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# *CCRE REPORTS 2005*

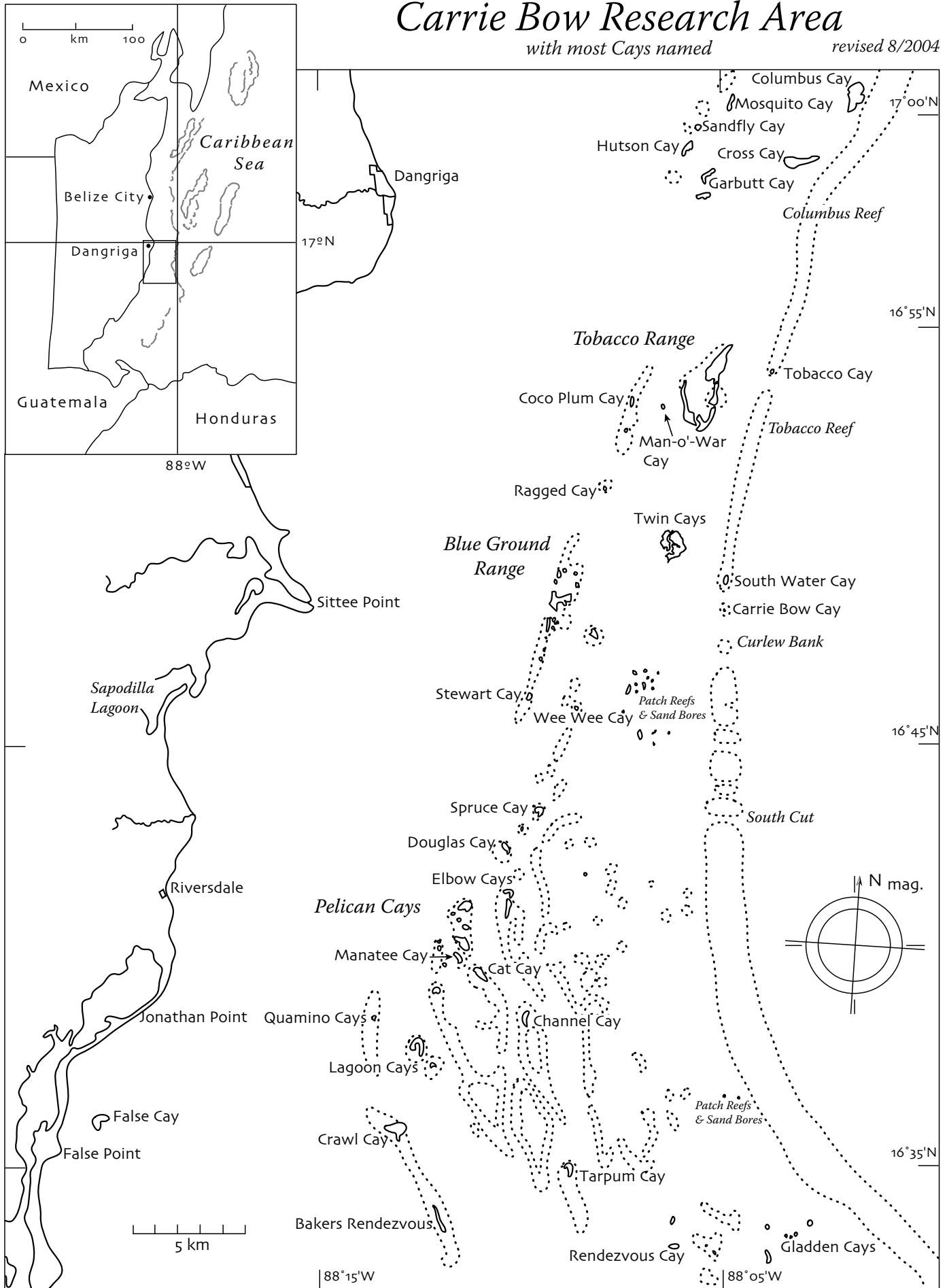
Caribbean Coral Reef Ecosystems • National Museum of Natural History

January 2006

# Carrie Bow Research Area

with most Cays named

revised 8/2004





*CCRE REPORTS*  
*2005*

National Museum of Natural History  
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# Research Projects

## Biodiversity and its Links to the Ecosystem

### Algae

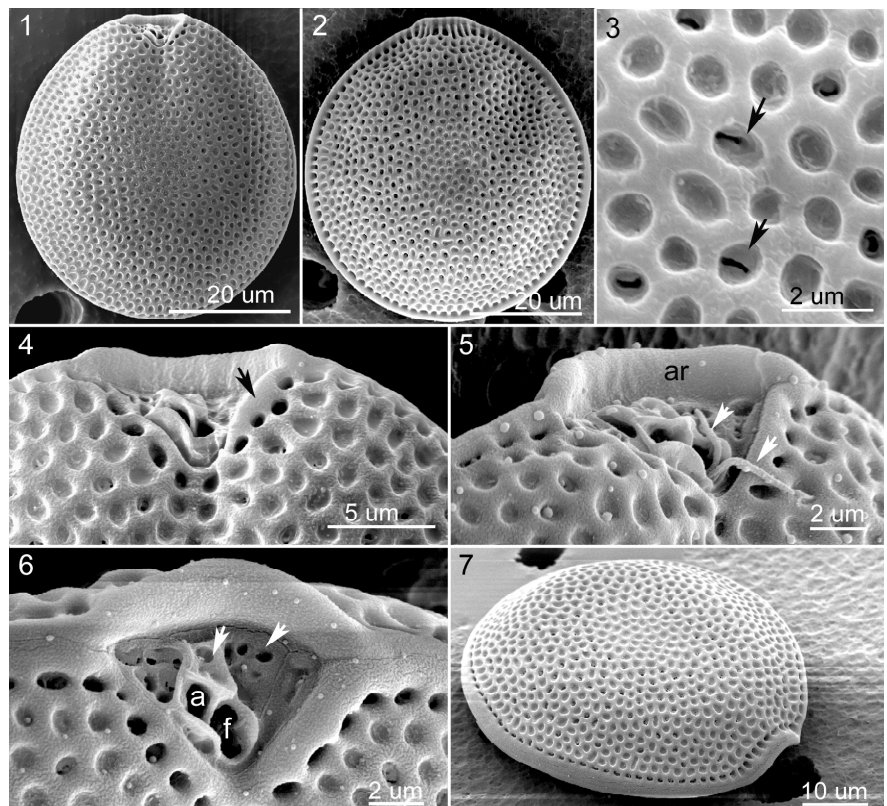
#### Phylogenetic analysis of 15 species of *Prorocentrum* (Dinophyceae) including two new species: *P. valentis* and *P. levis*

M. A. Faust

Benthic dinoflagellates are an important component of tropical, shallow marine phytoplankton communities where benthic *Prorocentrum* species frequently dominate. Many of these species produce various toxins that can adversely affect human and animal health. Despite what is known about the toxicity of these species, relatively little information exists about their diversity or ecology. Species in the genus *Prorocentrum* have long been recognized as being distinct from other dinoflagellate genera based on cell shape, thecal surface morphology and the anterior flagellar insertion. Historically, the species currently classified under *Prorocentrum* were divided into two separate genera, *Prorocentrum* and *Exuviaella*, which have been proven inconsistent. While assignment of species to the genus *Prorocentrum* is straightforward, establishing phylogenetic relationships among *Prorocentrum* species has proven difficult. This difficulty arises from their small size and morphological similarities and variations that cannot be easily classified into discrete character states for phylogenetic analysis. For this study, a Bayesian maximum-likelihood analysis of SSU rRNA gene sequences, along with morphological differences determined using SEM, were used to justify the establishment of two new species,

*P. valentis*, and *P. levis*, and determine their evolutionary relationship to all known *Prorocentrum* species for which comparable gene sequence data were available (Faust et al, 2005).

The two newly described species were included in a phylogenetic analysis of the fifteen known *Prorocentrum* SSU rRNA genes which fully supported the validity of the key morphological characteristics used to identify species. Key characteristics include the architecture of thecal plates, as well as the morphology and ornamentation of the periflagellar area. The analysis also revealed that the *Prorocentrum* species could be divided into two major clades. The presence of distinct clades implies a major evolutionary divergence among *Prorocentrum* species. Genetic divergence, however, did not consistently correspond with any known morphological, biochemical or behavioral characteristics, and consequently would not support the reestablishment of the genus *Exuviaella*.



*Prorocentrum valentis* sp. nov. 1. Areolated right valve surface. 2. Areolated left valve surface. 3. Areolae round to oval, and some have an oblong-shaped trichocyst pore in the center (arrows). 4. Periflagellar area is a V-shaped triangle with an adjacent raised margin (arrowhead). 5. Both flagella emerge from the flagellar pore (arrows); note flat apical ridge (ar). 6. In apical view, flagellar pore (f) is larger than the auxiliary pore (a); both pores surrounded by a protuberant apical collar. Several smaller pores are on the adjacent periflagellar platelets (arrowheads). 7. Cell shape in left valve view is a compressed oval, the intercalary band is smooth.

## *Gambierdiscus* linking taxonomy and genetics (Dinoflagellates)

M.W. Vandersea, R.W. Litaker, S.R. Kibler, M.A. Faust & P.A. Tester

*Gambierdiscus* species produce a variety of ciguatoxins that bioaccumulate in the food chain. Human toxicity is generally associated with consuming tainted fish such as barracuda or groupers. Much effort has been devoted to characterizing the structure and toxicity of ciguatoxins. However, the taxonomy and ecology of the dinoflagellates that produce these toxins is relatively understudied, largely owing to difficulties encountered when trying to discriminate species by light microscopy. SEM techniques can provide accurate identification and have been used to identify six *Gambierdiscus* species that exhibit pantropical distributions. SEM techniques, however, are too expensive, time consuming and non-quantitative to use in analyzing routine field samples. Having accurate distribution and abundance data may be crucial to interpreting temporal and geographic differences in toxicity. Currently, there are reports that toxicity of reef fishes varies both temporally and regionally. A fundamental question that arises from these observations is whether the variations in toxicity are related to dinoflagellate species distribution and abundance or to other factors such as environmental induction of toxin production. The former question can only be answered by having accurate abundance and distribution data for various species. Quantitative molecular assays afford an excellent means of obtaining this information.

Before these types of molecular assays can be developed, it is necessary to have genetically well defined cultures. Unfortunately, these cultures do not exist. To overcome this obstacle, we have established numerous ciguatera dinoflagellate cultures isolated from the barrier reef system in Belize, Central America and from Florida. In addition, a *Gambierdiscus* species from a deepwater reef system off the coast of North Carolina has also been isolated. We have employed electron and fluorescent microscopy to analyze cellular morphology. Currently we are developing molecular methods to sequence a ~ 3000 bp region of the ribosomal gene complex for each species. The sequence data will be used to develop quantitative PCR methods to rapidly identify and quantify specific *Gambierdiscus* species in future field studies.

## Phylogenetic relationship to other dinoflagellates of *Coolia tropicalis* and two new *Prorocentrum* species isolated from Belizean barrier reef and oceanic mangrove systems

M.W. Vandersea, M.A. West, S.R. Kibler, M.A. Faust, R.W. Litaker & P.A. Tester

The mangrove islands of the south central lagoon of the Belizean Barrier Reef contain a large diversity of tropical dinoflagellate species. Sediment samples and samples of floating detritus were collected from Carrie Bow Cay, South Water Cay and Twin Cays, Belize (May, 2003) as part of ongoing field surveys that explore the dinoflagellate ecology and natural nutrient enrichment that occurs within these isolated barrier reef cays. Clonal cultures of *Coolia tropicalis* and two *Prorocentrum* species were established from these samples. Genomic DNA was extracted from each isolate and PCR-amplified using ribosomal DNA specific primers. The resulting SSU sequences were aligned with a representative sample of related dinoflagellate species using the CLUSTAL-W program. A maximum likelihood phylogenetic analysis of the aligned sequences was performed using the MrBayes 3.0 program. The resulting phylogenetic tree indicated that *Coolia tropicalis* was most closely related to *Alexandrium* species, consistent with similarities in *Kofoidian* plate tabulation of these two genera. The phylogenetic analysis also showed that the *Prorocentrum* species grouped into two distinct clades, suggesting this genus is polyphyletic. Ribosomal DNA sequence data and SEM analysis that revealed that the *Prorocentrum* cells are the undescribed species.

## Foraminiferans

### Endosymbiont bleaching and ecology of the foraminiferan *Sorites dominicensis*

S.L. Richardson

Endosymbiont bleaching in *Sorites dominicensis* was surveyed. Blades of the seagrass *Thalassia testudinum* were collected from two sites: the shallow reef flat off Carrie Bow; and Boston Bay, Twin Cays. These two sites differ in depth, water clarity, water movement,

and temperature. The seagrasses growing on the reef flat off Carrie Bow are often exposed during low tides, while in Boston Bay, the seagrasses remain mostly submerged during low tides, except for plants growing on the sand flats at the entrance to the Bay which were observed to be partially exposed in July 2005. The water on the reef flat off Carrie Bow is clear and shallow (~0.5 m), compared to the somewhat deeper water (~1 m) in Boston Bay, which is stained a reddish-brown color from high concentrations of tannin, leached from the surrounding mangroves.

Water temperature at both sites was monitored for approximately 3 days using Hobo StowAway Tidbit Temperature Loggers set to an interval of 40 seconds.

