefs are well exposed in a road section 3 km north of Shahmirzad.

The reefs are constructed by a consortium of the anthaspidellid sponge *Rankenella* and a presumed variety of microbes including the calcimicrobe *Girvanella*. *Rankenella* is otherwise known only from the Ordian-early Templetonian stage of the Northern Territory, Australia, where it is a level-bottom dweller. That stage is equivalent respectively to the late Toyonian-early Amgan and Longwangmiaoan-Maozhuangian stages of Siberia and China.

Unit 2 comprises fossiliferous interbeds of grey limestone/dolostone and yellow-brown marly shale, with desiccation cracks, bidirectional ripples and probable tempestites and hardgrounds. The stratigraphically lowest known appearance of *Rankenella* is in upper unit 2, in a single decimetre-thick limestone bed of abundant eocrinoid ossicles. Scattered, widely conical *Rankenella* are preserved upright in life position, suggesting attachment to a hardground. Sponges, ossicles, trilobites and hyoliths are encrusted by *Girvanella*, which also forms rafts and onkoids. Texture within this biostromal bed ranges from floatstone-rudstone to *Girvanella* boundstone, with evidence of microbial and oxea-bearing sponge-body automicrites.

The lower, reef-bearing portion of overlying unit 3 is massive, comprising pale grey stacked bioherms of similar texture and composition to the unit 2 *Rankenella* bed. In this interval, *Rankenella* adopts the entire range of co-occurring cup shapes from narrowly conical through to explanate. Clotted-peloidal biohermal mud is interpreted as automicrite. Substrate, peribiohermal and overlying sediment is commonly a bioclast rudstone rich in orthide brachiopod valves.

Sponges are contributors to bioconstruction in a reef tract toward the top of lower unit 3. Component bioherms of this reef tract are constructed by ramose *Rankenella* encrusted by thick coatings of *Girvanella* to form a *Rankenella-Girvanella* framestone with only minor lime mud pockets. Interstices are rimmed by one to two generations of columnar cement and occluded by coarse equant cement. By comparison with Early Cambrian reefs, *Rankenella* and *Girvanella* played the roles of archaeocyaths and calcimicrobes: framework/substrate and encrusting/binding respectively. In many Early Cambrian reefs, however, lime mud represents a much greater component, while calcimicrobes were capable of building massive framework unaided by metazoans.

Key words: *Middle Cambrian, Iran, calcimicrobe,* Rankenella, Girvanella, *reef, automicrite*

HOMEOBOX GENES EXPRESSED IN THE ADULT AND REAGGREGATING SPONGE

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Homeobox genes encode a large family of conserved transcription factors that control a range of important developmental decisions, and can be interspersed or organised in clusters (e.g. HOX genes) within metazoan genomes. A striking feature of many homeobox orthologues is how they are expressed in a similar fashion during the development of phylogenetically-disparate animals. In recent years, interest has developed in homeobox genes in the lower metazoans, such as platyhelminths, cnidarians, and sponges. Instead of studying embryological development, focus has often been on the role of homeobox genes during regeneration. Interestingly, these genes exhibit comparable expression patterns during the regeneration of lower metazoans as they do during normal embryological development of higher metazoans. In this study, homeobox genes expressed in the adult sponge (newly dissociated cells) *Iotrochota baculifera* (Demospongiae: Poecilosclerida: Myxillidae) and during reaggregation were identified by RT-PCR with degenerate primers and sequencing. As with regeneration in other taxa, investigating the molecular genetics of reaggregation in the sponge, apart from providing many practical advantages, gives us insight into the level of conservation between embryological and other developmental processes. To confirm the poriferan origin of the homeobox genes isolated, the identical procedure was applied to another sponge and some homologous genes were obtained. As sponges

appear to be monophyletic with the rest of the Metazoa and the first lineage to diverge during metazoan evolution, the study of their homeobox genes provides insight into the initial role of metazoan-specific homeobox genes in governing the multicellular state and cellular differentiation.

Key words: *reaggregation, development, homeobox genes, regeneration*

GENETIC CONFIRMATION OF THE SPECIFIC STATUS OF TWO SPONGES OF THE GENUS *CINACHYRELLA* IN THE SOUTHWEST ATLANTIC.

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(PRESENTED BY NICOLE BOURY-ESNAULT)

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Cinachyrella alloclada and *C. apion* can be readily distinguished in the Caribbean area by the presence of buds only in the former and by their spicule types and sizes. However, in Brazil both can reproduce by budding, and also have identical chemical profiles in lectins, fatty acids, and steroids. It was decided, thus, to verify whether *Cinachyrella alloclada* and *C. apion* were indeed different biological species or just morphotypes of a single phenotypically polymorphic species on the Brazilian coast. Samples were collected in the intertidal zone of Pituba beach, Salvador, Brazil, and studied independently by morphological and allozyme analyses. There was a high congruence between morphology and allozymes and eleven (out of 19) gene loci were diagnostic of each species. *C. apion* had smooth oxeas of only one size class, protriaenes of two sizes, anatriaenes of one size, small sigmaspires and raphides. *C. alloclada* presented smooth oxeas of two or three size classes, protriaenes and anatriaenes with one size, and sigmaspires with a similar size to those of *C. apion*. The unbiased genetic identity between the two species was very low (I=0.28) as often found among congeneric sponge species. The consistent morphological and genetic differences observed between the two putative species confirm that, in spite of their high chemical and reproductive similarity, they are distinct biological species. This indicates that, at least in these species, evolutionary rates for allozymes and secondary metabolites are clearly unrelated.

Key words: Cinachyrella, *allozymes, Southwest Atlantic, genetic identity, morphotypes*

DISTRIBUTION OF SPONGE SPECIES OFF THE NORTH JAMAICAN COAST

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Concerning the sponges, the Caribbean is one of the better known seas of the world. Although we can distinguish many species we do not know much about their ecological differences. For most species, we know that they are sessile filter feeders but do not know their specific demands. In the present study we mapped sponge distributions around Discover Bay, Jamaica on selected sample areas and looked for a correlation of sponge distributions with measured or described data from sample areas.

Mapping followed the method of Braun-Blanquet, using nine estimation classes and covered 1659 m^2 down to a depth of 40m. The data underwent multivariate analysis using the programs MULVA, PPS and CANOCO. Methods applied and programs used were developed for plant sociology. Depths down to 107m were surveyed for sponges using mixed gas and limited to collecting and photographing, due to limited bottom time and a reduced number of dives performed. The data set is split in two groups; the distinction being shallow and deep water.

Ordination of shallow water data was carried out with the program CANOCO using RDA. There are two possibilities for figuring results, either using species as axes in multidimensional space in order to

get sample plots, or using samples, in order to get species plots. Distances in sample plots represent a measure for the similarity of species occurring on these areas. Two sample areas with similar species of similar cover are plotted close together. If the complete data set is figured in this way, sample areas and species form three large groups which can, comparing with field notes, be assigned to three large scale habitats, which show the biggest differences in species composition. These habitats are: reef, lagoon, and undersides of plate corals, and as subsets of the data were analyzed further. The undersides of plate corals were not analysed further due to the few representatives.

The lagoon subset shows different clusters of sample area and species groups, which may be interpreted as: blue hole, *Thalassia* sea grass beds, backreef, and protected backreef eg. the sponge *Xestospongia carbonaria* occured only together with *Thalassia* seagrass. Analyzing the reef areas subset (the largest), distribution of sample areas looks continuous. If using depth as a third axis it becomes obvious that distribution follows a depth gradient. A similar result is observed in a distribution along an inclination gradient. Both have more or less a similar direction because steep walls generally occur more in deeper water. It should be pointed out that the depth and inclination gradients become visible only after omitting sample areas from other habitats and those from reefs which grow in front of river mouths.

On 13 deep dives to a maximum depth of 107 m, using trimix below 30m and nitrox as travel mix, 60 different species were collected, 25 of them previously undescibed and several others previously not known from Jamaica, personal impressions are that the inventory of occuring species is far from complete. If orders are compared, striking differences between shallow and deep water are visible (comparison of shallow to deep).

Halichondrida double their relative species number from 17% to 38%. Haplosclerida gain about 5%. Lithistida occur with one species. The declining orders are: Hadromerida from nearly 14% to 5%. Homosclerophorida from 5% to 0. Spirophorida from 2% to 0. Astrophorida from 7% to 3%. Dendroceratida from 4% to 0. Poecilosclerida and Dictyoceratida maintain about the same number of species.

Differences between orders or families show very coarse trends only. For a better ecological understanding we think it is necessary to make a comparison at the species level, because even within one genus species may show completely different responses to the change of depth or inclination of substrate. Although depth and inclination are important in our investigation, it seems clear that there are distribution determining factors which were not taken into consideration in the present study.

Key words: *Jamaica, mapping of sponge distribution, depth comparison*

ANTIMICROBIAL ACTIVITY OF SPONGES FROM SOUTHERN BRAZIL, ATLANTIC COAST

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Within the research program of biologically active natural products, we have investigated different species of marine sponges collected by scuba diving in the South-western Atlantic region, near the coast of South-Brazil, aiming at the evaluation of their potential as a source for new drugs. In order to achieve this goal, the antimicrobial activity of five species, *Tedania ignis* Duchassaing & Michelotti, *Pseudaxinella reticulata* (Ridley & Dendy), *Polymastia janeirensis* (Boury-Esnault, 1973), *Batzella* sp. and *Petromica* sp. were analysed. The sponge species found in this particular region constitute a group of shallow-water Demospongiae in which a pharmacological usage has not previously been evaluated. Aqueous extracts obtained by grinding and maceration for 30 min following freeze-drying, as well as organic solvent extracts (toluene: methanol 3:1 v/v) were prepared from organisms frozen since the harvesting. Antimicrobial activity was evaluated by the agar diffusion method on paper disks (6 mm diameter). Each sponge extract was tested for growth inhibition of five bacteria species: *Escherichia coli* (ATCC 25922), *Staphylococcus aureus* (ATCC 6538P), *Staphylococcus epidermidis* (ATCC 12228), *Bacillus subtilis* (ATCC 6633), *Micrococcus luteus* (ATCC 9341) and two yeast species: *Candida albicans* (ATCC 110231) and *Saccharomyces cerevisiae* (ATCC 1600). None of the tested extracts analysed by this method could inhibit the growth of these microorganisms, an exception was the *Petromica* sp organic extract which showed an inhibitory activity for *Bacillus subtilis*. The same extracts were analysed for antitumour activity by *in vitro* inhibition of proliferation of different tumour cell lines. Some of the extracts showed promising results. The preliminary antimicrobial assay can be useful for pointing out sponge species to be further analysed.

Key words: *antimicrobial activity, Atlantic coast, southern Brazil, biologically active natural products, antitumor activity*

PROPAGATED ELECTRICAL IMPULSES IN A SPONGE

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Previous work has shown that *Rhabdocalyptus dawsoni*, a hexactinellid sponge, can arrest its feeding current following mechanical or electrical stimuli. Although a propagated impulse was suspected as the signal triggering arrests, numerous attempts to record such an event failed, due to the porous character of the tissue and extreme fragility of the surface membranes. Using a new approach, which involves dissociating sponge tissue, letting it reaggregate, and grafting it back on to the sponge as an autograft, we have found it possible to record propagated electrical impulses. The grafts fuse with the trabecular reticulum, a syncytial tissue that penetrates all parts of the body, including the flagellated chambers, and are eventually absorbed into the sponge. But for a while they form solid lumps that can be used for attachment of suction recording electrodes. Impulses are all-or-none events evoked by single electrical shocks that propagate diffusely through the entire preparation at 0.27 ± 0.1 cm·s at 10°C, presumably in the trabecular reticulum. The preparation shows an absolute refractory period of 29 s, and is relatively refractory for a further 95-100 s. Intracellular recordings have not been carried out but the wave form recorded extracellularly is suggestive of a conventional, overshooting spike. Pharmacological evidence suggests that it is sodium-based. Thus, despite its long refractory period and low conduction velocity the system is functionally equivalent to the through-conducting nerve nets and excitable epithelial conduction systems of other animals.

Key words: *hexactinellid, conduction, electrophysiology, sponge behaviour, pumping*

RANDOM AMPLIFIED POLYMORPHIC DNA (RAPD) ANALYSIS CAN REVEAL MICRO- AND MACROEVOLUTIONARY PATTERNS IN PORIFERA

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Sponge classification is problematic, due in part to lack of good morphological markers. One way to solve this issue is to generate molecular markers, which could contribute as extra characters. Polymorphic DNA sequences can be used for this purpose with the advantage that variation can be detected between individuals or species enabling one to analyse genetic variability at the population level and to determine evolutionary relationships. The random amplified polymorphic DNA (RAPD) polymerase chain reaction (PCR) technique is performed using arbitrary sequence oligonucleotide primers on a genomic DNA template. We have selected six primers based on their amplification profiles using genomic DNA from 14 sponge species. The total number of amplified scorable bands per primer varied from 8 (AP5/8) to 20 (OPS-17/6). The level of polymorphism was measured in 44 individuals of 5 populations of *Hymeniacidon heliophila*, between those populations and between 6 species of the genus *Mycale*. In a *H. heliophila* population from Praia Vermelha Beach (Rio de Janeiro State), the average number of polymorphic bands was 73% and of monomorphic bands was 27% for the six primers. Two bands, one monomorphic and one polymorphic, were isolated from two different individuals of two populations of *H. heliophila*, labelled and used as probes in Dot blots containing genomic DNA from 18 species of sponges and 15 individuals from the 5 populations of *H. heliophila*. From this experiment we obtained one marker that is specific to Southern Brazilian *Hymeniacidon* species. Using two single primers and one combination of two primers (double) and genomic DNA from *Mycale* (*Aegogropila*) *aff. americana*, *Mycale* (*Aegogropila*) *escarlatei*, *Mycale* (*Arenochalina*) *laxissima*, *Mycale* (*Carmia*) *microsigmatosa*, *Mycale* (*Mycale*) *arenaria* and *Mycale* (*Zygomycale*) *angulosa*, we generated band patterns with an average number of polymorphic and monomorphic bands equal to 91% and 9%, respectively, which are used as a measure of variability in *Mycale* species and as characters to construct phylogenies. The RAPD technique is shown to be a powerful tool to generate and identify population or species-specific molecular genetic markers for sponges and to assess different evolutionary aspects of their biology.

Key words: *molecular markers, RAPD technique, genetic diversity, PCR, polymorphic, monomorphic*

WIDE PHYLOGENETIC SPECTRUM OF HETEROTROPHIC MICROBES ASSOCIATED WITH THE SPONGE *DISCODERMIA DISSOLUTA* CHARACTERIZED BY MICROBIOLOGICAL AND MOLECULAR TECHNIQUES.

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Sponges are well known to harbour large numbers of heterotrophic microbes within their tissues. Studies to determine the diversity of these associated microbes have been carried out on only a few shallow water sponge species. We have cultured prokaryotes from the deep sea sponge, *Discodermia dissoluta*, and characterised them by standard microbiological identification techniques. A subset of these isolates has been characterised using molecular systematics techniques. Molecular phylogenetic analyses of partial DNA sequences of the small subunit (SSU) rRNA gene from 15-20 of these microbes were generated and show a highly diverse phylogenetic range of microbes associated with this sponge. Presence/absence differences between uncultured microbial sequences obtained by PCR amplification of whole tissue with cultured isolates were also evident. Identification of the isolates representing true sponge symbionts remains undetermined. The implications of these data for general sponge ecology and evolution will be discussed.

Key words: *microbial symbionts, SSU rRNA, molecular phylogenies, biodiversity, PCR*

LIMITS ON THE BATHYMETRIC DISTRIBUTION OF KERATOSE SPONGES: A FIELD TEST IN DEEP-WATER

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The keratose sponges (i.e. those in which the mineral skeleton is replaced by a collagenous skeleton) are generally restricted to shallow-water habitats, but the causes of this distinct bathymetric pattern remain unclear. Sharp pycnoclines at the depth of the upper slope may hinder colonisation of deep-waters because of thermal stress or reduced light and particulate food below the pycnoclines. It is also possible that oligotrophy and loss of symbiotic cyanobacteria below the pycnocline may lead to a nutritional stress. Using manned submersibles in Exuma Sound, Bahamas, we determined that the pycnocline lies between 70 and 100 m. We transplanted individuals of 2 keratose sponges (*Aplysina fistularis* and *Ircinia felix*) from their natural habitat on a shallow-reef (4 m deep) to 3 depths (100, 200, 300 m) within or below the pycnocline to investigate mortality, and changes in body size, shape and histology as a function of depth. We also recorded changes in populations of photosynthetic and heterotrophic symbiotic bacteria, as well as the parasitic polychaete *Haplosyllis spongicola*. By transplanting individuals of *A. fistularis* bearing buds for asexual propagation (fistules) and individuals of *I. felix* brooding embryos, we also tested the viability of reproductive propagules in deep-water environments.

We found that, although these 2 sponges do not naturally occur at depths below 40 m, 60% of *A. fistularis* and 40% of *I. felix* survived at 100 m for 12 mo. No *A. fistularis* survived at 200 m, whereas 30% of *I. felix* did. All sponges transplanted to 300 m died within 2 mo. Water temperature was the most likely cause of sudden mortality at this depth. There were no significant differences in growth between individuals at the slope and controls on the shallow reef. Cyanobacteria were lost in individuals of *I. felix* that survived at 100 and 200 m, and these sponges repositioned oscules and formed chimney-like processes, probably to enhance water flow through the sponge and compensate for nutritional stress. By contrast, cyanobacteria were still abundant in individuals of *A. fistularis* surviving at a depth of 100 m, and these sponges did not change shape significantly, apart from the loss of fistules.

It appears, therefore, that the loss of cyanobacteria and the increasing oligotrophy with depth do not set the lower bathymetric limits of species. Removal of sponge tissues by the parasitic polychaete *H. spongicola* also appears not to aggravate significantly, the nutritional stress experienced by sponges transplanted to deep water, at least to the extent that it may restrict the bathymetric distribution of the host. Despite the fact that only the species *I. felix* was heavily parasitised and that parasites survived within hosts at all depths, there was no significant difference in survival with depths between sponge species. A TEM examination of the mesohyl did not reveal significant cytological differences among sponges transplanted to various depths. At all depths, surviving individuals of both species showed archaeocytes engaged in phagocytosis and digestion of cyanobacteria and/or heterotrophic bacteria. Similarly, collencytes and spongocytes were apparently secreting collagen, indicating that temperatures at 100 and 200 m do not inhibit the formation of the skeleton. Sponge recruitment derived from either asexual or sexual propagules was never observed at slope depths.

Since adult sponges survived when they were artificially transported to deep waters, the inhibition of larval dispersal or settlement success (perhaps caused by a the sharp decrease in temperature with increasing depth) emerges as the most plausible explanation for the shallow-water confinement of these keratose sponges.

Key words: *keratose, bathymetric distributions, ecology, symbionts, infauna, slope megafauna, environmental parameters*

AN EXPERIMENTAL APPROACH TO THE ECOLOGICAL SIGNIFICANCE OF MOVEMENT IN SPONGES

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It has been known since the late 40's that sponges are capable of movement on substrates. However, the potential ecological role of this ability remains virtually uninvestigated to date. In the present laboratory study, we documented motion in the encrusting Mediterranean sponge *Scopalina lophyropoda* as a function of food availability, density of neighbours, water flow, burying by silt, and body size. For all experiments, we used small sponge individuals (1-2 mm^2) that developed from regenerating tissue fragments after attachment to acetate sheets that served as substratum.

In the first experiment, we examined the combined effects of food availability and density of allogeneic (histologically incompatible) neighbours on the movement response of 320 small sponges. Sponges were arranged in either high-density groups (individuals 0.5 cm apart) or low-density groups (individuals 2 cm apart) of 16 individuals each, then half of the groups were starved and half were fed with a general culture of marine bacteria for 8 wks. We found that sponges moved at a speed that ranged from 0 to 1.178 mm d^{-1}, with a peak during weeks 3 and 4 and very low motion rates after week 5, covering a total distance that ranged from 0.25 to 18 mm. Starvation was important in inducing movement: starved sponges covered distances significantly larger than fed sponges, irrespective of the density of neighbours. We assessed how much the path of each individual deviated from a straight line, and found that sponges described significantly more sinuous paths when moving through high-density groups compared to low-density groups, irrespective of food availability. Paths throughout high-density groups were particularly sinuous because crawling sponges frequently made contact with their neighbours, then stopped and resumed movement in a different direction to avoid prolonged contacts.

In a second experiment, we investigated directionality in the movement of an isolated sponge that was placed 2.5 mm in front of a semicircular barrier made of conspecific individuals. We expected movement towards barriers made of isogeneic (histocompatible) conspecifics, since contact with them will involve tissue fusion and an advantageous increase of size in the resulting individual. We also expected movement away from allogeneic barriers to avoid the imminent contact with allogeneics, which are competitors for food and space. We assessed directionality in 3 wk movements by measuring the angle that the mean vector of the path followed by a sponge formed with respect to the line defined between the initial position of the sponge and the centre of the neighbouring barrier, which served as 0° angle. We found that movements were not random, but directional. Mean direction was not significantly different from angle 0° in both treatments, indicating that movement was always towards the neighbouring barrier, and that sponges can perceive the presence of neither isogeneic nor allogeneic neighbours before physical contact is established.

