WORKSHOP REPORT

Proceedings of the SERDP Coral Reef Monitoring & Assessment Workshop

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DoD EPA DOE SERDP

Strategic Environmental Research and Development Program

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FORWARD

These proceedings summarize the SERDP Coral Reef Monitoring and Assessment Workshop and reflect the opinions and views of workshop participants, not necessarily those of the Department of Defense (DoD). This document will be available in PDF format at www.serdp.org.

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DoD sponsored this workshop through funding awarded by DoD's Strategic Environmental Research and Development Program (SERDP) and Environmental Security Technology Certification Program (ESTCP) with the Rosenstiel School of Marine and Atmospheric Science (RSMAS) at the University of Miami hosting/ supporting this workshop. These proceedings document the presentations, conversations and results of the workshop

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EXECUTIVE SUMMARY

From 2003 to 2009, SERDP funded the development of two technologies for assessing and monitoring coral reef health: 1) high-resolution (millimeter scale) video-mosaicing technology, capable of rapidly surveying and providing a permanent visual record for benthic areas over 100s of square meters in size (University of Miami) and 2) advanced bio-optical techniques for non-destructive assessment of selective natural and anthropogenic stresses using fluorescence induction and relaxation sensors (Rutgers University).

A SERDP-sponsored workshop was held at the Rosenstiel School of Marine and Atmospheric Science, University of Miami Nov 18-19, 2008. The goals for the workshop were to: (1) understand the DoD client perspective on benthic community/coral reef assessment and monitoring needs; (2) understand other potential user perspectives (i.e., in addition to DoD) regarding their coral reef monitoring and assessment needs and how the two SERDP-developed technologies may help address those needs; and (3) identify how the two approaches/technologies are complementary to each other and how they might be integrated to meet end-user needs.

Presentations by DoD personnel, representatives from governmental and non-governmental organizations/offices actively involved in coral reef management and research, and the research teams from the University of Miami and Rutgers were interspersed with active discussion. Key findings include the following:

1) Federal policy mandates that DoD characterize, assess, and monitor underwater benthic communities at Air Force, Army, Marine Corps and Navy bases in order to document compliance with national policy and to ensure that DoD operations do not lead to natural resource degradation, particularly with respect to coral reefs. DoD is looking for technologies and methodologies that will enable the collection of coral reef data with less dive time, that have the ability to reproduce data collection transects reliably year after year and provide a rapid deployment capability to document coral reef groundings. DoD is also interested in exploring how emerging technologies may foster new opportunities to develop productive partnerships between the Navy and other organizations.

2) Workshop participants were in agreement that metrics collected by current monitoring and assessment strategies conducted by the agencies are, in general, adequate to meet present mandates. However, there was also consensus that present methods of data collection are time consuming, labor intensive, and not standardized, thereby limiting the number of sites that can be monitored, comparison between studies, and the speed with which data can be provided to coral reef managers. There was also broad interest from all agencies in developing methodologies that reduce dive time, improve cost efficiency and provide repeatable data specifically from those agencies involved in field monitoring and assessment of coral reefs. Specific challenges and needs expressed by the agencies include developing capabilities for detailed mapping with improved capabilities (resolution and accuracy) and in-situ testing of physiological health of coral organisms. The improved methodologies would support expanded coral reef ecosystem level monitoring, monitoring of deep reefs, studies of infection patterns of coral disease

and non-destructive methods for determining coral reef physiological status and prospective health assessments of coral reefs.

3) There was consensus_regarding the usefulness of landscape mosaics and FIRe technologies for advancing coral reef monitoring and assessment practices. The mosaicing technique offers potential for more efficient methods of monitoring coral cover, colony size, mortality, bleaching and disease, population structure, extent of injury and recovery patterns, and documentation of coral reef ecosystem metrics. There was consensus that the FIRe technique also provides capability for in-situ monitoring for sublethal effects from stressors and for identifying the cause(s) of detrimental change. There was also agreement that the transition of both technologies to the end-user community would be valuable and should be pursued.