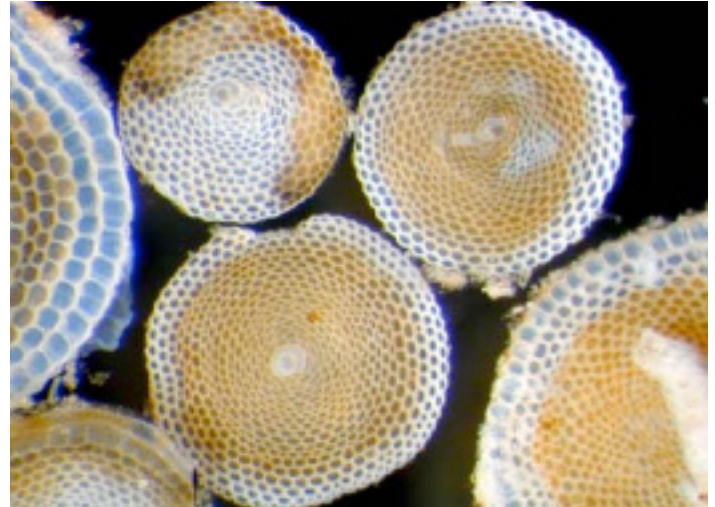
*Thalassia* blades were examined under the dissecting microscope, and all attached individuals of *S. dominicensis* were examined for evidence of bleaching, removed from the blades, and stored on a cardboard (paleontological) microslide. The dinoflagellate endosymbionts of live specimens impart a distinct yellowish-brown coloration to the cytoplasm. Bleached individuals have a mottled appearance due to the presence of large patches of light- or white-colored cytoplasm, interspersed with patches of healthy cytoplasm. Individuals that were totally bleached (completely white cytoplasm) were assumed to be dead. Individuals undergoing reproduction by multiple fission, possessed a central zone of colorless cytoplasm, surrounded by a ring of yellowish-brown cytoplasm in the peripheral chambers of the test where the juveniles begin to form. Bleaching and mortality rates were observed to be much higher in the Carrie Bow population compared to the population in Boston Bay at Twin Cays.

The dinoflagellate endosymbionts in soritacean foraminiferans are members of the *Symbiodinium microadriaticum* clade, other members of which include the zooxanthellae in corals and *Cassiopea* jellyfish. Numerous specimens of *S. dominicensis* were picked from seagrass blades collected from both localities, preserved in EtOH, and given to Dusty Kemp, a graduate student at the University of Georgia, for genotyping.

The relative abundance of *S. dominicensis* in the epiphytic foraminiferal biotas at each site will be determined from species counts of all epiphytic foraminiferans on four shoots of *T. testudinum* collected from Boston Bay and Carrie Bow Cay. The species identifications and counts were made in the lab on Carrie Bow Cay in July; data analysis is still in progress.

Artificial seagrass substrates (plastic aquarium grass) were attached to PVC pipes and left out at two

sites (vegetated and unvegetated sediments) near the boat dock off Carrie Bow Cay, and subsequently collected by Val Paul (late August). From a preliminary examination of the blades, it appears that epiphytic foraminiferans are more abundant on the substrate set out at the vegetated site, but a quantitative study has not yet been completed.



Live *S. dominicensis* specimens from Belize showing distribution of dinoflagellate endosymbionts in cytoplasm. Actively feeding specimens usually lack endosymbionts in outer chambers where digestive vacuoles are found. Top left & right: Specimens with bleached patches of cytoplasm. Lower center: Healthy specimen.

## Sponges

### DNA barcoding in sponges: A complementary tool for taxonomy

*S. Duran, K. Rützler & V. Paul*

Because of the limitations of morphological characters in sponges, molecular methods for diversity assessment are a new approach to taxon recognition. We report here our first steps towards a molecular taxonomy system in sponges that we believe will help in distinguishing and identifying sponge species.

In all, 65 species from different Caribbean regions and habitats were sequenced using the procedures described in Duran *et al.* (2004) and showed different cytochrome oxidase subunit I (COI) sequences, except for 5 species in the genus *Aplysina* (Verongida, Aplysinidae) and 2 species in the genus *Verongula* (Verongida Pseudoceratinidae). COI sequences from 38 species were either identical or very similar to other sequences from the same species. Furthermore, with the exception of *Aplysina* and *Verongula* species, COI sequence

differences between closely related species were far greater than differences within specimens of the same species.

lum where morphological characters are scarce and of limited use because of their plasticity.

A database called DNATaxPor (DNA Taxon-

 <b>DNATaxPor</b> Smithsonian Marine Station at Fort Pierce		<a href="#">Go to Species List</a>	
<b>Sample Code</b>	K12	<b>Genus</b> Ectyoplasia	<b>Species</b> ferox
<b>Subspecies</b>		<b>Color</b> orange	
<b>Lineage</b>	Porifera, Demospongiae, Poecilosclerida, Microcionina, Raspailiidae		
<b>Taxon authors</b>		<b>Reference</b>	
<b>Identified by</b>	Sandra Duran		
<b>Location</b>	Florida	<b>Sample Site</b>	11 foot mound, Keys
<b>GPS Coordinates</b>	N 24° 43.395 - W 80° 51.683		<b>Depth</b> 5m
<b>Sample Date</b>	30-5-2004	<b>Abundance</b>	Common
<b>Picture</b>			
<b>Voucher</b>	<input checked="" type="radio"/> yes <input type="radio"/> no	<b>USNM</b>	<b>Sequenced</b> <input checked="" type="radio"/> yes <input type="radio"/> no <input type="radio"/> failed
<b>COI Sequence</b>	GGATCGAAAAACGTAGTATTAAAGTTTCTATCCGTTAATAACATTGTAATC		
<b>Sequence Code</b>	DA		
<b>Haplotype</b>	Hap37		

Sample sheet representing the DNATaxPor Database.

It appears that a COI barcode will enable the separation of most related species of sponges. For two genera (*Aplysina* and *Verongula*) the COI showed very low levels of divergence between closely related species, some species within the genus shared the same haplotypes. The diagnosis of species is particularly difficult when specimens are young. Moreover, hybridization is often common when the ranges of recently arisen species overlap, further complicating identifications. Such newly emerged species are sometimes referred to as subspecies, or members of species complexes, to indicate their close genetic similarity. Some of these complications may apply to species in the genera *Aplysina* and *Verongula*. We believe that DNA barcodes will be a complementary tool for sponge taxonomists and will help elucidate the true limits between species in a phy-

lomy in Porifera) is being created in order to have all information from samples organized and linked, such as, species, location, picture, voucher number, and sequence. Our final goal is to publish the database on the World Wide Web.

## Cnidaria

### Systematics of hydroids (Cnidaria: Hydrozoa) of the Pelican Cays, Belize

*D.R. Calder & L. Kirkendale*

Studies on the hydroids of Belize are limited, and the primary focus of such work to date has been



on ecology rather than systematics. Initial work on the group in the area was undertaken by Spracklin (1982), who provided an inventory of species in the vicinity of Carrie Bow Cay. Subsequently, a series of studies on hydroids was undertaken in the Twin Cays area by one of the authors of this proposal, with an account of a new genus and species (Calder, 1988), an analysis of hydroid abundance and distribution in the mangal (Calder, 1991a), an assessment of hydroid substrate associations (Calder, 1991b), and investigations on factors regulating vertical zonation of the abundant intertidal species *Dynamena crisioides* (Calder, 1991c). Shortly thereafter, Kaehler and Hughes (1992) reported on three species of epiphytic hydroids on seagrasses near South Water Cay, the Belizean coast, a new species of syllid

The biodiversity of hydroids in mangrove ecosystems was studied at Twin Cays and the Pelican Cays, Belize, during the fall of 2005. Particular emphasis was given to the reproductive biology of *Symmetroscyphus intermedius*, gonophores of which were previously unknown. The species is particularly common on seagrasses.

An investigation was also undertaken on the nematocyst complement of the common mangrove jellyfish, *Cassiopea xamachana*, using specimens collected at Twin Cays. Nematocyst types were examined, and measurements of them were made, in specimens of different sizes, and from different parts of a given medusa.

## Revision and taxonomic study of the *Eudendrium* (Cnidaria, Hydrozoa) species of the Caribbean area

S. Puce, C.G. Di Camillo & S. Tazioli

The worldwide distributed genus *Eudendrium* is one of the richest among anthoathecate hydroids in terms of number of species.

During the period at Carrie Bow several hydroids belonging to different species (e.g. *Eudendrium capillare*, *Eudendrium* sp., *Zanclaea alba*, *Zanclaea* sp. 1, *Zanclaea* sp. 2, *Myrionema amboinense*, *Sphaerocoryne* sp., *Nemalecium lighti*, *Millardiana longitentaculata*, *Halecium nanum*, *Pennaria disticha*) were collected.

Two *Zanclaea* species (*Zanclaea* sp. 1 and *Zanclaea* sp. 2) was particularly interesting. After the collection, these two colonies and the just released medusae were cultured in laboratory for several days in order to ob-

serve their morphology and the polyp ethology.

*Zanclaea* sp. 1 was collected on mangrove roots at Cat Cay (Pelican Cays) growing on the bryozoan *Rinchozoon* sp. The polymorphic colony presents three kind of polyps: gastrozooids with trophic functions, gonozooids from which medusae arise and dactylozooids defending the colony. The reticular hydrorhiza creeps on the bryozoan skeleton and is covered by perisarc forming a small cap at the base of the polyps. The gastrozooids are 1.5 mm in height and are characterized by 6-8 oral capitate tentacles surrounding a white hypostome and 35-40 aboral capitate tentacles. Gonozooids, very small (about 0.4 mm in height), show 4-5 oral capitate tentacles and about 10 aboral capitate tentacles. Dactylozooids are very contractile and completely deprived of tentacles. Medusae buds arise from the middle portion of the gonozooids in clusters of 3-4. During the medusae maturation, the gonozooids may reduce to blastostyles. Cnidome: macrobasic euryteles (20-21 x 10 µm undischarged capsule; 20 x 7.5-10 µm discharged capsule) are distributed on the hypostome, abundant on the dactylozooids and scattered on the hydrorhiza; large stenoteles (12-12.5 x 10-10.5 µm undischarged capsule) are rare on the tentacle capitation and abundant on dactylozooids and hydrorhiza; small stenoteles (6-6.5 x 5 µm undischarged capsule) abundant on the tentacle capitations, dactylozooids, and hydrorhiza, rare on the hydranth.

The just released medusa shows a bell shaped umbrella, about 450 µm in diameter. The cylindrical manubrium and the four perradial exumbrellar pouches reaching 1/3 of the subumbrellar cavity. Two opposite orange coloured perradial bulbs with a long tentacle each, two opposite non-tentacled perradial bulbs. Tentacles with 30-40 cnidophores (25 x 15 µm) guitar shaped, covered by long flagella. Cnidome: macrobasic euryteles (4x6 µm undischarged capsule), 2-3 per each cnidophore; large stenoteles (10 x 9-10 µm undischarged capsule) 15-19 in each exumbrellar pouches; small stenoteles (5x5 µm undischarged capsule) in the distal portion of the manubrium; rare isorhizas (7 x 5 µm) scattered on the umbrella. This is probably a new species.

*Zanclaea* sp. 2 was collected on mangrove roots at South Eastern Cay (Pelican Cays) growing on the orange coloured bryozoan *Trematoecia aviculifera*. The reticular hydrorhiza creeps on the bryozoan skeleton and is covered by perisarc forming a small cap at the base of the polyps and by a thin calcareous layer secreted by the bryozoan. The non-polymorphic colony

is constituted by just one type of polyp showing 4-5 oral capitate tentacles surrounding a white hypostome and 20-25 aboral capitate tentacles. Medusa buds arise from the proximal portion of the polyps in clusters of 2-3. Cnidome: large stenoteles (15-17 x 12-15  $\mu\text{m}$  undischarged capsule) are abundant on the tentacle capitulation and rare on the hydranth; small stenoteles (7.5 x 5  $\mu\text{m}$  undischarged capsule) abundant on the tentacle capitulation, rare on the hydranth.

The just released medusa shows a bell shaped umbrella, about 500  $\mu\text{m}$  in diameter. The cylindrical manubrium reaching 1/4 of the subumbrellar cavity. Four perradial exumbrellar pouches are reduced to nematocyst knobs. Two opposite white coloured perradial bulbs with a long tentacle each, two opposite white coloured non-tentacled perradial bulbs. Tentacles with 15-28 ogival cnidophores (30-35 x 10-15  $\mu\text{m}$ ), covered by long flagella. Cnidome: macrobasic euryteles (9 x 7  $\mu\text{m}$  undischarged capsule; 7 x 6  $\mu\text{m}$  dischaged capsule, shaft about 50  $\mu\text{m}$ ), 2-3 per each cnidophore; large stenoteles (13 x 12  $\mu\text{m}$  undischarged capsule) 2-5 in each exumbrellar pouches; small stenoteles (7.5 x 5  $\mu\text{m}$  undischarged capsule) in the distal portion of the manubrium; isorhizas (5 x 5  $\mu\text{m}$ ) abundant, scattered on the umbrella.

The observation at the stereomicroscope of the living colony of *Zanclaea* sp. 2 revealed different trophic strategies performed by the polyps.

## Annelida

**A new sponge-associated species, *Syllis mayeri* (Polychaeta: Syllidae), with a discussion of the status of *S. armillaris* (Muller, 1776)**

*L. Musco & A. Giangrande*

In the framework of a study carried out along the Belizean coast, a new species of syllid, *Syllis mayeri* was found harboured by the sponge *Ircinia strobilina*. The description of this new taxon is compared to similar congeneric species and to Mediterranean material from the southern Italian coast ascribed to *S. armillaris*.

The new species is characterized by some stout compound chaeta with psiloid appearance, which may be used for attaching to the host. Other diagnostic features are antennae, tentacular cirri, and dorsal cirri of

the anterior portion of the body that are longer than in similar, compared species. Moreover, the mid-body dorsal cirri are typically spindle-shaped. A discussion



*Syllis mayeri* Musco & Giangrande, 2005

on the status of some sponge-associated taxa from tropical and subtropical seas attributed to the so-called cosmopolitan *S. armillaris* is also given.

## Bryozoa

### Cupuladriids in Carrie Bow?

*A. O'Dea*

Our Smithsonian Marine Science Network funded search for cupuladriids at Carrie Bow Cay this January was unfortunately unsuccessful. We made a total of 20 dives covering all the possible environments that exist close to Carrie Bow where cupuladriids could potentially occur. We sampled all areas of the muddy innerchannel leading up into the sand banks that flank the reef. We surveyed the sandy-muddy patches close to the mainland. We dove the deeper patches of muddy sand around Tobacco and Pelican Cays and we searched many of the cuts in the reef wall that collect sand and are free from coral growth. We were very lucky with good weather for diving and there was nowhere we were unable to survey within the limits of the small boats at Carrie Bow. I can confidently say that close to Carrie Bow, cupuladriids do not occur, probably because of the very close association of the sandy areas with reef and sea grass, and the inner channels' very fine mud. If another search for cupuladriids be made, I would recommend extending the survey further north

in Belize where sand banks collect in the inner channel. This project would require a larger boat than available at the Carrie Bow laboratory.