In a third experiment, we investigated differences in movement responses between sponges exposed to a slow, turbulent flow that left a thin layer of silt on the substratum and sponges exposed to a steady, directional flow that did not leave any silt on the substratum. We found that sponges under siltation covered distances significantly larger compared to sponges non-exposed to silt after 8 wk, and that movement was random with respect to flow direction in both flow conditions.

In a fourth experiment, we assessed motion as a function of sponge size by comparing the mean distance covered after 12 wks by large individuals (15 ± 4.5 mm^2) and small individuals (1.8 ± 0.7 mm^2). No significant difference was found. In summary, the results of the laboratory experiments strongly suggested that sponges in the field may use their capability of movement to prevent prolonged physical contact with competitors and to move away from sites with either limited access to food or excessive exposure to silt.

Key words: *sponge movement, spatial competition, sponge behaviour, environmental conditions*

A NEW DENDROCERATID SPONGE WITH RETICULATE SKELETON

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A new encrusting dendroceratid sponges characterised by a skeletal network made of upright primary spongin fibres transversally interconnected by secondary fibres is described from the Alboran Sea (Western Mediterranean). Primary fibres are 30-150 µm wide, unbranched or poorly branched, somewhat fasciculate, with a laminated bark and a pith containing foreign debris, and provided with a subcircular basal plate (300-700 µm in diameter). Secondary fibres are also laminated, but usually lack any detritic material, and occasionally form perforated plates around the point where they anastomose to a primary fibre. Secondary fibres are 20-40 µm wide, but they can reach 300 µm at the perforated plates. The reticulation of the skeleton is loose and irregular, so that some primary fibres are interconnected through their basal portions only, whereas others are interconnected along their apical portions. Some primary fibres, especially those located at the periphery of the sponge, are isolated lacking any transversal interconnection. The skeletal features of this new sponge are intermediate between those of the aplysinellid genus *Pleraplysilla* and those of the dictyodendrellid genus *Igernella*. Although the primary fibres resemble those of the genus *Pleraplysilla*, which also contain detritic inclusions in the pith and may anastomose to form fasciculate tracts or vague meshes in some species, the presence of an actual network of secondary fibres in the new species fits the current diagnosis of neither the genus *Pleraplysilla* nor the family Aplysinidae. Such a network would rather suggest a relationship with the genus *Igernella*. However, the new species lacks both spongin spicules and a basal continuous plate of spongin, which are two of the traits defining the genus *Igernella*. Therefore, assuming that the absence of both spongin spicules and a basal plate of spongin is not taxonomically relevant at the generic level, the new material could be assigned to *Igernella*. On the contrary, if the presence of secondary fibres is assumed to be polyphyletic, the new sponge could be included in the genus *Pleraplysilla*. We opted for the latter possibility because most of the recent chemical and cytological information supports the hypothesis that the acquisition of a secondary reticulate skeleton was probably a convergent process in Dendroceratida. Thus, we propose modifying the diagnosis of the genus *Pleraplysilla* to include species with an irregular network of secondary fibres to accommodate this new species, *Pleraplysilla reticulata* n. sp.

Key words: *sponge taxonomy, spongin skeleton, Aplysillidae, Dictyodendrillidae, Dendroceratida*

A NEW GENUS OF FRESHWATER SPONGES FROM THE LAKE TANA

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Twelve freshwater sponge genera unable to produce gemmules or statoblasts are known. Three of them belong to the family Lubomirskidae and are endemic of the most ancient lake of the world, the Paleocenic Bajkal Lake from where are reported *Lubomirskia abietina, L. baikalensis, L. fusifera, Baikalospongia bacillifera, B. dzhegatajensis, B. intermedia* and *Swartschewskia papyracea.*

The genus *Potamolepis* (*P. belingana, P. chartaria, P. leubnitziae, P. marshalli, P. micropora, P. pechueli, P. weltneri*) is restricted to the hydrographic basins of Central Africa and belong to the family Potamolepidae. The biology of these taxa is well known and their taxonomic status is not under debate in the present paper.

Other eight genera, characterised by scarce findings and data on life cycle, cannot be ascribed to extant families, therefore is compulsory to define their provisional grouping as *incertae sedis*.

The late Pliocenic Ochrida Lake, that displays a fauna composition with high phylogenetic affinities to the bajkalian one, hosts two endemic genera: *Ochridaspongia (O. rotunda* and *O. interlithonis)* and *Ohridospongilla (O. stankovici).* Two genera, *Echinospongia (E. brichardi)* and *Spinospongilla (S. polli),* are reported from the Pliocenic Tanganyika Lake; four other genera are known only from the type locality: *Balliviaspongia (B. wirmanni)* from Titicaca Lake (Middle Miocene); *Cortispongilla (C. barroisi)* from Tiberias Lake (Pliocene); *Malawispongia (M. echinoides)* from Nyasa Lake (Middle Pleistocene) and *Pachydictyum (P. globosum)* from Lake Poso.

In this paper is described the new genus *Makedia* whose type species is *Makedia tanensis* from the shallow-waters of the Pleistocenic Tana Lake in Ethiopia. Encrusting sponges were small and cushion-shaped under littoral stones. The skeleton is a regular network of oxeas from completely smooth to granulated and strongly spiny. None ectosomal differentiation is evidenced. Microscleres and gemmules are absent.

Summarising, all these freshwater sponge genera share the following characters: i) they are monospecific with the exception of *Ochridaspongia*; ii) they are known only from the type locality; iii) they do not produce gemmules or statoblasts; and iv) they inhabit tectonic or volcanic lakes with high percentages of endemisms. Apart from *Echinospongia brichardi* whose spiculation is very similar to those of *Potamolepis*, in the other eight genera the skeleton is a regular network of oxeas from smooth to variously spiny; the entire complex of traits shown by the *incertae sedis* freshwater sponge taxa (ISFS) is extremely homogeneous in contrast with their different taxonomic status at the genus level. As for Lubomirskiidae and Potamolepidae, the hypothesis of an adaptive radiation is suggested by the presence of congeneric taxa in different niches in a continuous environment; on the contrary, the morphological similarities of ISFS genera fit better an evolutive process of convergence.

Key words: *taxonomy, ancient lakes, endemisms, biogeography, freshwater sponges*

STUDY ON THE DISTRIBUTION OF BAIKALIAN SPONGES

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Freshwater sponges are classified into three families. Spongillidae, Potamolepidae and Lubomirskiidae. Spongillidae are cosmopolitan sponges and widely distributed throughout the world, Potamolepidae are found in lakes of Africa and South America, and Lubomirskiidae inhabit only Lake Baikal. One of the characteristics of Spongillidae is gemmule formation. Gemmules are asexual bodies with a structure in highly resistant resting stages. Most Japanese sponge species form gemmules, but a certain species which forms in the shallow zone of lakes, does not form gemmules in deeper zones. Therefore we have a great interest in the Lubomirskiidae of Lake Baikal which does not produce any gemmules. At present, the taxonomy of some Lubomirskiidae is in a chaotic state. Furthermore, the recent distribution of Baikalian sponges has not been recorded. We decided to collect as many Baikalian sponges as possible and to review their taxonomy and their distribution in Lake Baikal. About 700 specimens were collected, mainly from the entire littoral zone of Lake Baikal, but some specimens were collected from the Academishan ridge by a dredge survey, and others collected on diving surveys. Most of the specimens belonged to the family Lubomirskiidae, with a few belonging to the family Spongillidae. Lubomirskiidae were classified into three genera and eight species according to Rezvoy. Based on the results of our study, our Lubomirskiidae specimens were tentatively classified into 4 genera and 11

species. At the present, Lubomirskiidae are classified mainly by their spicules and skeletons not by the form of the sponges (changeable due to substrate and water current), oscula or colour (thought to not contain pigment cells). The colour of green sponges is owing to symbionts, zoochlorellae.

The Lubomirskiidae species were distributed throughout the entire littoral zone of Lake Baikal, except where the substratum was sand, mud or pebbles. On the other hand, Spongillidae species (*Spongilla lacustris, Ephydatia muelleri* and *Eunapius* sp.) were collected from only four stations. Lake Baikal may be an in appropriate habitat for Spongillidae species. Spongillidae lack of presence throughout the entire littoral zone may be due to; the amount of nutrients, wave action and water temperature. Regarding nutrients, certainly Lake Baikal is characterised by oligotrophy when compared with other lakes where many Spongillidae species live, but the limits within which the Spongillidae species can not live is unknown. Lubomirskiidae may be accustomed to poor food. Due to the weak and fragile bodies of Spongillidae, they cannot live in an area of strong wave action. But the wave action in deeper zones is weaker than that in shallow zone. In Lake Biwa, in Japan, Spongillidae species can live at a depth of 30m, where the wave action is weak. In Lake Baikal, wave action is also weak at such a deeper zone. But we could not find Spongillidae even at a depth of 30m. We compared the maximum temperature in Lake Baikal and Lake Biwa during the year at a depth of 30m. In Baikal, the maximum temperature is about 6°C. On the other hand, in Lake Biwa, it is about 10°C. The maximum temperature may be an important factor for the survival of Spongillidae species. More detailed information on differences in the two family habitats is necessary to resolve this problem, which is important in the analysis of spicules in old sediment.

The spicules of freshwater sponges are very stable in old sediment because they consist of silica components, such as diatoms, and we are now studying the spicules of old sediment obtained from drilling cores. Some spicules from sediments, believed to have been deposited there 4 500 000 years ago, were found about 180m under Lake Baikal. If Spongillidae spicules are found, we might hypothesise the lake's conditions as being similar to the Little Sea near Olkhon Island at present. If we examine spicules along the drilling core from surface to bottom, we might find successive changes in the circumstances. Furthermore, if we should find new spicules not seen in recent sponges, the new finding would help us in drawing up the phylogenetic tree of freshwater sponges.

Keyword: *freshwater sponge, Lake Biwa, Lubomirskiidae, Spongillidae, taxonomy, distribution, environmental conditions, sediment studies.*

AN ULTRASTRUCTURAL STUDY ON THE CONTRACTILE PINACOCYTE OF A FRESHWATER SPONGE

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Contractile cells in sponges were first observed in the oscular diaphragm of marine species, *Microciona* and *Tedania* (Bagby, 1961). They consisted of well-differentiated myocytes having myosin and actin filaments, whereas in the freshwater sponges, there are no reports of contractile apparatus or myocytes.

The oscular diaphragms and body walls of freshwater sponges are contractible when stimulated. Stimulation is effective whether it is osmotic, thermal, electric, etc. According to these observations on sponges we hypothesise that pinacocytes in the oscular diaphragm and body wall must bear the contractile apparatus, and we suggested in a previous report that these cells have a network of filamentous bundles. We subsequently attempted to structurally identify the contractile filaments of the cell using the following methods: 1. Observations under light microscopy; 2. Observations under electron microscopy; 3. SDS-PAGE; 4. NBD-phallacidin staining; 5. Anti-actin gold conjugation.

Pinacocytes in these sponges showed a flat and multiangular shape, measuring about 5μm in diameter and 0.1μm thick. Pinacocytes in the outer layer of the oscular diaphragms and body wall had many

bundles extending radially from the central nuclear zone to the peripheral region of the cell, whereas these bundles were not observed in the inner pinacocytes. Bundles were easily stained using NBD-phallacidin. Observations of thin sections showed these bundles are composed of many thin filaments of about 4-6nm diameter. These bundles ran straight in the contracted state, and were distributed in the basal region of the cell. Thin filaments in the bundle were clearly decorated with anti-actin gold conjugation. SDS-PAGE analysis of the diaphragm revealed a protein band of 45kD. These results support the idea that thin filaments of about 4-6nm in diameter in pinacocytes are composed of actin molecules.

A freshwater sponge, *Ephydatia fluviatilis*, has no myocytes but has contractile pinacocytes with actin bundles.

Key words: *freshwater sponge, contractile filaments, ultrastructure, actin, pinacocytes*

THE PHYLOGENETIC POSITION OF THE SPONGE *SPONGOSORITES SUBERITOIDES* DETERMINED BY ANALYSIS OF 28S RRNA GENE SEQUENCE.

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A number of problems exist in the phylogenetic consideration of the sponge *Spongosorites suberitoides* that cannot be resolved on morphological grounds alone. Placing the sponge in the genus *Spongosorites* divides this genus into two groups; a single shallow water species and many deep-water species. Described differences between these groups include; oxea size, aerophobic colour-change and surface texture. Further, *S. suberitoides* shows an affinity with hadromerid sponges such as colour in life, texture, arrangement of anastomosing choanosomal tracts and the lack of an aerophobic reaction. In its association with hermit crabs and gastropods it resembles the genus *Suberites*. It also shows similarities to species of *Aaptos* and some Polymastiidae. The DNA sequence of the five prime region of the 28S ribosomal gene of *S. suberitoides* is compared with DNA sequences from hadromerid and halichondrid species in a phylogenetic analysis to resolve the position of this species.

Key words; *phylogeny, morphology, DNA, 28S ribosomal gene*

PHYLOGENY OF LITHISTID SPONGES

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Kelly-Borges and Pomponi (1994) utilised partial 18S rRNA gene sequences to resolve relationships within lithistid sponges (Porifera: Demospongiae). While their results lent weight to the growing realisation that the Order Lithistida is polyphyletic, their conclusions were hampered by low levels of sequence variation. Our initial study sought to evaluate the resolution potential between regions of the 18S and 28S rRNA genes within a group of selected Porifera. Approximately 1,300 bp of the 18S rRNA gene and a 5' region of the 28S rRNA gene were compared with the data of Kelly-Borges and Pomponi (1994). Six taxa were selected which represented a gradient of relationships, ranging through the taxonomic levels of genus, family and class. We found that the 700 bp of the 28S rRNA gene presented the greatest potential for resolution of this group of porifera at the genus and family level, and that this molecular phylogeny is congruent with morphological hypotheses for the group. The study has progressed to include a number of other lithistid and non-lithistid taxa.

Key words: *Lithistida, 18S rRNA. 28S rRNA, phylogeny*

CLIONA LAMPA AND DISTURBANCE ON THE CORAL REEFS OF CASTLE HARBOUR, BERMUDA

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On reef regions of Bermuda, one of the most abundant populations of the boring sponge *Cliona lampa* occurs within Castle Harbour. Historically, dredging and landfill for the construction of an airfield (1941-1944) in the harbour caused changes in the coral reef community structure. The resulting increase in sedimentation and turbidity led to mass mortality, changes in species composition and age distribution of the coral (especially for *Diploria* sp.; Dryer and Logan 1978, Cook *et al*., 1994). We hypothesised another outcome of this disturbance was an increase in the distribution and abundance of *Cliona lampa* within Castle Harbour. If the sponge was able to invade the space made available by death of the coral colonies, it should be reflected in the extent and type of substrate infested. An *a posteri* field survey of Castle Harbour was conducted to determine the extent and substrate type (coral vs. non-coral in origin) of *C. lampa* infestation. A field experiment was conducted to test the ability of *C. lampa* to colonise *Diploria* sp. with and without live tissue coverage. Results from the field survey and experiment support our hypothesis. Following the disturbance, it appears the increase in substrate availability combined with the decrease in competitors occurred at a time and place that favoured colonisation by the sponge. In addition to algae and corals, sponges may be important to consider when examining alternative states following disturbances in coral reef communities.

Key words: Cliona, *coral reef communities, disturbance, Bermuda, colonisation*

THE PHYLOGENETIC HISTORY OF SPONGES IN PALAEOZOIC TIMES

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According to molecular biological analysis, the origin of the phylum Porifera dates back about 800 MY. The first sponges were probably aspicular, like the oldest known sponge fossils Palaeophragmodictyon from the Late Proterozoic Ediacara. Early Cambrian Sponge assemblages were dominated by Hexactinellida, but already in the Atdabanian also the Pinacophora (Demospongiae-Calcarea-taxon) were well represented. The Archaeocyatha can be considered as a stem lineage representative of this group. Early Cambrian Calcarea comprise of modern appearing forms as well as the exclusively Palaeozoic Heteractinellida and Polyactinellida, the latter group exhibits triradiate calcitic spicules, which are probably a constituent character of the taxon Calcarea.

Within the Demospongiae, the tetraxone is considered the basic spicular symmetry, from which the other spicula-types are derived. Oxyasters from the Early Cambrian, which are in the size range of megascleres and show well-developed central canals, may have developed from tetraxone mesotriaenes, whereas the large Middle Cambrian sigmata the may be derived oxeas. This means that the differentiation in mega- and microsclerocytes known from recent demosponges may have taken place at a later stage. The first desmata-bearing demosponges ("Lithistida") of the group Anthaspidellidae, Orchocladina, are from the Middle Cambrian and probably originated from reticulated monaxonid precursors close to the Hazeliidae. During the Late Ordovician, the chiastoclones developed from anthaspidellid dendroclones, and the Palaeozoic groups Tricranocladina and Sphaerocladina may be derived from chiastoclonellid ancestors. Contrary to widely accepted hypotheses, there seems to be no direct phylogenetic line from the Orchocladina to the modern Tetracladina, since the origin of tetraxial desmata from anaxial chiastoclones is very unlikely. The earliest true tetractones with definite axial canals are documented from the Permian Jereina, whereas the first phyllotriaenes of the modern spirasterophoran type are known since the Late Triassic. The "megamorine" Saccospongiidae probably

originated from a monaxonid group close to the Halichondritidae, but the Palaeozoic heloclones and megaclones as well as the elongate rhizoclones of the Haplistiidae are not considered phylogenetically linked to the modern Megamorina or Rhizomorina. Because of their skeletal architecture, the Palaeozoic Saccospongiidae and Orchocladina as well as theTricranocladina and Sphaerocladina, which probably evolved from the Orchocladina, are attributed to the Sigmatophora.

Mesozoic and Recent Rhizomorina are characterised by skeletons of exclusively rhizoclone desmata with sigmaspires as microscleres, but the latter are unknown from the fossil record and almost certainly have no connection with the sigmatophoran sigmata, which are known since the Cambrian. At the end of the Permian, the Palaeozoic "Lithistida", maybe with the exception of the Sphaerocladina, had all become extinct. Against widely accepted ideas, there is probably no phylogenetic link from the Tricranocladina to the modern Corallistidae (Dicranocladina). The Sphaerocladina, which have recently been documented also from the Early Palaeozoic, may have lead to the Mesozoic Neosphaerocladina, but no connection can be documented between these groups and Recent genera often attributed to the Sphaerocladina, such as *Crambe* or *Vetulina*. Lophocalthrops of the Plakinidae first occurred in the Early Carboniferous and are connected by transitional forms to the candelabra of the modern Homoscleromorpha, known since the Early Cretaceous. The Plakinidae spicules probably originated from the same type of tetraxones, which lead to the first dichotriaenes in the Early Carboniferous. Thus the Plakinidae/Homoscleromorpha are probably the sister group of the Spirasterophora, to which most modern "Lithistida" belong.

Key words: *phylogeny, Palaeozoic sponges, skeletal architecture, Calcarea, Demospongiae, Hexactinellida*

TOWARDS A PHYLOGENETIC SYSTEMATICS OF THE FOSSIL HEXACTINELLIDA

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The vast majority of all fossil hexactinellid taxa have been described from the Mesozoic. This is due to the rich occurrences of hexactinellids in the well-exposed Jurassic and Cretaceous strata of Europe, and to the generally larger preservation potential of the rigid Mesozoic hexactinellids compared to the predominantly non-rigid Palaeozoic ones. Nevertheless, most of the main hexactinellid taxa can be traced back to the Early Palaeozoic. Isolated hexasters of the Hexasterophora occur in the Early Ordovician, whereas the earliest Amphidiscophora are documented from the Late Silurian, and the first hexactinosans are known from the Late Devonian. However, the bulk of the Palaeozoic hexactinellid sponges, although well established as monophyletic groups, cannot definitely be attributed to any recent taxon and require an exclusively fossil-based systematics. The Early Palaeozoic Protospongiidae and Hintzespongiidae are derived from a reticulate hexactine-bearing ancestors, probably close the precursor of the Mattaspongia-Microstaura-group, which can be regarded as the adelphotaxon of the Hexactinosans. The Dictyospongiidae s.str., which are hexasterophorans, probably also originated from the Mattaspongia-stem lineage, and did the modern Sceptrulophora (Clavularia-Scopularia-taxon), which have recently been traced back to the Early Palaeozoic through the documentation of Ordovician scopules. The Brachiospongiidae, including the Stiodermatidae, may be attributed to the amphidiscophorans, because of the great similarity in skeletal architecture between Strobilospongia and the modern Hyalonematidae. However, the systematic affinity of many Palaeozoic lyssacine hexactinellids which appear or were in fact primitive, including most Early Cambrian genera such as Quadrolaminiella, Solactinella and Hyalosinica, is still uncertain, and these taxa have to be classified within the probably non-monophyletic grouping "Rossellimorpha".