4) The overall consensus was that the two technologies are complementary, but not necessarily synergistic, to each other. Integration of the two technologies onto a single platform could be useful in the future to some in the user community, but, in the short term, integration would not be necessary to benefit from the capabilities of the separate technologies when deployed separately

5) It was suggested that the developed technologies, in particular the FIRe fluorometry, be employed and validated at a non-DoD test site with a known stressor environment. As an example, the NOAA site(s) in Puerto Rico might be used for this purpose.

6) Based on widespread participant interest for using mosaics, paths for commercialization of the technology were discussed. Two strategies were considered: 1) licensing the technology to a commercial software company such that individuals could buy software to produce their own mosaics; and 2) commercializing a service under which mosaics would be produced on a fee-per-mosaic basis. Participants generally seemed to favor Option 2, but recognized that an informed decision would require a cost benefit analysis.

SERDP Coral Reef Monitoring and Assessment Workgroup

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LIST OF ACRONYMS

CREMP	Coral Reef Evaluation and Monitoring Project
DoD	Department of Defense
DoN	Department of Navy
DIDSON	Dual Frequency Identification Sonar
EPA	Environmental Protection Agency
ESTCP	Environmental Security Technology Certification Program
FIRe	Fluorescence Induction and Relaxation technique
Fo	Minimum quantum yield of fluorescence
Fm	Maximum quantum yield of fluorescence
(Fv/Fm)	Photosynthetic efficiency
GPS	Global positioning system
MMS	Minerals Management Service
NEPA	National Environmental Policy Act
NMFS	National Marine Fisheries Service
NOAA	National Oceanic and Atmospheric Administration
NPS	National Park Service
NWR	National Wildlife Refuge
RSMAS	Rosenstiel School of Marine and Atmospheric Science
SERDP	Strategic Environmental Research and Development Program
TNC	The Nature Conservancy

ACKNOWLEDGEMENTS

The Coral Reef Monitoring and Assessment Workshop sponsors wish to thank the RSMAS and Rutgers Teams who helped formulate the agenda, identify appropriate participants, and determine priorities. Special thanks to Dr. Pamela Reid and her team for hosting the workshop, and to Ms. Cheryl Kurtz for keeping the minutes and drafting the workshop proceedings.

The sponsors would like to acknowledge Ms. Meghan Dick and Ms. Brooke Gintert for the logistical and onsite support provided and their dedication in organizing the field trip of Emerald Reef (cancelled due to large swell conditions).

Finally, the sponsors wish to thank all the event's participants (Appendix A- List of Participants), without whom this workshop would not have taken place.

INTRODUCTION

From 2003 to 2008, SERDP funded the development of two technologies for assessing and monitoring coral reef health: (1) high-resolution (millimeter scale) video-mosaicing technology, capable of rapidly surveying and providing a permanent visual record for benthic areas over 100s of square meters in size (University of Miami); and (2) advanced bio-optical techniques for non-destructive assessment of selective natural and anthropogenic stresses using fluorescence induction and relaxation sensors (FIRe, Rutgers University).

A SERDP-sponsored workshop was held at the Rosenstiel School of Marine and Atmospheric Science, University of Miami, Nov 18-19, 2008. The goals of the workshop were to: (1) understand the DoD client perspective on reef assessment and monitoring needs; (2) understand other potential user perspectives (i.e., in addition to DoD) regarding their coral reef monitoring and assessment needs and how the two SERDP-developed technologies may help address those needs; and (3) identify how the two SERDP approaches/technologies might be complementary to each other and how they might be integrated to meet end-user needs.

This report summarizes the workshop activities, including: (1) background presentations by SERDP (DoD) and DoN presenting the Navy perspective; (2) other agency perspectives on coral reef monitoring practices; (3) descriptions and demonstrations of SERDP-funded technologies; (4) group discussion of current practices, evaluation of SERDP-developed technologies, and potential overlay of SERDP technologies on current practices and needs; and (5) summary and results.