## Crustacea

### Taxonomy and population structure of commensal leucothoid amphipods

J. D. Thomas & K. Klebba

*Taxonomic and ecological studies:* James Thomas and graduate student Kris Klebba continued their study of commensal amphipods at Carrie Bow in September. They expanded their sampling of potential sponge and bivalve hosts to an additional 12 stations in the vicinity of Carrie Bow and Pelican Cays. A number of new records for species-host relationships were recorded from a variety of sponge, ascidian, and bivalve mollusk species. All specimens were sorted, and processed for molecular genetic analyses. The September 2005 collections produced two new species, and, possibly, one new genus of leucothoid amphipods. These data, combined with previous research, led to the description of 10 new leucothoid amphipod species from the region under this study. Taxonomic descriptions of eight of these taxa are currently in press. When feasible, species names were taken from the Garifuna dialect, the black Caribe language.

Belize collections have allowed the assignment of new host data to both new and leucothoid species. One new species, currently in press, was previously thought to inhabit a single bivalve mollusk host, the rough file clam *Lima scabra* Born, 1778. The study has now recorded four new bivalve mollusk hosts for this species (*Americardia media* Linnaeus, 1758; *Lithopaga antillarum* Orbigny, 1846; *Lucina pennsylvanica* Linnaeus, 1758; and *Mytilopsis leucopheata* Conrad, 1831).

Distinct patterns of host and commensal occurrences were documented from three habitats in the Carrie Bow Cay Region. The Pelican Cays region was most diverse, followed next by patch reefs in sand bore habitats. Shallow and deep fore reef localities were less diverse but yielded occurrences and taxa often found nowhere else in the study area.

*Population structure:* In the molecular genetics part of this study, our initial focus was on exploring connectivity among geographic isolates of commensal

leucothoid amphipod species inside the branching vase sponge *Callyspongia vaginalis*. This sponge is common in both Florida Keys and Belize marine environments. We used variation in a 422 bp segment of the mitochondrial cytochrome oxidase subunit I (COI) gene to infer patterns of gene flow among six geographic locations in South Florida and the Caribbean, including 3 sub-localities in Belize, that is, Glover's Reef, Blue Ground Range, and Pelican Cays. A total of 42 haplotypes were identified from the 171 animals sampled.

Leucothoid amphipods exhibited highly restricted levels of gene flow between the Florida reefs and Belize Reefs. The very large ST value (0.98) and genetic distance between the Florida and Glover's Reef populations for *L. ashleyi* n. sp. (79 mutational steps; 20.3%) indicates a substantial barrier to gene flow between these locations.

Results from mitochondrial DNA sequence analyses indicate considerable genetic connectivity between northern and southern portions of the Florida reef system, but a high degree of phylogeographic disjunction between Florida and Belize reefs for all three species independent of reproductive strategy.

## Pisces

### The mangrove forest fish communities of the Belize Cays: Part III

S. Taylor, C. McIvor, W. P. Davis & E. Reyjer

We have completed a third survey (April 28-May 10, 2005) of fishes utilizing the mangroves of the Belize cays, with a focus on Twin Cays. This effort was targeted to achieve the needed 'low water' seasonal comparison. Two personnel were on site for 4 weeks, and two others for only 2 weeks. A slight revision to methodology used in the first 2 surveys was implemented, with the addition of some additional gears, discontinuation of some gears, and termination of the visual surveys comparing cut vs. un-cut shorelines. We have now completed collection of representative fishes, crustaceans and mollusks for stable isotope analysis (by others) for the Trophic Network Analysis.

The anticipated low-water conditions were present and we did not observe inundation of transition mangrove habitats, so most sampling effort was aimed at fringe, internal channel, sinkhole and dwarf red-man-

grove-pond habitats. We have continued with the same methodology and gear-types, with the exception of throw-trap collections, but added a gill net and a series of larger traps (“oval traps”), which were fished in deeper waters of Twin Cays Main Channel. In addition, we fished the fyke net in a mangrove channel fronting directly on Twin Cays Channel, which was very productive, with large catches including several new species. The ‘sinkhole’ habitat was also specifically targeted this time, with a new species added from here. In addition, deployment of traps within undercut fringe shorelines along the Twin Cays Main Channel revealed several new species. An opportune advection of a large amount of *Sargassum* into fringe habitats resulted in the collection of several new species (fishes and crustaceans), which were collected by dipnetting directly in the *Sargassum*. We completed 973 trap-nights with all gear types, collecting 3,341 fishes and 839 macro-crustaceans. Physical data (salinity/temperature/DO) were collected daily at all sampling stations. Visual surveys for fish identification were conducted around Twin Cays, Peter Douglas Cay and the Pelican Cays, for a total of 42.5 person-hours. Between our captured specimens and species identified in visual surveys, we have now confirmed at least 135 species of fishes present in mangrove systems of the Belize Cays, and addition of 25 species since the 2004 survey. Several additional species have yet to be identified. We have processed another 180 samples (representing 32 species) for stable isotope analysis, and these specimens have been turned over to others for analysis for the Trophic Network Analysis. This analysis is complete but data have yet to be interpreted.

This sampling episode completes a comprehensive picture of the fish communities of the Belize Cays. We have also confirmed significant differences between the fish communities of mangrove shorelines where the trees are being cut vs. un-cut shorelines. Stable isotope data, when analyzed, should provide much needed insight into trophic networks within Belizean man-

groves, possibly answering some long-standing questions on this issue. However, we speculate that a significant fish community may be ‘missing’ from our collections, so-called ‘cryptic’ species that can only be taken with ichthyocides. The intricate nature of the substrates and prop root habitats probably prevents collection of these missing species by conventional means.



Taylor’s “crew” with boat and gear.

## An assessment of ichthyofaunal assemblages within the mangal of the Belizean offshore cays

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

We assessed ichthyofaunal diversity within Belizean offshore cay mangroves during three sampling events (2003-2005). A variety of sampling gears (n=10 types nets/traps) were deployed in pre-defined habitats: red-mangrove fringe, transition, dwarf red, internal creek, pond, sinkhole, and deep creek. A total of 2,586 gear sets were completed and 8,131 individuals collected, comprising 75 species. The most abundant species was *Poecilia orri*, followed by *Gerres cinereus* and *Gambusia yucatana*. However, 38.2% of gear sets (primarily small traps) had no catch. Data analysis for the different sub-habitats is continuing, but our data reinforce the difficulty of collecting in the mangal. In addition, we conducted extensive visual (snorkel) surveys around the fringe at a number of cays, tallying an additional 67 species. An overall total of 142 species from 55 families have therefore been documented from mangroves of the cays, a figure among the highest reported for oceanic mangroves in this biogeographic realm.

## Molecular systematics of a marine species flock, Caribbean hamlets

*E. Bermingham, M.A. McCartney, O. Puebla & L. Whitman*

The project started in September 2003 carried out by Oscar Puebla under the supervision of Bermingham, and in collaboration with McCartney and Whitman. Data analysis was performed at McGill University (Montreal, Canada) in fall 2003 and 2004. Computer simulations are presently in progress at McGill.

Field work was carried out in Carrie Bow Cay (Belize), Bocas del Toro

(Panama), Kuna Yala (Panama) and Barbados, taking advantage of Smithsonian Institution and McGill University facilities there. A total of 1,346 samples from eight *Hypoplectrus* color morphs were collected, photographed, and processed. A tissue sample of each fish was conserved for genetic analysis and all collected fishes were conserved in the Bermingham laboratory's fish collection at the Smithsonian Tropical Research Institute.

For genetics, five new microsatellite loci were developed in addition to the 5 previously existing loci (McCartney et al. 2003). A total of 428 samples, including seven color morphs, were genotyped at the 10 microsatellite loci. Clear genetic differences between *Hypoplectrus* color morphs were demonstrated for the first time. Genetic differences between populations were evidenced, as well for several color morphs, showing the potential for genetic differentiation within the Caribbean despite a planktonic larval stage.

A total of 179 transects covering 400 m<sup>2</sup> each were surveyed. Densities of all color morphs were assessed at each site. Transects were sampled in different sites of the reefs in order to test for ecological differences between color morphs. Such differences were put into evidence at several sites.

All collected fishes were measured in order to test for standard length differences between color morphs. Such differences were determined at several sites.

Dusk dives were made at all sites to observe spawnings and record the association of color morphs during spawnings. In all, 249 out of the 252 observed



Caribbean hamlet, genus *Hypoplectrus*.

spawnings were between the same color morphs, indicating strong assortative mating.

Computer simulations are presently being developed in an effort to understand the spatial dynamics of *Hypoplectrus* communities at the Caribbean scale.

More field work will be carried out in 2006 in Honduras, Nicaragua and Venezuela. More samples will be genotyped in and DNA sequence analysis will be performed to complement microsatellite data. Computer simulations will be significantly developed in 2006 and 2007.

## Chaenopsid blennies systematic and ecological research

*J. Tyler & D. Tyler*

In March 2005, we collected a few more specimens of a new species of *Emblemariopsis*. All were juveniles or young adults, and we still need some larger adults before we can properly describe this new species, possibly the last of new species to be found in the shallow waters around Carrie Bow and the Pelicans Cays.

During our sampling in 2005 we also found that the habitat of *E. ruetzleri* at Carrie Bow had shifted from holes in dead parts of large mound corals where they were observed previously to holes in small, low, live mustard corals. This was most surprising because for the past four years of sampling we have only found *E.*

*ruetzleri* in holes in dead parts of corals. In 2006 we wish to continue our sampling (with quinaldine) of all species of *Emblemariopsis* to see if there have been other shifts in microhabitat in other species and whether *E. ruetzleri* has returned to holes in dead versus live corals. These results will be incorporated into the paper that will describe the new species of *Emblemariopsis* for which we still seek larger adult specimens before completing the description of the new species.

## Paleobiology and Microevolution

### Ecological speciation in a mangrove-reef sponge

S. Duran & K. Rützler

*Chondrilla* cf. *nucula* Schmidt (Chondrillidae, Chondrosida) is a common Caribbean sponge that grows in both reef and mangrove swamp habitats. On coral reefs, *C. cf. nucula* encrusts rocks or hard corals in thin sheets of golden green coloration. In mangrove swamps it grows enveloping mangrove roots in a thick lobate mass of grey-green to chestnut brown coloration. Despite these morphological differences in growth form and coloration, the spicules and internal structure of both morphotypes are very similar, suggesting that they are different growth forms of the same species.

Our results showed a mosaic distribution of haplotypes in mitochondrial DNA sequences. Out of a total of 12 haplotypes, 5 are unique to mangrove habitats while another 5 are only found in reef habitats. Only two haplotypes were shared between the mangrove and reef habitats, by one individual each, suggesting very high levels of reproductive isolation between mangrove and reef populations. Analysis of the molecular variance statistically supports high genetic isolation between the two habitats ( $p < 0.05$ ).

Our data suggest that strong ecological barriers exist between reefs and mangrove swamps. Because of the high haplotype relatedness of the two morphotypes and the fact that at least two haplotypes are shared between habitats (hap10 and hap4), we suggest designating the two morphotypes as formae of the same species (*mangle* and *hermatypica*), subspecific entities commonly used in sponge systematics.

### Speciation in coral reef fishes: Intraspecific niche evolution and the generation of biodiversity in wrasses (*Halichoeres*)

P. Wainwright

I last visited Carrie Bow Cay in early July of 2005, with Michael Alfaro. We made significant headway in a study of speciation in fishes of the genus *Halichoeres* (wrasses). We collected detailed data on habitat use on all eight species that occur in Belize and we collected specimens for an in-depth analysis of habitat specialization and within-species diversification in *H. bivittatus*. Specimens were collected from the three major habitats occupied by this species and muscle samples are being used to measure RNA/DNA ratios and RNA/protein ratios as estimates of individual growth rates (fitness) to be correlated with a series of musculoskeletal traits that we are measuring from the formalin-preserved and cleared and stained specimens. This data set will allow us to measure the strength of natural selection acting on feeding characters in these habitats and help us understand how strong a diversifying force habitat adaptation is in this species.

### Two lineages of *Halichoeres bivittatus*, spawning of hamlets species, and status of *H. socialis*

L. Rocha



*Halichoeres socialis*.

The main objective of our stay at Carrie Bow was to collect juvenile and adult *Halichoeres bivittatus*. This species of fish has two deeply divergent genetic groups, one restricted to subtropical waters of Florida and Bermuda and the other in tropical locations across

the wider Caribbean. In my project I proposed to study if those two lineages are maintained by self-recruitment or ecological constraints. These alternative hypotheses can be differentiated by comparing the genetic architecture of adult and juvenile populations across the species' entire geographic range. If adults and juveniles are genetically similar, the self recruitment hypothesis is favored. However, if adults are similar from year to year but different from the juveniles of the same location, this indicates that natural selection is generating the observed genetic partitions. Preliminary results from collections carried out last year indicate that natural selection plays a large role.

Juvenile and adult fishes were collected with hand nets while scuba diving or snorkeling. Whenever possible, only a piece of the fin was removed, and the fish was returned alive to the Ocean. The tissue was preserved and taken back to STRI for analysis.

A second objective of the field trip was to observe and photograph the spawning of hamlet species (*Hypoplectrus* spp.). Hamlets are a group of fishes composed by about 10 species that share all morphological characters except color. Because of this attribute, they have been the subject of several recent genetic studies that tried to find genetic differences between the species. No genetic difference was observed, and a general conclusion was that all color-defined species were formed very recently in the evolutionary time. Even

though there is no genetic difference, the species recognize themselves as different and mate mostly with others of the same color. We observed spawning during our entire stay, and only saw spawning between specimens of the same color.