From the end of the Permian, all major Palaeozoic hexactinellid groups had become extinct, and the Mesozoic Hexactinellida are represented by modern forms, mainly Hexactinosans and Lychniscosans. "Lyssacinosa", which comprise the majority of Recent hexactinellid taxa, are not commonly found in the Mesozoic, but nevertheless there are some important occurrences, from which Recent genera can be

identified. *Regadrella* of the Euplectellidae is known with several species from the Cretaceous, and the first species of the Hyalonematidae, *Hyalonema cretacea*, has been described from the Campanian, but there are more unpublished Late Cretaceous representatives of this group and also of the Rossellilidae from the section of Arnager (Denmark) still to be described. The first definite lychniscosans are known since the Middle Jurassic, and the group reached its maximal diversity during the Late Cretaceous. Probably, this group did not arise from the hexactinosans, but as the adelphotaxon of some lyssacine group, maybe the Euplectellidae. Today the Lychniscosans have become almost extinct, so the exact systematic attribution of the Mesozoic families and genera to Recent ones is in most cases not possible. The same thing is true to many Mesozoic hexactinosans, although many Recent genera have now been identified from Late Cretaceous strata, which allows an approximation of the zoological systematics at least for the Late Mesozoic and Tertiary. However, still many Cretaceous and most Jurassic hexactinosans traditionally classified in the grouping "Inermia", such as the Casearia-Porospongia-group, cannot be definitely attributed within the Recent systematics, but have to be subject of a phylogenetic-systematic approach based on fossil representatives only.

Key words: *phylogeny, systematics, Hexactinellida, fossils*

THE GENUS *ERYLUS* GRAY, 1867: A REVISION OF BRAZILIAN SPECIES WITH DESCRIPTION OF A NEW SPECIES (ASTROPHORIDA, GEODIIDAE)

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Before this study the genus *Erylus* was represented along the Brazilian coast by the following species: *E. formosus* Sollas, 1886, *E. corneus* Boury-Esnault, 1973, *E. topsenti* Lendenfeld, 1903 and *E. oxyaster* Lendenfeld, 1910. The reexamination of these species and additional material under SEM analysis showed the necessity of revision, allowing the detection of new species.

The most of the examined material was dredged from 12.8 to 918m in depth carried out under the auspices of: Diretoria de Hidrografia e Navegação da Marinha (DHNM), Brazil; Departamento de Recursos Pesqueiros da Superintendência de Desenvolvimento do Nordeste (SUDENE), Brazil; Pontifícia Universidade Católica do Rio Grande do Sul (PUCRS), Brazil; Projeto dos Recursos Vivos da Zona Econômica Exclusiva (REVIZEE/NE), supported by Universidade Federal de Pernambuco, Brazil; Projeto dos Recursos Vivos da Zona Econômica Exclusiva (REVIZEE/SUL), supported by Fundação Universidade do Rio Grande (FURG), Brazil; "Programa Rio Grande do Sul-I" (PRGS-I), supported by Universidade de São Paulo e Governo do Estado do Rio Grande do Sul, Brazil; Projeto Talude, Fundação Universidade do Rio Grande (FURG), Brazil; Britanic Expedition H.M.S. "Challenger"; French Expedition "Calypso". Two specimens were collected by Scuba or Narghilé (0-30m). Dissociated spicule mounts, thick sections and preparations for SEM study were made using the methodology of Mothes (1996). The reexamination of material detected the presence of *E. alleni*, a Caribbean species with a southern limit at the coast of Rio Grande do Sul state (31°20'S – 48° 40'W) and three new species: one separated from *E. oxyaster* (Galapagos), which is probably its sister-species; another from *E. topsenti* (Azores), based on morphology of aspidasters, and a third, which possesses globose aspidasters similar to the sterrasters of *Geodia*. These globose aspidasters unite a species group which also includes *E. topsenti* Lendenfeld, 1903, *E. geodioides* Burton & Rhao, 1932 (Indian Ocean) and *E. polyaster* Lendenfeld, 1907 (South Africa). Moreover, currently under study are specimens of *Geodia* from the Brazilian coast possessing "sterrasters" identical to the aspidasters of *Erylus*. Therefore, the term "aspidaster", which characterizes *Erylus*, should probably be changed to sterraster because of priority.

Key words: *Astrophorida, Geodiidae,* Erylus, *revision, new species, taxonomy, brazilian coast*

THEONELLAPEPTOLIDES FROM THE DEEP-WATER NEW ZEALAND SPONGE *LAMELLOMORPHA STRONGYLATA*

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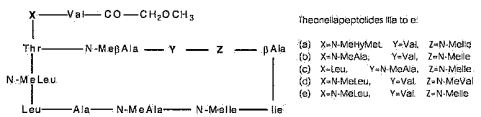
The deep-water marine sponge, *Lamellomorpha strongylata,* was collected by benthic dredging at 80 m on the Chatham Rise (200 km off the East Coast of the South Island of New Zealand). Besides the previously reported calyculins, calyculinamides and swinholide H, five new tridecapeptides, theonellapeptolides IIIa, b, c, d and e, were obtained.

The following strategy was used for determining the structures of the theonellapeptolides.

- the amino acids were established by GC/MS following acid hydrolysis and derivatization.
- methanolysis gave a linear peptide, which was sequenced by tandem mass spectrometry.
- isobaric residues were distinguished by 2D NMR experiments.
- detailed analysis led to the complete NMR assignment.
- the absolute stereochemistry of IIIe was determined by X-ray crystallography coupled with chiral HPLC.
- the stereochemistry of the other peptides were established by an LC/MS method.



Theonellapeptolides IIIb, c, d and e showed mild cytotoxicity against P388 cell line, but IIIa was very much less cytotoxic. This implied that the second residue from N-terminus (X) plays a key role in maintaining bioactivity.

A comparison with the known theonellapeptolide Id suggested that the crystal structure of IIIe is similar to that of Id although four residues are changed and the ring size is 36 in IIIe, not 37 as in Id.

The theonellapeptolides from the I and II series have all been isolated previously from Lithistid sponges, while those from the III group are nominally from a different sponge order. A key question still to be adressed is whether or not all three groups of peptolides have a similar, or comparable, symbiont origin?

Key words: *peptolides amino acids, nmr spectroscopy, lc/ms, gc/ms, chiral hplc, X-ray crystallography, symbionts, Lithistida,*. Theonella sp. , Lamellomorpha strongylata *New Zealand*

AN EVALUATION OF BIOCHEMICAL, MORPHOLOGICAL AND CYTOLOGICAL DATA SETS FOR THE PHYLOGENY OF THE HOMOSCLEROPHORIDA (PORIFERA, DEMOSPONGIAE)

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A cladistic evaluation of the phylogenetic information content of three independent data sets (allozymes, morphology, and cytology) is made in a phylogenetic analysis of ten Mediterranean species

representing four genera of the sponge order Homosclerophorida (Demospongiae). Two aspiculate genera (*Oscarella* and *Pseudocorticium*) and two genera with siliceous skeletons (*Plakina* and *Corticium*) were studied to evaluate the validity of the traditional classification with two families Plakinidae and Oscarellidae, respectively, with and without skeleton. *Discodermia polydiscus* (Tetractinomorpha) was used as the outgroup. Consistency indices, taxonomic congruence, character congruence and randomisation tests were used to analyse congruence within and between data sets.

The allozyme data set produced poorly resolved and highly incongruent trees, probably reflecting saturation of homoplasies due to high genetic divergence between the species studied. Allozymes were informative only at the infrageneric level. Cytological data showed intermediate resolution and was the only set that supported the monophyly of the Oscarellidae, although it also supported the paraphyly of the genus *Plakina.* Cell composition was more informative for the aspiculate species and at the deepest branches, being complementary to the morphological set. The morphological and combined analyses were both completely resolved and well supported, except at the most basal branches. These sets suggested topologies which differed only in the placement of *Pseudocorticium jarrei* or *Corticium candelabrum* as the most basal ingroup branch. The allozyme, morphological and combined sets supported the non-monophyly of the Oscarellidae. The morphological and the combined sets were considered more consistent and accurate than the other sets, but the monophyly of Oscarellidae supported by the cytological set cannot be completely discarded.

Key words: *Homosclerophorida, phylogeny, allozymes, morphology, cytology*

ANTIMICROBIAL ACTIVITY OF CARIBBEAN SPONGE EXTRACTS

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Marine sponges produce a diversity of unusual chemical compounds, but the ecological functions of these metabolites remain largely unknown. Organic extracts from 33 Caribbean sponges were assayed against a panel of 8 marine bacterial strains to determine if sponge secondary metabolites have ecologically significant antimicrobial effects. The test panel was comprised of an opportunistic pathogen (*Vibrio parahaemolyticus*), a common fouling bacterium (*Deleya marina*), and strains isolated from seawater and healthy and necrotic Caribbean sponges. Extracts were tested for antibiotic activity at concentrations that were volumetrically equivalent to those found in sponge tissues (i.e., whole-tissue concentrations). Bioassay results revealed that 16 species extracts (48% of those tested) exhibited antibiotic activity against at least one bacterial isolate and that the necrotic sponge isolates were the most sensitive test strains (inhibited by 40% of the extracts). Extracts from *Amphimedon compressa, Amphimedon erina, Aplysina lacunosa, Ptilocaulis spiculifera* and *Teichaxinella morchellum* inhibited the largest numbers of test strains and exhibited the most potent antibiotic activities with values frequently exceeding that of the control antibiotic (Gentamicin). The pattern of antimicrobial activity was different for 15 of the 16 active species indicating that diverse taxa do not produce similar antibacterial metabolites. In total only 23% of the extracts/bacterial interactions tested generated antimicrobial activity indicating that conspicuous members of the Caribbean sponge community do not generally produce broad-spectrum antibacterial metabolites. All the extracts from species that exhibited antibacterial activity also deterred feeding by reef fish in a previous study, suggesting that some secondary metabolites may have evolved with multiple functions. Stevensine, a compound from *Teichaxinella morchellum* known to deter feeding by predatory reef fishes, exhibited weak antibacterial activity, suggesting that this potent feeding deterrent is not solely responsible for the antimicrobial activity detected in the crude sponge extract.

Key words: *antimicrobial, antibiotic, secondary metabolites, chemical defense, ecology*

A TAXONOMIC KEY TO THE MARINE SPONGES OF NORTH CAROLINA.

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A taxonomic revision of the marine sponges of North Carolina and adjacent areas was compiled and arranged into a key for identification purposes. Although no new data were reported, corrections were made with regards to taxonomy, nomenclature, jargon, and descriptions. There were 50 genera and 70 species reported. Of these, 9 have been previously described from higher latitudes, 45 from lower latitudes, 6 are endemic to North Carolina, 6 are abundant along the entire eastern seaboard, and 4 are reported in areas other than the western Atlantic. The most recent survey of the sponges of North Carolina was done in 1960 by George Wells, *et al.*. Since then, 30 percent of the taxonomy adhered to by Wells has been revised. Fourteen genera have been assigned to new orders, there have been 4 corrections at the generic level and 5 corrections at the species level. An extensive glossary, including figures, is included for ease of use.

Key words: *systematics, taxonomy, North Carolina, key*

THE RELEASE OF ALLELOCHEMICALS BY THREE TROPICAL SPONGES (DEMOSPONGIAE) AND THEIR TOXIC EFFECTS ON CORAL SUBSTRATE COMPETITORS.

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Three sponge species (*Dendroxea* sp., *Amphimedon* sp. and *Halichondria* sp.) from Mactan Island, Philippines, were shown to release allelochemicals directly into the water. These allelochemicals were demonstrated to be toxic to one or more scleractinian coral species (*Porites* sp., *Pocillopora* sp. and *Acropora* sp.) and, for two of the sponges tested, to one hydrozoan coral (*Millepora* sp.). The five coral species tested were both numerically and spatially dominant organisms at the study site. The toxicity tests included exposing corals bought into the laboratory to water that had been conditioned by the sponges. The responses of each coral species to each sponge allelochemical varied. *Dendroxea* sp. allelochemical was found to be highly toxic (51-75% average tissue death) to both *Pocillopora* sp. and *Acropora* sp., and had only a weak effect (11-25% average tissue death) on *Porites* sp. *Amphimedon* sp. and *Halichondria* sp. were only weakly toxic to *Porites* sp., and moderately to weakly toxic to *Millepora* sp. Neither sponge was toxic towards the other coral species. *Montipora* sp. was not affected by allelochemicals from any of the sponges. Species composition and the occurrence of dead coral around 18 to 25 individuals of each sponge species were recorded. No particular coral species was regularly found adjacent to the sponges. In all cases, although dead coral was noted in many positions around the sponges, those locations in the direction of the current always consisted of dead coral space. This might support, although not confirm, an allelopathic effect. The influence of allelochemicals on the small scale and large scale spatial structuring of coral reefs is discussed.

Key words: *allelochemical, coral reef, toxicity, spatial competition*

OPTIMISATION OF THE GROWTH MEDIUM FOR AN *IN VITRO* SPONGE CULTURE

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The search for useful natural products within the phylum *Porifera* has yielded many compounds with potential value for the pharmaceutical industry and the anti-fouling industry. However, supply of sufficient sponge material has hampered the further development of these compounds. Biotechnological production of sponge biomass may bridge the gap between discovery and commercial production of these sponge metabolites.

In this study, the tropical Indo-Pacific demosponge species *Pseudosuberites andrewsi* (Kirkpatrick) was used as a model organism for the development of a method for *in vitro* sponge cultivation. We managed to maintain a culture of this species for an unlimited period under the following conditions: a continuous flow of artificial seawater, containing 100-250 µM sodiummetasilicate as a source of silica for the sponges, a reasonably constant temperature (annual variation between 23 and 28°C) and a nearly constant salinity and pH. Batch cultures of the microalgae *Chlorella vulgaris* and *Rhodomonas* sp. were regularly added as a food source. It was observed under the microscope that these algae were actually taken up by the sponges.

Three growth experiments were carried out in which explants of *P. andrewsi* were fed continuously with either *Chlorella*, or *Rhodomonas*, or a combination of both algae. Differences in growth rate between these explants will indicate a quality difference between the three food regimes. Two-dimensional growth of the explants was monitored by photography. Preliminary results suggest that growth can be considerable (up till 30% per day). However, daily variation and the variation between different explants was much larger than differences between the three food regimes. Some explants did not grow at all. Experiments are under way with larger explants, of which the underwater wet weight will also be monitored as an alternative measure for growth.

We are currently studying how we can enhance the growth rates of the sponges. An important factor in this respect is the quality of the food source. Generally, an important quality aspect for algae as food in aquaculture is their fatty acid composition. Especially Poly-Unsaturated Fatty Acids (PUFA's) are vital for many physiological functions in living organisms. Most animals are hardly able to synthesise PUFA's, and obtain these compounds from their food. Hence, the PUFA-composition of the algae that are fed to the sponges should be in agreement with the metabolic requirements of the sponges. Samples of both sponges and algae were taken to determine their fatty acid composition. Preliminary results suggest an important role for 18:1 oleic acid as a building block for PUFA's in *P. andrewsi*.

Key words: *In vitro cultivation,* Pseudosuberites andrewsi, *growth measurements, fatty acids*

RELATIONSHIP BETWEEN SPONGES AND A TAXON OF OBLIGATORY INQUILINES: THE SILIQUARIID MOLLUSCS.

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The associations among sponges and marine invertebrates are manifold and of different type, but just a few of them may be considered as compulsory. Some mesogasteropod molluscs are adapted to live embedded into a matrix, which can be sediment, coral or sponge tissue. The latter is the case of Siliquariid molluscs which can be considered as obligatory inhabitants of sponges, because no free living species are known, and they were always recorded either embedded or partially covered by a host

sponge. Siliquariidae is a small family (two genera and about fifteen species) with a circum-tropical and temperate distribution. The scant information till now available on their association with Porifera is limited to not more than 15 records, mostly by spongologists, because malacologists usually store dried specimens with accurately polished shells, with a few, if any remnants of the host sponges.
We have analysed in this study about 50 sponge specimens hosting Siliquariids coming from the Mediterranean Sea, the Eastern Atlantic, New Zealand, Philippines and New Caledonia at depths ranging from 10 to 440 m.

A close species specific association has not been till now ascertained, however Siliquariid molluscs are hosted by a restrict number of sponge families. It seems probable that the characteristic required by the host molluscs is a compact and rigid sponge skeletal structure, resulting from a radial organisation or from a high spicule density. Siliquariids, in addition, are mainly deep-water organisms and are so associated to the sponges which are most frequently present in that environment.

When Siliquariid larvae settle on the sponge surface their larval shells (protoconchs) are soon partially overgrown by the host. As soon as the mollusc begins its development it opens a slit all along its length. The relationships between the sponge aquiferous system and the mollusc occur through this slit, and have been studied by x-ray photographs and resin injections into the system in order to obtain casts.

The associated animals may show different growth rates. The mollusc, however, is able to conform to the sponge development by transversally cracking the shell and varying its curvature. The main shell aperture, in fact, leading to the mantle cavity, has to be maintained on the sponge surface. The water flow enters this aperture, is filtered by the gills and then expelled through the slit. Castings have demonstrated that the mollusc is able to modify the shape of the longitudinal slit adapting it to the exhalant canals. The slit becomes transformed into a series of contiguous holes that communicate with the exhalant canals. This behaviour was also observed in two *Tenagodus* species hosted by the same *Spongasorites* specimen, which, notwithstanding their different size, modify the slit and hole dimension in order to maintain alike the outflowing area to match with the sponge aquiferous system.

It seems clear - at this stage of the study - that the main advantages of this form of associated life as protection against predators, defense from sediment clogging of the slit, increased feeding efficiency are in favour of the molluscs. Minor benefits, apart from an increased water outflow and energy saving, can be inferred for the host sponge. The latter, however, seems not to be negatively affected by the Siliquariid presence, and is able to maintain, through its plasticity, its original skeletal structure.

This form of strict and integrated association between filter-feeders may be correctly interpreted as commensalism, because a direct food uptake by the sponge, from a water flow already passed through the mollusc ctenidia and contacting just the exhalant canals seems improbable, whilst even the sponge certainly gets some advantages from the remarkable increase of water circulation all around its body.

Key words: *Siliquariids, association, commensalism, adaptations, behaviour*

PREDATION ON CARIBBEAN SPONGES: THE IMPORTANCE OF CHEMICAL DEFENSES.

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The conventional view has been that the impact of predation on Caribbean reef sponges is minimal: generalist predatory fishes are deterred by sponge spicules and chemistry, while the few spongivorous fishes are "smorgasbord" feeders that circumvent chemistry by eating small amounts of many different sponge species. New data suggest that this traditional view needs to be re-examined. Generalist predatory fishes are deterred by chemistry, but not by structural elements, toughness, or nutritional quality of sponge tissue. Spongivorous fishes are not smorgasbord feeders, but instead choose to eat chemically undefended sponge species. Transplantation experiments reveal that the grazing activity of spongivorous fishes restricts certain sponge species to refugia, including cryptic habitats on the reef and

mangrove and grassbed environments, where these fish are absent. Chemical defense plays an important role in the ecology of sponges on Caribbean reefs.

Key words: *chemical defense, predation, Caribbean, ecology*

RESOURCE PARTITIONING BY CARIBBEAN CORAL REEF SPONGES: IS THERE ENOUGH FOOD FOR EVERYONE?

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Sponges are known to graze primarily on the ultraplankton fraction (plankton < 5 µm) of the water column community and have been implicated as the primary coral reef consumers of ultraplankton. On Pacific reefs, 90% of the ultraplankton is removed from water that passes over a reef and it has been suggested that this is the result of sponge grazing. In the Caribbean sponges are the dominant benthic invertebrate. Concurrent with this high biomass the sponge community is very diverse with morphologies ranging from encrusting to massive. The high abundance and species diversity of sponges coupled with oligotrophic conditions common to coral reefs could require partitioning of food resources between the sponges. In addition, height above the bottom is theorised to significantly impact food availability and thereby grazing.

I characterised the diet and retention efficiency of three cooccuring species of sponge at Chub Cay Reef, Bahamas ($25^{o}22'82"$ N, $77^{o}51'93"$ W). The erect tube sponge *Callyspongia vaginalis*, the mounding sponge *Spongia tubulifera*, and very small *Aplysina fistularis* were conspicuous and common members of the benthic community and had mean heights above the substrate of 22.5, 7.0, and 1.2 cm respectively. Ambient and exhalent current water samples were collected by snorkelers and analysed for ultraplankton using flow cytometry. *C. vaginalis* retained only *Synechococcus*-type cyanobacteria with an efficiency of 90%. In contrast, the diets of *S. tubulifera and A. fistularis* were more reflective of the overall water column community consisting of heterotrophic bacteria, *Prochlorococcus*, *Synechococcus*-type cyanobacteria and autotrophic picoeucaryotes. *S. tubulifera* had retention efficiencies of 41, 29, and 86% for heterotrophic bacteria, *Prochlorococcus*, and *Synechococcus*-type cyanobacteria respectively. Retention efficiencies were highest for *A. fistularis*, the smallest sponge, with 96% for heterotrophic bacteria, 95% for *Prochlorococcus*, 99% for *Synechococcus*-type cyanobacteria and 100% for autotrophic picoeucaryotes.