BACKGROUND

Program Overview - Dr. John Hall, OSD: SERDP/ESTCP

SERDP is DoD's environmental science and technology program, planned and executed in partnership with the Department of Energy and the Environmental Protection Agency, with participation by numerous other federal and non-federal organizations. To address the highest priority issues confronting the Military Services, SERDP focuses on cross-service requirements and pursues high-risk/high-payoff solutions to DoD's most intractable environmental problems. SERDP's investments range from basic through applied research to exploratory development needs in the areas of Environmental Restoration, Munitions Management, Sustainable Infrastructure, and Weapons Systems and Platforms. SERDP's Sustainable Infrastructure initiative supports research and development (R&D) efforts to (1) sustain the use of DoD's lands, estuaries, ocean space, and air space; (2) protect its valuable natural, cultural, and infrastructure resources for future generations; (3) comply with legal requirements; and (4) provide compatible multiple uses of its resources.

ESTCP is DoD's environmental technology demonstration and validation program. ESTCP seeks to promote the use of innovative, cost-effective environmental technologies that target DoD's most urgent environmental needs, including range

sustainment, through demonstrations at DoD facilities and sites. ESTCP selects lab-proven technologies with broad DoD application for rigorous field trials. These demonstrations document the cost, performance, and market potential of the technology. ESTCP technology demonstrations address DoD environmental needs in the Environmental Restoration, Munitions Management, Sustainable Infrastructure, and Weapons Systems and Platforms focus areas. These technologies provide a return on investment through improved environmental performance, reduced liability, and direct cost savings, while supporting and maintaining military readiness. Successful technologies supported by ESTCP often have commercial applicability.

DoD/Navy Perspective - Ms. Lorri Schwartz, NAVFAC HQ

DoD is authorized to manage natural resources on property under its control. Major drivers are the Sikes Act, Clean Water Act, Clean Air Act, Marine Mammal Protection Act, Endangered Species Act, and various Executive Orders including EO 13089 for Coral Reef Protection. Currently, 46 military facilities and ranges are located in areas with coral reef resources within DoD's jurisdiction. Additional Navy marine resource protection projects include:

- Artificial reef creation - sinking the retired aircraft carrier ex-ORISKANY
- Clean up of tires from the failed Osborne artificial reef
- Reference database for Natural Resource Managers containing scientific literature about DoD coral reef sites
- Beach clean-up projects in Hawaii
- Active reef ecosystem protection through NEPA, assessment, monitoring, and research/demonstration

SERDP/ESTCP plays a role in assisting in natural resource management by supporting the development of novel technologies for the assessment of benthic habitats, supporting routine activity planning, and providing high-quality data to support compliance requirements. The Navy is looking for technologies and methodologies that will enable the collection of data needed to support its mandate with: (1) reduction of costly field and dive time; (2) increased reproducibility and reliability year after year; and (3) flexibility to modify assessment plans based upon an expert's evaluation of site conditions at the time of survey. Moreover, to meet DoD needs, sampling method and data verification procedures need to be widely accepted by both the resource management agencies and the scientific community. DoD is also need of a rapid deployment capability to document coral reef groundings. DoD is also interested in exploring how emerging technologies may foster new opportunities to develop productive partnerships between the Navy and other organizations. The two coral reef assessment technologies presently funded by SERDP (video/image mosaics and coral fluorescence) are examples of the potential for developing these types of partnerships. Both of these projects have previously interacted with Navy (e.g., AUTEC) and other partners (e.g., NOAA) to start development of joint coral monitoring programs for the efficient and effective assessment of coral status and trends. Finally, upcoming DoD projects that will likely influence coral reef status in the affected jurisdictions and may potentially benefit from the application of these SERDP-funded projects include the installation of the Fort Kamehameha Sewer Outfall in Hawaii and marine infrastructure projects in Guam.

AGENCY PERSPECTIVES

To obtain a better understanding of the work currently being done in coral reef monitoring and assessment, presenters were chosen from a variety of governmental and non-governmental organizations/offices actively involved in coral reef management and research and asked to prepare a presentation covering the following information:

1. What is your agency's mandate with respect to reef monitoring and assessment?
2. How are coral reef monitoring and assessment activities structured within your agency (e.g., offices, groups)?
3. Who are your most common partners (e.g., other agencies, academics)?
4. What methods and technologies are currently used in your agency for coral reef monitoring?
5. What currently limits your ability to fulfill your mandate?
6. What future reef monitoring and assessment activities are planned by your agency?