A third objective was to check on the status of the populations of *Halichoeres socialis*. This wrasse was recently described, and as far as we know, it is endemic to the mangrove islands on Belize's barrier reef.

Its very restricted geographic distribution makes this species extremely vulnerable. We did one trip to the Pelican Cays, where *H. socialis* is relatively abundant, and I photographed for the first time the terminal male phase of the species. During the original scientific description, the authors could not find terminal males.

## The effects of biogeographic variation in recruitment on the ability of species to invade coastal marine ecosystems

*R. Osman, G. Ruiz, A. Hines & R. Whitlatch*

We contend that the basis of recruitment patterns is the availability of a critical resource, space, and how it is affected by spatial and temporal variations in the rates of settlement, growth, and mortality of all species. It is the variation in these processes that we see affecting diversity patterns as well as the potential for



Spawning of hamlet, *Hypoplectrus* sp.

communities to be invaded. Therefore, the long-term goals of our research are determining how the availability of space influences regional and local variation in diversity recruitment, growth, and mortality rates of native species, how communities may constrain the success of invaders through their influence on resource availability, and if and how invading species differ from natives in their abilities to capture

this critical resource.

To date most laboratory analyses of recruitment samples are complete through December 2005 for all sites except Belize. Data have been entered into spreadsheets for the Florida, Long Island Sound, Chesapeake Bay and San Francisco Bay sites.

We now concentrate on the live Florida samples. Prior to analyzing preserved samples from tropical Atlantic sites, we want to familiarize ourselves with the identification of what we hope is a similar fauna in Florida. Because most of our taxonomic identifications are made on very small new recruits and juveniles, we must often use subtle or incompletely developed characteristics to identify taxa, which is best done with live-material.

## Reproductive and Developmental Biology

### Molecular signatures for sex in the Placozoa

*A.Y. Signorovitch, S.L. Dellaporta & L.W. Buss*

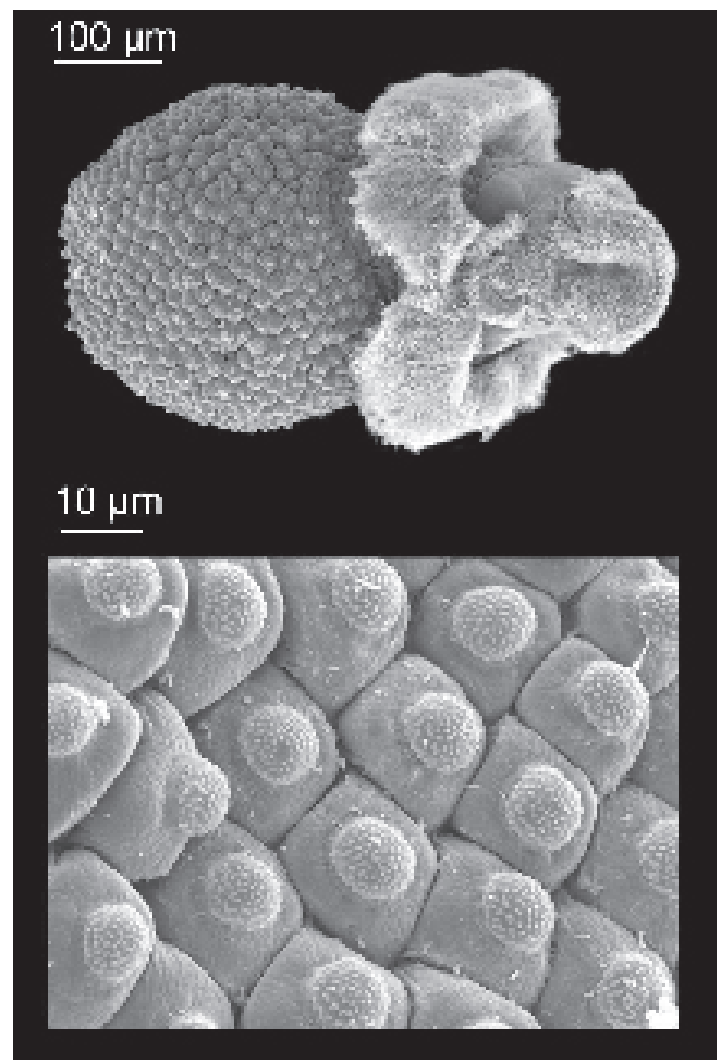
Placozoans, the simplest free-living animals, have never been observed to reproduce sexually. Here we describe molecular evidence for sexual reproduction within one clade of the Placozoa. In a population sample of ten individuals, within-individual and overall nucleotide diversity were similar to each other and consistent with levels observed in sexually reproducing species. Intergenic recombination as well as the sharing of alleles between hetero- and homozygous individuals was also observed. These hallmarks of sexual reproduction support the presence of a sexual phase in the placozoan life cycle and, for the first time, establish that sexual reproduction is indeed present in this phylum.

### Metamorphosis and population structure in Sipuncula

*A. Schulze, M.E. Rice & G.Y. Kawauchi*

During our stay in April, we performed daily plankton tows outside of the reef crest to the east of the Carrie Bow Cay field station. We recovered approxi-

mately 10 different morphotypes of sipunculan pelagosphera larvae, distinguished by size, body shape, coloration and structure of the body wall. The most common larval type was the larva of *Apionsoma misakianum* with a distinct black head and white body. Up to 350 specimens were collected in a single tow. Sipuncula undergo a gradual metamorphosis from the pelagosphera to the juvenile stage. Several reagents (potassium chloride, cesium chloride, isobutyl methylxanthine, L-dopa), known to induce metamorphosis in a variety of invertebrate larvae, were tested on the *Apionsoma* larvae. Although results were not consistent, the most successful agent appeared to be potassium chloride at concentrations of 20-30 mM. In one experiment, 25% of the larvae showed signs of metamorphosis after nine days as opposed to none in the control. Other larval



Pelagosphera stage of *Apionsoma* larva; detail (below).

types were photographed and fixed for morphological and molecular studies. We are currently documenting sipunculan larval diversity with color photographs and scanning electron microscopy. In addition, we are using



DNA sequence data to match larval morphotypes with a database of adult sequences and thus identify the larvae to species.

A second objective of our visit to Carrie Bow Cay was to sample adult sipunculan specimens of the species *Golfingia elongata* from the mangrove root mats at Twin Cays. *G. elongata* is an extremely widespread species, known from arctic to tropical waters from intertidal to 590 m depth. It is possible that it actually represents a species complex. During several trips to Twin Cays, we collected 23 individuals which were fixed in ethanol for future molecular work.

## Genetic and morphological identification of fish larvae at Carrie Bow Cay, Belize

*C.C. Baldwin, D.G. Smith, J.H. Mounts & L. Weigt*

This report summarizes the results of our expedition to Carrie Bow Cay in April-May, 2005, the eighth in our ongoing study of the larvae of shore fishes of Carrie Bow Cay, Belize. This was our first spring-time visit to the island; previous trips were in June, July, August-September, October-November, and February-March. As in our previous visits, we collected larval fishes in a plankton net suspended off the dock on the ocean side of the island. The larvae were then photographed, some of them transferred to rearing cages, the rest preserved. Tissue sample for DNA analysis, begun on our visit last year, was continued, with the objective of obtaining samples from as many different fish species as possible. This was done partly to facilitate identification of larvae by matching profiles from larvae and adults, and partly as a contribution to the larger Barcode of Life project. This year, we devoted more time to collecting adult fishes, especially those small species that live cryptically within the reef structure. We obtained permission to use small quantities of rotenone, and this was critical in enabling us to attain our goals. In addition we used quinaldine, spears, hook and line, and a small seine.

Periods of light wind early in the field period allowed us to make several collections on the outer ridge. Later the trade winds set in, making it difficult to work on the exposed ocean side of the island. The wind, however, was favorable for collecting larvae, as it enhanced the current flowing over the reef crest. We had a dark moon for most of the period; this is ideal for collecting larvae at night. Collections were excellent. Although each collection yields at least some new

(for the project) taxa, we found the species composition of the larvae similar to those we collected previously during the summer period. The catches were characterized by large numbers of the goby *Ctenogobius saepepallens*, the dominant larva during the summer. One night when the wind was calm, we took the boat out and towed the plankton net outside the reef crest. Our catch was essentially the same in composition as the catches we normally get in the stationary plankton net. This demonstrates that our stationary net is catching a representative sample of the larvae that occur just outside the reef; that is, there is nothing out there that is not carried in over the reef crest by the current. On future trips, weather permitting, we will try to sample farther out to see if the composition changes over and beyond the outer ridge.

We obtained 507 DNA samples, many of which were from the cryptic fishes that we were able to collect with rotenone. All sampled specimens were photographed and almost all were vouchered and returned to the Smithsonian. The DNA samples will be analyzed this summer at the LAB. We expect these samples to provide evidence for identifying a number of problematic larvae and for sorting out some confusing species or species complexes. The cardinalfishes (Apogonidae) have proved especially vexing, and the DNA profiles may allow us to sort out species in that family.

We upgraded to a new digital camera this year, a 12-megapixel Fujifilm S3. This camera gives us a much finer resolution, especially in such difficult-to-see features as fin rays. A rechargeable battery pack was used, replacing the disposable AA dry cells.

## Patterns of occurrence of transitioning pelagic fish larvae carried across the forereef of Carrie Bow Cay, Belize

*K. Cole*

A total of 5,954 late-stage fish larvae (i.e. ready to settle out from the plankton) were collected on the forereef of Carrie Bow Cay during 2005. Using a standing plankton net deployed over a total of 69 h, 120 collections were made over three sampling periods: April; June; and December. Fifty-four percent of the collected larvae could be identified to species and an additional 12% were identified to a described type within a genus. The taxon most represented in these samples was the order Gobioidae. This taxon includes gobies, eleotrids and microdesmids, and constituted 47% of all larvae

collected in 2005 for this project. A total of 1,837 individuals, representing 31% of the entire years' collection total, consisted of a single gobiid species, *Ctenogobius saepepallens*. As this species is not visibly abundant in reef or lagoonal habitats in the vicinity of Carrie Bow

terior of Twin Cays, Belize. On the edge of these ponds, thick mats of flocculent detrital material (floc) derived in large part from senescent *Batophora* accumulate. This "compost" has been shown to stimulate the growth of the dwarf red mangroves of this area, by providing

phosphorus to this P-limited community (Feller et al. 2002), and appeared to increase colonization of artificial substrates by living *Batophora*. While photosynthesis and growth of some tropical macroalgae species are limited by the amount of available nitrogen, phosphorus limits the productivity of a number of others (Lapointe et al. 1987, Littler et al. 1988, McGlathery et al. 1992). Little is known about the nutrient and photophysiology of *Batophora*, so we undertook a series of experiments to investigate whether short term nutrient additions could improve maximum photosynthetic rates and growth rates. We also began a field investigation of the factors that lead to establishment of new *Batophora* patches.

Investigations were carried out in August 2003 and January 2004, in order to compare effects during the anticipated seasonal periods of high and low photosynthesis (Morrison 1984). Samples were collected several times from several different locations within Twin Cays, representing a range of water depths, flushing regimes, and natural *Batophora* abundance. At Aanderaa Flats, water depth is generally more shallow than at West Pond, and *Batophora* abundance is very patchy. In contrast, West Pond has some areas of very high *Batophora* abundance, and few dwarf mangroves, and other areas of higher dwarf mangrove and relatively lower *Batophora*. As dwarf mangrove density increases, *Batophora* abundance declines to zero (three transects were conducted in Jan. 2004). At Carrie Bow, samples were cleaned of detritus and placed in shaded aquaria along with additions of  $\text{NH}_3\text{Cl}$  and/or  $\text{NaH}_2\text{PO}_4$  to final concentrations of  $10 \mu\text{M N}$ ,  $1 \mu\text{M P}$ . Incubations lasted 24 h, from dawn to dawn. In August, additions were made only at the beginning of the experiment, while in January, additions were made every 4 h. After 24 h, samples were brought indoors, moved to ambient seawater (no nutrient addition), and the photosynthetic performance was monitored for the next 24 h by measuring  $\text{O}_2$  evolution or active chlorophyll fluorescence. Some samples were selected for measurements of growth over the next week in ambient seawater.

Addition of N led to no change in the tissue N



*Myrophus punctatus*, Family Ophichthyidae, Snake Eels.

Cay, the disproportionately large contribution of this one species to the ichthyofauna is surprising. The constancy of occurrence of *C. saepepallens* larvae in all three sampling periods suggests that this species may have a long, or continuous spawning season, while their numerical abundance in the ichthyofauna implies that this species plays a major role in energy flow in coastal systems of Belize. In contrast, the variation in proportional occurrence of fish larvae of most other known species suggests that for the majority of coastal fishes in Belizean waters, reproductive output is more episodic, and occurs during discrete breeding season(s).

## Ecology, Population Dynamics, and Ecophysiology

### Nutrient physiology of the macroalga *Batophora oerstedii*

L. Franklin

The green macroalga *Batophora oerstedii* occurs in dense lawns in quiet mangrove ponds in the in-

content. In contrast, incubation in 1  $\mu\text{M}$  P or 10  $\mu\text{M}$  N plus 1  $\mu\text{M}$  P led to a two- and three-fold increase in tissue P, in August and January. This suggested that the *Batophora* was indeed P-limited. However, no differences in photosynthetic performance among treatments was observed by either measure. There were no differences in maximum photosynthetic rates among sites or between seasons in these experiments. While samples did grow during the subsequent week, there was no difference in growth rate among treatments. In January, a high proportion (>90 %) of the samples became reproductive during the growth experiment period, and no growth occurred in these cases. This occurred regardless of nutrient treatment.

In August, a field experiment was deployed to test the effect of detrital material on the establishment of new *Batophora* patches. Six treatments were placed in 13 cm x 22 cm bags made from window screen: detrital biomass in form of: (1) *Syringodium* (seagrass) wrack, (2) coarse woody mangrove debris, (3) dried *Rhizophora mangle* leaves, (4) floc from the edge of West Pond, as well as two controls: (5) 1 cm x 1 cm diameter PVC pipe pieces and (6) empty bag. Ten plots were set up in the center of the open area of West Pond where there were few dwarf mangroves, but a relatively abundant *Batophora* lawn on the bottom. Bags were held perpendicular to the bottom, and orientated to receive early morning and late afternoon sun on the front and back flat surface. Tissue analysis of the *Syringodium* wrack indicated a P content similar to that of the floc, coarse woody debris and *R. mangle* leaves whereas yielded only about half.