It is interesting to note that food availability increased closer to the benthos. Ultraplankton abundance was extremely low (< 10^{4} cells ml^{-1}) 22 cm above the bottom and availability increased closer to the benthos such that an order of magnitude more ultraplankton cells were available to *S. tubulifera* and *A. fistularis*. Overall low abundance of food particles 22 cm above the benthos may be preventing effective capture by the choanocytes. Interspecies competition for food resources is most likely the cause of the resource partitioning found at this reef rather than competition between sponges.

Key words: *ultraplankton, feeding, resource partitioning, Caribbean*

MESO-CENOZOIC HISTORY OF THE SILICEOUS SPONGES WITH RIGID SKELETON

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Hexactinosa and Lychniscosa (Hexactinellida, Hexasterophora) and Lithistida (informal group of Demospongiae with fused choanosomal spicules called desmas) having fused skeleton, are among the

most common fossils in the rocks of certain ages. The oldest Hexactinosa are known from the Upper Devonian of Poland but the next fauna of this type is known from the Upper Triassic, while the oldest Lychniscosa occur in the Middle Jurassic (Bajocian) of southern Hungary. Today, Hexactinosa are still an important element of relatively deep-water environments mostly in tropical areas, but common also in colder waters. Lychniscosa, on the other hand, which are represented by numerous taxa during the Late Jurassic till Tertiary, are today a relict group represented only by 3 species and 2 genera occurring exclusively in tropical (but cold) deep-waters. Some lithistid groups (Megamorina, Rhizomorina) have clearly Paleozoic roots, while the rest (Tetracladina, Dicranocladina, Didymorina and Sphaerocladina) originated during the Mesozoic; among them only Didymorina are extinct.

The distribution of large faunas of bodily preserved (intact) fossils sponges during the Meso-Cenozoic is rather punctuated and limited to certain periods of time: the largest known periods for such faunas are associated with the Upper Jurassic, Upper Cretaceous and the Miocene rocks. The interpretation of the evolution of these sponges has been difficult due to such large gaps separating these faunas and the fact that at the genus level, comparisons are made of very different taxa. Such patterns of distribution have been interpreted, at least for lithistids, by Rigby (1983) as the result of "selective preservation and discovery, not one of original limited diversity and density". This interpretation has found partial support in more recent discoveries of the important Upper Triassic (China), Middle Jurassic (Spain, Hungary, India), Eocene (Spain, Italy, Turkey) and Miocene (Spain) faunas of such sponges in various parts of the world. Generally, however, the pattern of punctuated record of large sponge faunas is preserved. These new discoveries, combined with earlier known occurrences, clearly points to the importance of sea level in controlling such distribution: all large faunas of siliceous sponges with rigid skeletons are associated with the periods of high sea level during the Mesozoic and Tertiary (and it seems that this pattern is valid also for the Paleozoic as well in the case of Hexactinosa). This may be due to sponges colonising new, vast, relatively deep-water areas, which were not available during lower sea level periods; sponges of these groups existed only in relatively rare refugia, as in many cases today.

It has been long known that numerous Recent hexactinellid genera occur in the Upper Cretaceous rocks; the recently discovered sponge faunas strengthen this pattern by showing the presence of the genera known from the Upper Jurassic also in the Triassic, and other Jurassic and Cretaceoues genera in the Tertiary, thus pointing to the extremely conservative nature of these sponges at the genus level. The Eocene faunas of both lithistids and hexactinellids with rigid skeletons show close affinities rather with the Upper Cretaceous faunas, than with Recent ones, indicating that K/T boundary was relatively less important for these groups of sponges, than for other organisms.

Key words: *Hexactinosa, Lychniscosa, Lithistida, Mesozoic, Cenozoic, history.*

SETIDIUM OBTECTUM SCHMIDT, 1879 REDISCOVERED

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The rhizomorine lithistid genus and species *Setidium obtectum* has been established by Schmidt (1879) based on a unique specimen dredged off Havana from the depth of 128-240 fathoms. Its curious morphological character, namely the occurrence of numerous bunches of very long oxeas protruding from the inner (upper surface) of the cup, makes it a very characteristic form. As a result of its incomplete description, and the fact it had never been found again, its systematic position remained obscure.

Lendenfeld (1903) regarded it as a synonym of *Leiodermatium* Schmidt and placed it within the family Leiodermatidae Lendenfeld, which encompasses rhizomorine lithistids without microscleres. Van Soest and Stentoft (1988) placed *Setidium obtectum* among rhizomorine lithistids of the family Siphonidiidae Sollas, which is characterised by an ectosomal skeleton composed of desmas without zygosis and no microscleres.

Recently, new, rich material of this sponge has been discovered in a collection of sponges housed in the RSMAS (Miami University), dredged in several settings of the Caribbean. The investigation of this material, and its comparison with the holotype preserved in the Schmidt collection at the Museum of Comparative Zoology, Harvard University, permitted me to establish taxonomic position of this genus and species. Its choanosomal desmas are typical dorny rhizoclones, while ectosomal spicules are mostly amphioxes forming a dense tangential layer; they show all transitional forms to rhizoclones. Numerous dorny sigmaspire microscleres have been found both in the holotype and the new material. As a result *Setidium obtectum* Schmidt, 1879 should be attributed the family Scleritodermidae Sollas. The presence of sigmaspires and rhizoclones, as well as the ectosomal skeleton of amphioxes makes this genus very close to the genus *Scleritoderma* Schmidt 1879.

Key words: *Rhizomorine lithistids,* Setidium, *systematic position*

RESEARCH ON THE NATURAL DYNAMICS OF SOME STRUCTURALLY DOMINANT TROPICAL SPONGES AND OTHER SESSILE FAUNA

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We are measuring the dynamics (growth, mortality, recruitment, and reproduction) of large sessile fauna (including sponges, gorgonians, alcyonarians and corals) that dominate and provide structure on patches of seabed habitat in open shelf waters, 20-50 m depth, at several sites in the Great Barrier Reef region. Sites have been chosen to contain several representative patches of benthos habitat. An ROV is being used to document dynamics, including: mapping the large sessile fauna in habitat patches at each site; tagging several dominant species; measuring growth and mortality rates through time; observing the occurrence of new small individuals for measurements of recruitment; taking small biopsy samples to confirm taxonomy; and histological examination in the laboratory to determine reproductive strategies. By tagging a full size-range of individuals of each study species, we anticipate being able to estimate life-time growth curves in three years. Models of the dynamics of sessile fauna will be constructed and these will also estimate how fast seabed habitat might recover in new reserve areas. This study will also document the usage of living seabed habitat by key fish species.

Key words: *sessile fauna, dynamics, growth, mortality, recruitment, ROV, video, tagging*

THE IMPACT OF TRAWLING ON SOME TROPICAL SPONGES AND OTHER SESSILE FAUNA

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Following a replicated repeated-trawl depletion experiment, which removed 70–90% of the initial biomass of sessile fauna, non-destructive methods have been used to measure recovery of the structurally dominant species. To date, three post-impact surveys have been completed (1 month, 1 year, 2 Years) on 6 trawled swathes and 6 controls. These involved quantitative video observations from a tow-sled and from an ROV with positioning precision of ≤2 m. More than 80 taxa of sessile fauna were recorded and about 15 taxa, including sponges, gorgonians, and alcyonarians and hard corals, were abundant enough for analysis. The attributes measured for each organism were species, position, size (height, width, and area), and condition (proportion intact, dead, or encrusted). These data enabled analyses of density, size, condition and species composition, with high precision. We have demonstrated statistically that the methods are very powerful for detecting recovery on the trawled tracks, but the time series is, as yet, too short to identify any recovery. Nevertheless, in treatment vs control or before vs after comparisons, all taxa analysed for which sample size was adequate, showed significant impact due to trawling, for at least one or more of the measured attributes. For all benthos

taxa combined, the rate of decrease in benthos density with trawl intensity corresponded to ~10% per trawl, so that in areas trawled 15 times, the benthos density was only ~25% of that in un-trawled areas. This conclusion was very similar to the overall estimate of removal obtained from depletion-regression analysis of the catch obtained during the repeat-trawl experiment. The results also indicated the nature in which trawl impact was manifested for different organisms with different structure and morphology. The sea whips were most resilient to removal, though they could be damaged. Sponges and soft corals were relatively easily removed and hard corals were easily broken. The gorgonians were intermediate and variable in resilience. The interaction between these differences and trawl impact caused a marked change in, and degradation of, the community composition of the seabed habitat.

Key words: *sessile fauna, impact, condition, recovery, trawling, ROV, video, resilience*

SPONGE SHAPE AS A TAXONOMIC CHARACTER: THE CASE OF *SPONGIA OFFICINALIS* AND *SPONGIA AGARICINA*

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The extreme phenotypical plasticity is one of the main characterizing traits of Porifera at all organizational levels (West Heberard 1986, 1989; Gaino et al., 1995). Body shape is highly variable both in time and space particularly among demosponges living in shallow waters with high selective pressures exerted by fluctuations of water movement and light, by substrate stability and shape and by spatial competition (Bidder, 1923; Hartman, 1950; Reiswig, 1973; Wilkinson & Vacelet, 1979; Palumbi, 1986; Barthel, 1986, 1991; Pansini & Pronzato, 1990; Gaino et al., 1991; Pronzato & Pansini, 1994). Moreover external morph can be influenced, in some cases, by the age of the sponge and therefore by its life cycle (Barthel, 1986; Manconi & Pronzato, 1991).

Spongia officinalis is reported as being very variable both in colour and body shape not only by spongologists (de Laubenfels, 1948; Storr, 1976; Pronzato et al., in press), but also by fishermen that encounter difficulties distinguishing it from other species as *S. zimocca* or *Ircinia spinosula* and *Cacospongia scalaris*. On the other hand *S. agaricina* displays such a peculiar shape that it is easily identified also by students of zoology. Such a different degree of body shape variation in these two close species is an intriguing case.

The present paper aims to investigate the temporal and spatial evolution of body shape of *Spongia officinalis* and *Spongia agaricina* in order to ascertain if and what external morphological traits can have a diagnostic value to clarify their taxonomic status. The trait of body shape could considered as the result of several associated sub-traits as growth in height; growth in width; number and shape of oscules; distribution of oscules; presence and distribution of lobes; differentiation of inhalant and exalant area; presence and distribution of conules. A comparative analysis was performed between Eastern and Western Mediterranean populations of *S. officinalis*. The rarity of *S. agaricina* in the Ligurian Sea meant it was not possible to carry out a comparison with the studied Aegean population.

The following material was considered: *S. officinalis*: 56 specimens collected at 15 m of depth on hard bottoms by diving at Portofino; 63 specimens at 5-10 m depth on hard bottom around the island of Crete; 50 living specimens from Portofino : 25+25 sponges settled, respectively, at 8-10 and 20-25 m of depth. *S. agaricina:* 37 specimens collected, at 50-100 m of depth, on sand bottoms by gangava around the island of Kalymnos.

Living sponges were monitored by under-water photography from 1994 to 1995 in September, July and November to follow the temporal evolution of body shape. To avoid morphological variations linked to pumping activity and to rhythmic contraction/expansion processes, cleaned skeletons were studied at the laboratory. The identification of specimens was performed by SEM at the level of the skeletal net.

Results highlight that *S. agaricina* displays a constant body plan in spite of a wide size variation within the considered population. In the case of *S. officinalis* a constant body plan is displayed by living *sponges* within the same population at different depth. *Spongia agaricina*: body shape is relatively constant; cup-shaped with distal margin ondulati, lateral profile trapezio-like; height and max diameter (at the cup aperture) show an isometric growth; diameter at the base seems to be allometric with respect to the other growth axis and constrained by the substrate morphology; the fan-like shape is absent, with the exception of two specimens, in this population; the distal margin is constantly elliptic with a low difference among the two axis; the irregular growth of the distal margin seems not linked to sponge size. It is possible to hypothesize a shape shifting during ageing from an opened cup toward a tronco-conic shape (*un po' tirata*).

Keywords. *Bath sponges, body shape, variability,* Spongia officinalis, Spongia agaricina.

SPONGE FARMING IN THE MEDITERRANEAN SEA: NEW PERSPECTIVES

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Some Mediterranean sponges, belonging to the genera *Spongia* and *Hippospongia*, are harvested for commercial purposes. Recently, the synergetic action of a widespread epidemic8 and over fishing, strongly reduced the density of sponge banks, drawing these species to the brink of extinction. Moreover, the recovery of banks was incomplete and needs a long time. A simple solution to the problem is sponge farming. This activity has been tested since the beginning of the century; and recently, in Cuba and Micronesia Islands, sponge-culture was initiated for commercial purposes. Because of their active pumping, sponges are able to retain bacteria and organic matter (SOD) from the entire water-column in littoral marine environments; this ability could be basic for an integrated mari-culture. Floating-cages used in fish farming release a lot of organic wastes that can be recycled as a rich source of food for surrounding commercial sponge intensive cultures.

Our goals are: i) eutrophication control; ii) sponge production for commercial purposes; iii) reduction of the fishing effort on stressed natural sponge populations; iv) re-establishment of local wild stocks by natural dispersal of larvae from reared specimens. As for commercial sponges farming in eutrophic waters, some experiments were carried out in different Mediterranean sites (Ligurian Sea and Dodecanese Archipelago). The rearing system is made by small horizontal, modular structures moored to the sea bottom; each plant supports about 100 sponge fragments of 5-10 cm in diameter, strung on lines and spaced by plastic tubes. Three species were tested: *Spongia officinalis, S. agaricina* and *Hippospongia communis*. The last seems to be the most resistant, with recovery of the external membrane lasting about one week; a 10% mean mortality was observed. Within this Mediterranean sponge farming plant, it is possible with pluri-annual planning to produce some millions of sponges/year.

Another recent economic challenge involved an increasing number of sponge species producing several substances of pharmacological utility. An experiment on a group of five species (*Agelas oroides, Aximella damicornis, Petrosia dura, Ircinia foetida* and *Cacospongia mollior*) was carried out, in order to collect preliminary results about the response of these species to the farming. Our work took into account regeneration processes, by SEM observations, both in species characterised by a spicular skeleton, and in those with spongin fibres. Generally, the recovery lasted a few days with at least two different processes: i) sponges with pinacoderm cells with typical archaeocyte features migrate in the undifferentiated outgrowth region enabling the formation of exo- and baso-pinacoderm; ii) in horny sponges, a very high number of small globular elements are able to produce the external membrane. The well known chronic morphogenesis of sponges support the possibility of clonation for a group of species chemically interesting. Their high commercial value could exert, in the future, an excessive pressure on many wild populations. A controlled rearing system can however avoid the impoverishment of species whose distribution and population structure are poorly known.

Key words: *integrated mariculture, eutrophication control, wildlife conservation, SEM, recovery*

CHEMICAL DEFENSES OF THE CARIBBEAN SPONGES *APLYSINA FULVA* AND *A. INSULARIS*.

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Sponges of the genus *Aplysina* (Family Aplysinidae, Order Verongida) are important elements of Caribbean coral reef communities due to their abundance and the variety of habitats they thrive in. Chemically, they are unique due to the possession of an elaborate series of brominated metabolites derived from tyrosine. Numerous metabolites have been isolated and identified from *Aplysina* spp.; however, their ecological roles have rarely been assessed. *Aplysina fulva* and *Aplysina insularis* are closely related species, commonly found growing concurrently in shallow reef environments of the Caribbean Sea. Crude extracts from both species are strongly unpalatable to predatory reef fish. In order to evaluate the effectiveness of a chemically based defensive mechanism, we performed a series of careful extraction procedures to chemically characterise both species. Crude extracts were purified using Column Chromatography and High Performance Liquid Chromatography. Compounds were identified by NMR spectroscopy and by comparison with data from the literature. Purified compounds were tested at natural concentrations, in aquarium and field assays, to determine the metabolites responsible for the strong anti-feeding activity typical of these sponges. Similarities in the chemistry of *A. fulva* and *A. insularis* and their implication in chemical defense are discussed.

Key words: Aplysina, *chemical defense, bromotyrosine derivatives, predation*

NEW HEXACTINELLIDS FROM THE MENDOCINO RIDGE OFF OREGON, USA.

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Two hexactinellid sponges collected by submersible from the Mendocino Ridge by Dr. Andrew Carey Jr., are representatives of new taxa of Hexactinellida. The first, a nearly complete specimen missing only the basal attachment is a soft cup on a hollow stem, 36cm total height, collected from 2102 m depth. Because it bears distinctive drepanocome spicules and is supported on a hollow stalk it was immediately suspected to be a close relative, if not a specimen, of the poorly known species *Trachycaulus gurlitti* Schulze (Family Euplectellidae, subfamily Corbitellinae) obtained by the Challenger Expedition in mid south Pacific. Comparison with the very fragmentary type specimen of *T. gurlitti* in the British Natural History Museum has shown that the two cannot belong to the same genus. The descriptive name chosen for this form, *Nubicaulus* n. gen., is combined from nubis = cloud and caulus = stem. The species, *N. careyi*, named after its collector and constituting the type species of the genus, is excluded from related genera by the presence of spirodiscohexasters and aspidoplumicomes, but the absence of floricomes.

The second specimen, a sheet, 21 x 28 cm, by 1 cm thick, represents a part of a larger fan- or cup-shaped specimen (video and collection records not yet available at this writing). It bears a dense marginal fringe of sceptres, pentact pinules with distinctive cylindrical pinulus, coarsly-thorned mesohexactins, large primary pentacts, uncinates and two categories of amphidiscs. The lower part of the specimen, and thus the important basal spicules, are unfortunately missing. The specimen agrees in general characters only with those of the genus *Poliopogon* (Amphidiscophora, family Pheronematidae), to which it has been assigned. It does not conform to the detailed characters of any of the 3 presently recognised members (2 species plus one subspecies) of this genus, and is designated as a new species, *P. mendocino*.

These new taxa will be deposited to the collections of the California Academy of Sciences in San Francisco, California.

Key words: *Hexactinellida, new species, new genus, Mendocino Ridge, submersible*

"MUD MOUND" STRUCTURES AND CORALLINE SPONGES FROM THE OSPREY REEF (QUEENSLAND PLATEAU, CORAL SEA, AUSTRALIA)

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The Osprey Reef is located at the north western tip of the Queensland Plateau. The Osprey Reef represents an open oceanic reef platform on a drowned carbonate platform (Queensland Plateau). The metamorphic basement was drilled in a depth of about 450 m below sea floor (cf. Betzler et al. 1995).

The reef caves of the Osprey Reef were studied during two one-week expeditions in 1995 and 1996 with the RV Gwendoline May. At the Osprey Reef a very patchy distribution of coralline sponges was observed. At some dive sites the caves and even the reef internal surfaces (RIS) were free of them, a few hundreds of meters away, the walls of the caves and the RIS were covered with coralline sponges. The reasons for this very patchy distribution are not clear at the moment. The structure of the cave community is equal to that of the GBR. The caves with an abundant coralline sponge fauna are mainly located at a water depth between 15 and 20 m.

At all sites, *Astrosclera* was the most dominant sponge. As at the outer barrier reefs (GBR), it sometimes lives in semi-dark conditions and is colored red or green. Its size never exceeds 5 cm. *S. (Acanthochaetetes)* was rare at Osprey Reef compared to the GBR.

We found two new species of colonial variations of the 'sphinctozoan' sponge *Vaceletia* at the Osprey Reef (Reitner & Wörheide 1995; Wörheide & Reitner 1996). These new types are common in most of the caves and are mostly associated with *Vaceletia crypta*. They only occur in the darkest parts of the caves.

This type of coralline sponge shows similarities to Permo-Triassic reef building sphinctozoans.
In a depth between 250-300 m aggregates of medium sized mound structures were observed. These structures are located at the northwestern steep escarpment of the reef and have a of length ca. 10-20 m, a width of 1-2 m, and probably 0,5-2 m height. The surface is rigid and sometimes overgrown with sponges and gorgonians. Between the elongated mounds groove systems are developed where reef sediments are transported. Little sediment 'snow' is fixed by the mound surfaces. These mound structures are comparable with micritic sponge reefs known from Mesozoic reef sites.

Key words: coralline sponges, mud-mounds, Vaceletia, *reef-building sphinctozoans, Osprey Reef.*

NEW APPROACHES TO THE BIOMINERALIZATION PROCESSES OF CALCIFIED SKELETONS IN CORALLINE DEMOSPONGES

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Biomineralization of calcareous basal skeletons in coralline sponges is a strongly phylogenetically convergent character. However, the basic mineralization process is ancestral and exhibits similarities with mineralization processes known in bacterial biofilms and organomineralization via controlled taphonomy. The main biocalcification events in the phylogenetically distinct taxa *Vaceletia* sp., *Astrosclera willeyana*, *Ceratoporella nicholsoni*, and *Spirastrella (Acanthochaetetes) wellsi* are discussed. *Vaceletia*, a demosponge with a thalamid basal skeleton, exhibits the most ancestral mode to build a calcareous skeleton via controlled taphonomy. The stromatoporoid *Astrosclera willeyana* intracellularly forms eggshaped aragonitic asters in a first step which grow together via an epitactical process. The chaetetid *S. (Acanthochaetetes) wellsi*, phylogenetically the most evolved coralline sponge taxon, forms its unique high-Mg calcitic skeleton in extracellular acidic organic mucilages in the presence of collagen. In all cases the mineralization is controlled by acidic matrix proteins. All aragonitic basal skeletons are characterized by high amounts of Sr and U. In *Ceratoporella nicholsoni* an increase of Mg, Sr and U in the old skeletal parts is observed using ICP-MS based geochemical analyses. The decrease of P concentrations is probably linked to the collaps of the intracrystalline matrix proteins. In *Ceratoporella* two distinct Ca-binding matrix proteins are observed which are enriched in the amino acids asp (20 mol%) and glu (15 mol%) (20kd, 80kd). The uppermost growing zones exhibit relatively light δ13C 3,8 and δ18O -0,4 values in average.

