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CCRE REPORTS 2007-08

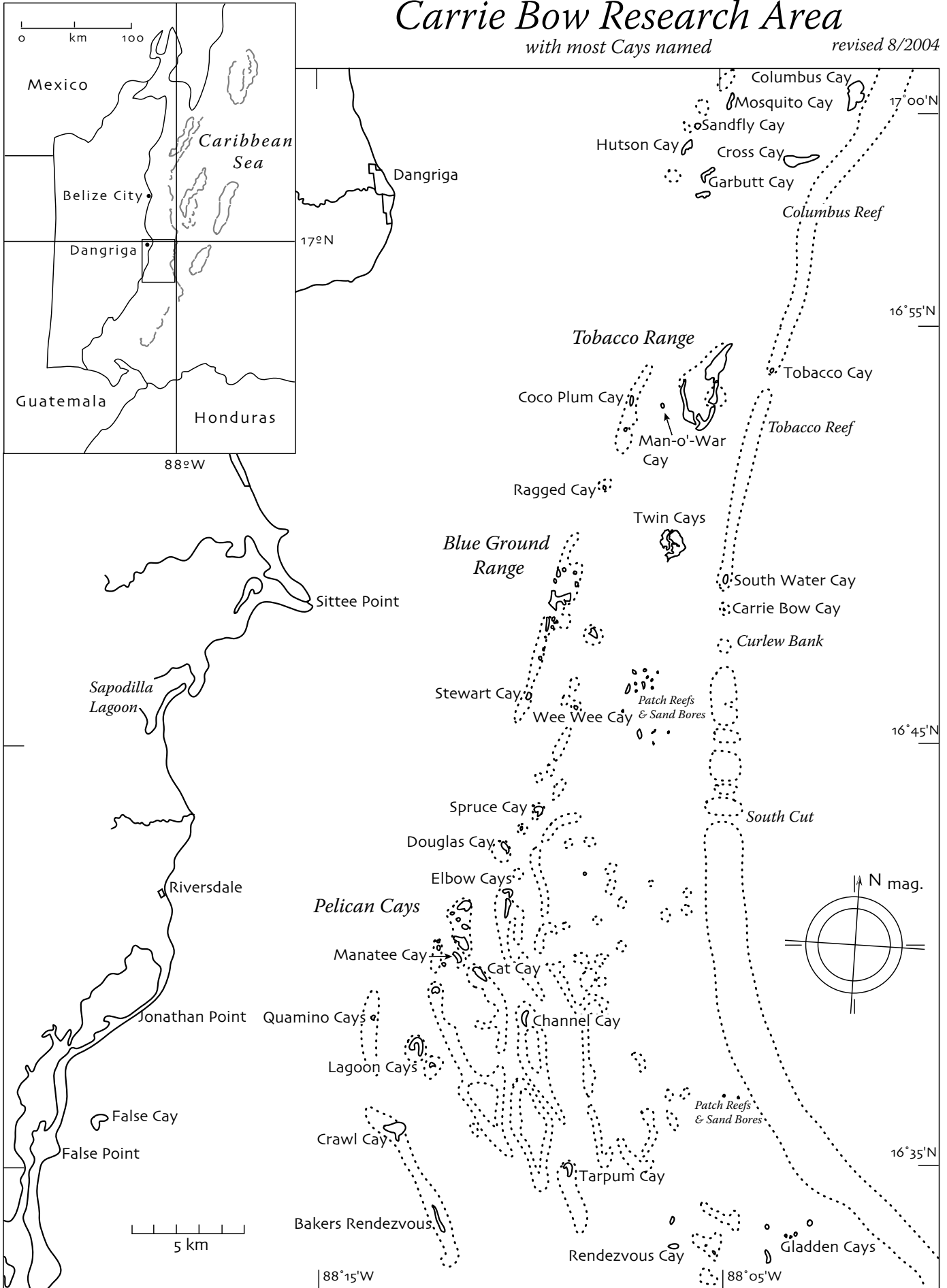
Caribbean Coral Reef Ecosystems • National Museum of Natural History

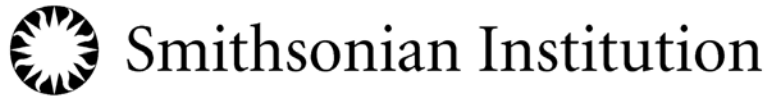
October 2008

Carrie Bow Research Area

with most Cays named

revised 8/2004





CCRE REPORTS
2007-2008

National Museum of Natural History
Caribbean Coral Reef Ecosystem Program
Washington, D. C. 20013-7012

October, 2008

Table of Contents

| | |
|---|----|
| CCRE News 2007-08 | 1 |
| Flashbacks..... | 3 |
| Acknowledgements | 5 |
| Research Projects | 6 |
| Biodiversity and its Links to the Ecosystem | 6 |
| Algae | 6 |
| Porifera..... | 10 |
| Annelida | 13 |
| Nemertina | 15 |
| Crustacea | 16 |
| Pisces | 19 |
| Paleobiology and Microevolution | 22 |
| Reproductive and Developmental Biology | 22 |
| Ecology, Population Dynamics, and Ecophysiology | 28 |
| Species Interaction and Behavior | 39 |
| Processes across Ecosystems | 46 |
| Publications 2007-08 | 51 |
| Participants 2007-08 | 53 |
| Photograph & Art Credits | 56 |

CCRE News 2007-2008

Nearly 120 scientists and their assistants worked at the Carrie Bow Marine Field Station during the report period. Almost as many groups of students, marine science teachers, and tourists from Belize and abroad visited for tours and demonstrations given by station managers or on-site researchers. VIP guests included National Museum of Natural History Acting Director Paul Risser and his wife (August 2007), hosted by CCRE co-founder and reef geologist Ian Macintyre (Dept. of Paleobiology); and the newly appointed Belize Minister of Natural Resources and his staff (May 2008), guided by conservation biologist Melanie McField (Smithsonian Marine Station, Ft. Pierce) who explained irreversible damages to cays in the Pelican archipelago caused by clear-cutting of mangroves for real-estate development. Less welcome visitors were two hurricanes in 2007, Dean (August) and Felix (September), but fortunately the damages to Carrie Bow were restricted to heavy beach erosion.

CCRE-associated researchers made 35 presentations at the Smithsonian Marine Science Symposium in Washington, hosted by the Office of the Undersecretary for Science (Michael Lang, organizer) in November 2007. The papers were mainly on biodiversity, geology, community ecology, and habitat conservation; a good number of them are now in preparation for publication in the recently re-established Smithsonian Contributions to Marine Science (planned for 2009). Two of the abstracts are reproduced below because of their general nature.



The Hon. Gaspar Vega, Deputy Prime Minister and Minister of Natural Resources and the Environment, in conversation with Mike Carpenter on Carrie Bow Cay.

CARIBBEAN CORAL REEF ECOSYSTEMS: 35-YEARS OF SMITHSONIAN MARINE SCIENCE IN BELIZE (KLAUS RÜTZLER)

In the late 1960s, a group of marine scientists from the Smithsonian National Museum of Natural History, Washington, founded a long-term Caribbean coral-reef field program, now known as Caribbean Coral Reef Ecosystems (CCRE). The core group consisted of botanists, zoologists, paleobiologists, and geologists. We were looking for a study location of high geological and biological diversity and minimal anthropogenic disturbance, suitable for long-term research. We settled on the tiny island Carrie Bow Cay on the barrier reef off Southern Belize and established a field station there in February 1972. A great variety of richly populated habitats, from mangrove to fore-reef, occurs within a distance of less than one mile. The Belize mainland coast and three offshore atolls are in easy reach by small boats. Each year, up to 120 Smithsonian staff and associated scientists, with assisting students and technicians, engage in the study of reefs and nearby mangroves and seagrass mead-

ows. Our expertise is “whole-organism” biology, involving systematics, evolution, paleobiology, ecology, and ecophysiology. Field research is complemented by use of the rich resources of the Smithsonian home base. Today, the CCRE program is member of the Smithsonian’s Marine Science Network, which includes costal laboratories in Panama, Florida, and Maryland. Field studies are mainly conducted by diving or wading and observations documented by samples, experimentation, and photography. Three small boats provide transportation to research sites. An oceanographic-meteorological monitoring station on Carrie Bow Cay records parameters such as tide, temperature, radiation, wind, and precipitation; data are made accessible through the Web. Additional sensors and recorders are applied in situ where required. We are also following the CARICOMP protocol for monitoring reef, mangrove, and sea-grass communities. The decline of reefs worldwide is accelerating and focus and resources are urgently needed to improve our understanding of biodiversity, community structure and dynamics, and environmental processes that control the ecosystem. The Caribbean is “our” American tropical sea, to which we are connected by weather, ocean currents, and marine resources, as well as by cultural and economic exchange. Fortunately, we were able to document the diversity and complexity of the originally pristine barrier reef complex near Carrie Bow Cay for more than 35 years and in over 800 publications. Despite considerable progress made by the CCRE and other research groups in the Caribbean, there are still many gaps in understanding the components and processes of coral reefs and related systems. Newly advanced methods, such as molecular techniques, will have to be applied and focus put on climate change and other stress factors responsible for the increasingly common occurrence of algal blooms and devastating invertebrate diseases. These topics and more need our full attention to help guide resource management and conservation efforts and preserve the esthetical and economic value of our reefs.

CARRIE BOW CAY AUTOMATED MET-OCEAN MONITORING: A DECADE IN SUPPORT OF SCIENTIFIC RESEARCH ON THE MESOAMERICAN BARRIER REEF (THOMAS B. OPISHINSKI)

In 1997 an environmental monitoring system was established at the Carrie Bow Cay (CBC) Marine Research laboratory in Belize to address the fundamental need for meteorological and oceanographic measurements. Many factors including operational environment, remote location, data accessibility, power restrictions, autonomous operation, and available communications for data transfer influenced the design criteria. In many ways the design criteria established for the system by CCRE in 1996 were both pioneering and vital to the long term success of the system. It was one of the earliest monitoring systems to process and transfer real-time data from a remote geographic location to a web site for public access. To our knowledge it is still the only auto-mated system continuously monitoring both oceanographic and meteorological conditions on the outer Mesoamerican Barrier Reef. The system continues to provide a baseline set of data used to examine long term trends, short term and seasonal cycles, and episodic events. This data has proven invaluable to management efforts for regional organizations and research studies for both Smithsonian scientists and an increasing number of organizations worldwide. To meet expanding needs of the users, continuous efforts are taken to improve and add functionality to the environmental monitoring system and supporting web site. This paper provides a general overview of the system, samples of new data offerings (e.g. statistical weather summaries, tidal forecasts and shoreline surveys), samples of data (illustrating trends, extreme weather events and seasonal dependencies), and examples of several projects that have utilized the data from Carrie Bow Cay. Also included is an introduction to some of the features of the newly designed web site (projected late 2007) that will provide interactive data analysis for the user, forecasting and real-time quality control of data.

Flashbacks

- 1971 • National Museum of Natural History's I.G. Macintyre (geology & sedimentology), W. Adey, P. Kier, T. Waller (paleobiology), A. Dahl (botany), A. Antonius (postdoctoral fellow, invertebrate zoology), M. Rice, and K. Ruetzler (invertebrate zoology) found the program Investigations of Marine Shallow Water Ecosystems (IMSWE).
- 1972 • IMSWE search party identifies Carrie Bow Cay on the barrier reef of Belize as ideally located and affordable site for long-term, collaborative field research on tropical coastal ecosystems
• Establishment of principal reference transect across the Belize barrier reef just north of Carrie Bow Cay.
- 1974 • Hurricane Fifi destroys laboratory structures, uproots coconut trees, and reduces the surface area of Carrie Bow Cay by about one third, to 0.4 ha.
- 1975 • EXXON Corporation provides grant for study of the coral reef ecosystem at Carrie Bow Cay.
• Marine and terrestrial post-hurricane surveys. • Establishment of all-manual meteorological station.
- 1976 • Refinement and calibration of profiles and maps with the aid of vertical aerial photographs taken by Royal Signals Detachment helicopter • Introduction of aerial photography by helium balloon for community mapping
• Submersible tide recorder installed at Carrie Bow Cay concrete dock.
- 1977 • Field trip to Carrie Bow Cay by participants of the Third International Coral Reef Symposium. • Aerial and underwater surveys expanded to cover the entire barrier reef of Belize • Geology team drills first cores to determine reef history • EXXON's The Lamp publishes article on company-sponsored research at Carrie Bow Cay ("Where seaworms glow..").
- 1978 • Hurricane Greta destroys Carrie Bow Cay field station.
- 1979 • Post-hurricane survey and rebuilding of laboratory with several improvements • Count of participating scientists and of published scientific contributions both pass the 50 mark; 23 scientific institutions are now collaborating with NMNH.
- 1980 • EXXON Corporation funds new initiative: comprehensive study of a western Atlantic mangrove swamp ecosystem, now known as SWAMP (Smithsonian Western Atlantic Mangrove Program)
• Mapping of Twin Cays, principal site of SWAMP, by aerial photography and ground truthing.
- 1981 • Initiation of Art in a SWAMP project where scientific illustrators and scientists collaborate in analysis and pictorial rendition of mangrove communities in time and space • Employment of H. Edgerton underwater time-lapse camera with strobe light (on loan from the inventor) to record day-night activity in benthic communities
• Vibracoring at Twin Cays to determine internal structure and development.
- 1982 • Publication of *The Atlantic Barrier Reef Ecosystem at Carrie Bow Cay, Belize, 1: Structure and Communities*. Smithsonian Institution Press (K. Ruetzler & I.G. Macintyre, eds.). 1983 • New weather protected and enlarged seawater system for laboratory experiments installed on Carrie Bow Cay • Series of extremely low tides at noon time were observed to have catastrophic effects on reef and mangrove organisms.
- 1984 • First automated weather station installed at Twin Cays • Cooperation with Belize Government identifying coastal marine areas suitable for natural resource conservation • Busiest year since program start: 8 months continuing laboratory operation for 45 research staff.
- 1985 • First year of operation of Caribbean Coral Reef Ecosystems (CCRE), a new program of the National Museum of Natural History. It replaces the old IMSWE project and supplements the ongoing SWAMP program which is supported by a renewed annual grant by the EXXON Corporation.
- 1986 • Renovations on Carrie Bow Cay to accommodate dry-laboratory space, added living quarters, and boat, diving, and laboratory equipment • Mangrove vegetation map for Twin Cays completed • Published scientific contributions pass the number 200.
- 1987 • Record visitation of Carrie Bow laboratory, 120 total: 90 scientists and assistants; others dignitaries, including the Prime Minister of Belize, Smithsonian administrators, and media people working on documentaries and news-related productions • Continued facility renovation, including addition of solar photovoltaic system, large seawater tank, twofiberglass whalers, fluorescence microscope, and time-lapse video recorder with underwater camcorder.
- 1988 • Mangrove workshop for 37 EXXON-SWAMP scientists at Solomons, Maryland, entitled *A Mangrove Ecosystem: Twin Cays, Belize*.
- 1989 • Science as Art exhibit at the Smithsonian's S. Dillon Ripley Center displays scientifically important and aesthetically pleasing products from SWAMP mangrove research, such as community drawings, paintings, photographs, and sculpture-like epoxy casts of soft-bottom animal burrows • Vandalized and malfunctioning weather

- station reconditioned and relocated to the Carrie Bow field laboratory • Mounting problems with anthropogenic stresses at research sites, such as heavy tourist visitation, garbage dumping, and clear-cutting mangrove trees.
- 1990 • CCRE-SWAMP program represented at first Caribbean Coastal Marine Productivity workshop, Jamaica, CARICOMP is a program for Caribbean-wide monitoring of environmental quality in reefs, mangroves, and seagrass meadows.
- 1991 • Belize Forestry Department helps stopping disturbances to SWAMP research sites. Belize Department of Natural Resources reviews legislation with intention of declaring Carrie Bow Cay - Twin Cays area protected research site • CCRE-SWAMP program staff participates in developing Belize Tropical Forestry Action Plan and helps designing Institute for Ecology to be based in Belmopan.
- 1992 • CCRE-SWAMP researchers produce video documentary on mangrove swamp biology • Unprecedented, severe problem with hydrozoan stings to snorkelers and divers in the Carrie Bow area traced to microscopic siphonophorans • CCRE-SWAMP staff and Belize Fisheries Department and Agriculture representatives conduct first workshop for Belize high-school teachers entitled Mangrove Conservation through Education • CCRE-SWAMP lecture series started in Belize City, co-hosted by Belize Audubon Society • CCRE officially joins the CARICOMP network and initiates monitoring program.
- 1993 • Belize Ministry of Natural Resources grants rights to Twin Cays for mangrove research • Launching of new 8 m (25 ft) research vessel *Physalia*, funded by a grant from the U. S. National Science Foundation, extends research radius over most of central and southern Belize • Ivan Goodbody pioneers surveys of Pelican Cays, a tunicate heaven at SSW of Carrie Bow. 1994 • Start of collaborative surveys and experimental projects in the Pelican Cays • Pelican Cays workshop, co-hosted by Candy Feller (SERC), at Edgewater, Maryland.
- 1995 • Finalized lease with the Villanuevas of Placentia to southern portion of Northeast Cay, Pelican group, to establish a field base for future studies • Malcolm Spaulding develops plans for new integrated environmental sensing system with radio- telemetry link to the University of Rhode Island's COASTMAP network.
- 1996 • Installation by Tom Opishinski of self-contained Endeco-YSI-Campbell monitoring station of meteorological and oceanographic parameters and hookup to Internet • Visit of field party from 8th International Coral Reef Symposium, Panamá.
- 1997 • Celebration of the 25th birthday of the Carrie Bow Marine Field Station • New U. S. National Science Foundation grant allows purchase of a second 8-m (25 ft) boat to back up the heavily used *Physalia* (under construction) • International team of seven expert systematists conduct workshop at Carrie Bow Cay to quantify the unusually high sponge diversity of the Pelican Cays • Number 500 reached of CCRE scientific contributions • Carrie Bow Field Station, including laboratories, weather station, kitchen, and living quarters is consumed by an accidental electric fire which was apparently sparked by a short in the wiring and aided by dry, termite-riddled lumber and strong northerly winds. Luckily, no-one was hurt.
- 1998 • Island clean-up and design for new field station completed. Construction work initiated but delayed by flooding and coastal erosion from hurricane Mitch • Completed editorial work on CD-ROM containing over 100 representative CCRE scientific papers that resulted from research at Carrie Bow Cay • Cosponsored Smithsonian (STRI) exhibit *Our Reefs –Caribbean Connections* in Belize City. Contributed large poster describing 25 years of CCRE coral reef research in Belize • Serious coral bleaching and die-off on reefs off Carrie Bow and Pelican Cays observed, partly caused by hurricane Mitch.
- 1999 • Rededication ceremony for the new Carrie Bow Marine Field Station, in August • BBC team (Bristol, UK) films segments for its *Blue Planet* TV series, including (with E. Duffy) eusocial shrimps living in sponges.
- 2000 • Publication of *Natural History of Pelican Cays, Belize*, in *Atoll Research Bulletin* (Macintyre & Ruetzler, eds, 2000) • Replacement of environmental monitoring station lost in the 1997 fire • Initiation of Twin Cays Biocomplexity Study funded by an NSF grant (to I. Feller & colleagues).
- 2001 • Completion of 3-room cottage over the eastern shore of Carrie Bow Cay • Hurricanes Michelle and Iris (October) barely miss Carrie Bow Cay, causing some damage to buildings and heavy beach erosion and devastate (Iris, in particular) large areas in southern Belize • Signing of MoU with Belize Fisheries Department officially acknowledging the Carrie Bow Marine Field Station as a nationally recognized laboratory • Publication of Golden (50-year anniversary) issue of *Atoll Research Bulletin* recognizing prominent coral reef scientists through their autobiographies, several of them participants in the CCRE Program.
- 2002 • Founding of the Smithsonian Marine Science Network (MSN), incorporating the CCRE Program and the Carrie Bow Marine Field Station • Number 600 reached of CCRE scientific contributions • Ranger Station established on southeast Twin Cays by Belize Fisheries Department to oversee South Water Cay Marine Reserve.
- 2003 • Cristián Samper, recently appointed director of the Smithsonian's Natural History Museum, visits the Carrie

- Bow station in July, makes dives on the barrier reef, and snorkels in mangroves habitats • Hurricane Claudette threatens Carrie Bow (July) and necessitates temporary evacuation • Smithsonian Secretary Larry Small visits the Carrie Bow lab in December and dives on the reefs • Twin Cays Mangrove Biodiversity Conference is held at Ft. Pierce, Florida (December), convened by Klaus Ruetzler, Ilka Feller, and Ian Macintyre, and cosponsored by Valerie Paul of the Smithsonian Marine Station at Ft. Pierce.
- 2004 • CCRE Postdoctoral Fellowship established • Hurricane Ivan causes substantial coastal erosion of Carrie Bow Cay • Atoll Research Bulletin volume dedicated to Twin Cays Mangrove Biodiversity goes to press • Number 700 reached of CCRE scientific contributions • Carla Dietrich takes over from Michelle Nestlerode as CCRE research assistant • Addendum to MoU with Belize Fisheries Department signed, clarifying intellectual property rights and issues of bioprospecting sponge in particular • CCRE Program Administrator Marsha Sitnik (recently, administrative advisor) retires.
- 2005 • A total of 13 hurricanes formed this season that came to a close on November 30. Three category five hurricanes, namely: Katrina, Rita and Wilma, caused substantial coastal erosion and damage to the, Carrie Bow facilities. All together, the record number of 25 named storms in the Caribbean area broke the previous record (from 1933) of 21 named storms • An external scientific review of the CCRE Program was conducted and resulted in a strong endorsement of the program's mission and accomplishments • Over 50 new CCRE scientific contributions were published.
- 2006 • The first Belize National Marine Science Symposium, cosponsored by Belize Fisheries and Forestry departments and the Hugh Parkey Foundation, took place and CCRE was represented with 4 talks and 8 posters, including a review of 35 years of Smithsonian Marine Science in Belize • CCRE hosted the U. S. Ambassador and 35 Embassy staff for a picnic, including a tour of the Carrie Bow lab facilities • More than 130 Smithsonian Associates, North Carolina teachers, and members of the Sierra Club visited Carrie Bow for guided tours of facilities and ongoing projects • A film crew for a Discovery channel in The Netherlands worked at Carrie Bow to document Gordon Hendler's work on newly discovered brittle-star light-sensing organs • The CCRE program and the Carrie Bow Marine Field Station, along with all other Smithsonian marine programs and facilities, took part in an external review ordered by the Smithsonian Undersecretary for Science; The efficiency and scientific productivity of the program and its field station received excellent marks.
- 2007 • Hurricane Dean strikes Northern Belize and Yucatan, Mexico (August), Felix passed over Honduras south of Belize (September); both cause major beach erosion at Carrie Bow Cay but no damages to buildings.
- 2008 • The Belize Minister of Natural Resources and his staff visit our facilities and tour the Pelican Cays to view damages caused by mangrove clear-cutting in this part of the Southwater Cay Marine Reserve.