Minerals Management Service - Mr. James Sinclair

Mineral Management Service (MMS, Under the Dept. of Interior) focuses primarily on offshore resource recovery (oil, gas, sand, sulfur and alternative energy sources). MMS is a resource regulation agency, with a focus on the impacts of resource recovery (oil, gas, sand) on natural habitats. MMS takes an active role in the protection of coral reefs and fish communities in the habitats impacted by resource extraction (e.g., Flower Garden Banks, northwest Gulf of Mexico). The types of habitats and communities protected by MMS include: live bottoms (coral reefs, soft-sediment communities, hard-bottom), potentially sensitive biological features, topographic features, and chemosynthetic communities. Methods that MMS currently uses to assess coral reefs and associated communities include video transects, photo quadrats, colony growth surveys, visual fish/urchin/lobster surveys, and water-quality surveys. In the future, MMS hopes to identify areas within their governance that need additional protection and characterize their baseline characteristics to be used for future impact analyses.

National Park Service- Dr. Benjamin Ruttenberg

National Park Service (NPS, under the Dept. of Interior) conducts status and trends assessments of coral reef habitats in support of the management responsibilities of individual National Parks in the U.S. that have coral reefs within their jurisdictions. Biscayne National Park, the Dry Tortugas, and U.S. Virgin Island parks at St. John and Buck Island (St. Croix) are primary focal points for coral reef assessments conducted by the NPS. The Florida/Caribbean Office (FLACO) is the only NPS office that supports monitoring efforts in the Florida and Caribbean regions. The office has no regulatory oversight over the parks; the data collected are provided directly to the Parks and regulators for their use. Typical methodology for reef assessment within NPS includes: visual surveys, video transect surveys, and photo surveys. Surveys are conducted at both random (Index) and permanent (Extensive) sites. The main indicator of coral reef condition recorded is percent cover of the main benthic organisms (corals, sponges, and

algae). These methods used have been shown to be repeatable and statistically robust, and can be used to generate habitat maps. In the future, NPS desires to look at deep-water corals at sites like Buck Island National Monument and Salt River Canyon, St. Croix). This expanded effort will require modified methods (e.g., mixed gas, ROVs) due to the logistic challenges associated with deep diving. In addition, NPS would like to expand its mapping capabilities (habitat and bathymetry); conduct circulation modeling w/larval transport information; and conduct research on coral diseases, ocean acidification, and lionfish eradication. NPS is presently working on a new, integrated standard coral reef monitoring protocol for coral reefs, fish, and seagrass communities. Some of the new techniques that NPS would like to integrate into its protocol include LIDAR, high-definition videography and drop cameras for monitoring deep water sites.

U.S. Fish & Wildlife Service – Mr. Bret Wolfe

U.S. Fish and Wildlife Service (also under the Dept. of Interior and in conjunction with the National Wildlife Refuge Systems) enforces the Endangered Species Act (ESA) and the continued protection of listed species. Jurisdictions with coral reef resources include: Great White Heron NWR and Key West NWR, Navassa Island (Haiti), Midway Atoll NWR, Hawaiian Islands NWR, Guam NWR, Johnson Island NWR, Baker Island, Howard island, Jarvis island, Kingman Island, Palmyra Atoll, and Rose Atoll. USFWS partners with the U.S. EPA Water Quality Program, NOAA, USGS, and the Moore Foundation on several coral reef projects. Current methods used to assess coral reefs are diver-towed visual surveys, photo quadrats, video surveys, and visual fish surveys. In the future, USFWS would like to see improved monitoring technologies, develop more partnerships, conduct more research cruises (especially at remote refuges), improve present understanding of invasive species, and find better ways to enforce fishing regulations and stop illegal fishing.