Astrosclera differs in many aspects from the phylogenetically closely related *Ceratoporella*. The newly formed aragonite asters are depleted in 13C (δ13C 3,5) in comparison with the mature basal skeleton. The spherulites are enriched in Sr, P, Li and Mo. In the youngest cementing area of the spherulites the content of 13C increases to δ13C 4,03. *Astrosclera* exhibits two acidic matrix proteins, a small one (17kd) which is controlling the spherulite growth and a large one (120kd) which probably controlls the cementation process.

The young portion of the basal skeleton of *Vaceletia* n.sp. differs from the aragonitic basal skeletons of the other coralline sponges. Mg and P is extremly enriched and the carbon isotope composition is relatively light (δ13C 3,8). *Vaceletia* exhibits 5 acidic matrix proteins.

Acanthochaetetes has the most evolved basal skeleton which exhibit only one acidic matrix protein. The basal skeleton is a Mg rich calcite (19-20mol% MgCO3). The uppermost growing zones exhibit light δ13C values (2,65). The simultaneously growing inner cements have δ13C 3,03. Based on the measured geochemical and isotopic data a vital effect during the early formation of the basal skeletons is very probable. The old or mature basal skeleton portions exhibit in all cases signals of a mineralization in equilibrium with the ambient sea water.

Key words: *coralline sponges*, , Vaceletia, *reef-building sphinctozoans*, Acanthochaetetes

NEW COLONIAL *VACELETIA*-TYPE SPHINCTOZOAN FROM THE PACIFIC

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Three new types of a Recent colonial sphinctozoan coralline sponge are presented. All types show close relationships to the taxon *Vaceletia*, a non-colonial form from Indo-Pacific reef caves. The first two types were discovered in shallow water reef caves of the Osprey Reef, which is located on the northern Queensland Plateau in the Coral Sea. The sponges are common in these caves. The third type of colonial sphinctozoan was found only at two localities at the North Astrolabe Reef and Great Astrolabe Reef of the Fiji Islands. This variation shows similarities with a previously described deep water variation of *Vaceletia* from New Caledonia.

The first two variations of colonial *Vaceletia* from the Osprey Reef show more show more similarities to the cryptic, non-colonial form *Vaceletia crypta* from reef caves of the Great Barrier Reef and reefs of the Indo-Pacific than to the deep-water colonial species described by Vacelet (1988) and Vacelet et al. (1992) from New Caledonia. The third variation from the Astrolabe Reefs of the Fiji Islands is more similar to this deep water variation from New Caledonia. All three variations will be described elsewhere in detail, investigations are in progress.

The discovery of these three new colonial variations from shallow water reef caves of the SW Pacific shows that the environment of this colonial sphinctozoan sponges is not restricted to the deep water

Sphinctozoan sponges were reef building organisms in the Permo-Triassic. They are chambered calcified sponges with morphological similarities to Cambrian Archaeocyaths. The *Vaceletia*-type of coralline sponges occured first in the middle/late Triassic. The sphinctozoans were considered to be rare scince the end of the Triassic. They were thought to be extinct scince the end of the Cretaceous until the "living fossil" *Vaceletia* was discovered by Vacelet (1979) in the Indian Ocean.

The solitary, non-colonial form *Vaceletia crypta* has no reef building potential and is only sparesly found in the darker areas of Indo-Pacific reef caves.

The newly found colonial variations of *Vaceletia* from shallow water reef caves retain a colonial growth mode and a reef building capability. They provide therefore clues for the understanding of the modalities of skeletal construction and biocalcification as well as the ecology of Permo-Triassic sphinctozoan sponges.

Key words: *coralline sponges, mud-mounds*, Vaceletia, *reef-building sphinctozoans, Osprey Reef.*

A STRANGE SUBERITID DEMOSPONGE FROM A MARINE HIGH ALKALINE CRATER LAKE (SATONDA ISLAND, INDONESIA)

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The Satonda crater lake is up to now the only known "marine" lake with an increased alkalinity compared to seawater. Therefore, the lake contains a decreased amount of Ca^{2+}. Its pH values about 8.5-8.6. The lake was originally filled with freshwater, which is evident from peat deposits (3,150 ^{14}C-yrs BP). Shortly after the lake was rapidly filled with seawater and a marine fauna had established.

Large input of organic matter has caused intense oxygen consumption and, as a result, the bottom water of the lake became anaerobic. Thus, an intense sulfate reduction produced high amounts of bicarbonate ions. The lake became stratified into three water bodies with various salinities separated by two pycnoclines. The surface water body is oxygenated and exhibits brackish conditions. Some portions of the highly alkaline water penetrates through the uppermost pycnocline and increases the alkalinity in the upper oxic water body (alkalinity pump). The upper water body, in addition to having a slightly increased alkalinity (4-5 meq/l), is characterized by a decreased salinity (32 ‰). The algae/microbialite reefs exhibit a vertical development which started with a serpulid framework, followed by loose crusts of the calcified red alga *Peyssonnelia* and thalli of the green alga *Cladophoropsis* calcified by cyanobacteria (microstromatolites). The top calcified layer is formed by a network of *Lithoporella*, *Peyssonnelia* and microbialites. On the top layer the living reef community is located. This special hydrochemical situation gave rise to a very specific and endemic development of the biota. Cyanobacteria and heterotrophic microbes exhibit large diversities in contrast to just one sponge taxon (*Suberites*/*Polmastia* n. sp.). Cyanobacteria of the taxa *Pleurocapsa*, *Phormidium*, *Calothrix*, *Spirulina*, *Microcoleus* and Microcystacea are common.

Sponges are represented by different morphotypes of the hadromerid taxon *Suberites*, which is characterized by tylostyle megascleres only. The dermal layer of the sponge is constructed of plumose bundles of short tylostyles (150-200 µm), the choanosomal spicules are randomly orientated and much larger than the dermal ones (300-500 µm). Most of the observed sponges exhibit a lateral, encrusting growth habit and therefore show a well developed exhalant canal system. The exhalant system is differentiated into star-shaped units ('astrorhizae'-pattern). In each unit the main exhalant canals conjugate in one large osculum. A second type of this sponge type is observed. It exhibits a more or less erect growth habit and does not show any star-shaped outer exhalant system. The sponge is forming tubes with a central osculum also known from the taxon *Polymastia*, which is phylogenetically closely related to *Suberites*. The spicular inventory and spicule arrangements are more or less similar. A lot of color variations are visible from dark green, brown, yellow/brown, and yellow. The different colors are related to microorganisms within the soft tissue. The dark green color is restricted to specimens in extremely shallow water (20-50 cm) and is related to the unicellular green algae.These algae are part of the plankton and filtered by the sponge. The algae lives within the mesohyle of the sponge. The brownish color variation is restricted to few specimens from deeper water (18-20 m) and is related to a higher amount of still unknown mesohyle bacteria (Pl. 2/3). The symbiotic bacteria of the sponge itself are rare and very small (less then ! µm – nanobacteria) (Pl. 2/5). Size and abundance are comparable to those observed in the marine hadromerid coralline sponge *Acanthochaetetes*. In many cases the encrusting sponges form very thin films (ca. 50 µm) growing in interspaces of dead red algae heads. The sponges penetrate large spaces of the dead portions of the algae reef surfaces. They prefer light protected areas, except the algae-bearing specimens. The steep slopes of the red algae reefs are entirely covered by a dense curtain of *Cladophora* colonies down to 15-16 m. Sponges are growing underneath this curtain where light amounts range from 200 to 300 lux. The depth limit of the sponges, noticed in 1993, was 20 m short above the pycnocline.

The investigated sponges are particle feeders. Within vacuoles of archaeocytes remains of diatoms were observed. In all observed cases the sponges are growing on active heterotrophic biofilms. There is a close relationship between the biofilms and the sponge, because ostia are very common in the basopinacoderm (Pl. 2/5). We assume that the biofilms release metabolic products consumed by the sponge. This behavior may explain the enormous lateral growth of thin sponge sheets. Of further significance for the sponge is the ability to build resting bodies. The observed resting bodies are located in small protected cryptic niches between coralline algae or small caverns of 200-500 µm. The resting bodies are hemispherical or sack-shaped and filled with archaeocytes/ thesocytes. The sponge fauna is perfectly adapted to this extreme environment. Referring to additional ultrastructural studies we assume that these sponge types are new taxa restricted to this special environment.

Key words: *suberitid demopsonge, alkaline crater lake, Indonesia*

ORIGIN AND EARLY FOSSIL RECORD OF SPONGES-A GEOBIOLOGICAL APPROACH

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The Porifera are Precambrian active filter feeding metazoans which exhibit a reproductive strategy as known from the Eumetazoa. However, most morphological characters of the sponges differ from those of the Eumetazoa. The constituent character of the taxon Porifera are aggregates of choanocytes which demonstrate a phylogenetic relationship with the protozoan taxon Choanoflagellata. Sponges have various amounts of symbionts that control metabolic processes. As a hypothesis sponges originated from biofilms which were associated with choanoflagellates. First remains of sponges are known from Middle Proterozoic (1,8 by) blackshales (biomarker C30-sterane). First spicules and entirely preserved sponge bodies are known from the Late Proterozoic (Ediacaran, various microbialite reefs). In the early Cambrian all main groups of sponges, including the Calcarea, do exist. During the Phanerozoic six major sponge events are noticed. The first one is represented by the development of the Archaeocyaths in the Lower Cambrian. The occurrence of typical stromatoporoids started in the Ordovician. Rigid hexactinellids are known since the Late Devonian. First modern demosponge taxa occurred after the Late Devonian extinction event. Modern type of coralline sponges, e.g. Ceratoporellida, occured in the Permian and have a diversity optimum in the Late Triassic. The last significant development is seen in the Jurassic - starting point of the fresh water sponges - when some marine taxa (Haploscerlida/Poecilosclerida, ?Hadromerida) have moved into fresh water environments. Sponges were important in reef buildung and are still reef dwelling organisms. Their importance as main reef buildung organisms decreased in the Late Jurassic/Lower Cretaceous when fast growing modern zooxantelle corals became more and important.

Key words: *symbionts, reef-building sponges, fossil sponges*

TAPHONOMY AND PRESERVATION POTENTIAL OF SPONGE TISSUE

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The preservation potential of sponge tissues is mainly controlled by the sponge related bacteria. Hybridization of the associated microbial populations in few modern demosponges (*Petrosia & Chondrosia*) showed that the majority of bacteria are members of the gamma-subclass of Proteobacteria. Using highly specific oligonucleotide probes for detecting sulfate-reducing bacteria, distinct signals were found scattered in native sponge tissue of both investigated sponges. Also other fermentative bacteria are involved in the degradation of sponge tissue. Sulfate reducing bacteria may control the calcification of the sponge tissue during degradation increasing the carbonate alkalinity. Therefore isolated pyrite crystals are common in mineralized (automicritic) sponges tissues. In the surrounding sediment, pyrite is absent or rare. The sponge tissue automicrites are often dark coloured due to statistically distributed very fine pyrite crystals (ca.1µm). Besides the small pyrite, larger crystals often exhibit patchy concentrations or they are arranged in rows. Pyrite formation is probably linked with sulfate reducing symbiontic bacteria in the sponge mesohyle. During early decaying processes of the sponge tissue the internal sponge space is becoming entirely anaerobic which favours the growth of the sulfate reducing bacteria.

This process may explain the rapid calcification of sponge tissue in modern marine microbialites and ancient sponge mud mounds. In mud mounds siliceous sponges contribute to buildup development with considerable amounts of sponge body related micrite produced in place. These sponge container automicrites form during the biodegradation of soft tissues, resulting in various "classical" microbial fabrics. The initial formation of carbonate crystals is controled by reactive organic compounds (macromolecules) during conditions of elevated carbonate alkalinity (ammonification). The resulting carbonate microfabrics correlate with different soft tissue precursors (mesohyle). The mesohyle structure varies from bacterio container (minipeloidal), bacteria bearing/rich in choanocyte chambers (peloidal) to bacteria poor or syncytial structures (aphanites). Intermediate reactive states of organic matter lead also to the in-situ preservation of non-rigid demosponges, which are recognized by spicular architecture, spatially restricted occurrences of unsorted spicules or even by spicule bearing minipeloids, peloids or aphanites. Principally non-spicule bearing sponges should be recognized by the outer, e.g. nodular shape of microbial fabrics. Organically induced automicrites (organomicrites) are high-Mg calcites with an inorganic signature of $\delta^{13}C$ (+3 to 4). The enhanced identification of an autochthonous sponge fauna within mud mounds provides new insight into the nature and origin of these structures. Semiquantitive data of Cretaceous mounds reveal that 50-80 % of mound micrites were produced in place from which up to 60 % of automicrites can be related to metazoans. Therefore, the origin of reactive organic matter is the crucial point to evaluate the pure microbial vs metazoan character of Paleozoic and Mesozoic mud mounds, as well as Precambrian micrites within biostromal and biohermal deposits.

Key words: *mud-mounds, micrites*

EXPRESSION OF HOMEOBOX-CONTAINING GENES IN FRESHWATER SPONGES

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In metazoans, homeobox-containing genes play an important role in spatio-temporal organisation and cellular differentiation in the course of development. They have been identified in organisms from sponges to humans. The study of homeobox-containing genes in sponges is of particular interest as they are considered to be the most primitive metazoans and could contribute to the comprehension of the evolution of Hox genes.

As reported previously, we have isolated and characterised two homeobox- containing genes, *EfH-1* and *EmH-3*, from *Ephydatia fluviatilis* and *E. muelleri* respectively (Coutinho *et al.*, 1994; Richelle *et al.*, 1996). These genes present a great homology of sequence and structure with *prox2* of *E. fluviatilis* (Seimiya *et al.*, 1994). However the first exon is 54bp longer in *E. muelleri* (Richelle *et al.*, 1996).

In the present work, we demonstrate, by Southern-blot hybridisation and RT-PCR, the existence of these genes as well as of *prox1* (Seimiya *et al.*, 1994) in three additional freshwater sponge species: *Spongilla lacustris, Eunapius fragilis* and *Trochospongilla horrida.*

To understand the role of the identified homeobox-containing genes in the development of freshwater sponges, we have undertaken the study of their temporal and spatial expression.

To pinpoint the moment at which these genes are expressed, total RNA was extracted at different stages of development from gemmule germination to the completion of functional sponges and from sponges transferred to natural environment for 3 weeks. RNA was amplified by RT-PCR with specific primers; EmA 1 (Actin gene from *E. muelleri*, Ducy, 1993) was used as a control.

EfH-1 and *EmH-3* are expressed differentially in the course of development in *E. fluviatilis* and *E. muelleri*. The level of expression increases from undetectable transcript levels in gemmules to higher levels in fully developed sponges with a peak of expression just after hatching. This pattern of expression is consistent with a role in cell

differentiation. On the other hand, *prox1* is expressed at nearly the same level throughout development. It is worth noting that *EfH-1, EmH-3* and *prox1* are still expressed in sponges transferred to the field for three weeks. EmA 1 is strongly expressed at the different stages of development, but at a slightly lower level in the gemmules.

Similar experiments performed with hydroxyurea-treated sponges that are devoid of choanocytes give the same results.

Experiments are in progress to determine in which cell types, *EfH-1, EmH-3* and *prox1* are expressed. For this purpose, two cell fractions were isolated on discontinuous Ficoll gradients from fully functional sponges. One is composed of pure archaeocytes, the other contains pinacocytes and choanocytes.

Preliminary results indicate that *EfH-1, EmH-3* and *prox1* are expressed in archaeocytes at the same level as in unfractionated cell populations. In contrast, in pinacocytes and choanocytes, the level of expression is lower and similar to that found in gemmules; however, EmA 1 is expressed at the same level in all the cell populations.

Key words: *homeobox-containing genes, expression, cell fractionation, freshwater sponge, RT-PCR*

THE SPONGES OF PARAISO NEARSHORE FRINGE REEF, COZUMEL, MEXICO

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Although sponges as a group are an easily recognisable life form, in ecological studies the identification of individual species of this phylum can be problematic. The objective of this study was to identify and describe the sponge species of the Paraiso nearshore fringing reef off the island of Cozumel, Mexico. A survey of sponges living within an 80 meter x 40 meter permanent study site was conducted using underwater video. Sponge tissue samples were also collected. A field guide based on morphological characteristics was compiled describing 42 different sponges, representing 9 orders, 18 families and 21 genera of the class Demospongiae. Comparing the results of this study with earlier descriptions of the diversity of this sponge community indicate the importance of correct sponge identifications for accurate evaluation of changes in reef community structure. The results of this study suggest that regional identification guides are necessary for life forms such as sponges that have a plastic morphology that can be dramatically affected by environmentally induced variables.

Key words: *taxonomy, species list, reefs, biodiversity, Cozumel, Mexico, field guide*

SPONGE ASSEMBLAGES AS BIOINDICATORS OF ANTHROPOGENIC DISTURBANCE

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Sponge assemblages were found to undergo rapid and marked changes in their distribution and abundance following the discharge of sewage into sub-tidal temperate reef habitats. Changes in the structure of sponge assemblages varied over a number of spatial and temporal scales depending on the quantity and quality of sewage effluent. The response of individual species was also found to vary. Sponges and other components of sessile encrusting communities were thought to be poor bio-indicators of sewage pollution because of their relatively slow growth rates. We have found that many temperate reef sponge assemblages can respond rapidly to both physical and biological disturbance, whilst responses to anthropogenic disturbance can occur within three months. The mechanisms by which sponge assemblages are altered by sewage disturbance are thought to include decreased light, increased nutrients and increased silt loads.

Manipulative experiments are now being used to examine the effects of these processes on sponges that have shown adverse effects to sewage disposal. Six species were initially tested for their ability to be successfully translocated using concrete mooring blocks with the sponge attached within a mesh bag. Successful translocation was measured as continual growth over three months with no tissue deterioration. Of the six species tested, 3 were found to show no adverse effects of physical translocation, they were *Cymbastela concentrica, Callyspongia* sp., and *Desmapsamma kirkii.* The sponges *Spirastrella* sp., *Ceratopsion* sp. and *Holopsamma arborea* were all found to deteriorate over the trial period. Based on these experiments the sponge *Cymbastela concentrica* was chosen and translocated into a nearshore sewage outfall at a depth of 18m. The design of the experiment involved using four treatments: (1) sewage site, (2) control site 1, (3) control site 2, (4) disturbed control site (sponges translocated back into the same place). Within each of the treatments, six individual sponges were translocated. Prior to translocation, the sponges were weighed to the nearest gram using a field balance. At the same time, six specimens of *C. concentrica* were collected, weighed, and their reproductive status and photosynthetic pigment concentrations determined. The sponges were retrieved after four months and growth rates, reproductive status and changes to symbiotic algae populations are now being assessed. The practicality of using sponges as bio-indicators in sewage outfall monitoring studies will be discussed.

Key words: *bio-indicators, sewage, disturbance, subtidal, reef, ecology*

FRESHWATER SPONGES (PORIFERA: SPONGILLIDAE) OF THE GUANACASTE CONSERVATION AREA, COSTA RICA: A PRELIMINARY SURVEY

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In August and December of 1996 and March of 1997, I conducted a survey of freshwater sponges in the Guanacaste Conservation Area (GCA) in northwestern Costa Rica. The GCA occupies 110,000 hectares and includes Pacific dry forest, cloud forest and Atlantic rain forest. My objectives were to find out what sponges occur in the GCA, their distribution and preferred habitats. Sites were chosen to represent aquatic habitats from each biome in the GCA. At each site I measured water temperature, pH, specific conductance and current velocity. A unique aspect of this project entailed the measurement of particulate organic carbon (POC). POC can be useful for estimating food availability and has not been used to describe habitat preferences of freshwater sponges. To estimate POC, I used methods outlined by Wetzel and Likens 1979 *Limnological Analyses* W.B. Saunders Company.

Listed in order of decreasing frequency, I found the following taxa: *Radiospongilla* sp., *Dosilia* sp., *Corvomeyenia* sp., *Spongilla cenota, Trochospongilla* sp., and unidentifiable colonies without gemmules. The *Radiospongilla* species is currently being described in a separate paper (Poirrier, in prep.) and prior to this survey, *Spongilla cenota* was known only as far south as Florida and Mexico and therefore represents a significant range extension and a new record for Central America. The other genera are of uncertain taxonomic position and will be considered in a future paper. Sponges in the GCA were restricted to temporary, slow moving streams and ponds in dry tropical forest. These habitats have a drought season of up to six months and have POC content greater than 560 µg/l. No sponges were found in the clear, fast, permanent streams of the cloud and rain forests. Due to the high volume of fast flowing water, POC values are extremely low in these streams. These data suggest that natural aquatic habitats within evergreen tropical forests do not provide adequate food for freshwater sponges and that more favourable habitats are found in the dry tropical forest biome. This may be an important point for the conservation of Central American freshwater sponges, because dry tropical forest is considered the most endangered of all tropical ecosystems.