Evidence from previous mangrove fertilization experiments (Feller *et al.*, unpublished) indicated that *Batophora* colonizes window screen material preferentially in the presence of floc, as compared to no floc. By January 2004, there was no evidence of colonization of the bags, though reproductive thalli were observed in the field in both August and January. The bags were periodically examined by members of Feller's lab. Recently, I have learned that there is good colonization of at least one of the treatments in each plot, prompting to return to collect these samples and have a tissue P assay performed on representative thalli to finish this work.

## Coral reef, seagrass meadow, and mangrove sponge distribution and diversity patterns: Influence of predators

J. Wulff

Sponges of 45 species, representing 10 demersal sponge orders and 3 distinct but linked habitats (coral reefs, seagrass meadows, and mangrove roots), have been presented to predators typical of each of these habitats in order to disentangle patterns in palatability that are related to habitat vs. sponge taxon vs. predator taxon. All experiments were done in the field, in situations that were as natural as possible. Pieces of living sponges, with surfaces fully healed before exposure to predators, were made available to angelfishes, parrotfishes, trunkfishes, and starfish for 24 hr periods. The hypothesis that many sponge species that are well defended against predators with which they coexist are palatable to predators with which they do not normally coexist is well supported by the data. Coexistence with predators was the most reliable predictor of adequate defenses against those predators, but some patterns related to sponge higher taxon (from genus to order), over-ride habitat patterns. Predators in the same family were generally similar in which sponges they were able to consume.

Although this study has demonstrated that predators exert substantial control on habitat distributions of many Caribbean sponge species, other aspects of the environment are clearly also important. The next step in learning how sponge species are sorted into habitats is to determine what factors control species that are immune to predators, how competition and abiotic factors interact with predation to influence habitat distribution, and how sponge species might be able to alter their habitat distribution patterns if predators continue to be lost from tropical coastal marine habitats. To this end, I have initiated a grand reciprocal transplant study involving dozens of sponge species that are typical of mangroves, seagrass meadows, or coral reefs. I am comparing growth and survival of each species in each habitat, hoping to elucidate a hierarchy of factors that control habitat distributions of these common, ecologically important invertebrates.

## Impact of environmental stress on reef and mangrove sponges

K. Rützler, S. Duran & C. Piantoni

Sponges (Porifera) in mangroves have adapted to a wide range of environmental parameters except for extended periods of exposure to freshwater or air. Many marine mangrove islands are located in the shallow backwaters of coral reefs in Belize and elsewhere in the Caribbean and have a mean tidal range of only 15 cm. They are densely populated by sponges, mostly attached to subtidal red-mangrove stilt roots and peat banks lining tidal channels. Some species are endemic to mangroves, others are immigrants from nearby reefs. Mangrove endemics endure environmental hardships, such as occasional exposure to air during spring tides,

temperature and salinity extremes, fine sediments, even burial in detritus. Reef immigrants into mangroves enjoy protection from spongivores that do not stray into the swamp but they eventually succumb to environmental stress. There is evidence exemplified by the common demosponge *Chondrilla* aff. *nucula*, that sponges flourishing in both mangrove and reef habitats may develop separate ecologically specialized and reproductively isolated populations. Such processes can lead to genetic modifications and thus serve as mechanisms for ecological speciation. Because *Chondrilla nucula* Schmidt was first described from the Mediterranean Sea, it was long suspected that the western Atlantic population may be a separate species. New morphological and molecular evidence prompt us describing it under a new name, with two ecological forms, one from mangroves and

lagoons, and the other from open reefs.

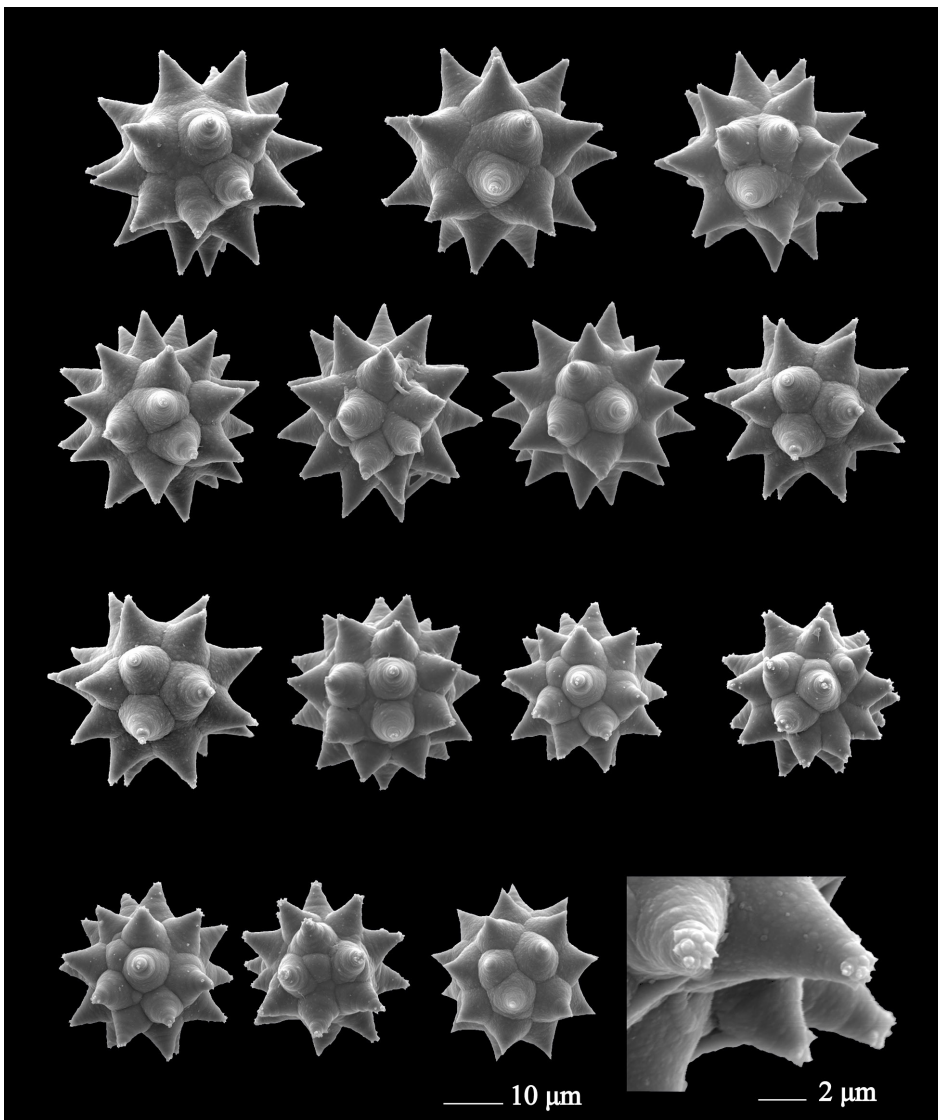
### Community dynamics on mangrove roots, and the morphological strategies of mangrove-inhabiting sponge species

J. Wulff

As tempting as it is to infer processes influencing distribution, abundance, and community dynamics from observed patterns, time series data often provide surprises, and this project is devoted to applying such surprises to development of our understanding of sponge ecology and evolution. This project about sponge-dominated communities on mangrove roots has 3 parts:

*Community dynamics:* I've completely censused (by drawing maps and measuring volume of every sponge) 10 mangrove roots or root clusters at each of two sites (Hidden Creek and Sponge Haven) at t=0 mo and t=15 mo, for comparison

with similar data from Bocas del Toro, Panama, and the Florida Keys. I hope to continue to re-do these censuses at about yearly intervals.



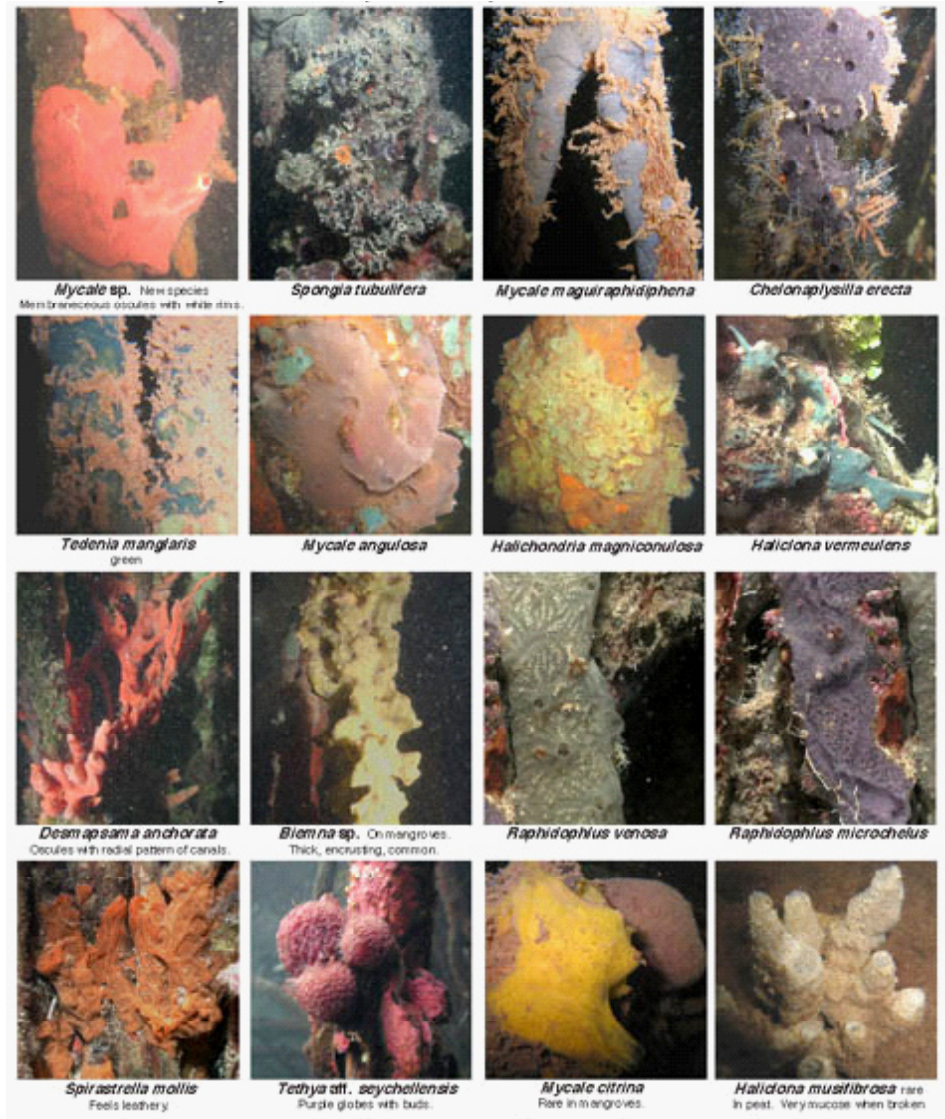
SEM micrographs of spicules (spherasters) of *Chondrilla caribensis*, mangrove form; inset: magnified tips of microspined rays.

*Community assembly:* By following settlement, growth, interactions, and mortality on pvc pipes suspended among the mangrove roots, I've been tracking the changes in species composition, diversity and cover over time. The first pipes I suspended are now indis-

tistinguishable from adjacent mangrove roots, and I'm following subsequently suspended pipes to see how deterministic the progress of community assembly is, or is not. Two more years of following the pipes that are now in the field should allow all of them to achieve their "climax" composition, if this is actually a meaningful concept for this community.

*Morphological strategies of mangrove sponges:* Different sponge species play very different roles in the mangrove root community, some of which can be predicted consistently by examining the morphology of the sponges. I'm accumulating data on the place in succession and microhabitat occupation of various species in order to compare among sites to see if the roles played by particular species or morphological types are consistent. Complicating the strategies that appear to be morphologically determined are differences among species in efficiency of colonization, ability to tolerate occasional emersion, and growth rates.

how surveys of species assemblages, loss or gain of species, relative abundance of habitat-specific and generalist species, and the spread of "pest" species can provide useful insights for evaluating environmental health of a particular area.



Common mangrove sponges (Bocas del Toro, Panama).

## Sponges as indicators of reef and mangrove health

M.C. Díaz & K. Rützler

Sponges are important components of Caribbean coral reef and subtidal marine mangrove communities. In both these ecosystems, sponges generally surpass the biological diversity and biomass of the other sessile zoobenthos, such as hexacorals, milleporids, octocorals, and ascidians. Our data and data of colleagues from reef and mangrove (roots and peat) locations in Belize and Panamá show that sponges are sensitive to environmental change and stress. We will demonstrate

Is coral recruitment controlled by coralline algal facilitator species? If so: What facilitates the facilitators?

R. Steneck, W. Adey & V. Paul

This report details the work completed at Carrie Bow Cay by Robert Steneck and Suzanne Arnold (graduate student) from 13-15 June, 2005, and by Suzanne Arnold and Jeanne Brown (graduate student) from 17-31 August, 2005. In June, a total of 250 terracotta coral-

settlement plates were affixed to the fore-reef adjacent to Carrie Bow and South Water cays. Fifty plates were placed at each of the four 10 m sites. One 5m site was selected where 50 plates were attached, half of which were covered with 1/4" wire mesh cages to mimic the effects of algal overgrowth. Though time did not allow for formal surveys, *Titanoderma prototypum* was not observed, so no corallines were collected for chemical extraction.