Acknowledgements

Our research is hosted by the Belize Fisheries Department and we thank Ms. Beverly Wade and Mr. James Azueta for collaboration and issuing permits. The owners and staff of Pelican Beach Resort in Dangriga provided logistical support for our fieldwork; Therese and Tony Rath of Dangriga helped with local management, photography, and computer backup for our weather station.

Numerous volunteer managers helped run the field station and assisted in research activities; we greatly appreciate their many efforts: Jerry and Sandy Alanko, Sam Benson, Mike Carpenter, Earl David, Greg and JoAnn Dramer, Zack Foltz, Ed Hunt, Ed and Bonnie James, Joel Leavitt, Dan Miller and Claudette DeCourley, Joel and Linda Moore, Keith Parsons, Gary Peresta, Tom Pezzella, Bert Pfeiffer, Craig Sherwood, Jim Taylor and Tanya Ruetzler.

Back in Washington, we thank Marty Joynt and Carol Youmans (Department of Zoology) for fund management, administrative advice, and preparation of many documents. Michael Lang and Laurie M. Penland supervised and ran all aspects of scientific diving at Carrie Bow. We also thank the Smithsonian offices of the Undersecretary for Science and the Director of National Museum of Natural History for continued support. Numerous colleagues inside and outside the Smithsonian Institution contributed grant funds and home-base facilities to aid our program.

The CCRE program is supported by a Federal appropriation complemented by the Hunterdon Oceanographic Research Fund.

Research Projects

Biodiversity and its Links to the Ecosystem

Algae

Biodiversity of *Gambierdiscus* harmful dinoflagellates in the Belizean coral reefs and mangrove forests

M. Faust, P. Tester, W. Litaker, S. Kibler, M. Vandersea & C. Holland

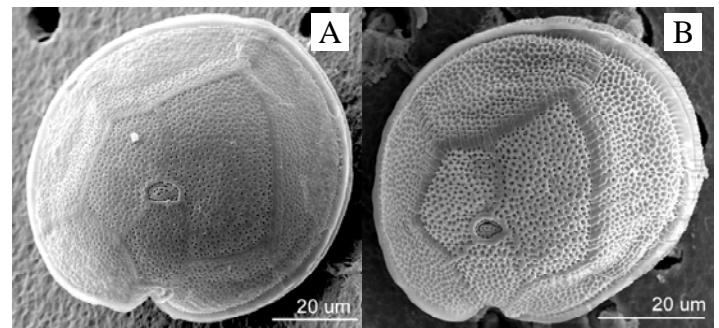
Research efforts at Carrie Bow have focused upon the distribution and ecology of dinoflagellates within the sheltered mangrove cay embayments and surrounding oligotrophic habitats in the central lagoon of Belize. The ecology and distribution of these dinoflagellates is of concern because some of these species produce the toxins responsible for causing ciguatera fish poisoning (CFP), the largest cause of non-bacterial food poisoning in the world. CFP adversely affects both human health and the development of fisheries resources in tropical regions. We have found that dinoflagellate populations in the embayments contain high concentrations of potential CFP-causing species and are supported by nutrients derived from the mangrove detritus. Single cell isolations have led to the identification of four new species in the CFP-associated genus *Gambierdiscus*, one new toxin-producing *Prorocentrum* species, and 2 new potentially toxic *Coolia* species. In 2007, we took advantage of the diverse array of habitats in proximity to the Field Station at Carrie Bow Cay to address the following two hypotheses: 1) Ciguatera-associated dinoflagellates are more abundant in naturally eutrophied or human-impacted sites and 2) *Gambierdiscus* species exhibit a preference for specific benthic habitats. Because CFP events are highly variable, both temporally and spatially, understanding species diversity, as well as nutrient and habitat requirements, may prove crucial in determining the conditions and species suites most predictive of CFP events.

Determining Dinoflagellate Abundances and Habitat Preference.

Because ciguatera dinoflagellates are often associated with the benthos and exhibit a very patchy

distribution, standard water-column collection methods used to quantify phytoplankton species were not applicable. Instead, we used small anchored squares of fiberglass window screens to which benthic species readily attach. The screens were deployed for 24h at various sampling sites (Figure), then transferred to ziplock plastic bags partially filled with ambient seawater and shaken to dislodge attached cells. Fixed samples of the resulting suspension were retained for cell abundance. Samples were also retained for DNA analysis and single cell isolation of ciguatera-associated dinoflagellates in the genera *Gambierdiscus* (Figure), *Coolia* and *Ostreopsis*.

Accumulation of cells on the screens indicated dinoflagellates were common (>7 cells cm²) in samples at all the embayment sites, with maximum abundance (>5000 cells cm²) observed inside Douglas Cay. Microscopic identification of the preserved cells from the 2007 screen samples are still in progress. However, preliminary data for the screens showed that the most nutrient enriched site, Douglas Cay was dominated by *Gambierdiscus* spp., *Prorocentrum* spp. and other dinoflagellates (DC, Fig. 1). At DC, the abundance of potentially toxic *Gambierdiscus* cells was noticeably higher than at open water, high energy sites such as Man-O-War Cay, Carrie Bow, and South Water Cay. The most abundant *Prorocentrum* species was *P. rathymum*, which also reached maximal densities inside DC. *Coolia* species were rare compared to previous years. The assemblage observed in DC was different than in the previous 2 years, when DC was marked by blooms of *P. mexicanum* (2005) and *Gambierdiscus* (2006). Our screen sampling also revealed several species of *Ostreopsis* were abundant in high energy, low nutrient environments on the windward side of DC, Carrie Bow Cay, Twin Cays, and South Water Cay. Representative samples are also being examined using SEM to provide additional information on the diversity of species present. These results require further investigation, but



SEMs of A) *Gambierdiscus caribaeus* and B) *G. belizeanus*.

suggest that *Gambierdiscus* and *Ostreopsis* may prefer different benthic environments and implies that sources of ciguatera toxins may be linked to specific habitats.

Because individual *Gambierdiscus*, *Coolia* and *Ostreopsis* species are very difficult to distinguish by light microscopy the screen samples are being evaluated using scanning electron microscopy, and species-specific PCR. These molecular assays are based on unique LSU rDNA sequences obtained from single cell isolates derived from samples collected in previous field seasons. Assay specificity was confirmed by testing whether the primers sets amplified other known *Gambierdiscus* species and by cloning and sequencing PCR products amplified from field samples. The SEM and molecular evaluation is ongoing, but preliminary screening has confirmed the presence of *G. caribaeus* and *G. belizeanus*, but not *G. toxicus*. Stations that were negative for *Gambierdiscus* by light microscopy have proven negative using the PCR assays as well. These data do not rule out the possibility that other species of *Gambierdiscus* were present in the samples. Seven other *Gambierdiscus* assays are in the final validation stages and will be used to detect *G. polynesiensis*, *G. australes*, *G. pacificus*, *G. yasumotoi*, *G. ruetzleri*, *G. carolinianus*, and *G. carpenteri* in the field samples. In summary, the screen cell count data illustrated an abundance of dinoflagellates within lagoon habitats and showed that the screens were an effective way to quantify benthic dinoflagellates. *Ostreopsis* species appear to predominate other CFP-associated dinoflagellates

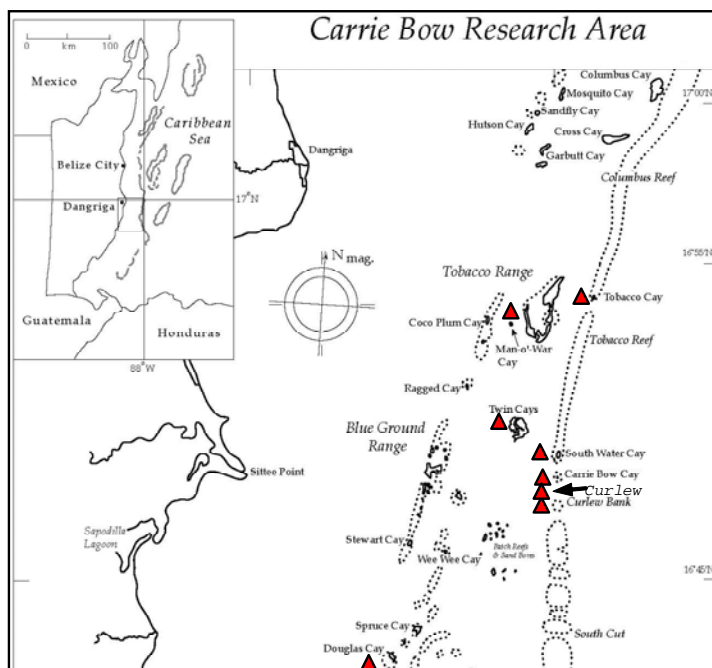
in high energy habitats. In contrast, *Gambierdiscus* are most abundant in high nutrient, low energy mangrove environments. We are concentrating on establishing single cell isolates of *Ostreopsis* to develop species-specific assays. The ability to specifically detect both *Gambierdiscus* and *Ostreopsis* will likely prove crucial for elucidating the mechanisms responsible for the temporal and spatial variability of ciguatera outbreaks.

Comparative diversity of ciguatera-associated dinoflagellates: Comparison of the Fort Pierce and Florida Keys regions with Belize

M. Faust, P. Tester, W. Litaker, S. Kibler, M. Vandersea, & C. Holland

A comparative survey of the Fort Pierce region was undertaken in July of 2008. Extensive sampling of the Indian River Lagoon system revealed no detectable *Gambierdiscus* cells even at the mangrove sites (Habitat III). In this respect the Indian River Lagoon is quite different than superficially similar sites in Belize. The channels opening to the ocean, however, had extensive macrophyte cover which consistently revealed low densities of at least two different *Ostreopsis* species. Live cells were returned to the laboratory and are now being cultured for future identification. Dives on the shallow carbonate sills offshore revealed low densities of *Gambierdiscus* and *Coolia* species despite prolonged upwelling and relatively cold water temperatures. Net tows were also collected in the Gulf Stream off Ft. Pierce allowing characterization of oceanic dinoflagellates and their diversity. Two harmful dinoflagellate species, *Ostreopsis siamensis* and the red tide-forming *Cochlodinium polykrikoides* were recognized. The collections also yielded *Ceratocorys horrida*, *Ceratium furca*, *C. carriensis*, *C. vulture* and *C. trichoceros*, *Dinophysis caudata*, *Pyrocystis noctiluca* and *P. lunula*, as well as the rare species *Amphisolenia bidentata*.

Given the low density of CFP-causing dinoflagellates observed at Fort Pierce we decided to collect samples from locations adjacent to Long Key, The Florida Keys in August 2008 to supplement data collected at the other two regions. Macrophyte samples collected to both the north and south of Long Key showed relatively abundant *Coolia* and *Gambierdiscus* cells but almost no *Ostreopsis*. Screen samples collected in a protected embayment revealed high concentrations of *Gambier-*



Study area surrounding Carrie Bow Cay with sampling sites marked (▲).

discus and *Coolia* cells. Although more extensive sample collection was curtailed by the arrival of tropical storm Gustav, a number of single cell isolates were established for SEM and molecular characterization and for representative cell cultures. In all, the 2008 field season has resulted in a total of 33 cultures of CFP-associated dinoflagellates. The collection of live material for culture has been crucial for both morphological and molecular characterization of new species and has provided the material necessary for physiological experiments and for toxin analyses.

Belizean coral-reef-associated mangrove forests: Habitats for motile, benthic, toxins-producing dinoflagellates

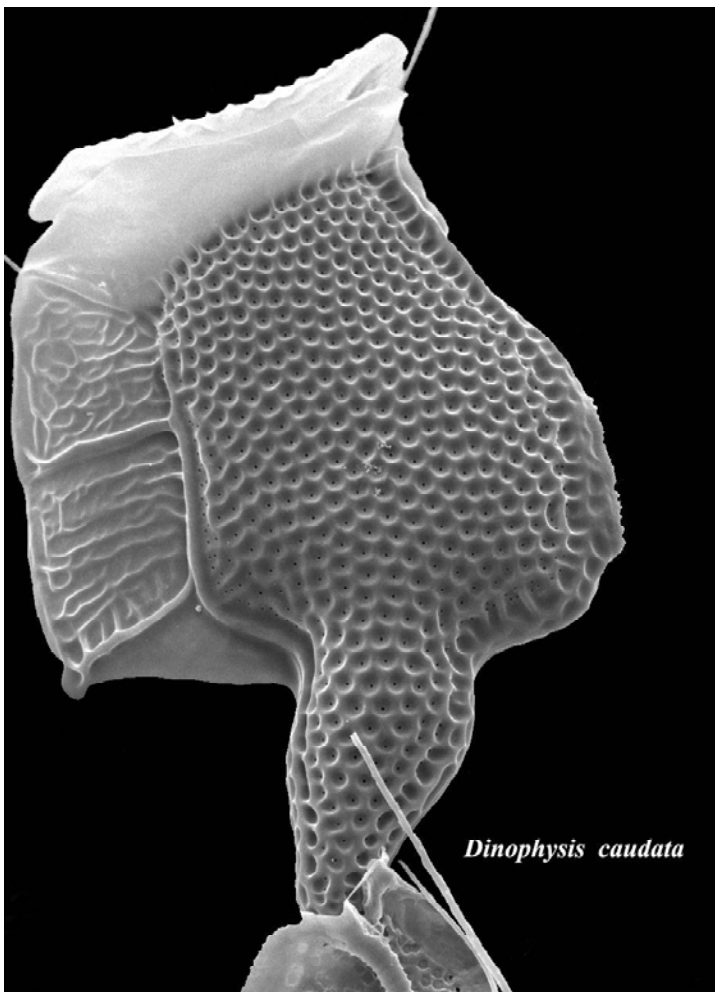
M. A. Faust

With the continuing decline in the world's coral-reef and mangrove natural resources, a question of public concern is how to assess the health of biological communities and ecosystems. The situation is especially problematic since the Atlantic barrier reef ecosystem

includes coral-reef-associated mangrove islands, shelters extensive mangrove lagoons, hosts tourists, and constitutes an important fishery resource for the country. Studies in the Pelican Cays, demonstrate that dinoflagellates are vulnerable and threatened by recent human activities of sediment runoff that caused water column turbidity and the loss of dinoflagellate species which are essential food source for fish and shellfish. Field research has identified numerous harmful dinoflagellates that thrive in sheltered mangrove cay embayments and surrounding oligotrophic habitats in the central lagoon of Belize. The ecology and distribution of these dinoflagellates is of concern because some of these species produce toxins. The aim of the research is to develop quantitative molecular assays, namely bar coding single harmful dinoflagellate species, some of which produce the toxins responsible for causing ciguatera fish poisoning, the largest cause of non-bacterial food poisoning in the world.

Biodiversity of dinoflagellates at elevated sea water temperatures in coral-reef mangroves in Belize

Field data suggest that elevated water tempera-



tures, 31° to 33° C, selectively changed dinoflagellate species composition, favoring *Ostreopsis* species in open water high energy, low nutrient habitats and *Gambierdiscus* species in low energy, high nutrient habitats. In previous years water temperature was lower ranging from 28° to 29° C, and rain fall provided additional nutrients to the sea supporting a broader range of dinoflagellate assemblages. Data indicate however that 2° to 3° C difference in water temperature, in the absence of rain, causes significant changes in the biodiversity of benthic and planktonic harmful dinoflagellate species. It is essential knowing the situation which may affect the species biodiversity of dinoflagellates caused by environmental variability in nature. The ecology and distribution of these dinoflagellates is of concern because some species produce the toxins responsible for causing ciguatera fish poisoning (CFP). Research established that harmful dinoflagellates are directly affected by elevated water temperature and the proximity of nutrient resources of either natural or anthropogenic origin in the habitats located in the central lagoon Belize.

Mixotrophy of harmful dinoflagellates in the Belizean coral reef-mangroves

Field research identified mixotrophy of the *Ostreopsis* species. Mixotrophy is a process when engulfed whole prey (microalgae, nannoplankton and protists) is found inside the cytoplasm of a dinoflagellate to supplement nutrients to a starving organism. Mixotrophy occurred in *O. labens*, *O. lenticularis*, *O. siamensis* and *O. ovata* assemblages dominating in plankton habitats: Southwater Cay, Man O' War Cay, Carrie Bow Cay and outside Twin Cays in high energy and low nutrient waters. These species exhibited cellular plasticity (cell volume increase of 30 to 50 %) during mixotrophic nutrition. Mixotrophy was observed infrequently in *Gambierdiscus toxicus*, *Prorocentrum belizeanum*, *P. hoffmannianum* and *P. arenarium* species in the nutrient enriched protected embayment of Douglas Cay. This phenomenon represents adaptation and survival of *Ostreopsis* species during absence of rain in oligotrophic warm ocean waters. Results indicate that the harmful dinoflagellate *Ostreopsis* species have the potential of being important grazers in the marine food webs when under environmental stress. This also implies that sources of biotoxins may be linked to specific habitats and need further investigation.

Taxonomy and phylogenetic relationship in the genus

Coolia, including two new species, *Coolia* sp. 1 nov. and *Coolia* sp. 2 nov. (Dinophyceae).

Two new *Coolia* species were identified. Both were morphologically distinguishable: *Coolia* sp.1 nov. has a small cell size, narrow 1' plate, pointed 3' plate, and a short apical pore; *Coolia* sp. 2 nov. have a medium size, broad 1' plate, and crescent-shaped 3' plate, and short curved apical pore. The morphology of the two new *Coolia* species differs from three known species, *C. monotis* Meunier (Type sp.), *C. tropicalis*, Faust, and *C. areolata* Ten-Hug et al. These species are supported by morphological differences and separate phylogenetic analyses of the small subunit rDNA genes. *Coolia* sequences included in phylogenetic analyses were derived from single cell isolates of *Coolia* sp.1 nov., *Coolia* sp. 2 nov. and *Coolia tropicalis*, and *C. monotis* sequences available in GenBank. The phylogenetic analysis of the SSU rDNA gene sequences showed that *Coolia*, *Gambierdiscus* and *Ostreopsis* formed monophyletic groupings consistent with three distinct genera.

Vulnerability of dinoflagellates in the Pelican Cays, Belize

Pelican Cays are delicate oceanic coral-reef-associated mangrove ecosystem and support high level of dinoflagellate abundance and biodiversity within the Belizean central lagoon. Pelican Cays are representative pristine oceanic cays situated in a remote region without major human perturbations. Dinoflagellate cells collected in Manatee Cay, Pond C in 1996 included 28 genera and 84 species; about 50% of total species were new reports in the Pelican Cays. Data in 2007 found only 6 genera and 17 dinoflagellate species. Field observations, as well recent aerial photographs, indicate that since January 2007 clearing of large tracts of land on several cays and destruction of the pond communities is extensive and continuing. Comparison of dinoflagellate species composition, before and after mangrove clearing, indicate dramatic loss in dinoflagellate species and a greatly changed microscopic food web, probably owing to high turbidity in the water column. Since taxonomic data of dinoflagellates in the Pelican Cays from 1996 to 2007 are available, the vulnerability of these organisms could be recognized. Because mangroves, like coral reefs and other tropical shallow-water communities exist near the limit of ecological tolerance of their inhabitants, dinoflagellates appear as significant indicators of changing environmental quality. Research data suggest Pelican Cays as an ideal research site that can provide an early biological warning system for

changing ecosystem water quality detrimental to the existence and survival of fish and shell fish.

Bar coding harmful dinoflagellate species

Gambierdiscus species are very difficult to distinguish by light microscopy. The morphology of individual cells is evaluated by SEM and species-specific PCR. These molecular assays are based on unique LSU rDNA sequences obtained from single cell isolates derived from samples collected in previous field seasons. Assay specificity was confirmed by testing whether the primers sets amplified other known *Gambierdiscus* species and by cloning and sequencing PCR products amplified from field samples. These molecular assays are based on unique LSU rDNA sequences obtained from single cell isolates derived from samples collected in previous field seasons. Assay specificity was confirmed by testing whether the primers sets amplified other known *Gambierdiscus* species and by cloning and sequencing PCR products amplified from field samples. Seven other *Gambierdiscus* assays are in the final validation stages and will be used to detect *G. polynesiensis*, *G. australes*, *G. pacificus*, *G. yasumotoi*, *G. ruetzleri*, *G. carolinianus*, and *G. carpenteri* in the field samples. The ability to specifically detect both *Gambierdiscus* and will likely prove crucial for elucidating the mechanisms responsible for the temporal and spatial variability of ciguatera outbreaks. This research is a collaborative effort with Dr. Pat Tester's group for identifying the morphology of dinoflagellate species at high resolution SEM microscopy and the genetics of single cell isolates with molecular taxonomy.

Porifera

Towards a DNA taxonomy of Caribbean demosponges: A gene tree reconstructed from partial mitochondrial CO1 gene sequences supports previous rDNA phylogenies and provides a new perspective on the systematics of Demospongiae

D. Erpenbeck, S. Duran, K. Rützler, V. Paul, J.N.A. Hooper & G. Wörheide

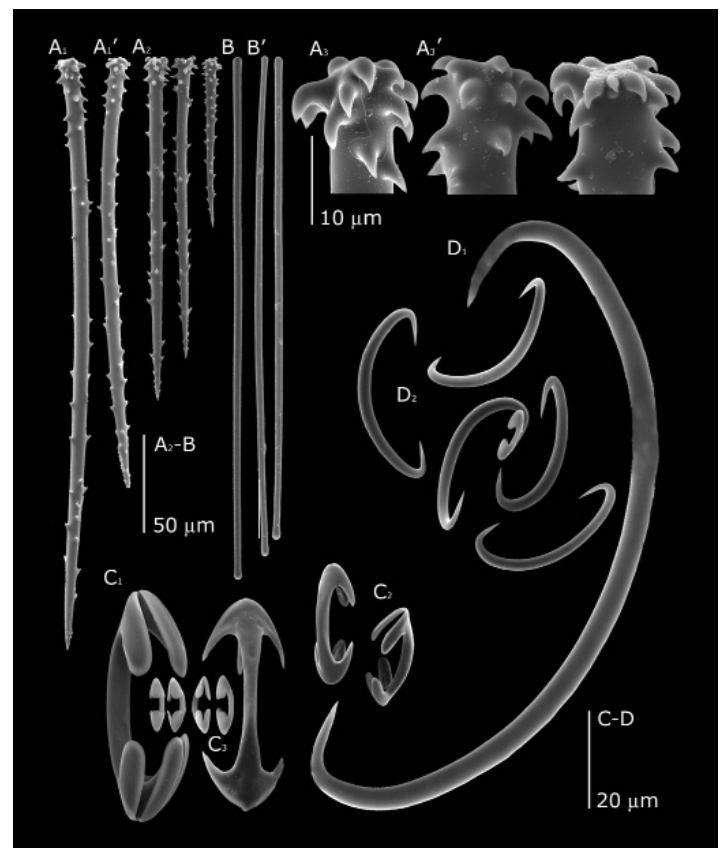
The most comprehensive cytochrome oxidase subunit 1 gene tree was published to date for demosponges based on new sequences. The CO1 barcoding

fragment is sequenced for 65 species from the Caribbean Sea, and its gene tree reconstructed. Although its deeper nodes are not particularly well-supported, the gene tree provides a variety of information for new phylogenetic patterns, as well as support for previously published 28S rDNA gene trees. In our analysis Halichondriidae cluster with Suberitidae, supporting previous 28S rDNA data. Chelae-bearing Poecilosclerida are monophyletic but most taxa lacking chelae in this dataset cluster more distantly. Haplosclerida are not resolved monophyletically under this fragment. While some species exhibit distinct barcodes, some genera contain species that share CO1 haplotypes.