Key words: *Costa Rica, ecology, habitat, POC, Spongillidae, taxonomy, conservation*

SEROTONIN IN PORIFERA? EVIDENCE FROM DEVELOPING *TEDANIA IGNIS,* THE CARIBBEAN FIRE SPONGE (DEMOSPONGIAE)

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Histochemical study of larvae and freshly settled juveniles of the Caribbean fire sponge *Tedania ignis* (Tedaniidae, Poecilosclerida) reveals evidence of serotonin-like immuno-reactivity, a possible indication for the presence of precursors of nerve cells in this species. Although sponges, one of the oldest metazoan groups, possess the greatest diversity of biologically active compounds of any marine phylum, the neuroactive substance serotonin (5-hydroxytryptamine, 5-HT) has been reported only once, in myocyte-like cells of *Sycon ciliatum* (Sycettidae, Calcispongiae).

Already in the earliest stages of its life, *Tedania ignis* is made up of two discernible cell types: monociliated cells arranged in quasi-epithelial fashion, covering the larva and the developing settled organism, and mesohylal cells (archaeocytes). In the adult sponge, several mesohylal cell types can be distinguished which form a complex connective tissue. Serotonin-like immuno-reactivity demonstrated by us occurs only in two cell types: in some archaeocytes of the parenchymella larvae, and in similar archaeocytes and in a second, bipolar cell type of the settled, juvenile sponge. The discovery of a neuroactive substance in cells of a developing sponge before and after metamorphosis provides new insights into the origin and evolution of nerve and muscle cells in the Eumetazoa. Further research is clearly needed and should include additional sponge species and approaches, including immuno-cytochemical and electro-physiological techniques.

Key words: *histochemistry, serotonin,* Tedania ignis, *larval development, evolution*

ISODICTYA BOWERBANK: POECILOSCLERIDA OR HAPLOSCLERIDA

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It has been argued recently that *Isodictya* Bowerbank should be transferred from the Poecilosclerida to the Haplosclerida. The reason for this is based on the observation that the skeletal architecture (Isodictyal reticulation) of *Isodictya* appears superficially homologous to some genera in the haplosclerid family Niphatidae. This hypothesis is re-examined by comparing the detailed skeletal architecture of a variety of species of *Isodictya* with respect to a range of poecilosclerids and haplosclerids having reticulate spongin skeletons (e.g. Niphatidae). The phylogenetic analysis of this data set produced two equally parsimonious trees of 39 steps with a consistency index (C.I) of 1.000 and retention index (R.I) of 1.000. The analysis indicates that *Isodictya* is monophyletic and that it is more closely related to *Esperiopsis informis* and *Cercidochelae lankesiri* (order Poecilosclerida) (which also form a monophyletic group) than the niphatid genera *(Niphates digitalis, Amphimedon compressa, Amphimedon* sp., *Niphates* n.sp., *Crebrochalina* sp.) from which it is clearly separated. Major characters that separate *Isodictya* from *Niphates* include the nature of the surface skeletal growths, the presence of chelae, the amount of spongin coring the primary fibres and the presence of small cigar-shape oxeas.

Key words: Isodictya, *Poecilosclerida, Haplosclerida, Phylogeny*

TAXONOMIC EVALUATION OF JASPLAKINOLIDE CONTAINING SPONGES OF THE FAMILY COPPATIDAE

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Much interest has been generated by the isolation of jasplakinolide or jaspamide, a novel depsipeptide originating from a variety of marine sponges. Various generic and specific assignations have been given to a group of specimens from the South and Indo Pacific, possessing a similar spicule composition and sharing parallel chemical profiles. We have carried out a comparative study of specimens (museum types and recent collections) representing all the species of involved in this controversy. Our goals are to: a) resolve the uncertainties arising from various taxonomic assignations of jasplakinolide containing sponges with a skeletal complement in accord with *Jaspis* Gray 1897, b) establish the conspecificity of at least two of the jasplakinolide producing sponges, and c) provide thorough characterisation of all *Jaspis* related species involved in this controversy: *Dorypleres splendens, Jaspis digonoxea* and *Jaspis johnstoni*.

Key words: *chemical profiles, jasplakinolide, Coppatiidae, Taxonomy,* Jaspis

BIOEROSION AND BIOCONSTRUCTION BY THE SYMBIOTIC SPONGE *CLIONA NIGRICANS* (PORIFERA, DEMOSPONGIAE)

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Cliona nigricans is a boring Atlanto-Mediterranean sponge which lives in symbiosis with zooxanthelle and which exhibits many different growth forms. It is distinguishable from the sympatric *Cliona viridis* genetically and by its morphological and ecological traits.

On the Gallinara Island cliffs (Ligurian Sea, Italy) this species (α, β stages) bores exclusively organogenous substrata negatively influencing the coastal biocalcification balance. This activity improves the water action on the benthic calcareous organisms, causing a continuous destruction of the bioherms. In this way, calcareous fragments roll down to the bottom where, with terrigenous sediments, they constitute a typical soft bottom characterised by the coastal detritic bottom biocoenosis.

In this particular habitat, large massive specimens of *C. nigricans* (γ stage) living partially burrowed, with evident inhalant papillae and high exhalant chimneys, incorporate large amounts (until 98% of the total DW) of organogenous fragments, selecting the greater size classes. As a result *C. nigricans*, with specimens of 200-500 cm^2 produces large extension of secondary hard substrata, which host a rich biocoenosis of sessile and vagile organisms, represented mainly by other sponges, hydroids, bryozoans and serpulids that, in turn, support nudibranchs, polychaetes, amphipods and harpacticoids. *Cliona nigricans* represents, therefore, a key-stone species in structuring the coralligenous Mediterranean cliffs considering the actions played at different levels: (i) the boring activity modifies the substratum that becomes suitable to host a rich non-boring infauna; (ii) the final result of the boring activity is the destruction of the calcareous substrata that roll down to the bottom; (iii) the same material is then gathered together by the large massive specimens that structure a new unusual biocoenosis.

A study carried out on the sedimentological features of the bottom indicates that the distribution of *C. nigricans* specimens is positively related to the distribution of the coarse sediment fraction. It is therefore probable that large γ stages of *C. nigricans* need to incorporate a particularly coarse organogenous substratum for growth.

Differences in the structure of the aquiferous system between boring and massive stages have been demonstrated by corrosion casts. Particularly evident is the arrangement of choanocyte chambers that, in boring forms, are clustered inside the boring chambers while, in the massive forms, are more homogeneously distributed in the sponge body. Corrosion casts have also clearly manifested debated boring activity of massive forms. In fact, several carbonatic fragments, particularly mollusc shells are bored and crossed by canals of the aquiferous system.

Both boring and massive forms of *C. nigricans* contain large populations of zooxanthellae. The density of these populations, evaluated by chlorophyll analysis of sponge papillae, is related to the solar radiation reaching the specimen.

Key words: *clionids, bioherm, detritic bottom, uptake, bioerosion, habitat, sediments, boring sponges*

RECOVERY AND GROWTH OF THE GIANT BARREL SPONGE (*XESTOSPONGIA MUTA*) FOLLOWING PHYSICAL INJURY FROM A VESSEL GROUNDING IN THE FLORIDA KEYS

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On February 2, 1997, the 187 meter (614 feet) container ship *Contship Houston* ran aground on the Florida reef tract near Maryland Shoal within the Florida Keys National Marine Sanctuary. This incident resulted in significant injury to coral reef resources over an area 650 meters (2,132 feet) in length. Hundreds of the Giant Barrel Sponge (*Vestospongia muta*) were damaged or destroyed as the ship approached the final grounding site, along with thousands of scleractinian corals and other reef organisms. A major coral reef restoration project is currently underway to address the physical and biological injury caused by the grounding. Over 3,000 broken and dislodged corals were reattached to the substrate within the inbound tract of the vessel, and large areas of rubble created by the ship's hull have been stabilised through a variety of techniques.

The purpose of this study was to assess the response of injured *Xestospongia* to the physical injury caused by the vessel grounding. As the vessel approached the grounding site, sponges which were in the path of the ship were subjected to various degrees of injury. This injury ranged from the minor breaking off of the tops of the sponges to the complete destruction of the sponge except for the basal tissue attached to the substrate. I located and marked 37 injured specimens with individual tags attached to plastic cable ties positioned tightly on the upper injured surface at two locations of each sponge. I monitored the sponges at two to three month intervals and measured upward linear growth from the cable ties. I also observed the condition and vitality of each sponge and the method by which the sponges responded to their injuries. All sponges were photographed at regular intervals.

During the course of the study, seven of the tagged sponges disappeared from the study site. Four of these were observed to have died from a wasting disease that was reported from numerous locations in the Florida Keys and the Caribbean. The causes of the disappearance of the other three were not directly observed. The 30 remaining sponges have survived and recovered from the direct physical injury at a minimum by healthy tissue regeneration of the damaged areas. The rate of upward linear growth ranged from zero to 4.48 cm over 13 months, with an average upward linear growth of 1.42 cm for all sponges. Eight specimens (27%) showed no upward growth over the observation period. The average growth rate for the sponges that did exhibit upward growth was 1.94 cm. The most significant period of growth was in the late summer and throughout the fall, which corresponds to the period of warmest seawater temperatures. Upward linear growth was correlated with the degree of injury, with the moderate or slightly injured specimens growing at a faster rate than the badly injured ones. Of the four sponges that died from the wasting disease, three had been categorised as badly injured, which may suggest that injured sponges may be more susceptible to disease than non-injured sponges.

Key words: *Giant Barrel Sponge,* Xestospongia muta, *sponge growth rates, recovery from physical injury, coral reef injury and restoration, Florida Keys, Florida Keys National Marine Sanctuary*

THE INFECTIVENESS OF A BIOERODING CLIONID SPONGE

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Sponges play a major role in reef bioerosion. Early impressions suggested that only dead coral skeleton was infected. Cores of *Cliona viridis*, a very abundant bioeroding sponge in the Great Barrier Reef, were removed with an underwater drill, allowed to heal and fixed onto living surfaces of 9 coral spp. at Orpheus Island. Sponge survival varied greatly. It was best on control surfaces of dead massive *Porites*, on live massive *Porites* and on *Astreapora myriophtalma*. It was least on *Lobophyllia hemprichii* and two branching *Porites* spp. Several individuals in 7 of the 9 coral species were infected within 8 weeks. The areas of infection varied widely. However, after removal of grafts, the sponges died, regardless of their size. The risk of epidemics by fragmentation of this sponge spp. is considered to be low.

Key words: Cliona, *bioerosion, Great Barrier Reef*

WANTED: THE NAMES OF COMMON BIOERODING SPONGES OF THE CENTRAL GREAT BARRIER REEF

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Bioeroding sponges have been well studied in the Mediterranean and the Caribbean Seas. Only few bioeroding sponge species are properly described from the Australian Great Barrier Reef. All other descriptions of Australian species are based on von Lendenfeld 1884-85 and de Laubenfels 1954. Detailed surveys in the central section of the Great Barrier Reef resulted in a large amount of new reference material suitable for revisions and new descriptions. Field descriptions and preliminary studies of spicule mounts made it possible to clearly distinguish 15 species from the rest of the samples, which are harder to categorise. Some of the sampled species appear to be sponges described previously and occurring in other oceans (*Cliona viridis, C. schmidti, Cliothosa hancocki* and *Aka mucosa*), some new species are likely to be endemic to the Coral Sea. Descriptions of the most common species are presented with preliminary names, distinguishing morphological features, and spicule assemblages with a request that participants assist by comparing with species they are familiar with or have observed elsewhere.

Key words: Cliona, *bioerosion, Great Barrier Reef, Coral Sea, taxonomy*

MICROBIAL INFLUENCED PYRITISATION OF MARINE SPONGES

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Although biomineralisation plays an important role in the history of earth, our knowledge about the involved processes is rather limited. In this context microorganisms take a significant part within these global processes. Pyrite crystals are often found in taphonomically mineralised sponge tissues but are absent or rare in the surrounding sediment. In terms of microbiology, the role of sponge-associated, anaerobic species such as sulfate reducing and fermentative bacteria is fairly unknown. During early decaying processes of the sponge tissue the internal sponge becomes entirely anaerobic which is a necessary prerequisite for the growth and metaboiical activity of the sulfate reducers. Therefore, pyrite formation is probably linked with sulfide producing bacteria. In the marine environment, sulfate reducing bacteria are most likely to play this role besides heterotrophic sulfidogenic bacteria.

In this study enrichment cultures of native sponge tissue of the mediterrean sponges *Chondrosia reniformis* and *Petrosia ficiformis* under sulfate reducing conditions were investigated using a combination of rRNA-targeted in situ probing and classical cultivation techniques. Recently developed specific oligonucleotide probes for sulfate reducing bacteria elucidated the occurrence and abundance of various sulfate reducing-species affiliated to different phylogenetic taxa within the *Desulfovibrionaceae* and *Desulfobacteriaceae.* Furthermore, 16S-rRNA based phylogeny revealed to hitherto unknown anaerobic bacteria inhabiting the sponge mesohyl. Additionally, the sulfate reducing activity was confirmed by established pysiological methods.

In living sponges, sulfate reducing bacteria were evenly distributed within the tissue. This indicates the presence of anoxic microniches, which allow not only the survival, but even the subsequent growth of these anaerobic bacteria. Further investigations of culturable sponge-associated sulfate reducing bacteria are currently carried out to investigate their ecology and ability to produce pyrit in culture.

Key words: *biomineralisation, pyritisation, anaerobic microbes, sulphate reducing bacteria, phylogeny, molecular biology, microniches*

CHONDROSIA RENIFORMIS: HITHERTO UNKNOWN BACTERIA

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Marine sponges as evolutionarily ancient metazoa are nowadays in the focus of great interest. However, the implication of closely associated bacterial populations within the sponge tissue is completely unknown. One of the sponges investigated to clarify this relationship is the Mediterranean species *Chondrosia reniformis.* Bacteria associated with this sponge were examined by aerobic and anaerobic cultivation, culture-independent methods as whole cell in situ hybridization, PCR assisted rDNA sequence retrieval and comparative sequence analysis. *In situ* hybridization of bacteria within the sponge tissue with fluorescently labeled rRNA-targeted oligonucleotides for the major subclasses of *Proteobacteria* revealed a great part of the population to be affiliated to the α-, γ- and δ-subclasses. Interestingly, no organism was found to be a member of the *b-Proteobacteria.* On the other hand, there are also many microorganisms that only gave signals with the universal probe for all bacteria, whereas group- or species-specific probes did not hybridise with these bacteria. Some of them are culturable and could successfully be characterised using the polyphasic approach. 16S rDNA-sequencing and subsequent analysis of the cultivated bacteria obtained from sponge-tissue showed that these metazoa are closely associated with a great diversity of hitherto unknown bacteria.

Further studies of culturable sponge-associated bacteria are currently carried out to investigate the ecology of sponge-associated bacteria. This survey will elucidate the physiological properties and, by molecular analysis, the phylogenetical affiliation of these organisms. Occurence and spatial distribution of even unculturable bacteria in sponge tissue will be analyzed by *in situ* hybridization with specifically designed rRNA-targeted oligonucleotides.

Key words: *bacteria, in situ hybridization, sequencing, phylogeny, PCR, microecology*

INTRASPECIMEN VARIATION OF SECONDARY METABOLITES IN THE SPONGE *OCEANAPIA* SP. AND EFFECTS TOWARDS GENERALIST AND SPECIALIST PREDATORS

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Oceanapia sp. from Truk Lagoon is a very distinctive sponge. It consists of an enlarged basal root that is completely buried in the sediment, a fistule that reaches above the sediment and a translucent capitum on top of the fistule. We isolated the two major compounds kuanoniamine C and D from the crude extract.

The concentrations of kuanoniamine C and D increased from the basal root to the translucent capitum. Initial experiments showed the lowest concentrations of the alkaloids in the basal root with 0.3% of dry mass for kuanoniamine C and 0.1% of dry mass for kuanoniamine D. The fistules showed a four fold increase of the metabolites. The highest concentrations were found in the translucent capitum with 5% of dry mass for kuanoniamine C and 1.75% of dry mass for kuanoniamine D.

The angelfish *Pomacanthus imperator* was deterred by the crude extract in laboratory feeding assays. The pure compounds kuanoniamine C and D revealed an even greater deterrent effect when tested against natural fish communities in the field.

Key words: Oceanapia, *secondary metabolites, predation, Truk Lagoon, kuanoniamine*

RELATIONSHIP OF SAND AND FIBRE IN THE PSAMMOCINIAN HORNY SPONGE

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Three new species of the genus *Psammocinia* are described from Cheju Island and Namhae Island, Korea. *Psammocinia jejuensis* n. sp., *P. mosulpia* n. sp. and *P. mammifornis* n. sp. belong to the family Irciniidae. The characteristic of this genus *Psammocinia* is that it has a large quantity of sand grains in fibres, matrix and the surface. *Psammocinia* sp. collected from Cheju Island has larger size sand than the three new species. In the choanosome, primary fibres are hardly found but the secondary fibres are abundant. Sometimes, short secondary fibres are connected with large sand grains, which have a bridge-like appearance. Large sand grains seem to support the sponge skeleton instead of the fibres themselves. The features that have been used to distinguish species are described with illustrations.

Key words: *Horny sponge,* Psammocinia, *Korea, primary and secondary fibre appearance, sand inclusions*

PATTERNS OF INTER AND INTRASPECIFIC GENETIC DIVERGENCE IN MARINE SPONGES

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Since the first molecular systematic studies on marine sponges in the 80s, a fairly large database has accumulated on levels of allozyme divergence between conspecific and congeneric sponge populations. The first findings seemed to indicate that sponge species were genetically more divergent than those of other marine invertebrates, possibly related to their high levels of genetic variation. However, an analysis of the 55 interspecific and 87 intraspecific pairwise genetic identity (I) values available to date indicates a more complex figure. Although the average of I over all interspecific comparisons ($\bar{I}$=0.42), is only slightly smaller than that found among other marine invertebrates ($\bar{I}$=0.54), the frequency distribution of I appears to be bimodal, with some genera consistently highly divergent (I<0.3; *Cinachyrella, Cliona, Spirastrella* and *Tethya*) and others within the normal range of gene divergence (0.4<I<0.8; *Chondrosia, Suberites, Petrosia, Plakina* and *Phyllospongia*). Furthermore, in some other genera (*Axinella, Chondrilla, Clathrina* and *Oscarella*), some species display low and others high levels of genetic differentiation in relation to their congeners. This pattern may reflect a large variance in the evolutionary age of genera in sponges, principally within the Spirophorida and the aster-bearing Hadromerida. In any case, this indicates that, although genetic identity levels are a very efficient way to discriminate sponge species, their use to evaluate taxonomic rank above that level is not recommended.

Key words: *cosmopolitism, genetic divergence, allozymes, molecular systematics*

PUSHING THE BOUNDARIES: A NEW GENUS AND SPECIES OF DICTYOCERATIDA

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Descriptive parameters used to segregate sponges into genera, families and orders are always subject to re-evaluation. 'Very white fan' is a rare foliose sponge usually found between 20 and 24 metres on reefs in the central Great Barrier Reef, Australia. The distinct thick, organised sand cortex and surface characters of the oscular and poral faces, and regular, simple primary fibres, form the basis for the diagnosis of a new genus. The lamellate form, brilliant white colouration and regular skeletal arrangement are diagnostic of the new species. 'Very white fan' has also required the establishment of a new family, Phyllospongiidae, to encompass the foliose dictyoceratid genera *Phyllospongia, Carteriospongia, Strepsichordaia, Lendenfeldia* and the new 'very white fan' genus. In addition to fibre structure, chemotaxonomic characters are used to confirm the placement of genera in the new family; - groupings of dictyoceratid families by sesterterpenoids have shown that Phyllospongiidae contain a unique series of bishomoscalaranes. Although the structure of the new sponge has caused a rearrangement within the Dictyoceratida, we do not regard the task of reassigning species and genera to be complete at this time.

Key words: *Dictyoceratida, sp.nov. genus nov, Phyllospongiidae n.fam.* Phyllospongia, Carteriospongia, Strepsichordaia, Lendenfeldia *chemotaxonomy, sesterterpenoids, bishomoscalaranes.*

POPULATION DYNAMICS OF A SPONGE/MACROALGAL SYMBIOSIS: POSSIBLE CAUSES FOR A PATCHY DISTRIBUTION AT ONE TREE REEF.