In August, at each site, transects were completed to quantify the abundance of live coral as an estimate of coral larval availability, and fish census and bite rate measures were completed to quantify the trophic structure of each reef. All settlement plates, top and bottom, were photographed underwater to quantify the rate of succession after two months. Additionally, half of the plates were analyzed under the microscope for newly settled corals and abundance of facilitator species. As expected, very little fouling was found to have occurred during this time, and only one newly settled coral was found at each of the following sites (i.e. 1 per 25 plates analyzed): Site 1 (10m depth-N 16 48.098, W 88 04.725), Site 3 (10m depth- N 16 48.769, W 88 04.686), Site 4 (10m depth- N 16 49.168, W 88 04.677), and Site 5 (5m depth- N 16 48.780, W 88 04.733), and two newly settled corals were found after analyzing 25 plates at Site 2 (10m depth- N 16 48.363, W 88 04.472). All recruits were of the genera *Porites* and *Agaricia*, and notably, *Titanoderma prototypum* was observed (<1%) on the undersides of several plates.

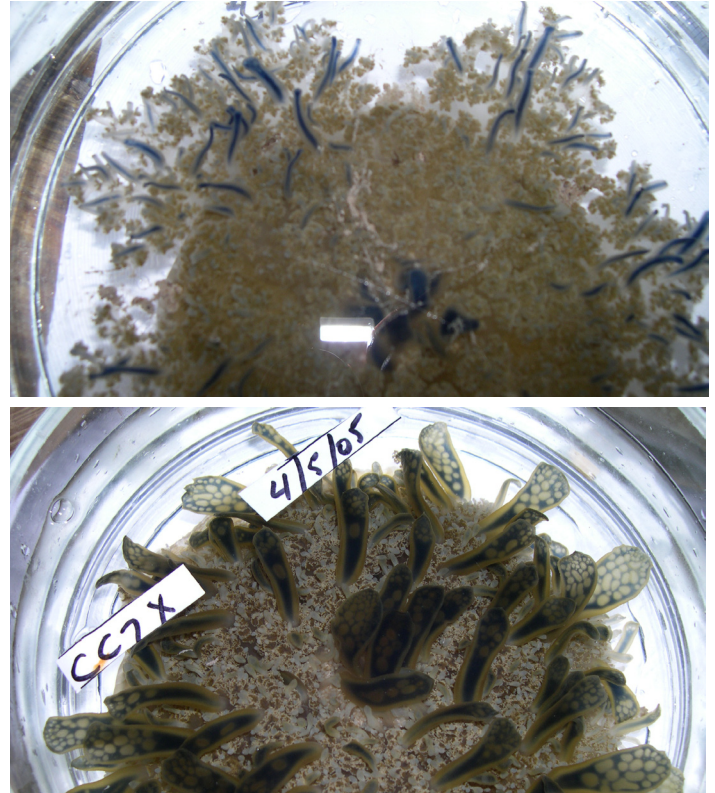
Also in August, species of coralline algae including *Titanoderma prototypum* were identified and collected for use in larval choice experiments with *Montastrea* and *Acropora cervicornis*. The details of this research are outlined in the report submitted by Dr. Paul.

## Abundance and distribution of the benthic tropical scyphozoan, *Cassiopea xamachana*, near Carrie Bow Cay, Belize

*D. Breitburg*

The goal of our pilot project begun in 2005 was to identify field sites and collect data on factors influencing the distribution and abundance of the little-studied tropical scyphozoan, *Cassiopea xamachana*, in the region of the Carrie Bow Cay field station. Becca Mor-

ris (gelatinous zooplankton technician at SERC) and I visited Carrie Bow Cay March 31 through April 6. We identified 12 potential sampling sites at Twin Cays with a range of *Cassiopea* densities. We collected animals at 4 sites, and photographed and described individuals to begin an assessment of morphological variation.



Morphological and color variation in *Cassiopea xamachana* from a single site at Twin Cays.

Our attempt to quantitatively sample *Cassiopea* was thwarted because our planned methods, which required handling animals while we were in the water, were not possible the severity of our response owing to this jellyfish's sting. It is not clear whether this particular population has a more severe sting than others (variation among populations has been documented, and both of us have handled *Cassiopea* before without any problems), or whether we have gotten sensitized from our exposure to the common Chesapeake Bay scyphomedusa, *Chrysaora quinquecirrha*. Nevertheless, we intend to collect data by using video recording during future research trips.

# Mangrove removal in the Belize cays: effects on fish assemblages in the intertidal and subtidal zones

D.S. Taylor, E.R. Reyjer, W.P. Davis & C.C. Mclvor

We investigated the effects of mangrove cutting on fish assemblages in Twin Cays, Belize, using two methods. We conducted extensive trapping with wire minnow traps within the intertidal zone in both disturbed and undisturbed fringing and transition (landward) mangrove forests. Catch rates were low - 638 individuals from 24 species in 523 trap nights. Cutting appeared to have no effect on species composition in either forest type (statistics using PRIMER, MDS followed by ANOSIM). Visual censuses from the subtidal area adjacent to fringing forests in two disturbed and two undisturbed sites gave different results. Here, observers recorded significantly more species and individuals in undisturbed sites (MDS, ANOSIM). Different results from the two methods (and habitat types) may be explained by two factors: a larger species pool in the subtidal habitat and more extensive mangrove clearing adjacent to the census sites.

## Species Interaction and Behavior

### A new ciliate/bacteria symbiosis: *Vorticella* sp. and sulfur bacteria on mangrove peat.

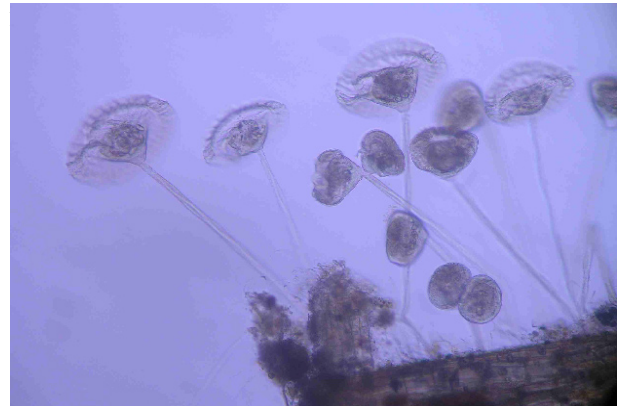
J.A. Ott

This study was inspired by discovering of another symbiotic ciliate (*Vorticella* sp.) cooccurring with *Zoothamnium niveum* on mangrove peat.

Quantitative samples were taken at Twin Cays from the surface of vertical peat walls along the main channel bisecting the island using cores with an area of 38 cm<sup>2</sup>. Under a stereo microscope 10 eyefields with a diameter of 1.2 cm (area 1.13 cm<sup>2</sup>) were examined on each sample and numbers of *Vorticella* sp individuals and colonies of the ciliate *Z. niveum* were counted. In a later stage peat pieces containing groups of *Z. niveum* colonies were selectively cut from the channel wall and observed under the stereo microscope for the presence of *Vorticella* sp.

Photos and videos were taken on. Leica S6D stereo microscope with a Nikon Coolpix digital camera and a Panasonic single chip camera and recorded digitally.

Specimens of *Vorticella* sp. were fixed in Bouin's flu-



Live *Vorticella* sp., enlarged ea. X130.

and Stieves fixative for taxonomic purposes and 100% ethylalcohol for sequencing of the 16S RNA gene of the symbiotic bacteria. Culture of *Vorticella* was attempted using Artificial Sulfide Producing Systems (ASPROS) in the laboratory.

From 15 core samples, 7 contained *Vorticella* sp., in numbers ranging from 165 to 1. Mean density was 1,5 individuals . cm<sup>-2</sup>. Distribution was highly patchy, with only 21 of the counted 150 eyefields containing *Vorticella* specimens. In contrast, 12 of the 15 cores contained *Zoothamnium* colonies and numbers of *Vorticella* sp. individuals were strongly correlated with numbers of *Z. niveum* colonies. *Vorticella* sp. specimens were found only on cores containing *Z. niveum*.



*Zoothamnium niveum* colony on mangrove peat. Smaller stalked kinds specimens of *Vorticella* sp., fine threads are filamentous bacteria.

From 32 groups of *Z. niveum* colonies, 21 were associated with *Vorticella* sp. Those *Z.*

*niveum* groups which were not associated with *Vorticella* sp. consisted only of a few small colonies. This supports the hypothesis that *Vorticella* sp. is a late colonizer of ephemeral sulfide sources. Culture experiments had only limited success, for reasons unknown; only a few specimens grew on the surface of the ASPROS. Therefore no data on growth speed and reproduction could be obtained.

Several specimens were transported live to Austria and were given, together with fixed specimens, microphotographs and films to Wilhelm Foissner, an expert on ciliates. He tentatively placed the species in the genus *Pseudovorticella*. For a description of the new species, however, more live material is necessary. A conspicuous feature – in addition to the coat of symbiotic bacteria – is the strongly reflexed posture of the extended zooids, giving them an umbrella-like appearance.

## Relationship with their hosts, competitive ability, and sex allocation in eusocial snapping shrimp

E. Tóth

Eusocial (or truly social) animals live in groups with overlapping generations, where colony-members help to rear the progeny of one or a few individuals and forego direct reproduction. Eusocial sponge-dwelling

snapping shrimp are the only known eusocial aquatic animals. Shrimp colonies, like ants and bees, consist of one or a few reproductive individuals (queens) and numerous smaller helpers. Social colonies live inside sponge canals, just as aggregating and pair-forming as other members of the clade with whom they often compete for the same sponge hosts. Because of their aquatic habitat, eusocial shrimp provide an important opportunity for testing theories of social evolution, which has been based on terrestrial animals. Understanding the social lives of these shrimp, however, is not easy because of the paucity of information on their basic biology.

My last visit to Carrie Bow Cay had three objectives. First, I wanted to know whether *Synalpheus* shrimp occupying sponge canals have a positive or negative effect on their sponge host. I sampled 16 small sponges, measured their wet weight and put them out in the lagoon, where the sponges normally do not occur, eight of them containing shrimp and eight of them not. I will collect the sponges in six months to see which group of sponges grows faster. A previous pilot study that lasted three months indicated that sponges are capable to survive being transplanted.

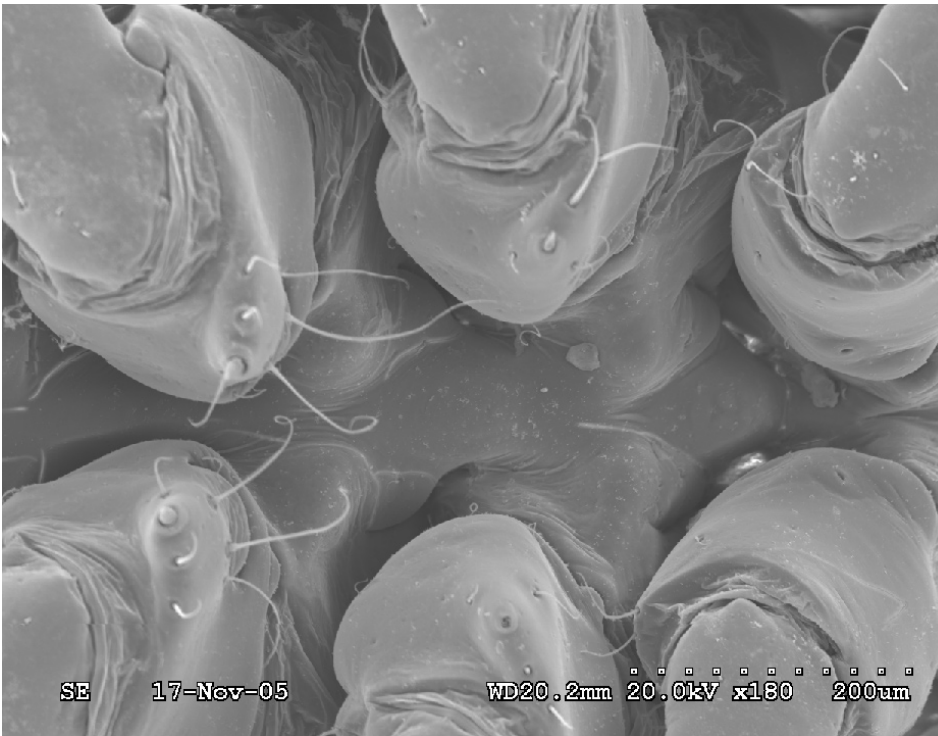
Secondly, I wanted to reveal whether competitive ability to occupy and monopolize sponges is better or worse in social shrimp compared to shrimp living in aggregations. I used one social species, *S. elizabethae*, and one aggregating species, *S. brooksi*, for this experiment. Both share the same sponge species, *Lysosodendoryx colombiensis*, but *S. brooksi* occupies about ten times more sponges than *S. elizabethae*. When size-matched individuals of each species were put in an arena, *S. brooksi* snapped significantly more often at *S. elizabethae* than vice versa. When size-matched individuals of the same species were introduced into an arena, aggression level was significantly lower than when individuals of the two species faced each other in the arena. This indicates that *S. brooksi* is more aggressive and probably can more forcefully fight for its host sponges than the social species, *S. elizabethae*. The finding that all *S. elizabethae* colonies that occupy sponges were very numerous indicates that once colonies reach a certain number of individuals they are able to compete with intruders of other shrimp species.

Lastly, I wanted to know what the sex allocation is in eusocial shrimp colonies. In many shrimp species, small animals without ovarian development are males and



Experiment with sponges in the lagoon.





SEM picture of hermaphrodite leg base of *S. filidigitus*.

as they grow larger they change into females. In terrestrial social animals, colonies contain also a lot of female helpers that lack ovarian development. In *Synalpheus*, sexing animals other than the reproductive female is difficult because they do not show external sexual characters. For this reason, helpers were assumed to be males, identifying males by default (no female with ovarian development). However, females release their eggs through tiny openings on the coxa of their third pair of legs while males release their sperm through small gonopores on their fifth pair of legs. I collected colonies of four social shrimp species, *S. elizabethae*, *S. regalis*, *S. chacei*, and *S. filidigitus* and preserved them. In collaboration with R. Bauer of the University of Louisiana at Lafayette we took scanning electron microscopy pictures of 45 animals of each species. The results suggest that helpers in shrimp colonies consist of both sexes, however, in *S. filidigitus* and *S. elizabethae* males might change their sex into a female that will not develop her ovaries. This finding contradicts what was assumed so far that helpers are males. This results will also facilitate future research and allow us for example to find out whether female helpers perform other tasks than male helpers.