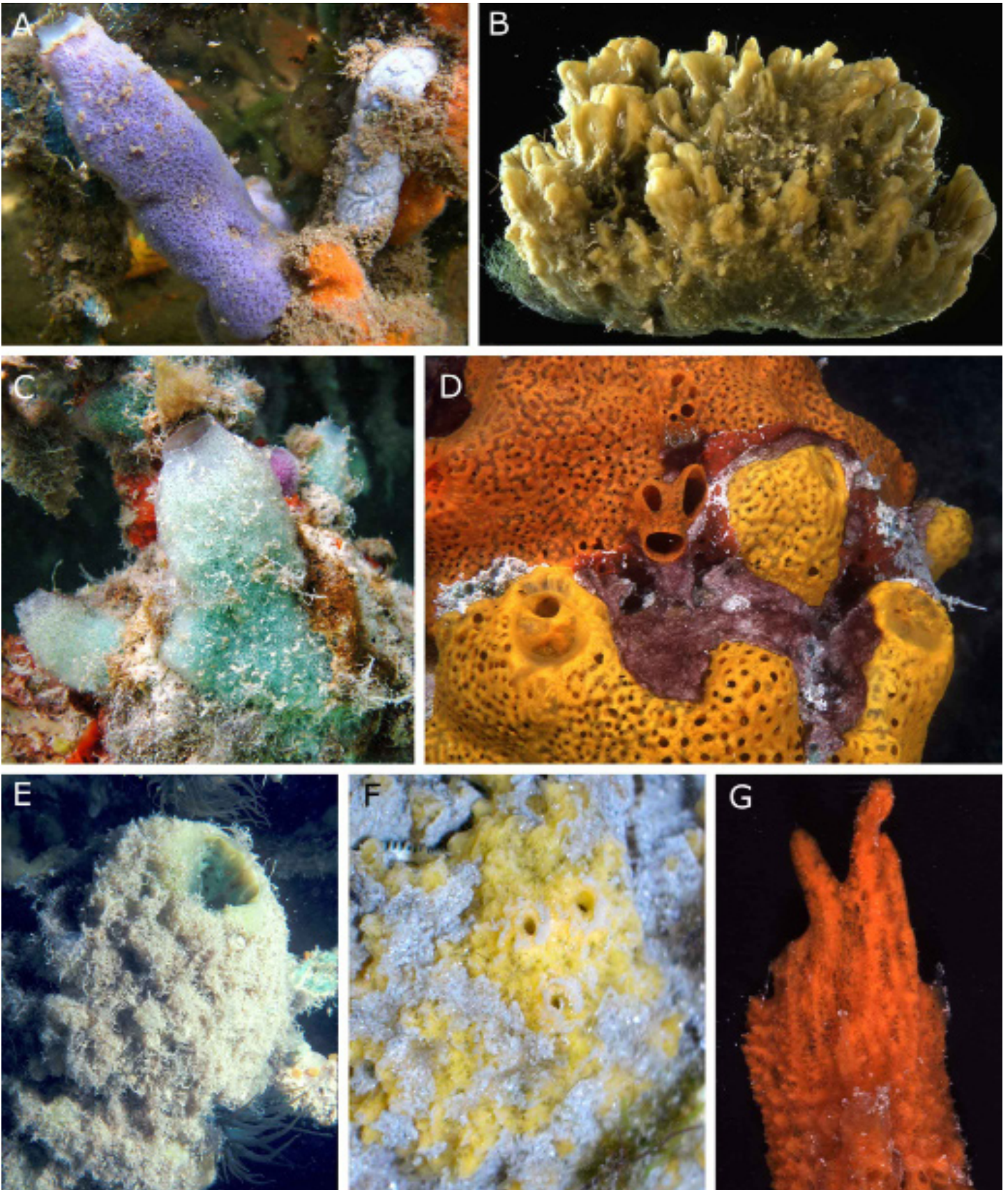
Lissodendoryx: rediscovered type and new tropical western Atlantic species (Porifera: Demospongiae: Poecilosclerida: Coelosphaeridae)

K. Rützler, C. Piantoni & M.C. Diaz

Seven syntypes of *Halichondria isodictyalis*, type species of *Lissodendoryx*, were rediscovered and studied. By choosing a lectotype and studying it along with the paralectotypes, we find that *L. isodictyalis*,



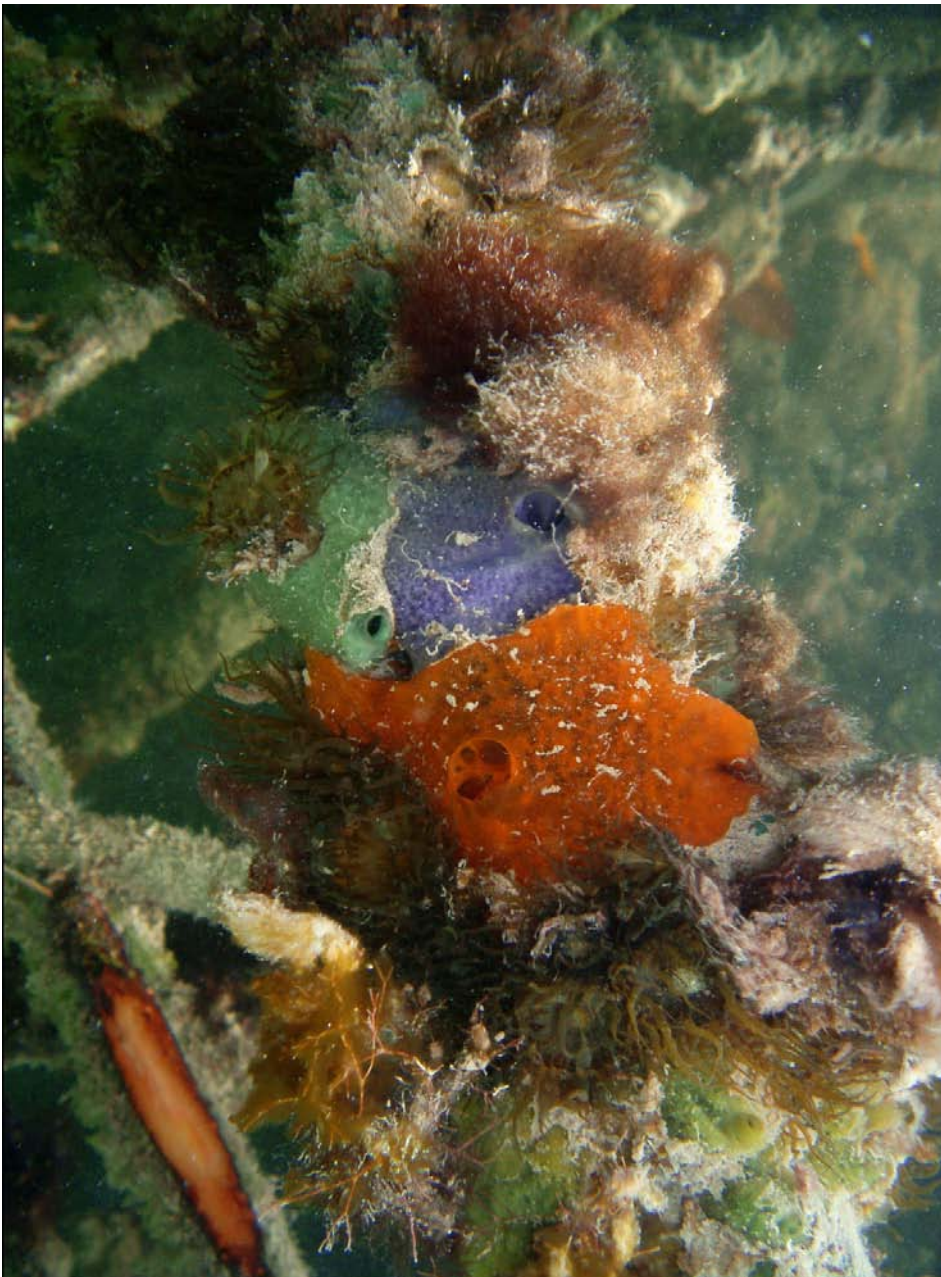
Lissodendoryx acanthostylota sp. nov.



Live specimens of *Lissodendoryx* species illustrating variations in shape and color. (A) *L. isodictyalis* (Carter) in situ; Twin Cays, Belize, mangrove; (B) Same species live in aquarium; Harrington Sound, Bermuda (photo: G. K. Jensen & W. E. Sterrer); (C) *L. carolinensis* Wilson in situ; Twin Cays (photo S. Duran & M. Becerro); (D) *L. colombiensis* Zea & van Soest, two color variants in situ; Twin Cays; (E) *L. spinosula* sp. nov. in situ; Twin Cays; (F) *L. sigmata* (de Laubenfels), var. nov.? in situ; Atlantic Florida; (G) *L. sigmata* (de Laubenfels), live in aquarium; Twin Cays.

subgenus *Lissodendoryx*, is a Caribbean sponge characterized by smooth megascleres, ectosomal tylotes and choanosomal styles, and one size-class of microscleres comprising arcuate isochelae and sigmas. Having determined these characteristics, we re-erect *Lissodendoryx* (*Lissodendoryx*) *carolinensis* (previously synonymized with *L. isodictyalis*), with the same smooth megascleres but two distinct size categories of microscleres, isochelae and sigmas; and we add a new western Atlantic species, *Lissodendoryx* (*L.*) *spinosa* sp. nov., with coarse spines on the megasclere terminals and with two size-classes of isochelae and sigmas. Other species in the region are *Lissodendoryx* (*L.*) *colombiensis*, with smooth tylotes and robust strongyles, and two categories of microscleres (isochelae and sigmas) accompanied by conspicuous raphids arranged in trichodragmas; *Lisso-*

dendoryx (*L.*) *strongylata*, with smooth tylotes and slim strongyles, one size-class each of isochelae and sigmas, rare and very thin raphids. *Lissodendoryx* *sigmata* is here assigned to the subgenus *Anomodoryx*, with smooth tylotes exclusively as megascleres, two size-classes of isochelae, and one or two sizes of sigmas; it may represent a species complex far more diverse than previously thought. To this subgenus we add another species, *Lissodendoryx* (*A.*) *amphispinulata* sp. nov., characterized by fine spines ornamenting both tyles of part of the tylotes. A third subgenus, *Ectyodoryx*, is represented by *Lissodendoryx* (*E.*) *acanthostylota* sp. nov., with smooth tylotes and finely spined acanthostyles in two size-classes, as well as two size-classes each of isochelae and sigmas as microscleres. All species studied alive occur in shallow lagoon habitats with mangroves and sea grass (*Thalassia*) but museum specimen records show that some may reach a depth of 60 m.



Biodiversity and abundance of sponges on the caribbean mangrove roots

M.C. Diaz & K. Rützler

Surveys of diversity and abundance of sponges in Caribbean mangroves were carried out between 2003 and 2007 with support of the Marine Science Network program. The results show that sponges are an important component of the red-mangrove (*Rhizophora mangle*) root epifauna, both in species richness and relative abundance. Sixty-five species were reported from Bocas del Toro (Panama) and 62 from Twin Cays (Belize) mangrove habitats. Three new Haplosclerida species with unusual (filamentous) cyanobacterial symbionts were discovered (genera *Haliclona* and *Xestospongia*) and are currently being described. Field guides for each fauna are being produced with the aim of facilitating the study of marine sponges by non-experts. A compari-



son of mangrove species composition from Bocas del Toro and Twin Cays shows a high similarity with other well studied Caribbean sites, such as Venezuela (65 species) and Cuba (43). However, since most Caribbean mangrove systems have not been rigorously evaluated taxonomically, a conclusive biogeographic analysis can not yet be attempted. The distribution of sponge species within each studied geographic region indicates that the majority (50-80%) presents a disjunct distribution, being restricted to one or a few sites within a particular locality. The relative abundance of major epibenthic taxonomic groups (such as, algae, cyanobacteria, ascidians, sponges, bivalves) shows great variations in both space (between localities of each region) and time (over the course of one year or more). Recommendations to be considered for future studies of mangrove sponges include, that surveys should cover long fringe distances to account for the disjunct distributions; and that ecological studies relying on abundance or changes in species composition should be based on one given locality and long-term because of large spatial and temporal variations in the relative importance of major taxonomic groups.

Annelida

Diversity of selected polychaetes families in Carrie Bow Cay and Twin Cays

G. Rouse

On my 2006 visit to Carrie Bow I mainly collected sabelid polychaetes for phylogenetic studies using molecular sequence analyses. The family Sabellidae was recently split into Sabellidae and Fabriciidae (Kupriyanova & Rouse 2008) and I focused my efforts at Carrie Bow and Twin Cays to finding a series of fabriciid species that were described from there by myself and Kirk Fitzhugh in the 1990s. I was successful in finding 5 of the 6 species present including specimens of an undescribed species in a new genus; picture attached. Sequencing is now complete and the phylogeny and species description will be published in the coming year.

Taxonomic status, abundance pattern and distribution of *Lysidice* and *Nematonereis* species (Polychaeta, Eunicidae) in the Western Caribbean Sea

M. C. Gambi, C. Vasapollo & K. Fauchald

Differently from other genera within the polychaete Eunicidae, the taxonomic status of *Lysidice* and *Nematonereis* of the

Western Caribbean Sea have not yet been evaluated. The cryptic habit of both genera (within dead corals, seagrass meadows and soft-bottoms) probably favoured their relatively high diversification especially in tropical areas, however, only a few species have been reported for the Western Caribbean.

Previous observation on material collected at CBC in a research stay of this team in November 2005, allowed to collect and confirm the occurrence of at least three new species of *Lysidice* whose morphological description is in progress, and which also showed a clear habitat selection: *Lysidice* sp. b and *Lysidice* sp. c were mainly associated to dead coral rubbles, while *Lysidice* sp. a and *Nematonereis* sp. occurred exclusively on the sheaths of the seagrass *Thalassia testudinum*. To confirm taxonomic status of such specie, increase the number specimens for the analysis of their genetic structure, and better define habitat selection and repro-

ductive biology, we needed to make additional observation and collect further material. In addition, for *Lysidice* sp. a and *Nematonereis* sp., associated to *Thalassia* as sheath borers, we performed a pilot study to first evaluate their abundance pattern and spatial variability in selected *Thalassia* meadows.

The aims of our research at Carrie Bow during this second study period of the team (3-17 October 2007) were:

1) collection of *Lysidice* sp. b and *Lysidice* sp. c in coral rubbles at different depths, to better define their morphology and genetic structure and relationships, and to test possible niche separation of the two species;

2) estimate the abundance pattern and the level of spatial variability of the sheath borers *Lysidice* sp. a and *Nematonereis* sp. in selected *Thalassia* meadows submitted to different environmental conditions.

Samples collection has been entirely conducted SCUBA diving by Gambi, Vasapollo, Miller and Keel, and lasted from 4th to 16th October for a total of 24 dives.

Coral dead rubbles were collected at various sites around CBC at depths from 1 to 20 m., both inside the lagoon (e.g., in front of Carrie Bow station, and at sand bores near Wee Wee Cay) and in the outer reef mainly in front of the CBC, at South Cut and Curlew bank. A few mature male specimens of *Lysidice* were fixed in 2.5% glutaraldehyde for electron microscopy analysis of gamete ultrastructure.

As for the two species associate to *Thalassia testudinum*, two meadows were selected, which both represent historical sites for long-term monitoring of *Thalassia* in the framework of the CARICOMP program: Carrie Bow Cay and Twin Cays. In each of these locations we accomplish a pilot study to clarify the distribution of borer polychaetes at different spatial scales. We operated according to a nested and hierarchical sampling design where in each location (CBC and TwC) two sub-sites (A and B) were identified at a distance of about 200 m each other; in each sub-site two stations (1 and 2) were established at a distance of 20-25 m each other; in each stations, 3 plots (1 m square) located at a distance of about 2-3 m each other, were considered as replicates. Within each 1 m square plot, between 30 and



Thalassia sheathborers.

40 shoots of *Thalassia* were sampled (mean 33 shoots per sample) and the shoot density was measured on a 30 x 30 cm frame; in each meadow 20 shoots were also analyzed for leaf morphology. All sampling stations were at 3-4 m depth.

A few other *Thalassia* meadows (sand borers near Wee Wee Cay, Man of War Cay, Twin Cays main channel) were considered for qualitative observations on borer occurrence in different conditions and depths (1 to 10 m). In the laboratory, all the *Thalassia* shoots collected were immediately checked for the presence of borer polychaetes; the collected animals were preliminarily identified at the stereomicroscope and then partially fixed in formalin 4% for morphological analysis, and partially dried for analysis of the stable carbon and nitrogen isotopes to define their diet.

As a whole, we were able to collect many specimens of both species of *Lysidice* associated to coral rubbles, including various mature, schizogamic female and male individuals. Part of these specimens were fixed in absolute alcohol for genetic analyses. As for the *Thalassia* borers we checked their presence in various meadows and found that they occurred in all sites considered but that were occasional at very shallow depth (1-2 m depth at Man of War and within the Twin Cays main channel), while they were particularly abundant at 10 m depth in meadows surrounding sand bores near Wee Wee Cay.

As for the two meadows selected for quantitative sampling, CBC and Twin Cays, in the various stations and replicates we analysed a total of 810 *Thalassia* shoots, finding 206 borer polychaetes, for a mean Index of Borer of 25% (IB = % of the shoots with borers over the total sampled).

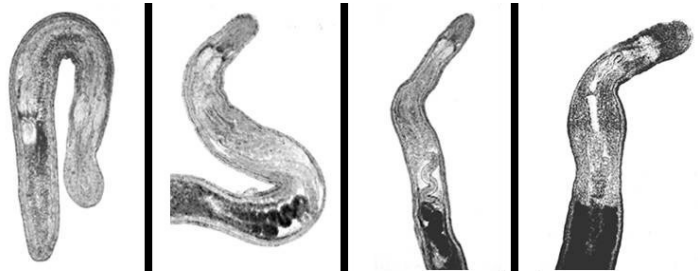
Abundance pattern was different in the two studied meadows, with Twin Cays stations showing significantly higher IB values than CBC ones. At Twin Cays IB values ranged between 20% and 38%, while at CBC they were lower, ranging between 20% and 5%.

Spatial variability among stations and sub-sites was evident at both meadows, but was higher at CBC, probably related to the fact that this meadow is characterized by a patchy distribution of shoots, it grows under more oligotrophic conditions, and showed lower shoot density (mean 394.4 shoot/m², at 3-4 m depth), respect to the Twin Cays meadow characterized by more uniform distribution, higher trophic conditions - due to the vicinity with large mangrove swamps - and slightly higher shoot density (mean 458 shoots/m² at 3-4 m depth).

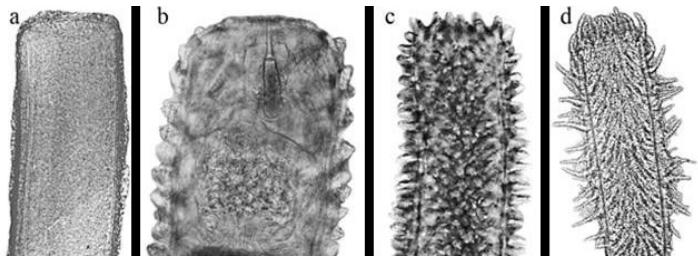
Phylogenetic and phylogeographic studies with the specialized interstitial genus *Ototyphlonemertes* (Hoploneurtea: Nemertea): Tiny worms take on big questions

J. Norenburg, S. Andrade, J.M. Turbeville & A. Tulchinsky

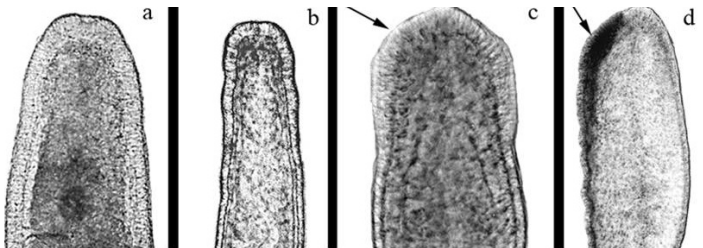
Ototyphlonemertes is a group of 21 recognized species and at least 30 more undescribed morphotypes that occupy the pore space of coarse marine sediments and share a number of features considered to be adaptations to that specialized mode of existence. The most conspicuous of these features is a pair of cerebral statocysts, which is a synapomorphy for the genus. There are two groups recognizable based on coordinated features of the statocyst and proboscis stylets – one group



Differences in some of gross morphological features evident even at low magnification.



Differing epidermal papillae on everted proboscis.



Caudal adhesive plate (arrows), primarily in surf zones; unique construction of adhesive and releaser glands.

has oligo-granular statoliths and smooth stylets, the other has polygranular statoliths and spirally sculpted stylets. However, monophyly of the genus or either of these groups had not been convincingly tested. The genus also can be divided into 6 “phylo-morphs” based on combinations of morphological features. The probability of convergent morphologies must be considered as significant, because the features that characterize the putative lineages also can be interpreted as having been strongly selected by the specialized mesopsam-mic habitat. Our goal is testing monophyly of the puta-tive lineages, as well as examining species boundaries. Specimens were collected from much of the world, with particular focus on the Western Atlantic Ocean, Carib-bean Sea and Eastern Pacific Ocean. We present results of phylogenetic analysis of sequence data from three genes for about 50 morphs, representing each of the 6 phylomorphs. Our results support monophyly of the genus but not reciprocal monophyly of the two groups de-fined by statolith structure. In addition to a phylogeny, we have sufficiently fine-grained data for some morphs to begin examining phylogeography.

Crustacea

History of the social shrimp: Insights from long-term research at the Smithsonian’s Caribbean field stations

J.E. Duffy, K.S. Mac-donald III, C.L. Morris-son, R. Ríos & E. Tóth

Snapping shrimp in the genus *Zuzalpheus* (formerly *Synalpheus*, in part) are abundant and diverse residents of coral reef ecosystems world-wide and include the only marine animals known to live in eusocial colonies. Research conducted at the Smithsonian’s field sta-tions at Carrie Bow Cay and in Caribbean Panama over nearly two decades has revealed much of the previously unknown

natural history, ecology, and evolution of these unique and fascinating animals. We have described nine new species and a new genus of alpheid, documented host associations in detail, reconstructed the group’s phy-logeny, observed behavior in captive colonies, and integrated these data to evaluate the ecological and evolutionary consequences of social life. Calibration of a molecular clock using three transisthmian species pairs suggests that Caribbean *Zuzalpheus* radiated rap-idly ~6 Mya during closure of the Panama seaway. The improved taxonomy has shown that host associations of sponge-dwelling shrimp are much more specialized than previously believed. Comparative studies reveal that eusocial life has had pervasive consequences for shrimp morphological evolution, life history, and ecol-ogy. For example, after controlling for phylogenetic re-latedness, eusociality is associated with smaller body size, and a switch in the direction of sexual dimorphism toward (paradoxically) relatively smaller females pro-ducing smaller clutches of eggs. Eusocial species also attain greater abundance and use a broader range of host sponges, supporting the hypothesis that sociality confers a competitive advantage in the crowded environment of the reef. Ongoing research uses social shrimp as a model for addressing general questions about animal social organization, focusing on how patterns of mating and dispersal influence the fine-scale genetic structure of social colonies, and how genetic relatedness in turn affects the balance between cooperation and conflict.