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Haliclona cymiformis is a tropical marine sponge that is found only in association with a red macroalga, *Ceratodictyon spongiosum*. This association is commonly found on coral reef flats where it is frequently the most prominent macroscopic organism. At One Tree Reef, populations of *Haliclona/Ceratodictyon* are generally restricted to the rubble banks just inside the reef crest that surrounds One Tree Lagoon. Only one population of the association is found in the centre of the lagoon. It is likely that the lack of rocky substrata in the centre of the lagoon limits the recruitment of the association into new areas. Sexual reproduction by the sponge has never been observed at One Tree Reef. At the rubble bank sites, populations of *Haliclona/Ceratodictyon* appear to be maintained by fragmentation and the size-frequency distribution is skewed toward smaller individuals. In the centre of the lagoon, clumps of the association grow to much larger sizes. Fusion experiments between individuals collected from different sites showed some histocompatibility, suggesting that existing populations of *Haliclona/Ceratodictyon* may have originated from the same parent population.

Key words: Haliclona cymiformis, Ceratodictyon spongiosum, *symbiosis, distribution, size-frequency, reproduction, fragmentation, histocompatibility*

PHOTOSYNTHESIS AND RESPIRATION BY THE SYMBIOTIC ASSOCIATION BETWEEN A CORAL REEF SPONGE AND ITS MACROALGAL SYMBIONT

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In the association between the haplosclerid sponge *Haliclona cymiformis* and the red macroalga *Ceratodictyon spongiosum*, the algal thallus comprises the bulk of the organism, while the sponge fills in the spaces between the algal branchlets and forms a thin layer around the outside of the association. The alga is exposed only at the very tips of the branches of the association. Measurements of photosynthesis and respiration of the symbiotic association have shown that this association makes a significant contribution to the primary productivity of the fringing reefs of One Tree Lagoon, southern Great Barrier Reef, in areas where few large primary producers (corals or algae) can live.

Light entering the branches of the association is rapidly attenuated and, as a result, the compensation and saturating irradiances are high; approximately 750 and 315 µmol photons m^{-2} s^{-1} respectively. Photoinhibition at higher irradiances does not occur. Maximum rates of photosynthesis reach 435 µmol O_2 mg chl a^{-1} h^{-1} during summer, while respiration consumes up to 220 µmol O_2 mg chl a^{-1} h^{-1}. These rates decrease by about half during the winter. Photosynthetic and respiratory rates were unaffected by changes in ambient oxygen concentration or by nutrient enrichment with nitrogen or phosphorus. Prolonged periods of heavy shading lead to an increase in pigment concentration in the alga, but no changes in maximum photosynthetic or respiratory rates were found when compared to control samples. There were no significant differences in the rates of respiration or photosynthesis between cultured *C. spongiosum* and the intact *Haliclona/Ceratodictyon* association, so it was not possible to formulate a model as to how respiration was partitioned between the partners in the association.

Key words: Haliclona cymiformis, Ceratodictyon spongiosum, *symbiosis, photosynthesis, respiration, oxygen concentration, nutrient enrichment, photoadaptation, partitioning of respiration*

THE ROLE OF EARLY LIFE-HISTORY STAGES IN DETERMINING ADULT SPATIAL PATTERNS OF ENCRUSTING SPONGES.

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We studied the abundance and spatial pattern of two Mediterranean encrusting sponges: *Crambe crambe* (highly toxic) and *Scopalina lophyropoda* (non-toxic) at three spatial scales (0.5, 1 and 2 m^2). We examined the reproductive output, larval behaviour and early recruitment in these species, and assessed the relative importance of these parameters in explaining the abundance and spatial patterns of adults. We also determined, in field experiments, whether the presence of adults induces or inhibits recruitment in these two species. We found that *C. crambe* was much more abundant than *S. lophyropoda* at the site studied in both number of individuals per square meter (67±2.7 vs. 10.2±2.1, mean±SE) and coverage (47±1.9% vs. 11.1±1.4%). At the smallest scale sampled (0.5 m^2), both species showed an aggregated pattern. Aggregation was also detected for *S. lophyropoda*, but not for *C. crambe*, at the scales of 1 and 2 m^2. The number of embryos incubated per cm^2 by *C. crambe* and *S. lophyropoda* was 76.2±12.5 and 14±1.7 (mean±SE), respectively. We estimated that the potential number of larvae of *C. crambe* released into the water column was about 20 times higher than that of *S. lophyropoda*.

Larval behaviour was monitored in the laboratory and in the field. Larvae of *S. lophyropoda* did not swim away from the release point. They maintained a vertical posture that minimised horizontal dispersal, and soon began crawling. In contrast, the larvae of *C. crambe* swam actively and had a comparatively delayed crawling phase. Recruitment of the two species in scraped quadrats surrounded by individuals of *C. crambe* and *S. lophyropoda*, and in controls (rocky areas with no sponges) was monitored weekly for a month. Recruitment of both species was higher in scraped quadrats surrounded by conspecifics. This effect was notably more marked for *S. lophyropoda* than for *C. crambe* recruits. The toxicity of *C. crambe* did not inhibit settlement of *S. lophyropoda* with respect to controls. The mean number of recruits per surface unit after one month (all substrates pooled) was ca. 3.5 times higher for *C. crambe* than for *S. lophyropoda*. This difference was smaller than expected given that larval production of *C. crambe* was ca. 20 times higher. This indicates that a significant proportion of *C. crambe*'s offspring did not contribute to the maintenance of the local population. The aggregated pattern of *S. lophyropoda* at scales ranging from 0.5 to 2 m^2 and its discontinuous geographic distribution may be partially explained by strong phylopatry of its larvae due to their poor swimming ability and limited dispersal. The dominance of *C. crambe* in littoral assemblages, its random distribution at scales larger than 0.5 m^2, and its ubiquity along the littoral are traits that are consistent with high reproductive output, the swimming behaviour of larvae which facilitates wide dispersal, and patterns of recruitment found in this study. Therefore, *S. lophyropoda* populations appear to be maintained by offspring supplied by autochthonous individuals while populations of *C. crambe* appear to be open, with a potentially significant flow of larvae between them.

Key words: *reproductive output, larval behaviour, settlement, recruitment, spatial patterns, encrusting, Mediterranean Sea*

SPATIAL AND TEMPORAL VARIATION OF THE NATURAL TOXICITY IN SPONGES OF A MEDITERRANEAN CAVE: IS THERE A TREND?

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Strong intraspecific variation of chemical defenses has been documented for marine seaweeds and, less often, for some benthic invertebrates. The causes and the extent of this variation still remain poorly studied.

We present here an extensive study on natural toxicity of sponges inhabiting a sublittoral cave at the Balearic Islands, (Mediterranean). We looked over spatial and temporal patterns at both community and species level.

First, we performed an exhaustive semi-quantitative census of the benthic species present along the cave walls and analysed the species/abundance matrix by cluster techniques. Three clearly different zones with distinct species assemblages came out from the analysis. We characterised the main abiotic factors of these zones by measuring irradiance, water movement, and particulate organic matter of the water. For the toxicity analysis, we collected at the three zones a minimum of three specimens per species (33 species, 291 specimens). We looked at seasonal variation in toxicity by sampling in June and November 1998. Toxicity was measured by the Microtox assay, which has shown a high performance for assessing natural toxicity in previous studies. We also ran sea-urchin bioassays over randomly selected samples to determine whether toxicity against marine bacteria correlates with toxicity against invertebrate cells. Correlation between both bioassays was always high. Whenever a given concentration of crude extract resulted in Gamma values (=toxicity units in the Microtox assay) above 0.5, it also featured some toxicity against sea-urchin embryos. Thus, we chose this value as a threshold to separate toxic from non-toxic sponges.

We found contrasting trends in the variation of toxicity along the cave with season. At the community level, toxicity values had a tendency to increase as irradiance and substrate occupation decreased (increasing distances from the cave entrance) in June. This trend reversed in November, when lower toxicity was found in the innermost zone. High variances prevented ANOVAs from detecting significant differences in mean toxicity between zones or seasons. In contrast, we detected significant changes (Chi-square statistic) in the number of toxic species among zones between seasons. At the species level, we found significant differences in toxicity among zones and the pattern of variation along the cave also changed seasonally.

Our results proved that spatial and temporal variability in toxicity is remarkably high in Mediterranean sponges. This variability, either genetically determined or environmentally induced, may have important ecological and evolutionary implications in benthic communities.

Key words: *natural toxicity, spatial and temporal variation, Microtox assay, caves, Mediterranean Sea, benthic communities*

NON-FEEDING PERIODS IN THE POECILOSCLERID SPONGE *CRAMBE CRAMBE*: AN ULTRASTRUCTURAL STUDY.

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The red, encrusting sponge *Crambe crambe* is a common species in the Mediterranean sublittoral. Its structure, biology, ecology, and defense mechanisms are well known from several studies in the last years (Becerro *et al.*, 1998, and cites therein). We report here the existence in this species of a resting, non-feeding period, describe the ultrastructure of this stage and discuss on its possible biological and ecological significance.

This sponge features a marked seasonality in its reproductive cycle, with larval release occurring in July and August. From the end of August until the end of October, some specimens appeared covered with a glassy cuticle. No inhalant pores or oscula were open through this cuticle, so that the sponges were not able to feed during this stage. In November, the surface of some specimens incorporated noticeable amounts of particulate matter and debris, contrasting with the usually clean and smooth surface of this species. Upon closer observation, these specimens were shown to feature a strong hispidation, with the spicules retaining entangled debris. We interpret this hispidate form as being an intermediate stage between the resting phase and the active period.

SEM examination of the surface during the non-feeding period confirmed the absence of inhalant orifices and the presence of an acellular cuticle instead of the usual exopinacoderm. In some specimens, microorganisms were adhered to the outside of the cuticle, while they were totally absent from the surface of active specimens. Through TEM observation, the cuticle appeared as a complex structure, about 0.1 µm thick, comprising a proximal dense layer formed by the aggregation of collagen fibrils, and an outer layer of granular material. Beneath this cuticular layer, a zone with few cellular components (mainly pinacocytes and spherulous cells in diverse degeneration stages) and negligible collagen fibrils extends. The choanosome appeared in a rather disorganised form, with some choanocyte chambers still recognisable, while others were largely disintegrated. Archaeocytes, sclerocytes, spherulous cells, degenerate cells and cell debris were abundant. Phagocytosis by archaeocytes was common.

At more advanced stages, the cuticle losses its structure and appears broken at many points. SEM images suggest that the cuticle is cast off, and a new pinacoderm develops underneath. Spicules, previously under the cuticle, are now exposed, thus giving rise to the hispidate phase. The surface is again perforated by functional inhalant orifices at this phase. Reactivation of the sponge filtering activity and release of the hispidation spicules to the milieu ensue.

The stages described here may be related to a reorganisation of the sponge canal system after larval release. It doesn't seem to happen, however, to all specimens and in all years; rather, this phenomenon had been only occasionally observed in previous years, but in 1997 it was especially intense and affected an estimated 5% of specimens at a time. It is not clear at present why this phenomenon has been so frequent in this particular year, although it may be related to the exceptionally high water temperatures during Autumn, 1997 in the Mediterranean area (the highest in the last 25 years).

Key words: *resting stage, ultrastructure, canal system reorganisation,* Crambe crambe, *Demosponges, Mediterranean Sea*

CARNIVOROUS SPONGE

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A movie, 15 min long, illustrates the carnivorous sponge *Asbestopluma hypogea* and the Mediterranean cave in which it has been discovered. The cave entraps a cold water mass, resulting in stable temperature conditions throughout the year and in colonisation by deep-sea animals. Time lapse cinematography made in an aquarium shows the capture and the digestion of the prey by the carnivorous sponge. Several other sponges from deep-sea origin are shown in their cave habitat including *Oopsacas minuta*, a representative of the hexactinellids and the recently described demosponge *Myceliospongia araneosa*.

Key words: *cave, Mediterranean, Cladorhizidae, carnivorous*

THE PLANKTONIC ARMOURED PROPAGUELES OF THE EXCAVATING SPONGE *ALECTONA* (PORIFERA, DEMOSPONGIDAE) ARE LARVAE: EVIDENCE FROM *ALECTONA WALLICHII* AND *A.MESATLANTICA* N.SP.

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The armoured propagules of the excavating sponges *Alectona* and *Thoosa*, which are the only stage of reproduction of sponges consistently observed in full planktonic conditions, have been considered as asexual because of a morphology unique among sponge larvae. Evidence of sexual origin is presented in *Alectona wallichii* and in a new species from the deep Atlantic, *A.mesatlantica*. Stages in spermatogenesis and oogenesis are present at the same time as embryos in various stages of development, from early segmentation to advanced embryos possessing a discotriaene and style skeleton. Thick collagen strands surround the blastomeres from early segmentation stages. Collagen strands and spicules appear when the embryo cells are still undifferentiated. The larva is unique among demosponge larvae by the absence of flagella and the presence of a larval skeleton including spicules that are unknown in the adult. The name *hoplitomella* is proposed for this special larva of alectonid sponge.

The uniqueness of the sexual development and the tetraxonial nature of the discotriaenes of the larva of *Alectona* both indicate that these sponges are not Hadromerida. The distinction of a family Thoosidae for the genera *Thoosa*, *Alectona* and *Delectona*, is supported, although the family is considered presently as *incertae sedis* within the Demospongiae.

Key words: *Demospongiae*, Alectona wallichii, Alectona n.sp., *sexual reproduction, embryology, planktonic larva, excavating sponges, bioerosion, phylogeny*

CHEMOSYSTEMATICS OF THE PORIFERA

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We reviewed all compounds described from the Phylum Porifera in an attempt to discover what level of reliability may be attached to chemistry data when applied in classification. To date (May 1998) more than 3500 different chemical compounds have been extracted from 475 species of marine sponges belonging to two of the three classes (Calcarea and Demospongiae), all major orders of Demospongiae, 55+ families and 165 genera (source: Blunt & Munro's MarinLit database, 1998 update). Previous studies revealed that several ordinal, family and genus patterns appear to exist, with unique types of compounds apparently restricted to discrete sponge taxa. If this were the case overall, then the impressive chemical dataset could be put to use in helping to solve persistent problems and disagreements over the classification of various sponge taxa.

However, the compounds may be produced either by sponge cells - and then may be regarded as proper characters of the sponges - or by microsymbionts which may be not at all species- or group-specific. Large numbers of proven or suspected microsymbiont compounds appear to be present from the lack of correspondence between sponge identity and compound structure (examples are macrolides and cyclic peptides which are found dottedly over most Demosponge groups, and are in fact known from various microbial organisms). Reported chemistry is distributed heterogeneously over the various groups, with highest numbers of compounds reported from Dictyoceratida (650+ compounds from 95 species), Dendroceratida (500+ from 48 species), Haplosclerida s.l. (660+ from 101 species) and Halichondrida s.l. (540+ from 86 species); other groups have intermediate (Astrophorida-Lithistida, Hadromerida, and Poecilosclerida) or very low (Calcarea) numbers of compounds. Despite previous claims of exclusive occurrence of particular compound types in discrete sponge groups, it was found that in most if not all cases compounds are distributed fuzzily. Concentrated occurrence of compound types in particular groups is common (examples are bromotyrosines in Verongida, furanoterpenes in Dictyo- and Dendroceratida, straight-chain acetylenes and 3-alkylpiperidine derivatives in Haplosclerida s.l.), but almost invariably there are also reports of these compound types from unrelated sponges. Furthermore, in the rare cases that a compound type is indeed restricted to a certain sponge group (for example pyrrole-2-carboxylic derivatives in Halichondrida s.l.), then such fuzziness reappears among the families within the group. Reasons for this fuzzy distribution may be either or all of the following: (1) parallel biosynthetic pathways leading to the same structure, (2) microsymbiont involvement (3) careless specimen handling (contamination by epibionts, confused labels), (4) wrong identification/classification.

Currently, the amount of fuzziness is such that direct use of chemistry (literature) data to solve classification problems or to erect new higher taxa, is inadvisable. Inconsistent occurrence of compounds cannot be dismissed without further study. Large scale voucher reexamination, or recollection and chemical analysis, as well as cooperative studies between systematists, microbiologists and bioorganic chemists, are necessary to demonstrate the properness of using chemical characters for classification.

Key words: *chemosystematics, marine sponges, classification*

BIOLOGY OF SPONGE NATURAL PRODUCTS

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This is to announce the EC-MAS 3 project, which started April 1, 1998. The biological and chemical aspects of selected sponge natural products (secondary metabolites) of interest to human use will be studied to obtain understanding of: (1) the cellular origin and possible microsymbiont involvement, and (2) the ecological significance of sponge secondary metabolites, and (3) the patterns in these processes enabling rationalisation of exploration for and exploitation of sponge secondary metabolites. The results will have a direct bearing on policy decisions concerning industrial production of sponge secondary metabolites, which are too difficult or too costly to synthesise. A major deliverable product of the proposed research will be the formulation of a standard protocol of research steps needed as a basis for such policy decisions.

The research will be structured in three phases: (I) exploration and pattern recognition, (II) testing of hypotheses using experiments with selected sponges, (III) protocol construction. Initially, investigations will be directed towards two sponge groups (Haplosclerida and Halichondrida) and towards a limited number of molecule types. Known secondary metabolite occurrence will direct exploration for related sponges and related secondary metabolites. For the experimental phase a choice for 3-4 target sponges will be made based on suspected production of secondary metabolites by own sponge cells (1-2 target sponges) or microsymbionts (1-2 target sponges). The biological aspects include: determination of the identities and phylogenetic relationships of bioactive sponges; within-sponge spatial distribution of sponge cells and microsymbiont cells; experimental observation of variability of biological activity of selected sponges in various environmental (biotic and abiotic) situations; identification of target microsymbiont cells; fractionation, isolation and culture of target sponge cells. The chemical aspects include: extraction, isolation and structure determination of selected bioactive compounds; development of qualitative and quantitative analytical methods for their spatial distribution in the sponge and for their distribution between and within different species.

Summary of methodologies: Sponges will be collected using SCUBA (shallow water) and/or dredges (deep water) and photographed upon collection. Various types of fixations of material will be made immediately after removal from the water. Voucher specimens will be studied for identity and phylogeny using routine morphological as well as molecular (18S / 28S rDNA) characters. Collected sponges preserved in methanol or as freeze-dried material will be extracted with methanol and dichloromethane. The primary extracts will then be tested for their biotoxicity using an invertebrate bioassay organism, the Artemia toxicity test, and several prokaryote and eukaryote bioassay organisms (bacteria, fungi, yeast). Cytological analyses will consist of two different approaches, one using glutaraldehyde-fixed material, the other using live sponges: (1) sponges will be fixed in glutaraldehyde, (a) for microsymbiont detection; thick sections will be stained with suitable fluorochromes and viewed by fluorescence microscopy and confocal scanning light microscopy. If microsymbionts are present, populations will be characterised by different parameters. Microsymbionts will be further identified by fluorescence in situ hybridisation using rRNA-targeted oligonucleotides as probes; (b) for sponge cell spatial distribution, samples will be postfixed in 1% osmium tetroxide and thin sections will be studied by Transmission Electron Microscopy (TEM). (2) Live sponges will be dissociated into single-cell suspensions. Recognition of secondary metabolite production will be realised using two advanced techniques: (a) cell fractionation into pure cell populations using continuous or discontinuous Percoll gradients; (b) symbiont-free sponge cultures, initiated either from pure cell populations or from dissociated sponge cell suspensions. Experimental observations will be made in situ using various types of manipulations (caging, artificial standard lesions, confrontation with substrate competitors, crude extract assays with substrate competitors and potential predators).

Key words: *secondary metabolites, microsymbionts, chemical ecology, exploration and exploitation*

THE FRESHWATER SPONGES FROM A NEOTROPICAL SAND DUNE AREA.

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The eastern part of Maranhão State is a sand dune field which stretches along 150km of the continent border and penetrates from 10km to 150km into the country side where it limits with the 'cerrado' (Brazilian savanna). The National Park of "Lençóis Maranhenses" was created in 1981 to protect the largest part of this area, which contains an amazing array of seasonal freshwater ponds nested among the dunes. The region is classed under Koeppen's AW climate with the rainy season extending from December to May and the annual mean air temperature ranging from 26.8°C to 27.2°C. The survey for freshwater sponges was performed from October 20th to 30th, 1995. At this time of that year the largest ponds (+- 50X20m) contained yet about 1/3 of their water. That condition was previously chosen as the one which would yield sponges with gemmules. Contact was kept with local residents who informed the team as to the water level, since dry and wet seasons do not occur exactly at the same period each year. The survey was carried out at the eastern and western borders of the National Park were the limiting sites with the savanna were more readily met. pH, temperature and conductivy of the water were determined for the ponds with higher water levels.

A transect departing from the mobile dunes into the savanna boundary disclosed a succession of ponds with different spatial fitting and water colours. The ponds situated amongst the mobile dunes are also mobile and have cristal clear waters whilst the ones approaching the savanna boundary are more stable ponds with increasing brownish colours and acid pH. Sponges were abundant and plenty of gemmules particularly in the two extremes of the "pond succession" i.e. the very clear ones and the brownish coloured ones thus proving correct the assumption that this huge seasonal aquatic system contained this fauna and that the sampling period was the one chosen.