## Causes and consequences of parrotfish predation on corals (*Porites* and *Montastrea* spp)

R.D. Rotjan & S.M. Lewis

Scleractinian corals are essential ecosystem engineers on tropical reefs, providing the backbone for one of the most diverse communities on earth. Caribbean parrotfish, long recognized as a major player in reef herbivory and trophodynamics, also feed on live corals, yet the consequences of such coral predation for reef communities have been little studied.

Our on-going studies at Carrie

Bow aim to examine several important and unresolved questions about parrotfish predation on scleractinian corals: (1) Do parrotfish predators select specific coral species, or certain colonies within a species?; (2) If so, what coral characters (including nutritional quality, nematocyst or chemical defenses) influence the likelihood of predation?; (3) What is the long-term impact of parrotfish predation on coral reproductive fitness (gamete production) or coral survival (growth and recovery)?

During this past year, we assessed direct predation by parrotfishes on the reef-building coral *Porites astreoides* and investigated possible hypotheses for why some coral colonies are selectively targeted. This work has resulted in a manuscript currently in press (Rotjan and Lewis, 2005) and two oral presentations, one at the Benthic Ecology Meeting in Mobile, Alabama, and one at the 10th International Coral Reef Symposium in Okinawa. We are currently working on developing mathematical models to predict parrotfish predation at the colony level.

In addition, in February 2004 we conducted a survey across reef habitats at CBC to quantify the specificity, incidence, and extent of parrotfish predation on scleractinian corals correlated with parrotfish density. We also looked at long-term changes in parrotfish abundance by comparing our results to similar data collected in 1982 (SML). We are currently preparing a manuscript for submission in October.

This year, we initiated studies designed to test several hypotheses that might help explain why certain

*Montastraea faveolata* and *M. franksi* coral colonies are targeted by parrotfish. We are in the process of conducting a large predator exclusion experiment on the outer ridge at CBC to relax, and then reintroduce, predation pressure, monitoring physiological changes in coral colonies over time. In this experiment, caged and control colonies have been monitored over 12 months for changes in nutritional quality (C:N ratio and spectral differences reflecting zooxanthellae density, following the methods of Edmunds *et al.*, 2003), symbiont community (zooxanthellae density, clade, and ITS2 type), defensive characters (nematocyst density, see Gochfeld, 2004) and skeletal hardness (Littler *et al.*, 1989). We anticipate that this experiment will be completed in Winter 2006.

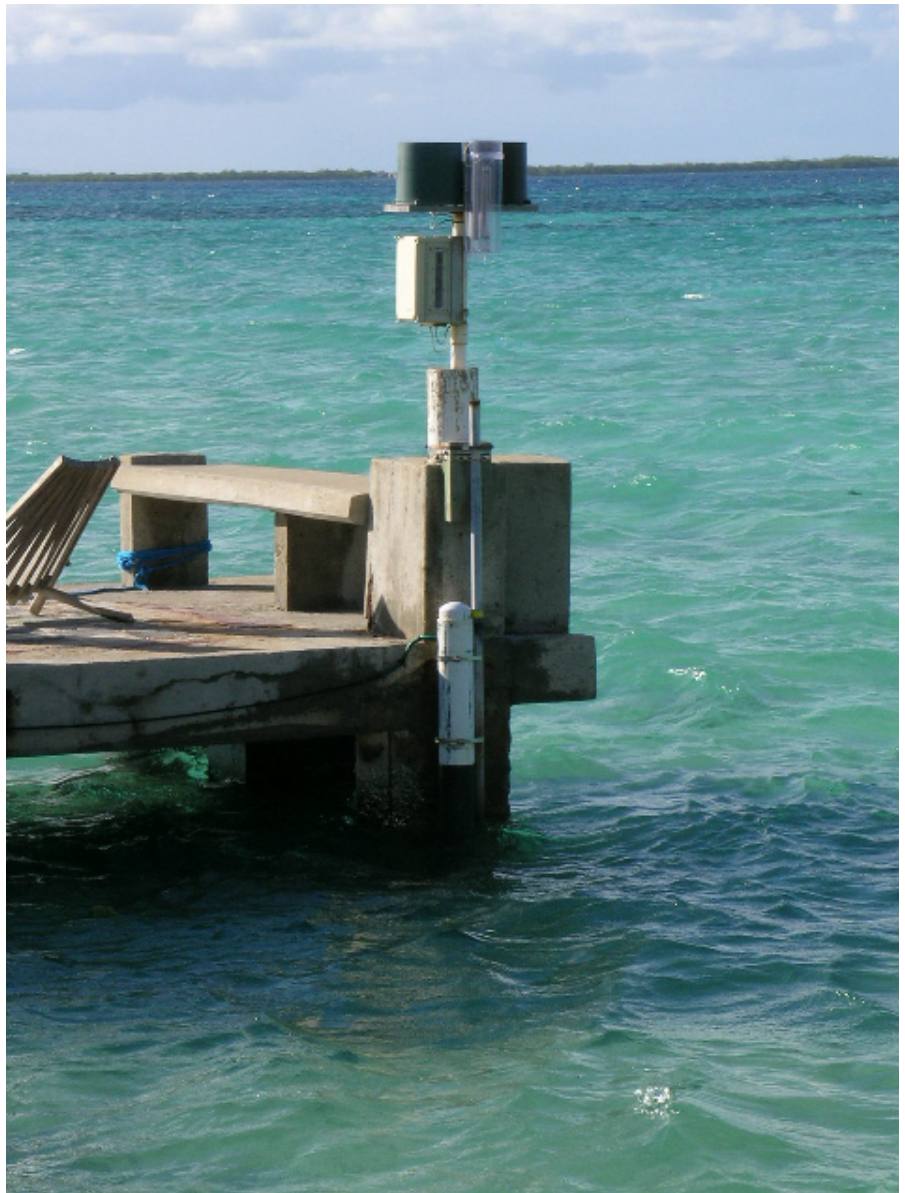
In September 2004, storm surge from Hurricane Ivan destroyed most of the predator exclusion cages we had deployed in August, just 1 month earlier. Despite this set-back, we fixed and re-set the experiment in October. We were also able to take advantage of this opportunity to examine how our experimental *Montastraea* colonies changed in response to hurricane stress, coincident with high seawater temperatures and seasonal bleaching. To this end, we teamed up with Brian Helmuth and other collaborators to investigate coral bleaching that occurred at CBC in October 2004. We found that *Montastraea* spp. corals that had been grazed by corallivorous parrotfishes showed a persistent reduction in symbiont density compared to intact conspecific colonies. Additionally, grazed corals exhibited greater diversity in the genetic composition of their symbiont communities, changing from uniform ITS2 type C7 *Symbiodinium* prior to bleaching to some mixed assemblages of *Symbiodinium* types post-bleaching. These results suggest that chronic predation coupled with coral bleaching can exacerbate coral stress and destabilize the coral-zooxanthellae symbiosis, and offer a novel explanation for reef-wide patterns of variable bleaching response and recovery. These data have resulted in a manuscript currently in review at the Public Library of Sciences – Biology (Rotjan *et al.*, 2005).

## Processes across Ecosystems

An automated real-time meteorological and oceanographic monitoring system supporting marine biological research and management on the Mesoamerican Barrier Reef: coral bleaching assessment

*T.B. Opishinski*

Recognized as the largest barrier reef complex in the northern hemisphere, the Mesoamerican Barrier Reef ecosystem extends a thousand kilometers from Mexico's Yucatan Peninsula southward to Honduras. The marine resource is shared by the populations of four countries (Mexico, Belize, Guatemala and Hon-



Weather Station at Carrie Bow Cay.

duras) and the significance of the reef is reflected in its status as a World Heritage site and the recent establishment of numerous MPA's. Heralded as a classic example of reef evolution, the ecosystem encompasses a rich diversity of reef structures (barrier, atoll, patch and fringe), habitat types (reef, mangrove forests, coastal lagoons, sea grass communities, open-ocean and deep-sea floor) and abundance of animal and plant biodiversity. The reef has been a focus of scientific research for decades as it offered researchers a pristine reef habitat largely unaffected by factors that contribute to unhealthy reef systems, however, increasing tourism, over-fishing, coastal development and natural events (hurricanes, bleaching) in recent years have severely impacted the health of the ecosystem. The scientific community has responded with expanded research, initiated new efforts to categorize and identify indicators of the reef's health, and established marine parks and MPA's to better manage the reef resources. Despite the increased initiatives there is a noticeable absence of continuous monitoring of abiotic and meteorological parameters that provide vital information for ongoing research and management efforts. The historical reasons for the lack of continuous environmental monitoring are not entirely clear, however, the remote location of the offshore reefs and corrosive environment previously contributed to the failure of several efforts to establish monitoring systems. Fortunately, scientists from the National Museum of Natural History (NMNH) of the Smithsonian Institution have long recognized the need for continuous measurements of oceanographic and meteorological data sets. Operating from the Caribbean Coral Reef Ecosystems marine research facility, which was established in the early 1970's and is located approximately 15km offshore on the barrier reef on Carrie Bow Cay, the Smithsonian researchers recognized that a unique approach was required to establish and maintain a long-term successful monitoring system while meeting the data requirements and creating a distribution network for a diverse array of collaborative research efforts. In 1997 an oceanographic-meteorological monitoring, management, and analysis system was established at Carrie Bow Cay. The system, to our knowledge, is the only automated system continuously monitoring both offshore oceanographic and meteorological parameters of the outer Mesoamerican Barrier Reef. Many factors including operational environment, remote location, data accessibility, power restrictions, requirements for unattended operation, and available communications for data transfer influenced engineer-

ing design criteria. The system has continued to provide a baseline set of data since it was established that serves as a tool for researchers to examine long term trends, episodic events, short term and seasonal cycles. It also allows comparative studies with other reef ecosystems to assess biodiversity and correlate environmental factors with biological phenomena and hazards (competition, parasitism, algal blooms, coral bleaching and invertebrate disease). The increasing degradation of the Mesoamerican Barrier Reef has highlighted the importance of the long-term oceanographic and meteorological measurements acquired at Carrie Bow Cay and proven invaluable to new and existing biological studies for both Smithsonian researchers as well as an increasing number of organizations from the region and beyond. To meet the expanding research, monitoring and management requirements, ongoing efforts have been implemented to continually improve and add functionality to the environmental monitoring system and supporting web portal. This paper describes in detail the design criteria of the system and includes supporting data and illustrations to demonstrate the reliability of the automated monitoring station and its importance to ongoing research efforts. Descriptions of the various



components that make up the system including an Internet-based data management, communication and web portal, a data analysis and presentation system with embedded geographic information system, and a commercial environmental data acquisition system and sensors are provided. A summary of specific biological research and monitoring projects that have utilized the data sets is presented. In addition, recent technological developments including the addition of a harmonic tidal analysis and prediction model and an intelligent processing module designed to identify conditions favorable for coral bleaching are presented.

## Carbon and nitrogen stable isotopic tracers of food sources for consumers in Belizean offshore mangroves

C.C. McIvor, M.L. Fogel, D.S. Taylor, W. Davis & E. Reyier

As part of an ichthyological survey of Twin Cays, Belize, we sampled fishes and crustaceans for carbon ( $\delta^{13}\text{C}$ ) and nitrogen ( $\delta^{15}\text{N}$ ) stable isotopes in multiple habitats (fringe forests, dwarf *Rhizophora mangle* forests, ponds, internal channels, sinkholes). Of 12 species with at least three individuals in a given habitat (individuals analyzed separately), eight species differed significantly in  $\delta^{13}\text{C}$  values, and five species differed in  $\delta^{15}\text{N}$  values in different habitats. Species using multiple habitats, therefore, are likely to derive both carbon and nitrogen from different within-habitat sources. Animals from hydrologically isolated sinkholes had  $\delta^{13}\text{C}$  significantly more negative than in other habitats, indicating either greater dependence on a carbon source from mangroves or from highly respired microbial carbon. Comparison of frequency histograms of consumer  $\delta^{13}\text{C}$  values at Twin Cays with those from other mangrove studies indicates a greater incorporation of a variety of carbon sources than previously reported.

## Nutrient limitation of *Batophora oerstedii* at Twin Cays

L.A. Franklin & I. Feller

Previous research by Ilka Feller on nutrient limitation of mangroves has suggested that detrital material, presumably derived from the green alga *Batophora oerstedii*, a commonly observed inhabitant of quiet tidal ponds in the interior regions of Twin Cays, is a major source of phosphate for *Rhizophora mangle*. It may also provide nutrients for newly established *Batophora* populations. However, little is known about the nutritional physiology of this alga at any stage of its life history. A pilot project to look at factors affecting recruitment of *Batophora* was initiated in August 2003, using mesh bags, filled with various biological and abiotic substrates (empty bags as controls), as settlement surfaces. No recruitment was observed until Nov. 2004. In May 2005, substrates were collected and the size of the *Batophora* population measured. Recruitment and

growth was extremely low on control bags and on abiotic substrates. Of the biotic substrates tested, biomass was highest for bags filled with coarse woody debris derived from *R. mangle*, with intermediate amounts measured on bags filled with leaves of *R. mangle* or *Syringodium* (seagrass). Surprisingly, growth on bags containing detrital material was also relatively low, though a number of these bags were no longer present at the end of the experiment, reducing our ability to detect a significant effect. One possible explanation for the low growth rate in this treatment compared to the other biological substrate treatments may be the shorter longevity of the detrital material, little of which remained in the bags relative to the other materials at the end of the experiment.

A second experiment to identify possible nutrient limitation of *Batophora* was carried out in May 2005. During periods of phosphate (but not nitrogen) uptake, chlorophyll fluorescence in several microalgae has been observed to decline, or quench, in a characteristic pattern (Beardall, *et al.* 2001, *Aquat. Sci.* 63:107-121). Using pulse amplitude modulated (PAM) fluorescence imaging technology, the pattern of chlorophyll fluorescence quenching during nutrient uptake was examined in individual *Batophora* thalli collected directly from the field. Similar to results obtained with the microalgae, chlorophyll fluorescence was quenched when phosphate (but not ammonium) was supplied, suggesting that *Batophora* in its natural habitat is under phosphate limitation. To our knowledge, this is the first demonstration that this technology will be useful for studying nutrient uptake in a macroalga. It also points the way for further nutritional studies of this alga.