Obvious invaders and overlooked infauna: Unexpected constituents of the Decapod crustacean assemblage at Twin Cays, Belize

D. Felder, P.C. Dworschak, R. Lemaitre, R. Robles, H.D. Bracken, A.M. Windsor & J. Felder

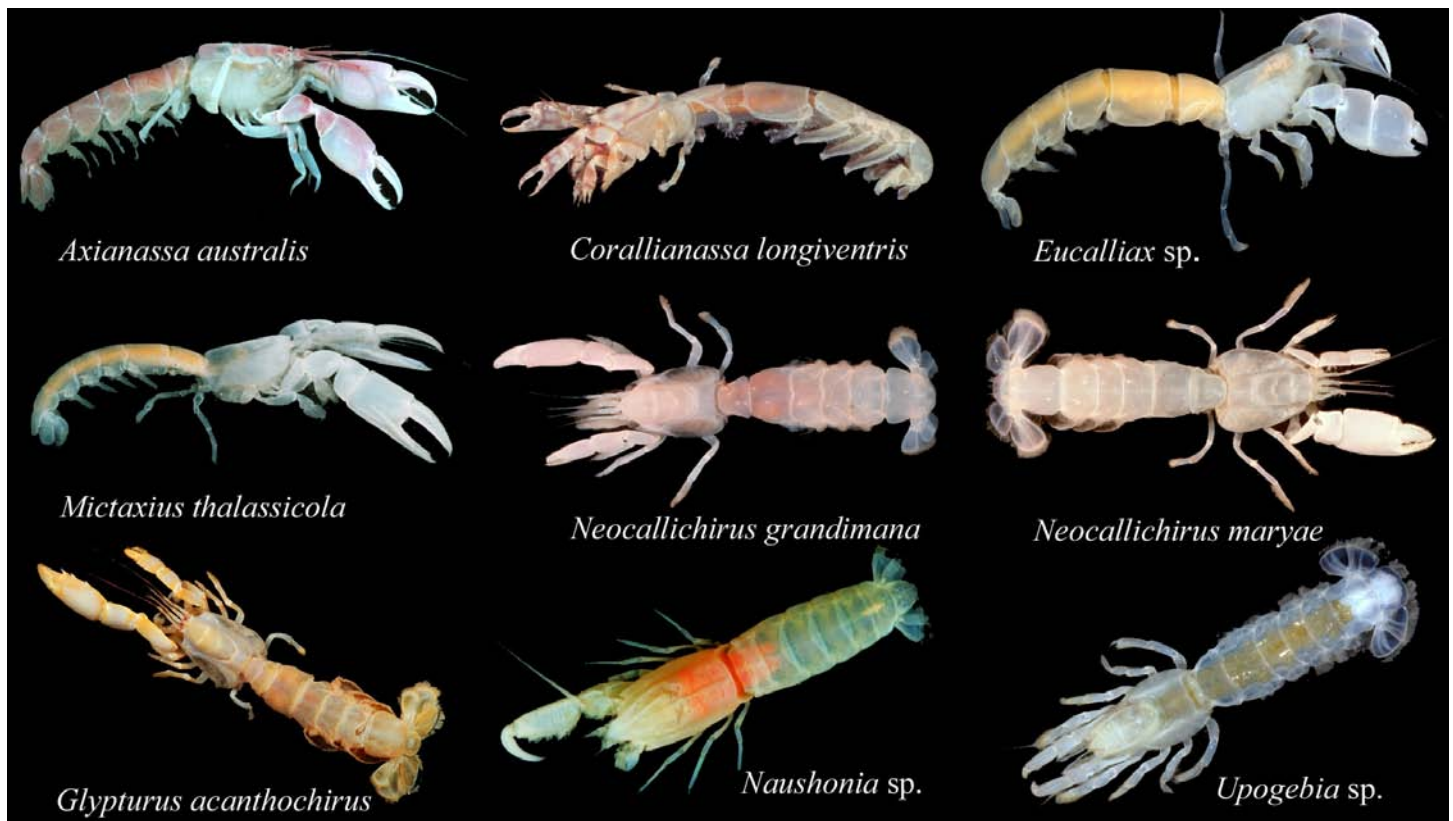
Decapod crustaceans in the vicinity of Carrie Bow Cay and Twin Cays, Belize, have been under study by ourselves and colleagues for over 25 years. In the course of investigations, new species have been discovered and large collections have been assembled, with many systematic problems yet to be resolved. Much of the effort has included photographic documentation of coloration in life, yielding characters of value in identification of problematic tropical taxa. Especially at Twin Cays, our measure of diversity has been markedly elevated by sampling in shallow subtidal muds with extraction corers (yabby pumps), and this has recently revealed species, genera, and families of thalassinidean decapods not previously known from the region. This also provided opportunity to explore ecological roles of callianassid burrowers, many of which are dominant bioturbators in intertidal to subtidal grassbeds of Twin Cays, producing conspicuous mounds of sediment

and constituting major infaunal biomass. By contrast, a familiar group of conspicuous brachyuran crabs and palinurid lobsters typically dominate macrocrustacean fauna of shallow rocky substrates. However, within the last four years, rocky habitats at Twin Cays have been massively invaded by the nonindigenous portunid crab, *Charybdis helleri*. In 2007, it was found to dominate cavities beneath coral heads in survey areas along the NE shoreline and the SW shoreline, possibly displacing populations of large *Mithrax*, *Menippe*, *Callinectes* and *Panulirus* previously found there in abundance. What we know of this fauna continues to change because of how we sample, but now also because of threats to faunal composition and stability.

New species and genera of fish-parasitic isopods from Carrie Bow Cay

E.H. Williams, Jr., L. Bunkley-Williams, & M.J. Dowgiallo

Bunkley-Williams and Williams (1981) named nine new species of large, external fish-parasitic isopods in the genus *Anilocra*. Two of these species occur at Carrie Bow Cay, Belize (*A. haemuli* on the French Grunt and *A. myripristi* on the Blackbar Soldierfish.



Representative thalassinideans distributed in Carrie Bow, Twin Cays and the Stann Creek District, Belize.

Many years ago the Smithsonian gave us a preserved, but undated, damaged isopod attached to a damaged damselfish possibly from Belize. In 2001, we searched for this isopod on the off shore cays, in 2006, MJD searched the on shore shelf area. Not until this year was MJD able to collect the host (Longfin Damsselfish) and the isopod (*Anilocra* n. sp.) at Carrie Bow Cay. Thus a longtime mystery was solved, but more specimens will be needed to make a proper description. Also the abundance and distribution of this parasite needs to be determined. MJD found an immature isopod on juvenile Longfin Damsselfish and adult female on the adult fish. This might suggest a different life history for this isopod as compared with other Caribbean *Anilocra*, but further study is necessary to confirm the life cycle.

A different new species of isopod was also collected from deepwater damselfish. This may represent a new genus as well as a new species. Williams and Bunkley-Williams (1985) previously described a new genus and species from Sergeant Majors collected at Carrie Bow Cay by Roger Cressey. This fish-parasitic isopod also occurred in Curaçao, Colombia, Panama, and the Florida Keys, and we have recently found it in Barbados.

Fish-parasitic isopods are an important detriment to the health of commercially important fishes (Bunkley-Williams and Williams, 1998) with strong manage-



Adult female *Anilocra* sp. on longfin damselfish.

ment implications in cultured fishes. Their presence in the western Caribbean has been poorly studied. A more comprehensive field study at Carrie Bow Cay could not only improve the knowledge of this important group of parasites, but provide additional specimens to allow the description of two new species and a new genus and possibly find other new isopods. The nine species of *Anilocra* named by Bunkley-Williams and Williams (1981) and the two to three new species to be described in this genus are being used to study the process of speciation. Our distribution studies have suggested the different species appear to be at different stages of speciation. Ongoing molecular studies of parasitic isopods at the University of Puerto Rico at Mayagüez will further elucidate the systematic relationships among and between this interesting group of parasitic isopods.

Studies of commensal Leucothoid Amphipods of the Carrie Bow Cay and Pelican Cays Region, Belize

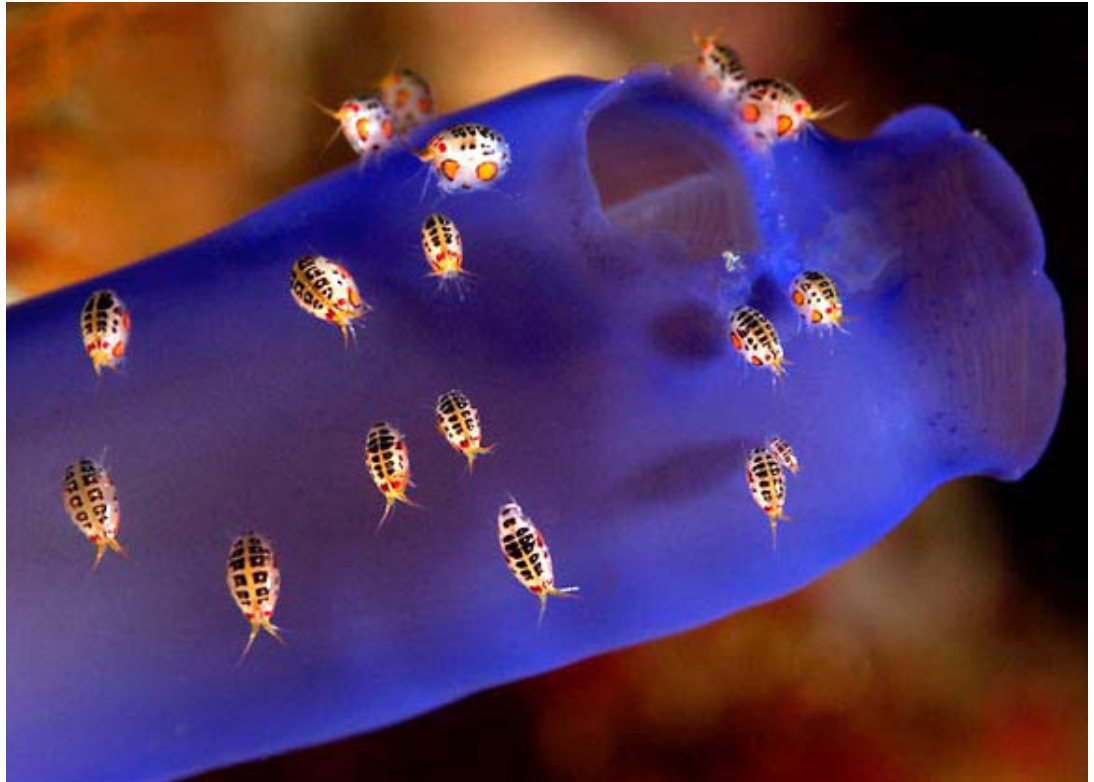
J. Thomas & K. White

As a result of our studies six new amphipod species in the genus *Leucothoe* from the tropical western Atlantic Ocean have been formally described and illustrated in recent publications. Five of these species are named using terms from the tradition-



Immature *Anilocra* sp. on juvenile of longfin damselfish.

al Garifunae language of Belize. Continued extensive field collecting and specialized underwater collecting techniques have documented 43 new invertebrate host records for these new taxa. Four of these new species inhabit interior canals of sponges; *Leucothoe barana* n.sp., *Leucothoe garifunae* n.sp., *Leucothoe saron* n.sp., and *Leucothoe ubouhu* n.sp. A remarkable new species, *Leucothoe flammosa* n.sp., nestles in the gills of seven species of bivalve mollusks. A single spe-



cies, *Leucothoe wuriti* n.sp., appears restricted to the branchial chamber of two species of solitary ascidians. Detailed illustrations and scanning electron microscopy has enabled comparison of ultrastructure details among all species. More precise taxonomic character morphologies are being developed as a result of these ongoing studies thus allowing improved taxonomic precision within the family Leucothoidae.

Pisces

The rich ichthyofaunal diversity within the mangal of the Belize offshore Cays

D.S. Taylor, E.A. Reyier, C.C. Mclvor & W.P. Davis

We assessed ichthyofaunal diversity within offshore mangrove cays in Belize during three, two-week surveys (2003, 2004, 2005). Nine sampling gears were deployed in pre-defined micro-habitats: fringe, transition, dwarf red mangrove, internal creeks, ponds, and sinkholes. Water quality data (temperature, salinity, DO) were taken during most collections. A total of 2,586 gear sets was completed and 8,131 individuals collected, comprising 75 taxa. Minnow trap data from the various micro-habitats tested indicates some overlap in assemblages. Significant differences in water quality were also noted, with the fringe presenting the best conditions and

sinkhole the worst. We also conducted extensive visual surveys around the fringe at a number of cays, tallying an additional 67 taxa. The fringe is the most diverse (128 taxa) and sinkhole least (12 species). An overall total of 142 taxa from 55 families has therefore been documented from the cays, and all but eight were found on Twin Cays alone. This figure is among the highest reported for oceanic mangroves in this biogeographic realm. Our comprehensive approach with a variety of gear-types in a wide range of micro-habitats, combined with visual observation, lends credence to the conclusion that most ichthyological species inventories for the mangal are commonly underestimates.

Taxonomic abundance and composition of the larval ichthyofauna located at the neritic transition on the forereef of Carrie Bow cay, Belize

K.S. Cole

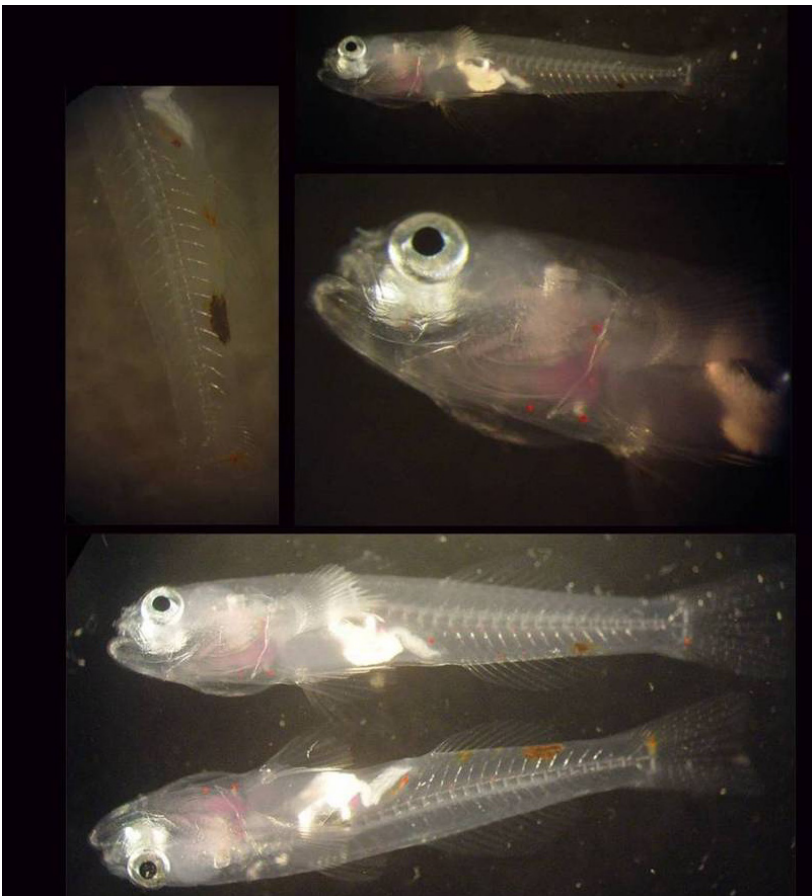
Carrie Bow Cay comprises part of the Belize barrier reef system which marks the transition from

oceanic to neritic epipelagic waters in this region of the western Atlantic. Prevailing north-easterly trade winds generate a north-to-south current that sweeps along the ocean side of the barrier system, providing a steady stream of ready-to-metamorphose fish larvae. Quantification of the larval ichthyofauna, based on repeated sampling of the Carrie Bow forereef over the last three years, has yielded an unexpected pattern of taxonomic abundance and composition of the larval ichthyofauna. Among the 15,000+ larvae collected, sorted, identified and counted, there was an overwhelming numerical dominance of relatively cryptic species. Larvae of two species, *Ctenogobius saepepallens* (the dash goby, Family Gobiidae) and the speckled worm eel, *Myrophus punctatus* (F. Ophichthidae) typically comprised over 50% of all collected specimens. Interestingly, both of these species live in burrows, are relatively inconspicuous, and would not be seen during traditional visual fish surveys. Given the nature of these findings, the ecological importance and relative contribution to energy flow for some cryptic fish species occupying coastal and reef habitats of Belize has likely been greatly under-estimated.

Does color pattern drive speciation in *Hypoplectrus* coral reef fishes?

E. Bermingham & O. Puebla

Theory shows that speciation in the presence of gene flow occurs only under narrow conditions. One of the most favorable scenarios for speciation with gene flow is established when a single trait is both under disruptive natural selection and used to cue assortative mating. We demonstrate the potential for a single trait, color pattern, to drive incipient speciation in the genus *Hypoplectrus* (Serranidae), coral reef fishes known for their striking color polymorphism. We provide data demonstrating that sympatric *Hypoplectrus* color morphs mate assortatively and are genetically distinct. Furthermore, we identify ecological conditions conducive to disruptive selection on color pattern by presenting behavioral evidence of aggressive mimicry, whereby predatory *Hypoplectrus* color morphs mimic the color patterns of non-predatory reef fish species to increase their success approaching and attacking



Family Gobiidae. *Nes longus*: early larval pigmentation.

prey. We propose that color-based assortative mating, combined with disruptive selection on color pattern, is

driving speciation in *Hypoplectrus* coral reef fishes



Unidentified fish larva.

Sympatric speciation by hybridization in a marine fish

L.A. Rocha

Mechanisms that lead to speciation remain among the most debated topics in evolutionary biology, and sympatric speciation is especially difficult to demonstrate in nature. Because of their peculiar biogeography and their rare ability to produce sounds, fishes of the genus *Haemulon* serve as a great subject for tests of speciation hypotheses. Collectively known as grunts, the genus is comprised of 19 nominal species and occurs in tropical and subtropical reefs along both sides of the Americas. Aiming to elucidate the phylogenetic relationships among the species of *Haemulon*, a combined total of ~2,000 base pairs from two mitochondrial genes (cytochrome b and cytochrome oxidase I), one nuclear intron (TMO-4C4) and one nuclear gene (RAG2) were obtained from all nominal species. Our data also indicate that the trans-isthmian *H. steindachneri* is composed of

two species, one in each side of the Americas, and we propose the revalidation of the Atlantic species. The closure of the Isthmus of Panama seems to have played a role in the diversification of *Haemulon*, however, many sister species pairs have completely overlapping geographical distributions, indicating that vicariance is not the only process driving speciation in this genus, and that sympatric speciation by sound recognition is possible. Finally, the species *H. carbonarium* seems to have originated through a hybridization event between *H. macrostomum* and *H. flavolineatum*. These three species form a strongly supported group in the phylogeny, however, mtDNA groups *H. carbonarium* with *H. macrostomum*, whereas nuclear DNA groups *H. carbonarium* with *H. flavolineatum*. A detailed morphological analysis shows that many morphological characters in *H. carbonarium* are intermediate between *H. macrostomum* and *H. flavolineatum*, indicating a probable hybrid origin for *H. carbonarium*. If supported by additional ongoing analyses, this will be the first case of sympatric speciation by hybridization in a vertebrate animal.

Paleobiology, Microevolution and Biogeography

Biogeographic variation in the recruitment of native and invasive marine sessile invertebrate species

R.W. Osman

The biogeographic variation in the timing and rates of recruitment of sessile marine invertebrates is being examined by contrasting recruitment to panel substrates exposed at sites in southern New England (Connecticut), Chesapeake Bay (Maryland and Virginia), Indian River Lagoon (Florida), and Belize. Panels were exposed for 1 week periods at several sites in Connecticut and for 2 week periods at all other sites. The general patterns observed were: 1) that recruitment rates were often inversely correlated with diversity with the highest rates seen in Chesapeake Bay and the lowest in Belize, 2) seasonal variability in recruitment varied latitudinally, 3) the timing of peak recruitment shifted latitudinally for some invasive species and this timing shift can be related to temperature, 4) annual and within-region variability in recruitment can be extremely high, and 5) this variability can be related to temperature for some invasive species but not for native species. The most dramatic pattern to date is for the bryozoan *Bugula neritina* which recruits in the winter in Florida and the late summer in Connecticut. From historical data it appears that it recruits in the late spring in North Carolina. The temperatures at these different recruitment times are similar among sites, suggesting that recruitment patterns reflect biogeographic variation in temperature. Within regional differences in recruitment can also be quite dramatic with sites separated by <1 km often differing by an order of magnitude in their recruitment. These differences were seen in all regions. In addition, differences between sites in species composition were more prevalent at low-latitude sites. The within-region

and biogeographic variation in recruitment suggest that both invasion success and the impact of climate change on recruitment and its consequences will be complex.

Reproductive, Molecular and Developmental Biology

Microbial ecology of corals: Investigating bacterial communities in early stages of Caribbean corals

K.Sharp

Coral cover and diversity worldwide are heavily threatened by environmental changes and outbreaks of coral bleaching and disease. Healthy scleractinian corals have been shown to harbor diverse assemblages of microbes, but neither the specificity of these associations nor the mechanisms that maintain them across host generations is well-understood. In order to implement effective management strategies and conservation concerning unhealthy and diseased corals, it is critical to learn more about the diversity and functions of bacterial assemblages in healthy corals. To date, there are only a limited number of examples in which the role of bacteria on coral health is well-understood. Some corals, including the Caribbean corals *Porites astreoides* and *Favia fragum*, brood larvae within adult colonies, releasing fully developed swimming planulae into the



water column. In this study, bacteria were found in the planula larvae of both *P. astreoides* and *F. fragum*. Molecular techniques were used to identify microbes associated with larvae, and sequence-specific oligonucleotide primers were designed to survey multiple samples for the presence of particular bacterial species. Fluorescence in situ hybridization (FISH) and microscopy were used to localize particular species within the larvae and determine relative abundance of certain groups of bacteria. Egg-sperm bundles released from *Montastrea* and *Acropora*, which, in contrast, reproduce via mass-spawning, did not appear to contain bacteria or archaea. These results reveal new insight into mechanisms by which marine invertebrates maintain complex microbial assemblages during embryogenesis and early development. In addition, the localization of bacteria in the larvae present the possibility for a bacterial role in larval settlement of some coral species.