The sponge community in the ponds also uncovered a quite interesting, though not ecologically unusual distribution pattern. Approaching the savanna boundary the community found included one or more of the following species, but never the five of them: *Metania spinata* (Carter, 1881), *Corvomeyenia thumi* (Traxler, 1895), *Dosilia pydanielli* Volkmer-Ribeiro, 1992, *Trochospongilla variabilis* Bonetto & Ezcurra de Drago, 1973 and *Radiospongilla amazonensis* Volkmer-Ribeiro & Maciel, 1983. In the sand-dune clear water ponds the only species found was *Corvoheteromeyenia heterosclera* (Ezcurra de Drago, 1974). However going towards the clear-water dune ponds some mixing of *C. heterosclera* with *R. amazonensis* and *T. variabilis* could be detected.

Recent litterature dealing with the study of Neotropical freshwater sponge communities structure has shown that the five species upper referred close to the savanna boundary were found together in seasonal ponds form the north to the south of the Brazilian savanna. So two characteristic communities are here dealt with: the one of the savanna ponds with five species and the one of the sand-dune ponds with one species.

The region extending between the areas occupied by these two communities thus biologically and physically configurates a savanna/sand-dune ecotone. To this particular ecotone the best fitting dynamic concept seems to be the patch concept as exemplified by Bretschko (1995). In spite of the fact that this concept was settled up for a river/land ecotone we think that if fits quite adequately the presently studied one.

Key words: *freshwater habitats, sand dune, savanna, ecotone, distribution, community structure*

THE PECULIARITIES OF SPONGE SPICULE COMPOSITION IN HOLOCENE - PLEISTOCENE SEDIMENTS OF THE UNDERWATER AKADEMICHESKY RIDGE OF LAKE BAIKAL

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Sponge spicules in Holocene - Pleistocene sediments of the top of the underwater Akademichesky Ridge were studied. In a bottom sediment core (480 cm high obtained with a gravity corer), the spicules of 3 genera of Lubomirskiidae sponges (*Lubomirskia, Baikalospongia, Swartschewskia*), the macroscleres and microscleres of *Spongilla* sp. and the macroscleres of *Ephydatia* sp. (fam. Spongillidae), were found. We consider the two unusual types of spicules an interesting find since these spicules have no analogy with the spicules of recent sponges. They are long and thin oxeas (360-700 µm x 12-16 µm) with small spines evenly distributed over the main part of spicules, and smooth and thick oxeas (224-280 µm x 24-40 µm). It is noteworthy that typical recent *B. bacillifera* spicules are absent from the core studied, and strongyles with small spines on their extremities are smooth. As a rule, *B. bacillifera* colonise various depths of Lake Baikal. At the same time the core contains the spicules of *Lubomirskia baicalensis* which inhabits usually the shallow waters. These spicules as well as the spicules of *Spongilla* and *Ephydatia* might be introduced here from the region of Ushkanii Islands. Spicule quantity and their morphological diversity reach their maximum during warm climate periods. Cold periods coincide with the absence of *Spongilla* sp. and *Swartschewskia papyracea* spicules present in sediments, and the number and diversity of other spicules significantly decreased.

Key words: *Holocene, Pleistocene, sediments, spicules, Lake Baikal*

PHYLOGENETICS OF THE EURETIDS (EURETIDAE; HEXACTINELLIDA)

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A long overdue phylogenetic analysis using 25 characters from 20 OTU's and two outgroups reveals several monophyletic groups within the Euretidae (Hexactinellidae) which may merit sub-family designation. The ordinal separation of Clavularia and Scopularia is disregarded on the basis of *Bathyxiphus subtilis* Schulze and *Claviscopulia furcillata* which contain both clavule and scopuline spicules. This analysis also suggests that an ancestral euretid closely resembling *Farrea occa* Schulze possessed a pre-scopuline spicule and that microsclere spicules and body form are prone to homoplasy and are therefore not stable phylogenetic indicators. This preliminary analysis will hopefully stimulate further research and interest into hexactinellid phylogeny and biology.

Key words: *Hexactinellida, Euretidae, classification, spicules, phylogenetics, homoplasy*

NITROGEN FIXATION IN SYMBIOTIC MARINE SPONGES: ECOLOGICAL SIGNIFICANCE AND DIFFICULTIES IN DETECTION.

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There has been considerable speculation and some evidence that coral reef sponges can fix atmospheric nitrogen through some of their microbial symbionts, particularly symbiotic cyanobacteria. Many Indo-Pacific coral reef sponges can satisfy much of their requirement for carbon energy compounds via translocation from photosynthetic symbionts and a similar mechanism has been invoked to explain how some sponges could supplement the low amount of available nitrogen in clear tropical waters. Attempts to measure N^2 fixation using the acetylene reduction test have proven technically difficult and, in our hands, given ambiguous results. However, fixation was demonstrated unambiguously with incorporation of the stable isotope ^{15}N into the amino acids glutamine, glutamate and aspartate of *Callyspongia muricina*, although at relatively low rates. The variability in measuring acetylene reduction in 23 sponge species is attributed to several factors: the number of cellular and matrix barriers that must be passed by acetylene and ethylene, the difficulty of maintaining sponges alive under experimental conditions, and possible toxicity of the reagents. This fixation indicates how sponge-symbiont associations can assist sponges occupy habitats that are not ideal for filter feeding animals.

Key words: *microbial symbionts, nitrogen fixation, ecology, sponge-symbiont associations*

LONG TERM SURVEY OF THE CALCIFICATION RATE IN JAMAICAN CORALLINE SPONGES

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Coralline sponges, also formerly called "sclerosponges", are known to build up a massive calcareous skeleton that can play a significant role as framework builders on modern coral reefs.

Ceratoporellidae are readily accessible by SCUBA diving in unusual cryptic habitats of the North Jamaican reef, such as submarine caves and tunnels where they provide us with the unique opportunity to study the growth rate of the calcareous skeleton of organisms which have had an important geological role in the past.

A technique was developed to record the growth rate of the skeleton of several species by marking the newly deposited aragonite with calcein, a fluorochrome stain. Specimens were labeled *in situ* in a reef tunnel at a depth of 28 m near Discovery Bay, Jamaica. Staining was performed up to five times from 1984 to 1997. Each time, small skeletal samples were removed from the periphery of the sponges, leaving specimens in their place. Sections, cut perpendicular to the surface and ground to a thickness of ± 10 µm, were observed by fluorescence microscopy. The annual growth rates of the skeleton were calculated from measurements of the linear extension between calcein stained lines along the growth axes.

Data indicate that the mean annual extensions vary in time, from one species to the other, and within and between specimens of the same species: from about 150 µm to 300 µm in *Ceratoporella nicholsoni*, and from 50 µm to 180 µm in *Stromatospongia norae*. Growth rates also appear to depend on the size of the specimens.

This first long-term study on sponges of the family Ceratoporellidae brings important information on the variability of their growth rate, which will have to be considered for further studies on the use of coralline sponges in paleoclimatic reconstruction.

Key words: *coralline sponges, sclerosponges, growth rate, calcein*

BIOGEOGRAPHY AND TAXONOMY OF THE REEF CAVE DWELLING CORALLINE DEMOSPONGE *ASTROSCLERA WILLEYANA* THROUGHOUT THE INDO-PACIFIC

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Astrosclera willeyana Lister 1900 is a pyriform-half spherical, predominantly bright orange colored, coralline demosponge. The habitat of *Astrosclera* is generally restricted to cryptic and light reduced environments of the Indo-Pacific, found mainly in reef caves, but sometimes also in the dim-light areas of cave entrances and overhangs.

The spicular skeleton of *Astrosclera* consists of megascleres; microscleres are absent. The basic spicule type is a sub-verticillate - verticillate acanthostyle, of the *Agelas* type, with a mean length of 80 µm. The spicule morphology and size is highly variable, depending on the geographic origin of the specimen.

Variability in spicule morphology of *Astrosclera* from different geographic localities was previously reported by several authors (Vacelet 1967a, 1977, 1981; Ayling 1982; Wörheide *et al.* 1997a), and Vacelet (1981) discussed the idea of more than one species of *Astrosclera*. Empirical testing of the question whether variation in spicule morphology represents geographic variation or separate species was undertaken in this study, examining the spicule morphology of specimens from 26 geographically distinct populations. Corroborative evidence from a restriction fragment length analysis of the ribosomal DNA was also undertaken for twenty specimens from five geographically distinct populations of *Astrosclera*.

Analysis of spicule morphology showed that variation was not random but specifically linked to geographical origin of the specimen. Six groups were recognised with similar spicule morphology (group with similar spicule morphology: GSSM's), based on spicule length, spination, proximal thickening, and abundance. These GSSM's comprise populations from adjacent localities. Based on these spicule morphology data the idea that there may be more than one species of *Astrosclera* was further supported, but analysis of ribosomal DNA did not lend support to this hypothesis. The RFLP-method of rDNA-Analysis was sensitive enough to detect species-level differences in sponges, as shown by comparative studies on other demosponges, but until further divergent characters are found, the different GSSM's should be regarded as one species. The GSSM's seem to represent geographic subspecies, whose genetic differences, expressed by different (non-random) spicule morphology, were not detected by rDNA-analysis. It is supposed that *Astrosclera* is currently in the process of species separation.

Each GSSM (or subspecies) is likely to have its own history with respect to radiation, isolation and evolution, and a model of the biogeographic and phylogenetic relationships of the GSSM's will be presented.

Key words: *biogeography, taxonomy,* Astrosclera, *ribosomal DNA, spicule morphology, phylogeny*

CLIMATIC CHANGES OF THE LAST 450 YEARS RECORDED IN THE SKELETON OF THE CORALLINE DEMOSPONGE *ASTROSCLERA WILLEYANA*

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Stable isotope time series of $\delta^{18}O$ and $\delta^{13}C$ were measured in successive growth layers of the largest and oldest *Astrosclera* ever found (diameter of 25 cm, max. age 550 yrs) from Ribbon Reef #10 (GBR). *Astrosclera* forms its skeletal aragonite in equilibrium with the ambient seawater, and represents, therefore, a high precision recorder of the isotopic history of the ambient seawater. $\delta^{13}C$ of surface water dissolved inorganic carbon in the northern Great Barrier Reef has apparently decreased continuously since the mid-16th century. The total decrease is 0.7 ‰. The major decline of 0.5 ‰ occurred during the industrial period of the 19th and 20th century, likely to be due to the increased release of CO_2 by deforestation and burning of fossil fuel during the period of industrialisation after 1850 (increased input of lighter carbon isotopes). The oxygen isotope history shows a slightly colder (and/or dryer) phase before 1850, which correlates with the Little Ice Age. A considerable shift to lighter values occurred during the 20th century (warming of SST). This may be due to an anthropogenic greenhouse effect. Most of the major climatic changes caused by ENSO/El Niño events, as reported by Quinn et al. (1987), as well as by large volcano eruptions (see LaMarche & Hirschbroek 1984) in the last four and a half centuries were recorded in the oxygen isotope record of *Astrosclera*.

Key words: Astrosclera, *growth layers, isotopes* $\delta^{18}O$ and $\delta^{13}C$, seawater

BIOCALCIFICATION IN THE INDO-PACIFIC CORALLINE DEMOSPONGE *ASTROSCLERA WILLEYANA* LISTER - THE ROLE OF BASOPINACODERM

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The aragonitic calcareous basal skeleton of *Astrosclera* is composed of 20-60 µm-sized aragonitic spherulites, produced by a combination of three processes. First, the spherulites are formed in large vesicle cells (LVC's) inside large vesicles in the ectosome. In a second process, after release from LVC's, basopinacocytes transport the spherulites to the tips of the skeletal pillars, where they fuse together by epitaxial growth; and in a third process, during upward growth, the soft tissue is slowly rejected from the lowermost-parts of the skeletal cavities and the remaining spaces are subsequently filled by epitaxially-growing aragonite fibres. In the second and third process, basopincocytes produce either the insoluble intracrytalline organic matrix, which does not consist of collagen, as well as the soluble intracrystalline matrix, which consists of highly acidic Ca^{2+}-binding mucus substances. Basopinacocytes control speed and direction of epitaxial growth in both of the latter two biocalcification processes. It is hypothesised that *Astrosclera* is able to control the rate of calcification by the regulation of its bacterial population. The mean growth rate of *Astrosclera* was measured at 230 µm per year. See Woerheide (1998) for a detailed description of soft tissue ultrastructure and its cellular composition.

Key words: Astrosclera, *X, skeletal development, calcification regulation, ultrastructure*

CHEMICAL AND ANTIMALARIAL INVESTIGATIONS OF SOME *CYMBASTELA* SPECIES

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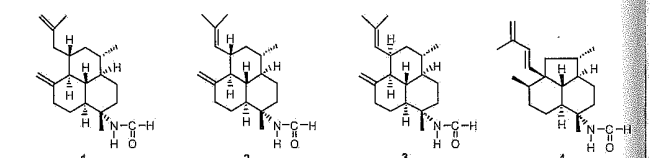
Cymbastela hooperi collected from Kelso Reef, the Great Barrier Reef, North Queensland, yielded a series of natural products, mainly diterpene isonitriles, which demonstrated significant *in vitro* antimalarial activity. As a result of these compounds being consumed in a number of bioassays it was considered desirable to have more of them so as to enable further and more detailed testing to be undertaken. As we had three *Cymbastela* species (2 x *C. concentrica* and 1 x *C. coralliophila*) and two samples of *Amphimedon* (*Cymbastela*) *terpenensis* in our sample collection it was decided to test them for their antimalarial activity and for their general chemical content. The results of these investigations provided further evidence that *Amphimedon terpenensis* is more appropriately *Cymbastela terpenensis*, and also led us to reinvestigate one of the semi-pure fractions from our original investigation of *Cymbastela hooperi*. This investigation led to the isolation of a number of new as well as some known diterpene formamides (**1-7**) with antimalarial activity. In the presentation a number of the afore mentioned areas will be addressed.



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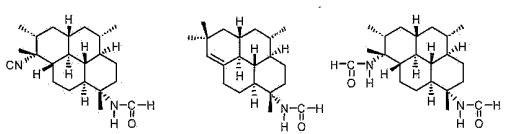

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Key words: *natural products, diterpene isonitriles, antimalarial activity,* Cymbastela, *chemical isolation*

RAPID CHANGE AND STASIS IN A CORAL REEF SPONGE COMMUNITY

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Four census of a sponge community on a shallow coral reef in San Blas, Panama, have revealed a combination of both extreme change and also apparent stasis over the 14 years between 1984 and 1998. Total biomass of the sponge assemblage varied little over the first 11 years, with the exception of the first few years after a hurricane decreased sponge populations in 1988. However, relative contributions to total biomass by the different species have changed to the extent that over half of the original species are now altogether absent from the censused area. Species lost were not necessarily those that had been rare initially, and the hurricane does not appear to have been responsible for the loss of species. The most striking pattern of loss is that keratose species account for a disproportionately large number of the species and also of the volume of biomass lost. The three most common species initially have remained the most common, but their relative abundance has shifted. The hurricane affected these 3 species differently, but the effects of subsequent infections by pathogens have been much greater. Pathogens appear to be species-specific, and act in a density dependent fashion such that whichever species is most common also has the highest percentage of individuals infected, and is doomed to lose its status as most common species.

Key words: *coral reef sponges, population dynamics, disease, environmental ecology*

A SPONGE THAT CHEATS ON DIFFUSE MUTUALISM AMONG OTHER SPONGE SPECIES

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The demosponge *Desmapsamma anchorata* is frequently found growing on other organisms, especially gorgonians and other sponges. For paired *D. anchorata* individuals of the same genotype and initial size, growth rates were lower and mortality rates were higher on carbonate substrata than they were on sponges of other branching species. The three sponge species that served as hosts in the experiments, *Iotrochota birotulata, Amphimedon compressa,* and *Aplysina fulva*, grow and survive better when they are intimately intertwined with each other, and do not therefore discourage other sponges from adhering to them. However, *D. anchorata* does not improve the quality of life for these species when it participates in associations with them. *Desmapsamma anchorata* grows many times more rapidly than the other species, and appears to accomplish this by skimping on skeletal quality such that it requires skeletal support produced by other organisms in order to withstand physical disturbances. In the early stages of its growth on sponges of other species, *D. anchorata* does not decrease growth rates of its hosts, but as it continues to grow, it can entirely overwhelm the other sponges, smothering and killing enveloped tissue. The extreme fragility of *Desmapsamma anchorata* makes it vulnerable to being swept away by physical disturbance, and this prevents it from becoming a chronic hazard for the other sponges. Intimate association with *D. anchorata* may provide one benefit to other sponge species, which is to facilitate reattachment of loose fragments. Because *D. anchorata* is able to reattach to carbonate substrata within one day, fragments of other species to which it is attached are anchored for the few additional days that they require in order to establish their own stable attachments to solid substrata.

Key words: *mutualism, parasite, sponge growth, sponge mortality, asexual fragmentation*

PRODUCTION OF BIOACTIVE FURANOSESTERTERPENE TETRONIC ACIDS AS POSSIBLE INTERNAL CHEMICAL DEFENSE MECHANISM IN THE SPONGE *IRCINIA FELIX* (PORIFERA: DEMOSPONGIAE)

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Marine sponges of the genus *Ircinia* (Porifera, Demospongiae) are known to produce several linear furanosesterpene tetronic acids - FTAs, with antimicrobial, cytotoxic and antitumoral properties. *Ircinia felix* is a common and abundant sponge in the shores of Santa Marta, Colombian Caribbean Sea, and contains FTAs in appreciable quantities, up to 4.5% of its ash-free dry tissue weight. FTA concentration was quantified by organic extraction and HPLC in individuals of *I. felix*, resulting in the following: (a) Peripheral tissues had greater FTA concentration than internal tissues. (b) Total body FTA concentration was inversely and significantly related to the ambient illumination where the individuals were found (with depth, and comparing shaded vs. well-lit locations and localities with different water turbidity). (c) There was no significant variation in FTA concentration through the time of study (June to December 1995). (d) In a temporal scale of 2 months, it was found that experimental shading induced a significant increase in total body FTA concentration. (e) There was a strong FTA increase (in a scale of 1 week to 2 months) when sponges were manipulated in depth transference experiments and when they were purposely injured. (f) However, intact or injured individuals did not exude measurable quantities of FTAs into the surrounding medium in the laboratory. Together, these results indicate that FTAs may have some adaptive value, but probably not mediating in external ecological interactions, but instead acting as allomonal internal suppressors and/or antibiotics. The shade-dependent production of FTAs suggest that these substances may prevent parasitising by photosynthetic *Aphanocapsa feldmani*-type endosymbionts, when the ambient illumination is below their compensation point; also, as the sponge turns more heterotrophic in lower light levels it may have an increased need of antibiotics in the choanosome to prevent bacterial food from becoming infectious. Finally, during wound healing, increased FTA levels may also act as internal antibiotic protection.

Key words: *furanosesterterpene tetronic acids,* Ircinia, *chemical internal defense, Caribbean, natural products, antibiotic*

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ANNOTATED CHECKLIST OF MARINE SPONGES (PORIFERA) OF THE INDIAN REGION

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An annotated systematic checklist of sponges from the Indian region is presented. The list is based primarily on (a) identification of sponges in the holdings of Zoological Survey of India by several workers; (b) cross-referencing of all available literature pertaining to this region; and (c) personal correspondence with experts in the field both from European and Oriental regions.

The groundwork for this checklist arises from the century old work of Bowerbank (1873), Carter (1880-1887), Dendy (1887-1916) and Schulze (1894-1904) followed by the present century workers Annandale (1911-1915), Kumar (1924-1925), Dendy and Burton (1926), Burton (1928-1937), Burton and Rao (1932), Rao (1941), Ali (1956), Thomas (1969-1993), and Pattanayak (1995), with some revisions of this fauna also made by Hooper (1996).

With the exception of several papers by Thomas few taxonomic studies have been undertaken in the Indian region over the past two decades. Consequently most of the liturature and species descriptions are relatively old, sometimes too brief, and sometimes entirely inadequate to differentiate between related species. It is also well known that sponges are notoriously difficult to identify and misidentifications are common. Unfortunately re-examination of many type or voucher specimens was not possible given that they are scattered throughout many different museums and institutions. The preparation of this checklist therefore relies heavily on the published literature, with only relatively few species yet checked from original material.

For the purpose of convenience the Indian region is divided into 9 zones: (1) Arabian sea: Off-shore waters; (2) Laccadive Islands; (3) Northern West coast: Gujarat and Maharashtra; (4) Southern West coast: Karnataka and Kerala; (5) Southern East coast: Tamil Nadu and adjacent islands in the Gulf of Mannar (6) Northern East coast: Andhra Pradesh, Orissa and West Bengal; (7) Andaman Islands; (8) Nicobar Islands; (9) Bay of Bengal: Off-shore waters.

In the species checklist each published record is accompanyied by one or more literarture citation, the locality and region of collection, as defined above.

451 species are included in the checklist, with representatives from **3 classes**, **17 orders**, **64 families** and **168 genera**. It is anticipated that this checklist will be both expanded (with current work being undertaken by the Zoological Survey of India), and revised (as type material becomes available to check species' identities, and as contemporary authors increase their published revisions.

Keywords: *Systematics, Indian sponges, sponge identification.*