## Dinoflagellate blooms in near reef mangroves, Douglas Cay and Twin Cays

M. Faust

The Belizean coral near reef mangrove habitats are ideal for investigating the distribution, ecology, species associations, and abundance of toxic dinoflagellates in an ecosystem of great typological diversity. Preliminary surveys found that although harmful dinoflagellates are found in low abundance throughout the Belizean barrier reef system, the greatest diversity and highest numbers of bloom-forming species appear restricted to mangrove cay embayments and similar environments (Faust, 2000, 2004; Morton and Villareal, 1998; Morton, 2000). Dinoflagellates dwell in plank-

tonic and benthic habitats protected from wind, and having restricted water exchange with the surrounding oligotrophic waters of the open barrier reef. The limited water exchange favors retention of dinoflagellate cells and trapping of nutrient-rich organic material, which is rapidly recycled. There is, however, a paucity of information about harmful dinoflagellate blooms that occur in tropical regions, including the Atlantic barrier, reef and mangrove ecosystems of Belize. In the study, the ecology, diversity and morphology of eight bloom-forming dinoflagellate species from two nutrient-enriched habitats, Douglas Cay and The Lair, Twin Cays, are described in detail and an evaluation of how well the prevailing conditions and dinoflagellate abundance agree with the prediction of Margalef's Mandala is presented (Faust *et al.*, 2005; Atoll. Res. Bull., submitted).

A comparison of the species composition showed that Douglas Cay contained coastal planktonic and offshore oceanic dinoflagellates (45 species), while The Lair at Twin Cay, contained mainly benthic dinoflagellates (27 species). A total of 19 bloom-forming species were observed in these locations during 3 two week studies. Approximately half of the bloom-forming dinoflagellates are known toxin producers. The congruence between Margalef's prediction and the distribution of dinoflagellates in these naturally eutrophic systems suggests that increased nutrient inputs in oligotrophic portions of the Caribbean will favor a shift in species dominance toward dinoflagellate species. The affect will be most pronounced in bays or other regions where turbulence is likely to be reduced. This species shift may have consequences for food web dynamics and the prevalence of dinoflagellate toxins in the food chain.

### A nutrient oasis in an oligotrophic sea: a model of natural eutrophication (continued)

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

The highly productive phytoplankton and bacterial communities associated with mangrove cay embayments in the Belizean central lagoon are supported by natural eutrophication. Hydrological measurements indicate that waters in the Douglas Cay embayment, and the Twin Cays Lair, Lair channel and Main channel are divorced from the surrounding lagoon. This

is caused by a combination of geomorphology, weak tides, and strong density gradients. These factors retard mixing between these mangrove fringe habitats and the surrounding lagoon. As a result, these sheltered mangrove habitats are characterized by elevated levels of  $\text{NH}_4^+$  and other nutrients, resulting in high levels of planktonic and benthic microalgae. Microalgal populations are typically dinoflagellate-dominated, including numerous toxic species (Faust 2004, Sunda 2005). Our research efforts have been directed toward testing the hypothesis that sheltered mangrove embayments like those at Douglas and Twin Cays act as reservoirs for toxic dinoflagellate species, most noteworthy of which are members of the genera *Gambierdiscus*, *Prorocentrum*, and *Ostreopsis*.

In 2005, we sought to examine the distribution of these dinoflagellate species at the mangrove cays surrounding Carrie Bow Cay. We sought to characterize how toxic dinoflagellate distribution relates to hydrography, light availability, and dissolved nutrients. In addition, we examined the effect of anthropogenic disturbance and nutrient input to reef and mangrove communities.

*I. The Main Channel, Twin Cays:* Data collected in May 2004 demonstrated the southern portion of Main Channel was characterized by outflow of mangrove waters at the surface and inflow of lagoon water along the bottom (Kibler *et al.*, submitted). Strong vertical stratification allowed reef-type biota to co-exist with organisms endemic to mangrove fringe waters. In contrast, the northern channel was characterized by shallow (< 2 m), poorly stratified waters that were divorced from the surrounding lagoon by shoals and a steep density gradient. Because of this restricted exchange, the northern part of the Main channel underwent rapid shifts in temperature and salinity due to strong solar radiation and intermittent rainfall.

In May 2005 we collected hydrographic data from 10 stations along the Main Channel. Vertical profiles showed a hydrographic structure very similar to that described in 2004. However, the 2005 data indicated inflow of lagoon water occurred along the bottom of both the northern and southern ends of the channel. This suggests the northern Main channel may not be as isolated from lagoon waters as previously thought. Instead, circulation may depend upon prevailing wind direction.

*II. The Lair and Lair Channel, Twin Cays:* Profile data collected in May 2004 demonstrated that high temperature and high salinity bottom water, trapped within the

Lair during the day, was exported from the Lair and Lair channel at night (Kibler *et al.*, in prep.). The majority of the phytoplankton resided in a narrow band near the bottom and exhibited vertical migration, suggesting phytoplankton move towards the bottom to avoid daytime photic stress.

Profiles collected 2005 data again showed export of bottom water from the Lair channel as well as high chlorophyll fluorescence along the bottom of both the Lair and Lair channel. This vertical distribution supports our hypothesis that photic stress can be an important control upon phytoplankton distribution in mangrove waters.

*III. Twin Bays, Twin Cays:* Observations made in May of 2005 suggested that inner Twin Bays supported a high abundance of toxic dinoflagellates. These cells were present in whole water samples and plankton tows but cell abundance was not quantified. In 2005, we collected samples to quantify the distribution of toxic dinoflagellates between inner Twin Bays and the mouth of the Main channel. Despite abundant phytoplankton,  $\text{NH}_4^+$  concentrations were relatively low, ranging from  $0.6 \mu\text{mol L}^{-1}$  at inner Twin Bays to  $0.2 \mu\text{mol L}^{-1}$  in outer Twin Bays. A bloom was observed in outer Twin Bays

the afternoon of 17 May 2005. Cell samples revealed the bloom was caused by a species of *Prorocentrum* that was normally present in low numbers. Clonal cultures of this species have been established and identification is awaiting DNA sequencing and morphological examination by SEM.

*IV. Embayment at Douglas Cay:* Data collected in 2003 and 2004 showed that the Douglas Cay embayment was characterized by high dinoflagellate abundance. Exchange with the surrounding lagoon was limited, resulting in retention of dissolved organics, bacteria, dissolved nutrients and phytoplankton cells.

In 2005, we collected samples to quantify distribution of toxic dinoflagellates and characterize  $\text{NH}_4^+$  and reactive phosphate concentrations. Ammonium concentrations ranged from  $0.6 \mu\text{mol L}^{-1}$  inside the embayment to  $0.3 \mu\text{mol L}^{-1}$  outside the bay. Although morning and afternoon  $\text{NH}_4^+$  levels were not significantly different,  $\text{NH}_4^+$  concentrations were significantly higher near the bottom, indicating flux of  $\text{NH}_4^+$  from the sediment. These data constitute the first direct evidence of autochthonous nutrient input at Douglas Cay. The abundance of dinoflagellates in other mangrove embayments may be ascribed to similar nutrient inputs.

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## *New web site address for the Environmental Monitoring Program at Carrie Bow Cay (T.B. Opishinski)*

The new address for the web site is <http://cbc.riocean.com> Each of the topics listed below is accessed from the web site's Table of Contents.

### New content:

1. 2006 Weather Statistics - View and download weather statistics for 2006. Weather statistics are available for viewing in HTML format or for download (PDF format) and for daily, weekly monthly and year-to-date time periods. Statistics are available for wind speed, air temperature, relative humidity, barometric pressure, solar radiation and rainfall rate. Wind speed and rainfall accumulation will be added to the list however they require different methods to process the data to yield meaningful information.
2. 2006 Tidal predictions - Tidal predictions for Carrie Bow have been generated for 2006. Predictions are calculated using a harmonic analysis model that utilizes past measurements of tidal elevation at Carrie Bow to forecast future tides for the Carrie Bow Cay area. The information is useful for planning the surveys and experiments that depend on the state of the tide. Water level measurements are overlaid on the forecasted tidal graphs for comparison.
3. Shoreline Surveys - Historical surveys have shown both seasonal and long-term changes occurring to the shoreline of Carrie Bow Cay. This page provides results of the latest shoreline survey of Carrie Bow Cay conducted in February of 2006 using GPS technology.
4. Four-Year Temperature Record - September of 2005 marked the 5th year since the monitoring system went into operation. The temperature record was constructed from data collected by the system.
5. 2005 Charts and Hurricanes - Although there were no direct strikes in Belize the effect of hurricanes in the region is observed in the environmental data. This section includes summary charts of water level and temperature for 2005. Charts are annotated with the names of hurricanes to identify their passing and to display the effects on water level and temperature.

### Updated content

1. Environmental Data Archives - The quality of the historical data sets contained in the archives and the format of the page has been improved.

Some of the changes include:

-The order of the tables containing the download links was reversed so that the most recent year appears at the top of the page with tables for previous years following in chronological order.

-Data may still be downloaded in ASCII text or Excel format.

-An extensive effort was taken to process and improve the quality of the historical data sets. This primarily includes removing invalid data (spikes, data contaminated by biofouling) and correcting for normal instrument drift based on pre- and post-calibration processes. \*\*\*In general the data included in the ASCII text files is raw or has a minimal level of quality control while the Excel files contain data that of higher quality.

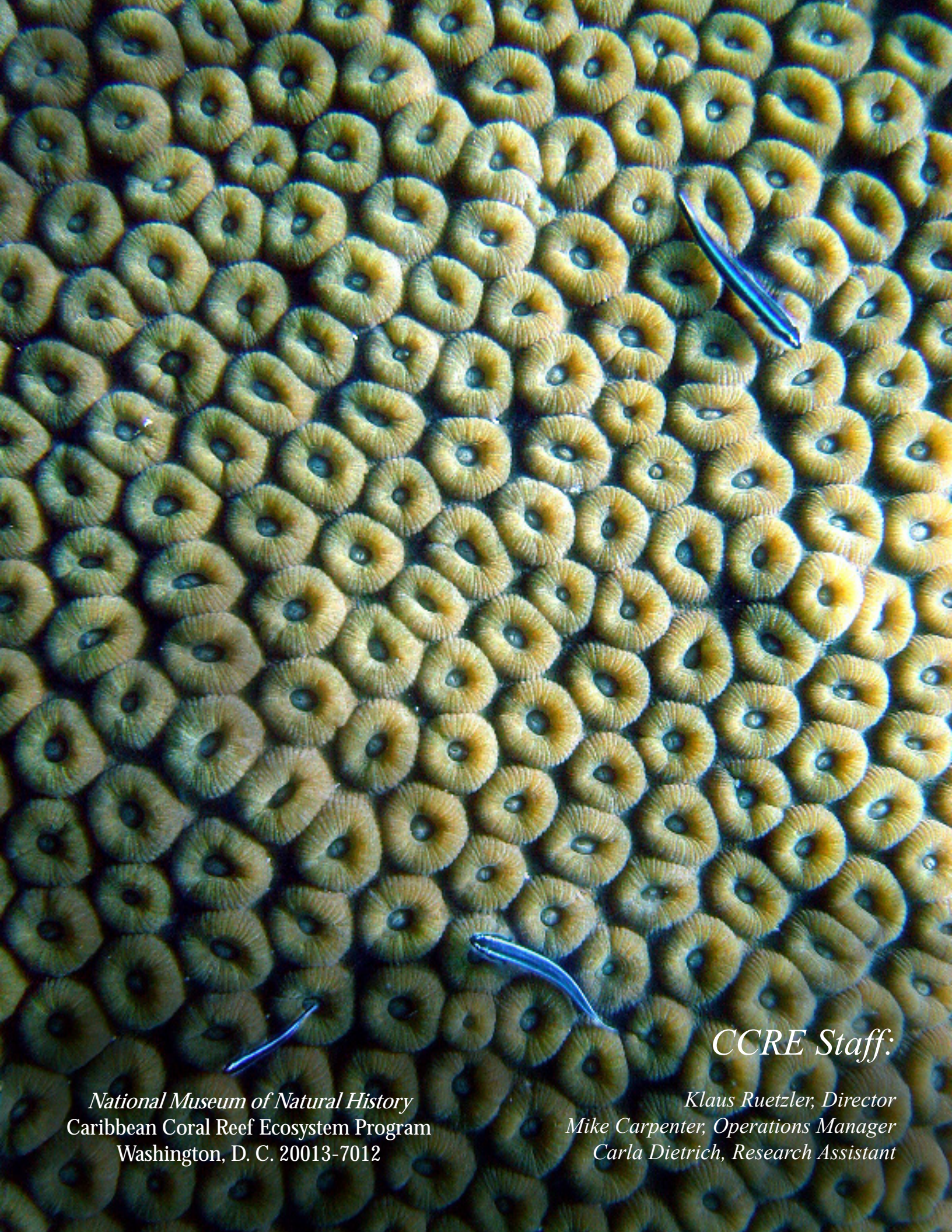
-The format of the ASCII text files has been changed to a tab-delimited format beginning in January 2006. The tab-delimited format offers increased readability in text editors and can be opened directly in Excel without having to using the import dialog.

-Graphs for each parameter have been added to the Excel archives.

2. Download Area maps - Maps that focus on southern Belize and the Carrie Bow Cay area are available to download. The maps are in PDF format and have been created from GIS images and hand illustrated research maps. Note that some of these files are large and can take a few moments to download to your web browser.

### Future content

1. Links/Publications - NOT yet available, the objective of this section of the web site is to have descriptions of projects, links to related web sites, papers and presentations that have utilized the Carrie Bow data available for viewing or download. It is meant as a means to share information and generate new ideas. We are looking for additional references and papers to include in this section and welcome all submissions. A number of you have provided brief descriptions of your projects in the past and an expanded description or published paper would be



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