Development and evolution of the musculature in sipunculan worms

A. Schulze & M.E. Rice

The taxonomy of the Sipuncula, a small phylum of benthic marine worms, is partially based on the number and degree of fusion of the introvert retractor muscles as well as the arrangement of the body wall musculature. The majority of sipunculan species develop indirectly, passing through a lecithotrophic trochophore and a planktotrophic pelagosphaera larval stage. Some species show abbreviated development in which the pelagic phase may be completely skipped. Here we examine myogenesis in four species that represent different developmental modes, using F-actin staining with fluorescent-labeled phalloidin in conjunction with confocal laser scanning microscopy. All examined species have smooth body wall musculature as adults and less than

the full set of four introvert retractor muscles. All go through stages with four introvert retractor muscles that eventually fuse into the reduced number in the adult. The circular and sometimes the longitudinal body wall musculature is split into bands that later fuse to form a smooth sheath. We have also reconstructed the ancestral states of the introvert and body wall musculature using Bayesian statistics. Our reconstructions suggest with high probability that the ancestral sipunculan had four introvert retractor muscles, longitudinal musculature split into bands and a smooth sheath of circular body wall musculature. We conclude that the plesiomorphic condition in sipunculans is four introvert retractor muscles. This condition is retained in the larvae of all sipunculan species examined in this study and by previous authors. We also found that crawling larvae have more strongly developed body wall musculature than swimming larvae which propel themselves by means of their metatrochal cilia.

Hybridization dynamics in the threatened Caribbean coral genus *Acropora*

N.D. Fogarty

Since the 1970's, Caribbean acroporids have decreased by 80-98% prompting their enlistment as threatened under the Endangered Species Act. Caribbean acroporids consist of two species, *A. palmata* and *A. cervicornis*, which form a hybrid (*A. prolifera*). Although hybrids were not given protective status, they may play an important ecological and evolutionary role in the fate of this reef-building genus. At sites throughout the Caribbean where both parent species are present, hybrid abundance forms a continuum from absent or rare to dense stands. It is unclear if this variation in abundance occurs as a result of differences in the strength of reproductive isolating barriers, variation in



Acropora prolifera (far right) is an hybrid formed by *A. palmata* (left) and *A. cervicornis* (middle).

success of asexual fragmentation, or differential fitness (i.e. susceptibility to disease, predation, and bleaching) of hybrids. Carrie Bow Island, Belize, has a population of at least 75 hybrid colonies. Molecular analysis at six loci showed that these colonies are composed of two genets that have fragmented and reattached in this shallow (< 1m) turbulent habitat. *A. palmata* and *A. cervicornis* gametes were collected in situ during a simultaneous spawning event. Fertilization and viability experiments revealed that there is no statistical difference in fertilization success, larval survival, or metamorphosis between conspecific and heterospecific crosses. Gamete comparisons of one hybrid colony with the parent species showed hybrids have larger eggs, less sperm and an intermediate number of eggs per bundle. Preliminary analysis suggests these hybrids host a different zooxanthellae clade that may allow them to persist in this extremely shallow, stressful environment. Despite low genetic diversity among the Carrie Bow hybrids, the population is robust due to the hybrid's ability to persist and reproduce asexually in a stressful habitat. Further, these hybrids are fertile and capable of producing an F2 generation or backcrossing with the parent species, which may lead to genetic introgression between the parent species.

Modular variation and phenotypic plasticity in the gorgonian coral *Pseudopterogorgia bipinnata* along the Western Caribbean

J.A. Sánchez, D.D. & N. Manrique

One of the most intriguing aspects of evolution is whether habitat-induced phenotypic variation can lead to genetically fixed morphotypes. The gorgonian coral *Pseudopterogorgia bipinnata* (Gorgoniidae: Octocorallia) provides a great opportunity to explore phenotypic plasticity and morphological variation in a coral reef organism. We studied the variation of different morphological traits including the different types of microscopic sclerites (0.1-0.2 mm of length variation), polyp aperture and spacing (1-20 mm), and branches and internodes (1-20 mm), which are all repetitive modules throughout the colony. In addition, we studied the genetic variation of the Internal Transcribed Spacer 2 (ITS2, rDNA) using a combined approach with Denaturing Gradi-

ent Gel Electrophoresis (DGGE) and DNA sequencing of the different copies found at each individual colony. We examined colonies from Carrie Bow Cay (Belize), Bocas del Toro (Panama) and Cartagena (Colombia), which included most reef habitats (1-35 m in depth) and several morphotypes. Only one feature did not change at all across habitats despite an order of magnitude difference in other features such as branch length and lesser, but significant, differences in the remaining traits. We distinguished three *P. bipinnata* morphotypes: shallow exposed, shallow to mid-depth (moderately exposed) and low water motion in deep-water (<20 m) morphotypes, which were independent of genetic variation. The three phenotypes did not exhibit clines and were seldom seeing side-to-side sharing the same environment at overlapping zones. Ecotypes can make some traits exhibit phenotypic plasticity but are not so extreme to make these traits fixed in the species genome. In need of further screening, genetic assimilation can be a viable event for octocorals, where habitat seems to be a conditioning factor for niche separations.



Plexaura flexuosa spawning.

Cellular biomarkers as a measure of sub-lethal stress in coral larvae

R. Ritson-Williams & V.J. Paul

As coral reefs across the Caribbean decline in coral cover, coral larval recruitment is a key process that will aid in the recovery of coral reef communities. Elevated temperature is known to inhibit larval coral recruitment, however few experiments have evaluated the mechanisms of temperature stress on coral larvae. Short term exposure of *Porites astreoides* larvae to elevated temperatures (+3.5°C for 4.5 h) induced significantly more reactive oxygen species and upregulated the enzymes catalase and superoxide dismutase compared to 27°C seawater temperatures. In addition the larvae had reduced settlement and increased mortality after the heat treatment. Our results indicate that even short term exposure to thermal stress can induce antioxidant enzyme activity and causes a lethal and sublethal reduction in recruitment of coral larvae.

Coral recruitment in the gardens of good and evil

R. Ritson-Williams, S. Arnold, R.S. Steneck & V.J. Paul

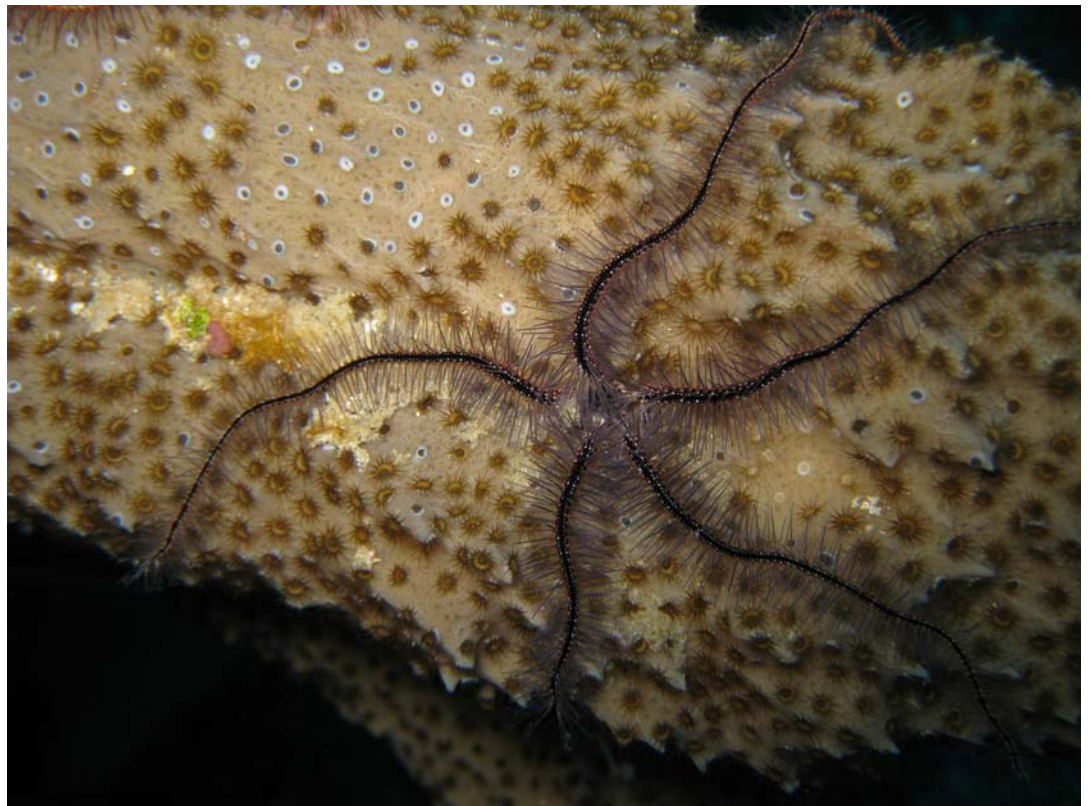
Coral reefs are losing coral cover on a global scale, but improving natural coral recruitment is one management strategy that could reverse this trend. In order to better understand where and why coral larvae settle we tested settlement preferences of corals in the field and in the laboratory. Our field research suggests that settling coral larvae select some biological substrates such as coralline algae, which they settle on and near. Of the three spawning coral species that we tested in the laboratory, each one had different settlement rates in response to different species of coralline algae.

Sponges, invertebrates and macroalgae, which can overgrow coralline algae in the field, are known inhibitors of coral recruitment. Increased herbivory may be a mechanism to shift the benthic community composition away from coral recruitment inhibitors towards facilitators such as coralline algae, thus providing managers a strategy for improving the benthos for higher rates of coral recruitment

Biology of Belizean brittle stars

G. Hendler & D. Pawson

Although ophiuroids are among the most abundant macro-invertebrates in Belizean waters, new species are still being described, and our understanding of even the most common and conspicuous species remains quite superficial. In order to further an ongoing study of their biology, preliminary observations were made on the larval behavior, skeletal regeneration, and parasitism of several ophiuroid species that were collected at Carrie Bow Cay, the Pelican Cays, and nearby mangrove islands. (1) The larva of one species was reared to metamorphosis and filmed, in order to document ontogenetic changes in its swimming behavior. (2) An ablation technique was developed, and applied to the investigation of skeletal regeneration in the post-metamorphic ophiuroids. Subsequently, scan-



ning electron microscopy of the experimental animals has shown that the procedure was successful and that the regeneration of ophiuroid disk ossicles can be monitored. (3) Two different species of enterozoic copepod parasites, one of which is undescribed, were discovered in two species of ophiuroids. Observations were made on the host specificity, distribution, behavior, morphology, and development of both species of copepods, and a manuscript on the results is being prepared.

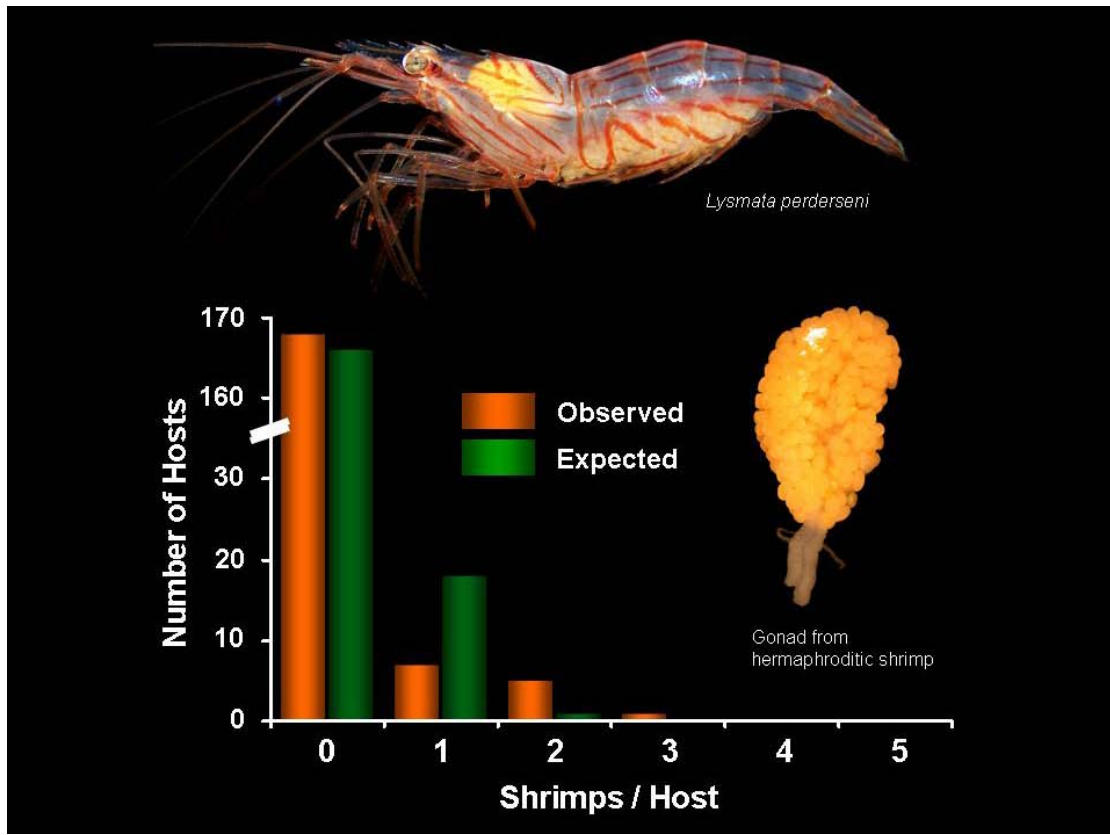
Testing the effect of group size on sex allocation in simultaneous hermaphrodites: *Lysmata* shrimps from the Caribbean as model system

J.A. Baeza

My research at Carrie Bow aims to understand the conditions favoring monogamy and simultaneous hermaphroditism and the partition of resources between sperm and ova in hermaphrodites. Theory predicts the evolution of hermaphroditism and monogamy when the environment limits encounter rates among conspecifics; with low encounter rates, paired hermaphrodites double their mating chances compared to organisms with separate sexes. Also, monogamous hermaphrodites should

produce the smallest amount of sperm necessary only to fertilize their partner's eggs while promiscuous hermaphrodites should produce copious sperm to profit from male mating opportunities. I have been exploring the ideas above with *Lysmata pederseni*, a symbiont of tube sponges found at patch and fringe reefs at Belize. Various *Lysmata* species are reported as protandric-simultaneous hermaphrodites; individuals invariably reproduce solely as males first and later in life as hermaphrodites. However, the sexual system of most of the species is unknown.

My ongoing research at Carrie Bow demonstrates that *Lysmata pederseni* is indeed a protandric simultaneous hermaphrodite. This species should feature social monogamy as mating system because its host sponge is scarce and small enough to support only few individuals, and because predation risk away from hosts is high. In agreement with this prediction, preliminary results suggest that this species inhabits more frequently than expected by chance as pairs in a single host. Mark and recapture experiments further suggest that this pairs might last up to three months. However, other pairs appear to be short term in nature lasting less than two weeks. The socially monogamous pairs might be comprised either by one male and one hermaphrodite or by two hermaphrodites. Whether the sexual phase of the mate affects pair formation and stability remains to be addressed by future studies.



Sex allocation theory predicts that the overall sex allocation of hermaphroditic shrimps (in terms of male versus female gonad mass) should be female biased; it permits hermaphrodites to profit from the female function that provides a greater fitness return than the male function. In agreement with this prediction, preliminary dissections suggest that the amount of resources allocated to the female gonad is much greater than that allocated to the male gonad in the studied species. Thus, so far

these results support sex allocation theory, one of the successful theoretical frameworks in evolutionary ecology.

Shrimps from the genus *Lysmata* feature a wide diversity of lifestyles, mating behaviors, colorations and symbiotic partnerships. Some live in groups, others solitarily, and others are socially monogamous. Some dull species dwell freely among rocks in temperate localities while other colorful species inhabit tropical sponges. Other strikingly brilliant species clean fishes. Because of this diversity, *Lysmata* shrimps represent ideal candidates to explore the role of ecological conditions in explaining evolutionary innovations (monogamy, hermaphroditism). Future studies on the species present at Carrie Bow promise a better understanding of the environmental factors favoring differing mating and sexual systems in marine invertebrates.

From larvae to lineages: Investigations of the shorefish diversity in the tropical Atlantic

C.C. Baldwin, D.G. Smith & L. Weigt

The identities of pelagic larval stages constitute the largest gap in our knowledge of the coral-reef fish fauna of the tropical Atlantic. This fundamental taxonomic information is necessary before larvae can be used in studies of, for example, evolution, fisheries biology, and ecology. Over the past 15 years, we have identified larvae of numerous Belizean fishes by rearing net-collected larvae at the Smithsonian's marine station at Carrie Bow Cay. More recently, we have begun matching larvae to adults using mitochondrial cytochrome oxidase 1 sequences (DNA Barcodes).

In March and June, 2008, as part of our shorefish-diversity study based on DNA Barcoding. We worked at three western tropical Atlantic locations in 2008: Belize, Curacao, and the Bahamas. At each site, we collected adult fishes using a variety of methods, including quinaldine, rotenone (where we had permission to use it), pole spears, hook and line, and by visiting local fish markets. We

took tissue samples for DNA Barcoding analysis from up to five individuals of as many species as possible, with an emphasis on species that we had not sampled before. We photographed fresh color patterns for every specimen from which tissue samples were taken and preserved the vouchers for most specimens (all but the very large, easily identified adults).

We have sampled fishes in Belize for DNA Barcoding for several years. The DNA data reveal genetic diversity of fish species in the area that is not always reflected in the current taxonomic classification. Most of the shorefish species in Belize, however, are broadly distributed geographically, and, in 2007, we conducted field work in the Ft. Pierce, FL, area as a first step in expanding our geographical coverage. This year, in addition to sampling fishes in Belize to continue building our database of DNA data from that locality, we conducted field work in Curacao and the Bahamas. Although Curacao was not part of our original proposal to MSN, discussions about our research with colleagues at the MSN symposium in November, 2007, indicated that it would be an excellent site for providing comparable habitats to those we sample in Belize.

Baldwin, Smith, and Weigt conducted the work in Curacao from March 11 to March 19, working out of the facilities at the Carmabi Research Institute. Curacao proved to be an exceptionally favorable place to work, as coral reefs begin only a short distance offshore, and mangrove habitats are present adjacent to the lab. We collected 456 samples for DNA Barcoding, and after the first pass of this material in the LAB, we have 407 DNA Barcodes verified. The remainder are schedule to be re-sequenced in September 2008.

From May 14 to June 5, we worked at Carrie Bow Cay. The field crew comprised (each working one to two weeks) Baldwin, Smith, Weigt, and volunteers





Zach Foltz and Julie Mounts. To increase our coverage of Belizean fish species, we chartered a boat to take us from Carrie Bow to Glover's Reef one day and part way to Turneffe another day. Despite being plagued with bad weather from a tropical disturbance the last week of our field period (during which time we focused on collecting larval fishes at night from a net suspended from the dock), we obtained tissue samples and vouchers from 478 specimens. After the first pass in the LAB, we have 454 DNA Barcodes confirmed.

In June (6/14 – 6/21), Baldwin had the opportunity to sample fishes in the Berry Islands, Bahamas, under the auspices of the Perry Institute of Marine Science, Lee Stocking Island. Tissue samples (and vouchers for most) were obtained from 265 fishes. After the first pass in the LAB, we have 243 DNA Barcodes verified.

Once neighbor-joining trees from the verified DNA Barcodes have been produced, we will compare data among the three locations as well as data we obtained previously from Florida. Results will dictate priorities in terms of locations for field work in 2009.

Ecology, Population Dynamics, and Ecophysiology

Nitrogen and phosphorus limitation to the growth and clonal reproduction of *Batis maritima*, a dominant understory plant in Florida and Belize mangroves

D.F. Whigham

Batis maritima is a halophyte that is a dominant understory plant in mangroves throughout much of the Caribbean. It occurs as far north as North Carolina on

the Atlantic coast and southern California on the Pacific coast. In many habitats *Batis* forms a continuous canopy cover that has the potential to influence the recruitment and growth of mangrove seedlings. Little is known, however, about this abundant and potentially important species in mangrove ecosystems. The goal of this initial project was to evaluate nitrogen (N) and phosphorus (P) limitation on the growth and asexual reproduction of *Batis* at MSN sites in Belize and Florida where mangrove growth has been shown to be limited by different nutrients. Fertilization studies at both sites demonstrated a strong response to P in Belize and a positive but less pronounced response to N fertilization at the Ft. Pierce site. A greenhouse experiment at SERC in which N, P and N+P were applied to *Batis* seedlings demonstrated the lack of N and P reduces growth compared to the presence of both nutrients. Preliminary interpretations are that sites at Carrie Bow have sufficient nitrogen to support a strong growth response to the addition of phosphorus. In Florida, there is sufficient phosphorus in the substrate to support *Batis* growth as the addition of N resulted in a significant, but relatively small growth response. The relatively small response to N addition in Florida may be due to salt stress as salinities at the sites where the study was conducted are annually 80-100 ppt, a level that requires halophytes to use much of the available N to maintain sufficient osmotic values to continue to be able to maintain an adequate water balance.

Aplysina red band syndrome at Carrie Bow Cay, Belize

K. Ruetzler, M.C. Diaz, D.G. Gochfeld, J.B. Olson, R.W. Thacker, & E. Villamizar

The main objective of our study is identifying the causative agent and pathology of *Aplysina* Red Band Syndrome (ARBS), an emerging infectious disease of Caribbean marine sponges first discovered in species of the genus *Aplysina* (Aplysinidae, Verongida) (Figure 1). Originally observed in the Bahamas in 2004 (Olsen et al., 2006), this disease is now believed to be widespread throughout the Caribbean. Our project enabled us to survey several reefs near Carrie Bow Cay, Belize, where we determined its presence and abundance and compared it with the occurrence of other stress or disease conditions affecting sponges, including space competition and predation, and the diversity and composition



of the reef communities. Our surveys included lagoon patch reefs and the shallow fore-reef, to 25 m depth, where we examined specimens of several sponge species seen with disease-like conditions in previous years, such as *Xestospongia muta*, *Geodia neptuni*, *Callyspongia plicifera*, *Aplysina archeri*, *A. cauliformis*, *A. fistularis*, *A. fulva*, *Ircinia campana*, *I. felix*, and *I. strobilina*. In July 2008, only the *Aplysina* species and *C. plicifera* showed distinctive disease conditions and samples were preserved for histological examination and microbiological, molecular, and chemical analyses. We established and photographed 40 quadrats of 1m² to monitor size, density, growth (of new recruits, in particular), and health of sponges over time. We also performed controlled nutrient enrichment experiments to assess whether the condition of diseased individu-

als may be compromised by anthropogenic nutrient input (pollution). Comparative studies of ARBS-affected sponges are underway with material collected in the Bahamas.

To monitor ARBS specifically, six 10 x 2 m transects were established (in addition to the afore-mentioned photo quadrats) on each of three patch reefs near Carrie Bow Cay. A combination of line intercept and band transects was used to quantify cover of all substrata, and abundance and diversity of sponges, corals and gorgonians (Figure 2).

Because filamentous cyanobacteria seem to be at least part of the disease process, the abundance of cyanobacterial mats and the substrata on which they were found were recorded, along with the condition of each *Aplysina* sponge encountered. On the survey reefs, ARBS was found to affect both *A. cauliformis* and *A. fulva*, but to a much lesser degree than at our study sites in the Bahamas. For example, we found ARBS on 1.9% of *A. cauliformis* in Belize compared to 5.4% in the Bahamas. Preliminary analyses suggest that ARBS abundance is not correlated with sponge, coral, or gorgonian diversity. Mats of red filamentous cyanobacteria were observed on corals, gorgonians, sponges (including *Aplysina* spp.), and abiotic substrata, but their abundance was not correlated with that of ARBS. Marked transects will be re-surveyed in 2009 to examine change in ARBS



prevalence over time.

We labeled and measured each ARBS-affected *Aplysina cauliformis* along our transects, as well as its nearest healthy neighbor (Figure 3). In total, we marked 18 pairs of the branch-like sponges by attaching numbered plastic tags with cable ties at their bases. For diseased sponges, we measured the length of healthy tissue from the cable tie to the lower margin of the band, the widths of the lower band, lesion, and upper band, and the distance from the upper margin to the tip of the branch. For healthy sponges we measured total length of branches from the cable tie. We also measured the distances to the nearest healthy and diseased neighbors and recorded the number of sponges within a 1 m radius that were healthy, or had ARBS or other lesions. These specimens will be relocated in 2009 and remeasured to monitor individual responses to ARBS over time.

A nutrient enrichment experiment was performed in the wet lab on Carrie Bow. Healthy sponges were placed in individual chambers with flow-through seawater and ambient light levels. Initial measurements of photosynthesis and respiration were made using light-dark bottles. Packets of Osmocote slow-release fertilizer were placed into half of the chambers and sponges left under conditions of low water flow. After 7 days, oxygen production and consumption was measured again and the sponges were measured and weighed. Samples were preserved for chemical and molecular analysis, and fixed for histological examination to assess changes in the sponge tissue and cyanobacterial symbionts. These analyses are currently underway in our laboratories and will be used to design new experiment to be conducted in 2009.

Nutrient over-enrichment differentially affects growth and herbivory in mangrove forests along latitudinal and tidal gradients

I.C. Feller & C.E. Lovelock

Mangroves form complex marine ecosystems with spatial differences in structural complexity, biodiversity, biogeochemistry, and hydrology that vary at local and regional scales. Although mangroves provide critical ecosystem goods and services, they are threatened globally by human activities including nutrient over-enrichment. Our goal was to determine if enrichment with nitrogen (N) or phosphorus (P) interacts with forest structure and latitude to alter growth, nutrient dynamics, and patterns of herbivory. We established a

series fertilization experiments across more than 2000 km and 18° of latitude from the Indian River Lagoon (IRL), Florida to Twin Cays, Belize to Bocas del Toro, Panama. At each site, we fertilized individual trees with one of three treatment levels (control, +N, +P) in two zones (fringe, dwarf) along transects perpendicular to shorelines and measured their responses at 6-mo intervals for 4 yr. Growth was measured as shoot elongation, and herbivory was measured as a function of folivory, loss of yield, and tissue mining. Results showed that all sites were nutrient limited, but patterns of nutrient limitation varied by zone and latitude. At IRL, growth was N-limited from the fringe to the dwarf forest; at Twin Cays, the fringe was N-limited, but the dwarf forest was P-limited; at Bocas del Toro, the fringe was N-limited, but the dwarf forest was both N- and P-limited. Nutrient enrichment had dramatic effects on herbivory that varied by treatment, zone, latitude, and species. Our studies show that responses to eutrophication of mangrove ecosystems will depend on site characteristics, the species considered, and the nature of nutrient limitation. Predicting how herbivores respond to nutrient over-enrichment requires an assessment of spatial



heterogeneity coupled with feeding strategies and species-specific behavior measured on multiple scales of response.

Assessment of coral reefs using herbivory/nutrient assays and indicator groups of benthic primary producers. The ecology of *Batis maritima* in mangrove

M.M. Littler & D.S. Littler

Rapid assessment protocols for determining and monitoring the status of any given coral reef are provided and include measuring: (a) standing stocks of functional indicator groups, (b) herbivore populations, (c) water-column nutrient levels, (d) tissue C:N:P ratios, (e) algal physiological-response assays, and (f) herbivory assays. These measurements can reveal quantitative tipping-point levels beyond which resilience to undesirable phase shifts begins to become critically reduced. Universal tipping-point approximations are reviewed for inorganic nutrients, and posited for the first time for herbivory. The relative roles of top-down and bottom-up controls in determining benthic community structure and the health of coral reefs are especially important management concerns. This paper specifically addresses the top-down effects of herbivory and bottom-up effects of nutrient enrichment on critical indicator groups; i.e. reef-building corals, crustose coralline algae, dense turf algae, and frondose macroalgae. A predominance of (a) massive corals and calcareous coralline algae relative to frondose macroalgae and algal turfs indicates a healthy spatially heterogeneous condition reflecting low nutrients and high herbivory. An abundance of (b) frondose macroalgae illustrates the least desirable condition of elevated nutrient levels and reduced herbivory, possibly reflecting pollution in concert with destructive herbivore fishing practices. High coverage of (c) coralline algae suggests healthy high herbivory levels, but problems with elevated nutrients that are inhibitory to some corals. Domination by (d) dense turf algae indicates desirably low nutrient levels, but an inadequate herbivory component. The experimental results demonstrate flaws in some of the previously published manipulative methods and provide insights for the improvement of in-situ nutrient studies on coral reefs. The fast growth and turnover rates of fleshy algae compared to other reef organisms highlight their value as early-warning indicators of reef degradation.

Recruitment, growth, and ecological relationships of mangrove root encrusting bryozoans in the Pelican Cays, Belize

J.E. Winston

Artificial substrata (plastic pipes) were used to initiate fresh mangrove stilt roots and study settlement of bryozoans. All pipes planted the year before were successfully retrieved and returned to the lab at CBC where the plastic “bark” was removed from each pipe and flattened so that it could be examined under the stereo microscope and its fauna and flora counted and categorized. The rectangular plastic sheets were dried retained. Voucher material was detached and preserved wet or dry.

The results of settlement experiments were similar in some ways to panels submerged on the reef, but surprising in other respects.

Similarities

Strong historical aspect. Recruitment on each pipe was different from others even at the same site.



Those pipes placed near potential parent colonies had most bryozoan settlement. Those pipes that were in the brightest settings had fewest (or no) bryozoans and most algal settlement.

Many of initial foulers were from the same groups that dominate early development of cryptic reef communities: crustose and filamentous algae, serpulids and spirorbids.

Differences

Bryozoans could grow to mature and reproductive size in 5 months or less. The greatest growth was achieved (as expected from other settlement studies) by *Schizoporella pungens*. The largest colony (3rd from left in photo) was 6.5 cm in width and 17 cm in length with hundreds of ovicells containing embryos.

Two cryptic species, *Stylopoma auranticum* and *Parellisina latirostris*, also reached reproductive size within 5 months. The largest single *Stylopoma* colony was 6.5 cm by 11 cm in size and contained at least 200 ovicells. It takes cryptic *Stylopoma* colonies more than 3 years to reach this size. On one of the Cat Cay pipes (see Cat Cay photo) 3 colonies had joined to produce a large (6.5 X 21 cm) compound colony, the lack of competition at the boundaries between them indicating that the initial recruits may have been half-sibs from a nearby parent colony.

How can such a rapid growth rate be achieved? A much higher food supply (phytoplankton) is the most

likely explanation, although further work in collaboration with a phytoplankton person would be needed to prove this. It is certainly not because of a lack of predation. The same kinds of organisms that damage or destroy bryozoan colonies through grazing or single zooid predation were present among the mangrove roots including, chitons, limpets, and micro-gastropods. There were almost completely skeletal colonies on the pipes, and marks of grazing. In studying large colonies from Cat Cay we found a new predator: a polyclad flatworm that was so cryptically colored to match its *Stylopoma* host that its presence could only be detected by a faintly blurry look to the colony surface, or the movement of the flatworm to a new spot on the colony.

Some of the round to oval newly skeletal surfaces the flatworm's feeding activities bared contained an array of tiny bubble-like eggs. These and the flatworms were given to Jörg Ott for further study.

Predation by fish is also possible, but it will take further study to determine how much there might be. For such work it would necessary to find a way to stay in the Pelican Cays or nearer to them than the CBC lab.

There is also an indication that bryozoan recruitment might be more seasonal than previously suspected. This was shown by the artificial roots and also by collecting submerged plastic debris and wood. In September *Hippopodina feegeensis* was recruiting actively. *Stylopoma* and *Schizoporella* were the most abundant recruits during the period of the settlement experiment (and larvae of colonies of those species continued to settle while the parent colonies were held in tanks in the wet lab).

During the second visit more *Lobophora* was collected and measurements made of the soft-bodied ctenostomes were made from living colonies. All *Lobophora*-dwelling bryozoan species have been now photographed through microscope or SEM. One paper from this project is in press, and the second is now being prepared.



Monitoring taphonomic-process initiative, and maintain bottom-up vs. top-down experimental plots; examining the status of the spur-and groove system after twenty five years; and work on the revision of Caribbean reef plants.

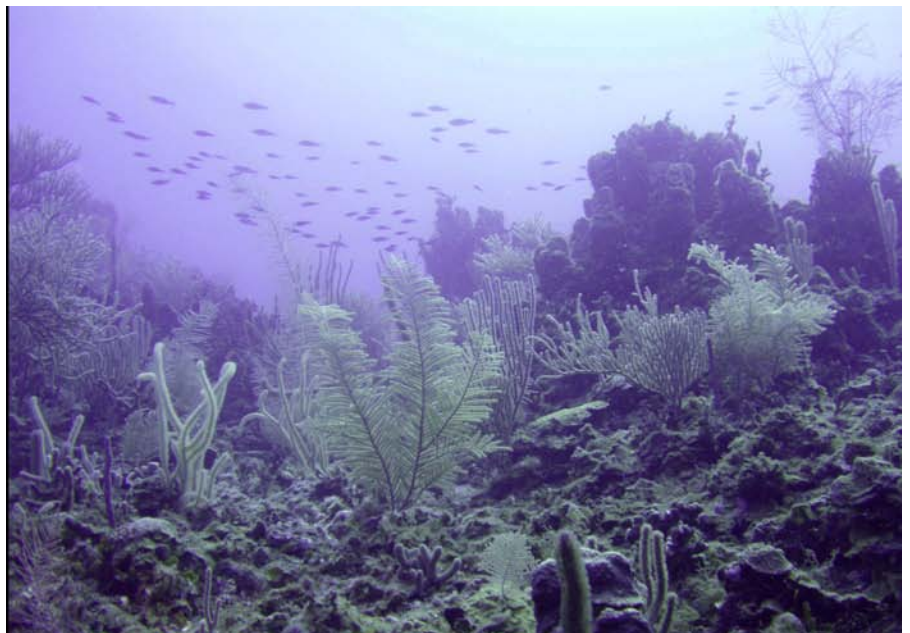
M.M. Littler, D.S. Littler, & B.L. Brooks

Eighteen experimental replicates (sections of massive *Porolithon* and *Porites*) were scanned and evaluated from 30 sets on the reef crest, 16 on the back-reef site and 16 on 15-m deep pinnacle site. This experiment is designed to address the “paradox of the coralline algae” which stems from the fact that although coralline algae are nearly always abundant (often dominant) in terms of cover on coral-reef systems worldwide, they only show up abundantly within fossil coral-reef deposits in relatively few isolated formations. We hypothesize that this disappearance during fossilization is due to differential taphonomic processes. Many rock-boring and limestone altering creatures abound on coral reefs. However, one of the most important groups of limestone altering organisms is the clionid sponges. Even though the calcite deposited by coralline algae is much denser/harder than the aragonite form of calcium carbonate produced by reef building corals, the former is precipitated within and between cellulose cell walls, which, even after death, may provide an energy source for the boring clionid sponges. Our preliminary results indicate that, although slow, this is indeed the case, potentially explaining the enigmatic/mysterious disap-

pearance or alteration of massive/extensive deposits of coralline algal in the fossil record.

Nutrient experiments – Due to the growing problems associated with eutrophication and overfishing throughout tropical and subtropical shorelines, the ecological responses of coral reefs and macroalgae to nutrient enrichment and release from predation have been repeatedly cited as priority areas in need of further research. The conflicting reports in a recent issue of *Science* (Jackson et al. 2001) and another study (Miller et al., 1999) refuting the nutrient component of the RDM, but using tree stakes as fertilizer (containing up to 6% of the algacide chlorine), have only served to “muddy the water”. The CCRE research we published recently (Littler et al. 2006b) reveals the flawed experimental evidence purported to refute the RDM. By avoiding the use of inappropriate sources of enrichment documented above (Littler et al. 2006a), we have completed ecologically sound experiments that provide new insights into the complex nutrient status of Caribbean coral reefs. These results (Littler and Littler 2006a) have already been used in what we consider to be a seminal paper (Littler and Littler 2006) to clarify our understanding of the ecology and sustainable management of coral-reef ecosystems worldwide.

This previous CCRE research using non-toxic, slow-release, Osmocote fertilizer to experimentally assess the role of nutrients on coral-reef communities paved the way for the critical experiments that were established and monitored during March-April 2004-2006 (Littler et al. 2006a). The team monitored and re-assessed the 30 previous pulsed Osmocote-treated and the thirty mini-reef replicates (15 experimental and



15 controls) set up in 2004 on the CBC back reef flat northeast of the laboratory in an area of documented moderate herbivory. All of these were re-photographed on two sides (Sept 2008) to provide 59 quantitative samples for photogrammetric analysis in the laboratory. The findings show a marked, but highly-complex, effect of nutrients in that increases in bottom-up nutrient controls and their interactions stimulate harmful fleshy algal blooms (that can alter the abundance patterns among functional groups, even under intense herbivory); conversely, elevated nutrients inhibit the growth of ecologically beneficial reef-building corals. The results demonstrate even further complexity in that nutrients also act directly as either limiting factors (e.g., physiological stresses) or as stimulatory mechanisms (e.g., growth enhancing factors), as well as functioning indirectly by influencing competitive outcomes. Herbivory directly reduces fleshy-algal biomass, which indirectly (via competitive release) favors the expansion of grazer-resistant reef-building corals and coralline algae. Because of the sensitive nature of direct/indirect and stimulating/limiting interacting factors, coral reefs are particularly vulnerable to anthropogenic reversal effects that decrease top-down controls and, concomitantly, increase bottom-up controls, dramatically altering ecosystem resiliencies.

During 2006, 5 experimental sets of 4 diffusers were moved and securely fastened to the zone of highest herbivory seaward of the reef crest. Changes in cover of each algal-group/coral interaction, as well as recruitment, were determined by photographic sampling of each of the four sides and top of the diffuser arrays, projecting the images in the laboratory and scoring changes in the percent cover of predominant taxa (Littler and Littler 1985). Another group of 4 sets that had been placed in the seagrass community just north of Carrie Bow Cay in a low herbivory area were likewise evaluated with photographic sampling for algal/coral interactions. As mentioned, the early results are rewarding and show an extremely complex set of competitive interactions that are specific to the individual species among functional groups and the presence of either SRP or DIN. One particularly exciting finding is that SRP alone appears to be responsible for blooms of certain harmful bluegreen algae (Cyanobacteria). We will continue to monitor critical successional/competitive interactions among these most important of reef-builders.

Collections and Field Guides – Our group collected over 197 numbered specimens in 2008 to aug-

ment the National Herbarium algal collections from various and diverse sites around CBC, Curlew, Twin Cays and South Reef. Over 834 in situ underwater images of the collected specimens were recorded and labeled in support a revised edition of “Caribbean Reef Plants”.

Molecular work – One hundred and sixty molecular samples were quick dried in silica gel and frozen for future DNA experimentation (all of these samples backed-up with matching morphological collections).

Comparison of settlement and recruitment among spawning and brooding corals in the Caribbean

V. Paul, R. Ritson-Williams, N. Fogarty, S. Arnold, & R. Steneck

Our objective was to better understand the settlement preferences of larvae from different species of Caribbean corals. During two trips to Belize (April and August, 2008) we collected larvae from six species of corals, including; *Acropora palmata*, *A. cervicornis*, *Agaricia tenuifolia*, *Favia fragum*, *Montastraea faveolata*, and *Porites astreoides*. For each species of coral larvae we found various degrees of settlement specificity in response to different crustose coralline algae species. Both *Hydrolithon boergesenii* and *Titanoderma prototypum* were preferred settlement substrate over more common CCA species for the coral species that we tested regardless of reproductive mode (spawning vs. brooding). The research this past year has been very successful, and we will be writing at least one paper in the coming year on the results of this work.

Investigating the role of nitric oxide in regulating bioluminescence in marine brittlestars

S. Lewis, R. Rotjan, G. Hendler, & T. Michel

Ophiopsila riisei, a common brittlestar on shallow Caribbean reefs, is capable of generating precisely-timed bioluminescent flashes. When disturbed, these brittlestars produce sequential bioluminescent flashes in photocytes along the arm spine neural ganglia, and this response has been shown to deter nocturnally active crab predators. We used several pharmacological approaches to investigate the possible role of nitric

oxide (NO) in regulating bioluminescent flash production by *Ophiopsila riisei*. We found no evidence that the NO-donor sodium nitroprusside (SNP) triggered bioluminescence in whole animals or in isolated arms. Similarly, the NO inhibitor nitro-arginine did not block bioluminescence triggered by mechanical stimulation. Thus, it seems unlikely that NO is involved in mediating bioluminescence in this marine brittlestar.

Bioenergetics of the tropical swarm-forming copepod *Dioithona oculata* with special consideration of the energy expenditure during feeding

A. Bochdansky

Animals tune their feeding rates in response to availability of food resources. This results in different types and shapes of their functional feeding response. A popular hypothesis to explain the shape of functional response is that the animal optimizes energy expenditure during feeding and foraging against energy gain, a notion that requires feeding to be sufficiently costly to impact its energy budget. Studies employing fluid dynamic models, however, suggested that energy expenditure during feeding activity represents only a negligible portion of the overall budget. Direct empirical tests of whether or not energy expenditure constitutes a significant portion of the budget have been elusive, mainly because zooplankton usually live at low densities in their environment and do not behave normally, or even die, when kept at the high densities required for metabolic measurements. The cyclopoid copepod *Dioithona oculata* is an exception. It naturally occurs in extremely dense

swarms between the prop roots of the red mangrove and therefore behaves normally when undergoing metabolic tests in experimental systems. In addition, this copepod is a strong swimmer, being able to maintain position in the chambers and thereby avoiding the outflow screens even at high flow rates. It was therefore possible -- for the first time -- to measure feeding and oxygen consumption simultaneously in a zooplankton species. During three weeks, 10-14 hour time series incubations were performed daily using freshly collected copepods from Twin Cays. Naturally occurring phytoplankton from the collection site was used as a food source and contrasted against filtered seawater. Food and oxygen levels were monitored by means of fluorometry and polarographic oxygen sensors. Clear differences in the oxygen consumption rates between feeding and non-feeding copepods were observed in all trials contradicting the current fluid dynamics models. The exact magnitude of the energy expended during feeding will



be calculated using data from elemental analysis (C, N, and P) of both food and copepods. These samples are currently being processed. The results of this study will be used to parameterize a general bioenergetic model representing the trade-offs between energy expenditure and energy gain during food acquisition.

The mangrove forest fish communities of the Belize cays: Phase III: Cryptic species and description of red mangrove fringe “hot-spots” of fish abundance

D.S. Taylor, C.C. McIvor, W.P. Davis, & E.A. Reyier

Although our original proposal called for continuation of fish collections using ichthyocides, we were unable to get permission from Belize Fisheries for this work. As a result, our ‘fall-back’ project was description of ‘hot-spots’, that is, those zones of the shoreline fringing red mangroves where fish tend to concentrate. Our goal was to physically characterize these zones utilizing a number of metrics as well as count density and diversity of fish species. We completed this fourth survey (March 12-March 26, 2008) of the Belize Cayes mangroves, with a focus on Twin Cayes and Tobacco Range. We also collected 134 samples for continuation of the stable isotope study (with other collaborators). Processing these remaining isotope samples should allow completion of this ongoing project. Finally, we cooperated with Conservation International by sponsoring three students from the University of Belize Resource Management program for three days of field training in our fish collection/census techniques.

Shifts in predation pressure with latitude

A. Freestone

While latitudinal gradients in diversity are one of the clearest patterns in ecology, with diversity peaking in the tropics, we know surprisingly little about the ecological and evolutionary processes that underlie this pattern. Many hypotheses have been proposed, and one in particular, the ‘biotic interactions



hypothesis’ is receiving increasing attention. This hypothesis predicts that higher speciation rates occur at lower latitudes due to stronger and more specialized species interactions. For example, predation pressure is assumed to be a primary factor in shaping tropical communities, but the community consequences of this assumption, specifically consequences for species diversity, have never been tested in a comprehensive, latitudinal scale experiment. Under the advisement of Richard Osman (SERC), Gregory Ruiz (SERC) and Mark Torchin (STRI), I am empirically testing the hypothesis that community structure at low latitudes will be driven by stronger and more variable (i.e. specialized) biotic pressures, specifically predation.

First to understand temporal variability of predation pressure in the tropics, I ran two predation experiments on sessile marine invertebrate communities at Carrie Bow and Twin Cays. I allowed naturally recruiting epifaunal communities to develop on settlement panels deployed with and without predator exclusions in seagrass beds. Using this experimental design, a short-term three-week predation experiment was initiated in early spring 2008 to examine effects of predation on recruitment and early stages of survival (Figure 1). While no effect occurred at Twin Cays, strong patterns were evident at Carrie Bow where panels with the predator exclusions were more diverse. I next ran a similar experiment for three months (April – July, 2008) and found striking negative effects of predation on community diversity at both Twin Cays and Carrie Bow (Figure 2). These results demonstrate that the effects of predators on community diversity of sessile marine invertebrates are strong and spatially variable. Effects were strong and consistent in seagrass beds near a reef system at both recruitment and early community development timescales. In seagrass adjacent to mangroves, these effects are slightly weaker at the recruitment stage,



but have an increasingly strong effect during later community development.

Next, to understand spatial variability of predation pressure



along a latitudinal gradient, I redeployed a three-month caging experiment at Carrie Bow and Twin Cays in July 2008 to run simultaneously with parallel experiments in Connecticut, Florida (SMSFP), Belize (Carrie Bow), and Panama (STRI). The experiment in Belize will be sampled in October and data can then be compared with results from the previous experiments in Belize to understand temporal variability in interaction strength as well as with the data from the other regions to understand spatial variability along the latitudinal gradient. This study will fill a critical gap in our understanding of how basic ecological processes shift across latitude, shaping diversity patterns.

The nutritional content of bottlenose dolphin prey from the shallow waters of Belize

K.L. West, O.T. Oftedal & C.W. Potter

Bottlenose dolphins (*Tursiops truncatus*) are commonly sighted in the shallow waters of Turneffe Atoll, Belize. This represents one of the few tropical marine environments where photographic identification and behavioral studies have been conducted for *T. truncatus*. The foraging behavior of dolphins in this area and fish species prevalence has also been previously described. Our study involved in the collection of 337 potential prey items near Turneffe Atoll for comparison in nutritional content between seasons and years. The prey obtained during 3 collection trips represented 5 orders, 19 families, 22 genera and 35 different species from Turneffe Atoll. A maximum of 6 replicates of each

prey species collected during a single sampling trip to Belize (totaling 260 samples) were analyzed for proximate composition, including a determination of dry matter, crude protein, fat, and caloric, calcium and phosphorus content. Comparisons indicated significant differences between seasons for the Blue-striped Grunt, White Grunt, Lane Snapper and the Schoolmaster Snapper. In the case of the Blue-striped Grunt, ash was greater during the winter. Dry matter was significantly greater in the winter for the Schoolmaster Snapper. Both protein and ash were greater in the winter in the White Grunt. All proximate components were significantly different among seasons when considering the Lane Snapper where dry matter, protein and ash were greater

in the winter while fat and caloric energy were found to be greater in the summer. In addition to determining the nutritional content of potential prey in Belize waters, biopsy darts of tissue were obtained from 15 dolphins during a field sampling trip in the summer of 2007. Dolphin biopsy samples and potential prey items will be analyzed for fatty acid composition and stable isotope signatures to provide insight into the diet composition of bottlenose dolphins in Belize.

Impacts of mangrove clear cutting and dredge filling at the Pelican Cays and Twin Cays ranges, Belize

K.L. McKee

Background: Clear-cutting of mangrove cays has greatly accelerated in Belize and other locations in recent years for development of tourist resorts, fishing camps, and “improved land” for resale. In addition to removal of mangroves, bottom sediments are dredged from adjacent seagrass beds and reef flats and pumped onto cleared mangrove areas to raise elevations sufficiently to support beach vegetation and buildings. Beginning in 1992, several sites at Twin Cays have been cleared and filled with dredge material. Most recently (2007-2008), several cays in the Pelican Cays range have been similarly disturbed. Even if such areas are ultimately abandoned and allowed to recover, recolonization by mangroves may be extremely slow, if it occurs at all. In the interim, the loss of mangroves may



in the region.

Findings: Disturbed mangrove areas on Twin Cays and Pelican Cays varied from 1.0 to 6.2 ha in size, accounting for 7 to 29 % of the island area (Fig. 1). Removal of mangroves and especially dredge filling significantly altered soil characteristics and decreased shear strength and aggregate stability of soil surfaces. The surface soil of reference forests was peat composed of a matrix of live and dead mangrove roots, filamentous algal mats, and trapped organic matter that retained its structural integrity even when disturbed

negatively impact island stability as well as the biota dependent on mangrove habitat.

by sampling. Bulk density was low (0.17 g cm⁻³) and water (68%) and organic (60%) contents were high.

Objectives:

The specific objectives of this study were to assess changes in soil stability and erosion potential on mangrove islands subjected to clear-cutting and dredge-filling activities. Surveys were conducted across disturbed and adjacent undisturbed, reference sites on each cay. The work focused on the designated marine preserves of Twin Cays and the Pelican Cays ranges, which have been highlighted as critical habitat for marine biodiversity



Despite its organic nature, the reference soil (peat) had high shear strength (overall mean: 0.084 kg cm²), which did not vary spatially. Core samples retained their shape and showed little or no slaking upon immersion in water and little loss of material upon repeated agitation. In contrast, the surface soil in disturbed areas was composed of inorganic, carbonate particles derived primarily from calcareous algae (*Halimeda* spp.), coral fragments, and shells (particle-size distribution analysis showed that >90% of the mass was sand or larger particles). This material had a high bulk density (0.72 g cm⁻³), low water content (30%) and low organic content (9%). Soil shear strength (0.044 kg cm²) and aggregate stability in disturbed areas was lower overall compared to reference areas and varied spatially. Many cores were friable and readily disintegrated when disturbed mechanically.

At a number of locations, artificial barriers had been installed along the shoreline where mangroves had been removed in an attempt to retard erosion (Fig. 2). Qualitative observations of shoreline retreat at such sites indicated that such structures were less effective at erosion control than were natural mangrove stands. Deep cores collected at both island ranges beneath disturbed and reference sites also revealed underlying deposits of peat (1.5 to 10.8 m thick), attesting to the role of mangroves in vertical building of these islands and their capacity to withstand sea-level rise.

Conclusions: Clear-cutting of mangroves and dredge-filling activities dramatically increased the potential for soil erosion at Twin Cays and Pelican Cays ranges. Removal of the “foundation species”, *Rhizophora mangle* (red mangrove) eliminated the primary peat-forming mechanism as well as a natural barrier against shoreline erosion. Although dredge filling temporarily raised elevations, shoreline erosion and the inexorable subsidence of deeper peat and sea-level rise will ultimately submerge such areas. Degradation of key biophysical components and critical habitat will eventually impact ecotourism activities dependent on a healthy, natural environment. Our work highlights the unsustainable nature of these activities and the consequences for stability of islands in this unique ecosystem.

Species Interaction and Behavior

The life cycle, phylogeography, and comparative mitochondrial genomics of Placozoans from Twin Cays

A.Y. Signorovitch

Placozoans are microscopic marine invertebrates that are distributed along tropic and subtropic latitudes. They possess only four somatic cell types, a dorsal-ventral polarity, no definite shape, and are the simplest known free-living animals. Here I describe findings of three separate studies that utilized placozoans collected from the mangrove island of Twin Cays, Belize. In the first study, the margins of Twin Cays were surveyed for placozoans during the summers of 2003 and 2004. Sampled isolates were haplotyped at the mitochondrial 16S rDNA and mapped to their collection sites along the island’s margins to form the basis of the first high-resolution phylogeographic study of placozoans. Twin Cays was found to be home to an unprecedented diversity of placozoans, including sympatric highly diverged species. The second study aimed at detecting molecular signatures of sexual reproduction through a molecular population genetics approach. Although never observed, it is now known that placozoans do indeed reproduce sexually as demonstrated by patterns of allele sharing between individual placozoans. Lastly, a select group of highly divergent Twin Cays placozoans was used in a comparative study of whole mitochondrial genomes (mtDNA). While the majority of animal mtDNAs are ca. 15-20 kb, all placozoans so far examined possess genomes well above this range, from 32-43 kb. Based on these data and other complete mtDNAs, phylogenetic analyses of the Lower Metazoa revealed that all members of the Phylum Placozoa belong to the earliest diverging animal group.

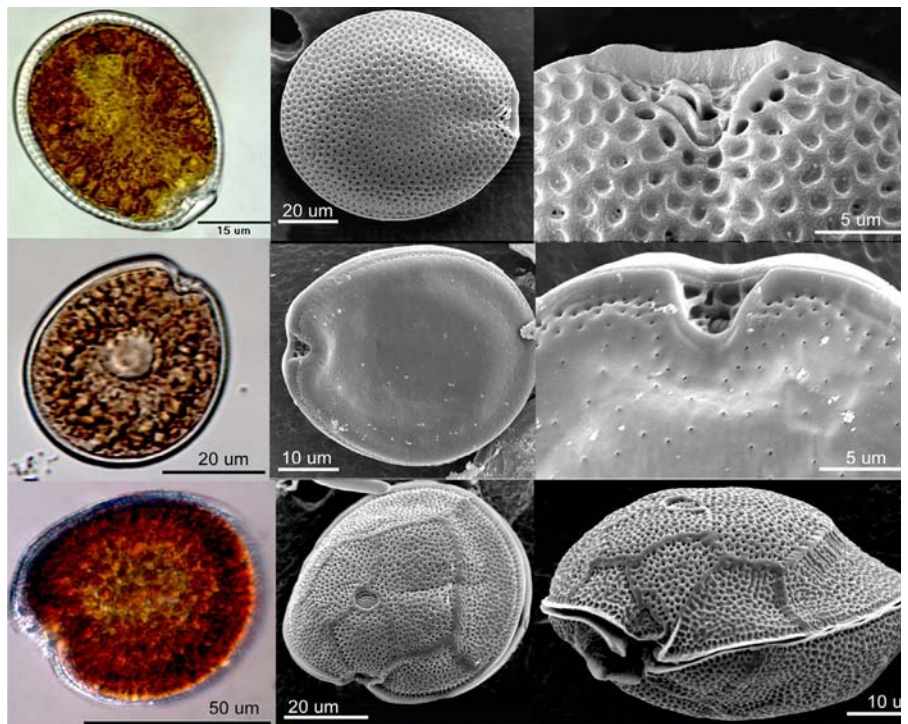
An overview of symbiont-bleaching in epiphytic foraminiferans

S.L. Richardson

Sorites dominicensis is a disk-shaped foraminiferan that lives attached to phytal substrates in tropical to subtropical, shallow-water habitats. This species harbors dinoflagellate endosymbionts (*Symbiodinium*

sp.) that are closely related to the zooxanthellae in corals and other cnidarians. Symbiont bleaching has been observed in field populations of *S. dominicensis* collected from turtle grass (*Thalassia testudinum*) meadows in the Florida Keys, the Indian River Lagoon, Florida, and Belize. The degree of bleaching for each specimen is assessed using a relative scale: healthy, pale, mottled, or white. Healthy individuals possess a distinct yellowish-brown coloration to their cytoplasm. Pale individuals possess a light, yellowish coloration to their cytoplasm. In both healthy and pale individuals, cytoplasmic coloration is evenly distributed throughout the foraminiferal test. Mottled specimens possess large patches of white cytoplasm interspersed with regions of healthy cytoplasm. Individuals are recorded as being totally bleached if they possess completely white tests. Field studies involve surveying several hundred individuals (~500) from each population, or collecting site, and recording the incidence of bleaching. Bleaching rates vary from 0.3-19%, with the lowest rates of bleaching to date observed in populations living in the tannin-stained waters of Boston Bay, within the mangrove islands of Twin Cays in Belize (7/07). The highest rates of bleaching to date were observed in the same population, a few days later after a period of intense rainfall. High rates of bleaching (18%) were observed in July 2005, in populations living on the reef flat off Carrie Bow Cay, a site that experiences subaerial exposure during extreme low spring tides, and water temperatures as high as 40 °C. Other environmental factors that appear to trigger bleaching in foraminiferans include: increased irradiance, exposure to UV and blue wavelengths of light, freshwater influx, and periodic disturbance by hurricanes.

CFP through food web concentration of toxins found in contaminated reef fish. CFP is the most commonly reported marine toxin related illness and can be fatal. CFP occurs circumtropically and it is endemic in the Caribbean region. Annually 3% of the population of the US Virgin Islands and 4.4% of the households surveyed in St. Thomas are affected while 7% of the residents of Puerto Rico have experienced at least one episode of CFP in their lifetime. While CFP is a threat to public health throughout the Caribbean, it is generally managed by local, traditional knowledge of the native fishers of the seasonality of occurrence and locations of local reefs known to be ciguateric. Also, the custom, some times formalized and sometimes not, of avoiding large, top predators like barracuda factors into the management of CFP. With increasing ocean temperatures the validity of local and traditional knowledge may fail to provide adequate guidance and increase the potential for CFP. A goal of our research is to develop quick, reliable, economical, and quantitative method of detecting



Ciguatera fish poisoning in the Caribbean

P. Tester, M. Faust & W. Litaker

Tropical dinoflagellates of the genus *Gambierdiscus* are known to produce toxins that cause ciguatera fish poisoning (CFP). Humans are susceptible to



Dinoflagellates, genus *Gambierdiscus*, causing ciguatera.

Gambierdiscus, in natural assemblages. Comparative distribution and abundance data from the Caribbean Coral Reef Ecosystems field station at Carrie Bow Cay, Belize and a CFP “hot spot” in the eastern Caribbean will be presented.

Interplay between dinoflagellate toxins, membrane sterol composition and parasitism by *Amoebophrya*

X. Bai & D.W. Coats

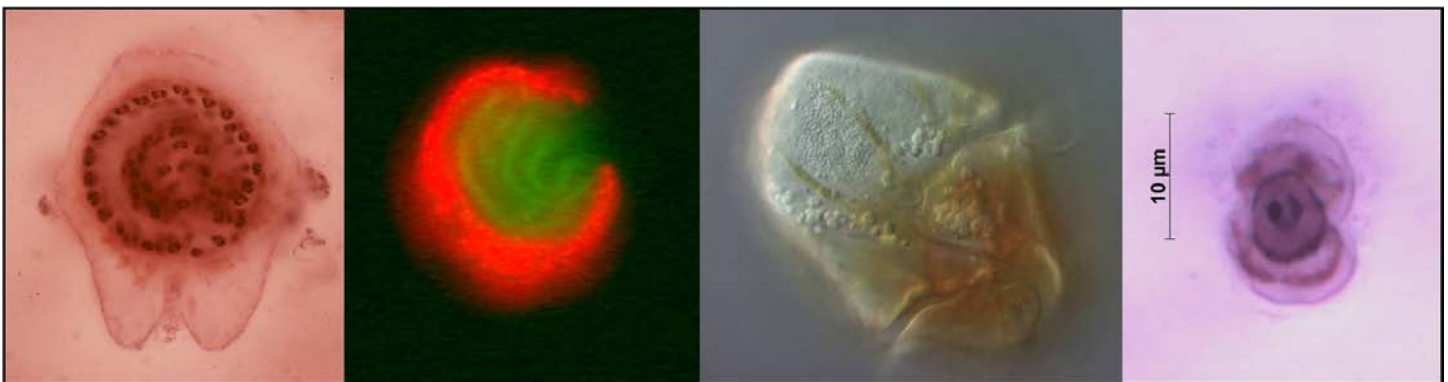
Parasitic dinoflagellates of the genus *Amoebophrya* infect many bloom-forming dinoflagellates, including several toxic species. These parasites can spread rapidly through host populations and have been linked to the decline of red tides. The fate of host toxins during bloom decline caused by parasitism is unknown. Equally unresolved is the performance of parasites in host strains that differ in toxin content. The ichthyotoxic dinoflagellate *Karlodinium veneficum* produces karlotoxins (KmTX) that permeabilize cell membranes, resulting in cell death through osmotic lysis. Membrane sterol composition appears to govern sensitivity KmTX, with a preponderance of 4 α -methyl sterols (gymnodinosterol) providing immunity to the toxin. Like its host, *Amoebophrya* sp. ex *K. veneficum* possesses gymnodinosterol and is presumed to be immune to KmTX. We examined the effect of purified KmTX on several host-parasite systems, the ability of *Amoebophrya* to infect non-toxic to highly toxic *K. veneficum*, and toxin levels of parasitized *K. veneficum* over the infection cycle. Addition of purified KmTX to culture medium had no effect on survival or infectivity of *Amoebophrya* ex *K. veneficum*, but caused mortality and reduced infection of *Amoebophrya* from other host species. Parasite prevalence was positively correlated with *K. veneficum*

toxicity, suggesting that *Amoebophrya* is more likely to control toxic blooms than non-toxic blooms. KmTX ml⁻¹ increased with growth of *K. veneficum* in control cultures, but declined in infected cultures as the parasite completed its life cycle. On a cellular basis, toxin content of infected and uninfected cultures differed little during the experiment, suggesting that the parasite does not actively catabolise host toxin. Rather, infection appears to promote degradation of toxins via death of host cells and subsequent bacterial activity. Thus, this parasite may limit the occurrence of toxic *K. veneficum* blooms in marine and estuarine environments, while simultaneously functioning as a pathway for dissipation of host toxin.

Life history and morphological strategies of sponges drive community dynamics on mangrove roots, with help from predators, competitors, and pathogens

J.L. Wulff

Descriptions of the rich sponge faunas inhabiting mangrove roots at various Caribbean sites are unanimous in pointing out the heterogeneity of species distribution and abundance patterns at all scales, from very local (e.g., adjacent roots) to regional. Abiotic factors have often been implicated by correlation, but the possibility that ecological interactions, and life history and morphological strategies of the sponges, also play key roles has not been examined comprehensively. I have been exploring the processes underlying these patterns by probing community dynamics with experimental manipulations and time-series censusing at sites in Belize (Twin Cays), Panama (near the Bocas del Toro station), and the Florida Keys (Long Key). Established communities on roots have been fully censused (by



Amoebophrya; stained, autofluorescing, and viewed by interference contrast.



Comparisons of chemical by mediated marine plant-herbivore interactions on Florida, Caribbean and Pacific reefs and consequences for reef communities

V.J. Paul, R. Ritson-Williams, L.J. Walters, I.B. Kuffner & M.A. Becerro

Overgrowth of corals by macroalgae and benthic cyanobacteria is of increasing concern on coral reefs worldwide. Algal species that have proliferated in reef habitats often contain natural products that deter generalist herbivores. On reefs in Guam, Florida, and Belize, we determined the palatability of common reef algae to reef fishes. In Belize, we also studied the feeding preferences of the sea urchin *Diadema antillarum*. Some macroalgae were not eaten by fishes even when left in areas of high herbivory for 1-2 days. Cyanobacteria were not readily consumed by reef fishes. Inter-specific variation was observed in the susceptibility of different species of *Dictyota*, *Halimeda*, and *Caulerpa* to herbivory. *Diadema antillarum* individuals were less discriminating than reef fishes and consumed some of almost all species offered to them. For example, fish were strongly deterred by some brown algal extracts, while the urchins readily consumed them. Herbivorous reef fishes and *D. antillarum* have different algal preferences (often driven by chemical defenses) and can have differential effects on algal community composition. Fish and urchins can control a mixed community of algae on Caribbean reefs better than either one alone. Due to the macroalgal dominance observed on reefs today, competition between corals and macroalgae is a topic of great interest. Past research has shown that adult corals are very good competitors with macroalgae, but it is at the early life-history stages, as larvae and new recruits, that corals are thought to be inferior competitors. We directly tested this hypothesis in controlled field and laboratory experiments with different species of coral larvae. Species of *Lyngbya* and *Dictyota* as well as extracts of some of these species caused either recruitment inhibition or avoidance behavior in coral larvae. On reefs experiencing increased algal abundance, the restocking of adult coral populations may be slowed due to recruitment inhibition caused by algal natural products

volume) yearly for 3 years, initially bare artificial roots (suspended pvc pipes) have been monitored for recruitment and subsequent community development for up to 5 years, and small, asexually generated (by razor blade) individuals of the 7–9 most abundant sponge species have been placed on artificial roots and subsequent community dynamics monitored for up to 2 years. Patterns of community development on initially bare pvc pipes suggest that trade-offs between recruitment efficiency and competitive ability may be responsible for at least some among-root heterogeneity. Even though community dynamics on pipes to which individuals of the most abundant species had been added were not influenced by the stochastic effects of larval recruitment, heterogeneity of sponge distribution and abundance among pipes was still very high. When sponge species were sorted into categories defined by different morphological strategies for gaining and holding onto substratum space, it became clear that presence or absence of representatives of these morphological categories influenced community dynamics substantially. Interactions with predators, pathogens, and non-sponge spatial competitors played surprisingly important roles in creating differences among pipes that began with identical inhabitants.

Molecular mechanisms of establishment and maintenance of a nematode/bacteria symbiosis

S. Bulgheresi

Establishment of RNA interference in the marine symbiotic nematode Laxus oneistus

To test the involvement of worm genes in symbiosis, I tried to suppress their function by the reverse genetic technique RNA interference (RNAi). I collected adult *L. oneistus* individuals and immediately soaked them into 4 mg/ml of *beta-tubulin* or *cytochrome c oxidase (cco)* or *mermaid* or *pesto* double strand RNAs (dsRNAs) or filter-sterilized seawater without dsRNA (essentially according to Aboobaker et al. 2004). *tubulin* and *cco* are house keeping genes and were used as positive controls i.e. their functional knock-out should be lethal. The Ca²⁺-dependent lectin Mermaid mediates worm-symbiont and symbiont-symbiont attachment (Bulgheresi et al., 2006), while Pesto is a highly hydrophilic protein which is thought to function as a Mermaid chaperone. We expected *mermaid* and *pesto* dsRNAs not to be toxic for the worms and to cause symbiont detachment, in case their mRNAs would be required for symbiosis maintenance. Although *beta-tubulin* and *cco* dsRNAs were reproducibly more, and more rapidly, toxic than *mermaid* and *pesto* dsRNAs, and cause the worms to stop moving, stretch and die within 24 hrs, also *mermaid* and *pesto* dsRNA seemed to be toxic for the worms. Moreover, I could RT-PCR amplify *beta-tubulin*, *cco*, and *mermaid* mRNAs in dsRNA-treated worms indicating failure of specific gene silencing and that worms' death was caused by RNA toxicity.

A totally unexpected, but reproducible, finding was the release of fertilized eggs by worms soaked in *beta-tubulin* or *cco* dsRNAs. This gave me the opportunity to follow, for the very first time, the earliest stages of *L. oneistus* development and, at the same time, to exclude vertical transmission of the symbiont from mother to offspring. Although I could not detect any symbiont on the eggshell of unhatched embryos by microscopic observation, we want to confirm this by PCR with symbiont 16S rDNA-specific primers.

Pharmacological inactivation of a L. oneistus tyrosine-kinase Abl₁ homolog

Because the *Abl₁* gene is thought to suppress immune response in the model nematode *Caenorhab-*

ditis elegans (Burton et al. 2006) and because an *Abl₁*-like gene is abundantly expressed in *L. oneistus*, I tested if its pharmacological inactivation causes symbionts' death/dissociation. I incubated freshly collected worms in 10 µM *Abl₁* inhibitor STI571/Gleevec in seawater or in seawater without inhibitor and microscopically observed the worms every 4-8 hrs for three days. I could not appreciate any difference between treated and untreated worms. This might be due to the inability of STI571/Gleevec to block *L. oneistus Abl₁* or to the fact that *Abl₁* does not suppress *L. oneistus* immune system.

Collection of water samples for the detection of the free-living forms of stilbonematid symbionts

Although environmental transmission of the stilbonematid symbionts has been hypothesized for decades, up to now, their presence in the environment has not been proved. I collected 10 litre of surface seawater in front of the Carrie Bow Cay dock and also 10 litre ca. one mile off the reef. 1 litre aliquots were filtered through 0.2 µm filters and the filters store in a dry shipper. Back in Vienna, DNA was extracted from some of the filters and used as a PCR template. We could amplify 16S rDNA of the symbiont of *Robbea* sp.3 (a stilbonematid exclusively found at Carrie Bow Cay), but also of those of *Robbea* sp.1 and 2, collected in Corsica (France) and Little Cayman (Cayman Islands), respectively. There was no difference between dock and off-shore water samples indicating that the free-living form of the symbionts can survive in very diverse habitats. Moreover, *Robbea* sp.3 symbionts were also detected on the filters by fluorescence in situ hybridization. Surprisingly, the positioning of the division plan in mitotic symbionts seems to follow environmental cues: while sticking to the host cuticle, the bacteria set their division plan parallel to their main axis, while free-living, they set it perpendicular.

Diversity and distribution of the genus *Paracatenula* (Platyhelminthes, Catenulida) associated with symbiotic sulfur-oxidizing bacteria

J. Ott & H. Gruber

Flatworms of the genus *Paracatenula* Sterrer and Rieger 1974 (Catenulida, Platyhelminthes) are part of the interstitial meiofauna at the oxic-anoxic interface



acters (e.g. immotile sperm) is corroborated by our analysis of 18S and 28S rRNA.

Symbiont phylogeny

Every species of *Paracatenula* so far analyzed harbors one strain of closely related *Alphaproteobacteria*, indicated by our 16S rRNA phylogenetic analysis and FISH. The symbionts form a single clade within the available 16s rRNA sequences, distantly related (all <90% sequence similarity) to *Rhodovibrio*, *Magnetospirillum* and the *Alphaproteobacteria* endosymbionts of oligochetes.

Symbiont metabolism

All species of *Paracatenula* appear whitish in incident light, a feature typical for many meio- and macrofaunal invertebrates symbiotically associated with SOP. Element analysis of the trophosome region of *Paracatenula erato* shows a sulfur content between 9% and 17%. With PCR based assessments we were able to recover two lineages of the symbionts' *aprBA* gene. Phylogenetic analysis places both genes among the two known lineages of *aprBA* from SOPs. Collaborating partners at the

department of microbial ecology at the University of Vienna were able to sequence the *rdsrAB* gene of two species of *Paracatenula* and place it among alphaproteobacterial SOP (Loy et al. 2008). The possession of the *rdsrAB*, the phylogenetic position of the *aprBA* genes and sulfur storage is a strong indication that the symbionts of *Paracatenula* are thiotrophic sulfur oxidizers.

in shallow water subtidal sands of tropical to warm temperate oceans. Adult *Paracatenula* lack a mouth and a gut lumen. Instead, a parenchymatic “trophosome” containing intracellular bacteria fills most of the body (Ott et al. 1982). We found wide variety of different species in surveys in the vicinity of Carrie Bow Cay field station, Belize Barrier Reef, measuring from 0,4mm to 6mm in length. 8 species including the two already described species *P. erato* and *P. urania* were found in more than one sample, two other species only on one occasion. *P. erato* and *P. sp. 1* and *P. sp. 2* were recovered from most or all sampling sites. *P. sp. 1* and *sp3* were locally high abundant, with more than 100 specimens found in single 10 cm² cores. The placement of the host within the Catenulida based on the few available morphological char-



Sympatric speciation by hybridization in a marine fish

L.A. Rocha

Mechanisms that lead to speciation remain among the most debated topics in evolutionary biology, and sympatric speciation is especially difficult to demonstrate in nature. Because of their peculiar biogeography and their rare ability to produce sounds, fishes of the genus *Haemulon* serve as a great subject for tests of speciation hypotheses. Collectively known as grunts, the genus is comprised of 19 nominal species and occurs in tropical and subtropical reefs along both sides of the Americas. Aiming to elucidate the phylogenetic relationships among the species of *Haemulon*, a combined total of ~2,000 base pairs from two mitochondrial genes (cytochrome b and cytochrome oxidase I), one nuclear intron (TMO-4C4) and one nuclear gene (RAG2) were obtained from all nominal species. Our data also indicate that the trans-isthmian *H. steindachneri* is composed of two species, one in each side of the Americas, and we propose the revalidation of the Atlantic species. The



closure of the Isthmus of Panama seems to have played a role in the diversification of *Haemulon*, however, many sister species pairs have completely overlapping geographical distributions, indicating that vicariance is not the only process driving speciation in this genus, and that sympatric speciation by sound recognition is possible. Finally, the species *H. carbonarium* seems to have originated through a hybridization event between *H. macrostomum* and *H. flavolineatum*. These three species form a strongly supported group in the phylogeny, however, mtDNA groups *H. carbonarium* with *H.*

macrostomum, whereas nuclear DNA groups *H. carbonarium* with *H. flavolineatum*. A detailed morphological analysis shows that many morphological characters in *H. carbonarium* are intermediate between *H. macrostomum* and *H. flavolineatum*, indicating a probable hybrid origin for *H. carbonarium*. If supported by additional ongoing analyses, this will be the first case of sympatric speciation by hybridization in a vertebrate animal

Does color pattern drive speciation in *Hypoplectrus* coral reef fishes?

E. Bermingham & O. Puebla

Theory shows that speciation in the presence of gene flow occurs only under narrow conditions. One of the most favorable scenarios for speciation with gene

flow is established when a single trait is both under disruptive natural selection and used to cue assortative mating. We demonstrate the potential for a single trait, color pattern, to drive incipient speciation in the genus *Hypoplectrus* (Serranidae), coral reef fishes known for their striking color polymorphism. We provide data demonstrating that sympatric *Hypoplectrus* color morphs mate assortatively and are genetically dis-

tinct. Furthermore, we identify ecological conditions conducive to disruptive selection on color pattern by presenting behavioral evidence of aggressive mimicry, whereby predatory *Hypoplectrus* color morphs mimic the color patterns of non-predatory reef fish species to increase their success approaching and attacking prey. We propose that color-based assortative mating, combined with disruptive selection on color pattern, is driving speciation in *Hypoplectrus* coral reef fishes.

Processes across Ecosystems & Conservation

History of reef-coral assemblages on the Rhomboid shoals of Belize

R.B. Aronson, I.G. Macintyre, A.M. Moesinger, W.F. Precht, & M. Dardeau

Coral assemblages of the rhomboid shoals of the Belizean barrier reef have undergone dramatic, historically unprecedented changes over the last several decades. Prior to the late 1980s, the flanks of the shoals exhibited a distinct biological zonation, with branching *Porites* spp. dominant in a shallow zone (0–3 m water depth); the staghorn coral *Acropora cervicornis* dominant in an intermediate zone (3–15 m depth); and large, plating agariciids and the lettuce coral *Undaria* (formerly *Agaricia*) *tenuifolia* dominant in a deep zone (15–30 m depth). *Acropora cervicornis* died off catastrophically from white-band disease after 1986 and was replaced by *Undaria tenuifolia* in the intermediate zone. Push-cores extracted from intermediate depths in previous studies showed that *Acropora cervicornis* was the dominant space occupant and primary framework builder for millennia prior to the phase shift to *Undaria tenuifolia*. Cores extracted from the shallow zone showed that *Acropora cervicornis* dominated until several centuries ago, when the tops of the reefs reached ~2 m water depth and branching *Porites* spp. replaced it. In contrast, three cores extracted from the deep zone in the present study showed that for millennia the subsurface coral assemblage, like the assemblage on the modern deep-reef surface, was dominated by large, plating agariciids and *Undaria tenuifolia*. Because white-band disease only affects acroporid corals, the unprecedented phase shift that followed the outbreak was confined to the intermediate zone. High sea temperatures in the summer of 1998 caused coral bleaching and mortality, especially of agariciids in the intermediate and deep zones, but to date this event has not left a geological signature in the Holocene recor.



Patterns of the water movement over the forereef at Carrie Bow Cay

K.H. Koltes & T.B. Opishinski

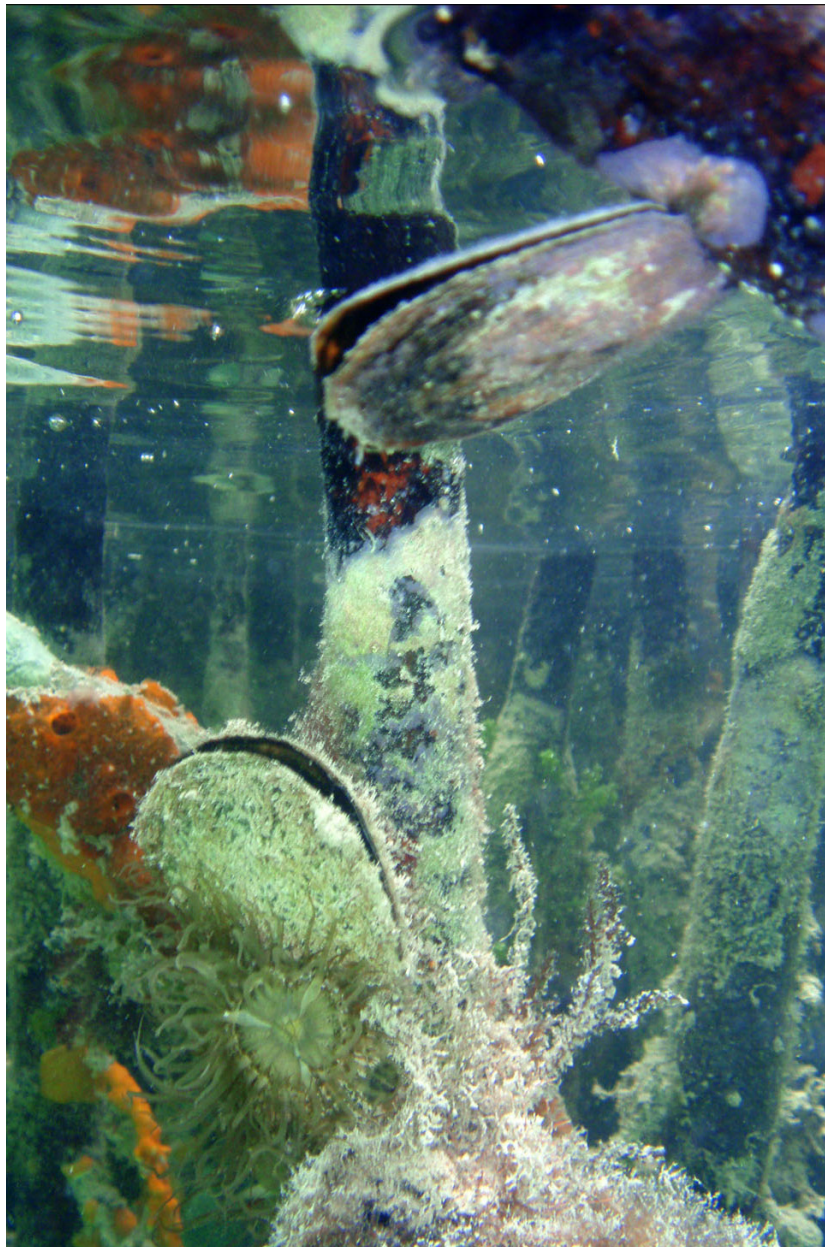
Since 1993, meteorological and oceanographic conditions, including water and air temperature, rainfall and turbidity, have been monitored as part of the CARICOMP program at Carrie Bow Cay (CBC). In 1997, an automated monitoring system was established at Carrie Bow Cay (under CCRE funding) that provides an independent set of weather conditions and water quality measurements. Qualitative observations following Hurricane Mitch and other major weather systems that produced heavy rainfall along the Belizean coast indicated that turbid, low-salinity water typically passes over the forereef at CBC within 1–2 days. Qualitative observations also indicated that these pulses were occurring with more frequency and intensity since CARICOMP monitoring began in 1993. To better characterize specific water masses on the forereef, light meters were installed at the CARICOMP site in 13.5m of water as a proxy for measuring turbidity (water quality). Light intensity (L/m^2) has been recorded on the forereef intermittently since Dec 2002. Light intensity was analyzed along with other CARICOMP measurements and data from the automated monitoring system. As expected, strong correlations exist between the light intensity and other forereef measurements as well as conditions mea-

sured on the shoreward side of the reef (e.g., temperature, incident radiation, rainfall, etc.). Under normal weather conditions, conditions on the forereef are fairly stable. Following major weather events, signatures of different water masses can be seen in the oceanographic records, including upwelling of deeper ocean water and/or input of riverine (coastal) water. The appearance of both water mass types correlates strongly with weather conditions.

Biophysical controls on habitat stability of Caribbean mangrove ecosystems

K.L. McKee

Habitat stability of mangrove and other coastal wetlands depends upon the capacity of the system to maintain soil elevations relative to sea level. Although some mangrove wetlands develop in alluvial habitats with abundant mineral sediment, mangrove systems in sediment-deficient settings are dependent upon biogenic processes of vertical land-building. Plants contribute directly to soil formation through accumulation of organic matter, but few data directly link specific biological processes to elevation change in coastal systems. This type of information is necessary, however, to accurately predict future responses of coastal wetlands to sea-level rise and interactions with other biophysical controls on soil elevation. Biogenic processes are particularly important for the oceanic mangrove island-type of setting common throughout the Caribbean Region. Work conducted along the Caribbean coastlines of Belize, Florida, Honduras, and Panama focused on processes of mangrove peat formation and plant production-decomposition in relation to modern rates of elevation change, vertical accretion, and shallow subsidence. Elevation change was measured with Surface Elevation Tables (SETs) along with root accumulation and surface accretion of organic and inorganic material (above marker horizons). To compare modern changes in elevation with past peat development, geological rates of accretion were determined using radiocarbon-dated peat cores. The findings showed that 1) peat has primarily formed through slow accumulation of mangrove root matter that resists de-



cay, 2) modern rates of elevation change varies with health and productivity of mangroves and in response to changes in nutrient availability, and 3) without inputs of mangrove roots and other organic matter, land submergence is inevitable as subsidence and sea-level rise continue. These findings have relevance for models predicting sea-level rise impacts to mangrove wetlands and show that damage to or removal of mangroves in this type of setting (oceanic mangroves) will impair the capacity of these systems to keep up with sea-level rise

Eutrophication and fisheries: A global perspective

D. Breitburg, D. Hondorp & L. Davias

Both nitrogen loadings and the number of coastal systems experiencing hypoxia have increased worldwide. Numerous studies have shown the potential for negative effects of hypoxia, but scaling up from effects at the local or individual scale to population, system-wide, and fisheries effects is not straightforward for mobile species. Cross-system comparisons of >35 estuaries and semi-enclosed seas in industrialized nations suggest that the relationship between nitrogen loading and fisheries landings is unaffected by the spatial extent of hypoxia. N loading and fisheries landings were positively related up to about 15,000 kg N km⁻² y⁻¹, the point represented by Chesapeake Bay. The positive relationship between N and landings of mobile demersal species was unaffected by hypoxic extent. The increased demersal:pelagic ratio in eutrophic systems reported in other studies is highly dependent on fisheries regulations, the increased catch of pelagics in some highly enriched systems, and a very high pelagic:demersal ratio in a single system – the Black Sea. Mean trophic level of catch and mean size of species in the fishery also did not differ between systems with and without extensive hypoxic or anoxic areas. Nutrient enrichment creates a spatial mosaic of prey-enriched and physiologically stressful habitats. Spatial averaging of enriched and degraded habitats, and preferential use of enriched habitat, may reduce system-wide negative effects. Tur-

bidity may reduce piscivore capture success as well as the abundance of macrophytes that provide a predation refuge. Fisheries exploitation also keeps most species below carrying capacity, potentially reducing the realized consequences of habitat loss. Our analyses suggest that improving water quality is likely to increase populations and fisheries landings only at the local scale and for particularly susceptible species. Such improvements may be especially critical in developing countries where discharge of raw sewage creates more severe and long-lasting oxygen depletion and human populations are more dependent on local resources.

Biogeography of marine invasion: Current status and future predictions

G.M. Ruiz, P. Fofonoff, B. Steves, K. Larson, L. McCann, A. Whitman Miller & A.H. Hines

Biological invasions are a significant force of change in coastal ecosystems. Invasions have occurred throughout Earth's history, but the scale and tempo has increased strongly in recent time due to global trade. Available data suggest there is a strong latitudinal pattern in recent marine invasions, with more non-native species documented in temperate marine communities than polar or tropical systems. This geographic pattern of invasion may reflect historical biases in search effort and taxonomic knowledge. Contemporary surveys suggest these patterns are robust across mid- and high-latitudes, when controlling for search effort. For example, a standardized survey of sessile invertebrate assemblages in estuaries of western North America found a



significant decrease in non-native species richness with increasing latitude (32° to 61° N). Several mechanisms may explain the observed invasion pattern across latitudes, operating alone or in combination, such as differences in (a) propagule supply, (b) biotic resistance to invasion, (c) environmental resistance to invasion, and (d) disturbance regime. To date, the relative importance of these mechanisms across geographic regions has not been evaluated, but each may be expected to change over time. Of particular interest and concern are the interactive effects of climate change and human activities on marine invasions, especially at high latitudes. Current climate change models pre-

dict not only an increase in sea surface temperatures but also a rapid reduction in sea ice in the Arctic. Combined with human responses, climate change is predicted to cause directional shifts in invasion biogeography, including increased invasion opportunity at high northern latitudes.

Decimating mangrove forests for commercial development in the Pelican Cays, Belize: long-term ecological loss for short-term gain?

I.G. Macintyre, M.A. Toscano, I.C. Feller & M. Faust

The unique, biologically diverse and delicate ecosystems of Pelican Cays, Belize, are in serious danger from sediment suffocation related to the recent clear-felling of mangroves for commercial development in what is currently designated Southwater Cay Marine Reserve. Field observations in the Pelican Cays in March 2007 revealed extensive clear-felling of mangroves and covering of exposed peat surfaces with sediment dredged from the adjacent seafloor to create false sand cays. On Manatee Cay, introduction of dredge spoils taken from the nearby seabed has resulted in fine sediment plumes spilling into the adjacent ponds, smothering the attached benthic communities on mangrove roots and burying *Thalassia* bottom communities. In addition, comparative studies of microalgal (phytoplankton) assemblages in a Manatee Cay pond before and after mangrove clearing indicate a dramatic loss in this group owing to high turbidity observed in the water column and signaling a serious impact to this aquatic ecosystem. Continuing clear-felling, burning and dredge and fill operations were taking place on Fisherman's Cay, with additional survey lines cut on Fisherman's, Manatee, and Cat Cays. A series of aerial photographic surveys from 2003 to 2007 document the extensive loss of mangroves on both Manatee and Fisherman's Cays. Additional clearing of mangroves has occurred on Northeast Cay, Bird Cays, and Ridge Cay resulting in a total of 15.3 ha or more than 29% on the mangrove community that have been destroyed to date in the Pelican Cays. The conversion of mangrove ecosystems for residential, tourism, and commercial uses is obviously widespread and increasing in Belize, as well as elsewhere in the global tropics. This pressure is having an adverse effect on the health of coral reefs

and biomass/viability of commercial fisheries, which, ironically, are essential for tourism, not to mention local livelihood.

The healthy Mesoamerican reef ecosystem initiative: An opportunity to enhance collaboration and application of environmental data

M. McField

The Healthy Mesoamerican Reef Ecosystem Initiative is a collaborative international initiative that generates user-friendly tools to measure the health of the Mesoamerican Reef (MAR) Ecosystem, and delivers scientifically credible reports to improve decision-making that effectively sustain social and ecosystem well-being. The Initiative is becoming recognized and respected as an independent and scientifically rigorous partnership that works to improve management decisions that affect the Mesoamerican Reef at the regional, national, and local level. The Healthy Reefs conceptual framework is built upon the fundamental elements of reef ecosystem structure (biodiversity, community structure, abiotics, habitat extent) and function (reproduction, herbivory, coral condition, reef accretion and bioerosion), while also integrating human stressors and social dimensions. Suites of indicators have been selected that measure these different components. Combinations of indicators can be evaluated to answer a wide variety of applied and basic research questions on multiple spatial scales. One example of using these indicators to answer practical management questions is the evaluation of impacts of the 2005 Bleaching event in Belize. Water temperatures surpassed 30°C for much of the summer, resulting in a coral bleaching index of four degree heating weeks, the highest since 1998 (which reached an index of eight). Approximately 30-40% of the corals bleached but the passage of eight tropical storms intermittently contributed to lowering water temperatures and reducing the cumulative thermal stress. In summer 2006, we joined forces with TNC and WWF to carry out the largest reef assessment ever conducted in the MAR, involving over 330 sites (141 in Belize). The Belize survey assessed over 5,614 corals, finding minimal coral mortality (<1.5%) or disease (<2%). However, the mean coral cover remains low (about 10%) showing no signs of recovery from the recent declines. The partnership is now working to syn-

thesize these and other indicators into an annual report card for the region.

Biotic turnover on reefs of the Caribbean and eastern Pacific: Holocene surprises and future projections

R.B. Aronson, I.G. Macintyre & W.F. Precht

Although coral reefs are degrading globally at present, widespread mass mortalities of dominant, framework-building corals first occurred in the Caribbean and eastern Pacific in the 1980s. Real-time observational data can be combined with millennial-scale paleobiological records to explore the geological implications of ecological phase shifts in both regions. Populations of acroporid corals died off catastrophically from disease in the Caribbean during the 1980s–90s, and their loss depressed coral cover regionally. Coring studies in Belize and Jamaica revealed that the *Acropora*-kill, and the ensuing replacement of *Acropora* by coral taxa that are not framework builders, were unprecedented events in at least the last 3000–4000 years. Rates of vertical reef accretion have been slowed or halted over the last 25 years. In the eastern Pacific, populations of *Pocillopora damicornis* were bleached and killed by the 1982–83 El Niño event. Coral mortality and subsequent bioerosion of reef frameworks suggested that centennial-scale recurrences of strong El Niños are responsible for the slow accretion rates of eastern Pacific reefs. Off the Pacific coast of Panamá, *Pocillopora* recovered rapidly after 1983 in some places. Where it did not recover the *Pocillopora* rubble was colonized by *Psammocora stellata*, which is not a framework builder. Coring studies in progress in the Gulf of Panamá are showing that *Pocillopora*-kills and shifts to *Psammocora* had occurred previously during the past 3000–4000 years; however, *Pocillopora* growth was suppressed continuously for centuries, depressing vertical reef accretion for far longer than the return time of individual, strong El Niño events. These depressed rates of reef growth can be used to parameterize models

of vertical accretion under long-term scenarios of biannual to annual coral bleaching, predicted to commence in the next several decades.

The dynamic hydrology of an overwashed mangrove island

D.W. Urish, R.M. Wright & W. Rodriguez

The tidally induced hydrodynamics in an overwashed mangrove island, in conjunction with the topography, greatly affect the ecosystem and vitality of the resident mangrove systems. This is evident from a 20 year study on the small archipelago of Twin Cays lying along the Belizean Barrier Reef. This study concentrates on the interior hydrology of West Island of Twin Cays, a 21.5 ha island characterized by a vigorous fringe of red mangrove (*Rhizophora mangle* L.) and a depressed central portion. The dominant water feature is a large shallow pond interconnected by channels in direct communication with the surrounding reef lagoon. Sparse dwarf red mangrove dominant in this pond. Poor flushing creates water temperatures ranging from 25°C in the winter to 40°C in the summer. High evapotranspiration creates a hypersaline condition of 45 ppt salinity in summer. In winter with the infusion of fresh rain water, the pond changes to a brackish water of less than 5 ppt. Additionally, of primary significance to the vitality of the mangrove ecosystem is the duration of the flooding-exposure cycle, viz. the hydroperiod. The temporal and spatial characteristics of the interior flow system were investigated using extensive field measurement and the employment of animated computer graphics produced from dye flow studies. While for the past 8000 years the mangrove growth has managed to keep up with rising sea level, the future is in doubt because of anticipated greatly increasing sea level rise rates. The islands of Twin Cays, with its history of comprehensive observational research, remains an important location for observing and measuring changes in the mangrove systems as they occur in a world of dramatic coastal change.

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