U.S. Environmental Protection Agency - Dr. William Fisher

The EPA's Office of Research and Development (ORD) is focusing on biocriteria development and ecosystem services research. Biocriteria are authorized by the Clean Water Act and allow states to define the expected condition of aquatic resources (such as coral reefs) and enforce changes in watershed management if those expectations are not met (impairment). ORD is conducting research to assist states and jurisdictions in the development of biological indicators and long-term bioassessment monitoring programs to support implementation of regulatory biocriteria. Their most recent research on coral reefs has resulted in the drafting of the EPA Coral Reef Rapid Bioassessment Protocol, which focuses on stony corals. The proposed survey methodology relies on visual surveys conducted by trained divers who collect three core measurements (species identification of coral colonies, size, and percent living tissue). These metrics are combined to calculate multiple indicators that are sensitive to human disturbance such as total live coral cover and surface rugosity. Indicators for regulatory purposes must respond to human disturbance and be detectable beyond natural variation. ORD is now beginning to look into other assemblages, such as soft corals, sponges, fish, and invertebrates for responsive indicators. In a separate but related program, EPA is developing a strategy to incorporate coral reef ecosystem services into local management and regional policy decisions. All too often, decisions in coastal zones and watershed

areas are made without considering the effects of these decisions on coral reef communities and the many services the reefs provide (e.g., shoreline protection, tourism, fisheries). The new program will work toward the valuation of reef ecosystem services and tools to ensure that the value of these services is included in the decision equation. The ultimate purpose of the research is to better inform decision-makers of the system-wide consequences of different options (trade-offs).

NOAA Southeast Fisheries Science Center- Dr. Margaret Miller

NOAA SE Fisheries Science Center (under the Dept. of Commerce) is responsible for monitoring of coral reef fish and invertebrates, coral condition, and coral population dynamics, as well as assessing the status of protected species, and conducting reef restoration activities. Dr. Miller's research focuses on coral population status and coral restoration. Techniques commonly used by SE Fisheries are: stationary visual censuses yielding multi-species/size/abundance data for reef fish; coral surveys using visual and photographic methods; reef habitat characterization using acoustic techniques; visual surveys of mangroves; surveys of mangrove fish populations using sonar (DIDSON) and photo-video sampling. The metrics of coral reef condition commonly recorded include coral cover, colony sizes, partial mortality, abundance of coral predators, and prevalence of diseases and bleaching. Limitations that hamper SE Fisheries' ability to conduct reef assessments are classical trade-offs between in-water time/effort and data quality, spatial and temporal coverage, and sampling frequency. Moreover, visual and photographic methods provide limited ability to census coral recruits (1 mm), resulting in a general lack of information on recruitment, and growth and mortality of the early life stages of corals. Finally, an overall challenge in the field of coral conservation is the lack of coral health/disease diagnostic techniques.

NOAA Center for Coastal Monitoring & Assessment - Mr. Robert Warner

NOAA's Center for Coastal Monitoring and Assessment (CCMA) is composed of two branches, the Biogeography Branch (BIOGEO), headed by Dr. Mark Monaco, and the Coastal and Oceanographic Assessment, Status and Trends Branch (COAST) headed by Dr. John Christensen. CCMA involvement in coastal monitoring is diverse, with projects that assess estuarine and coral reef resources in Florida and the Caribbean, and the assessment of Marine Protected Areas. This office also administers the U.S. Mussel Watch Program, and evaluates environmental contamination throughout the nation's coastal regions. Working in close collaboration with partners, the Biogeography Branch maps and monitors coral reefs residing within United States jurisdiction. Techniques and methods used by the Biogeography group to map and monitor status and trends of submerged resources include visual fish surveys, visual/photo quadrats, and remote sensing methods. Some of the tools commonly used include photogrammetry, imaging spectroscopy, collection and analyses of IKONOS Imagery, LIDAR, and multi-beam acoustic data. In constantly seeking ways to improve, CCMA is interested in such areas as new benthic characterization tools; improved underwater positioning systems; acoustic methods for fish surveys; and AUV platforms/sensor payloads. NOAA's Coral Reef Conservation Program (CRCP) has recently refined its focus to three topics involving the impacts to coral reefs from fishing, land-based pollution, and climate change. CCMA's

two branches are currently working closely on projects, with their partners, to assess the effects of chemical contamination on the health of coral reefs in the Caribbean.

NOAA Marine Sanctuaries – Mr. Bill Goodwin

NOAA – Marine Sanctuaries manages the Florida Keys National Marine Sanctuary (FKNMS) in accordance with the Marine Sanctuaries Act. The physical damage caused by vessel impacts on shallow habitats is a major source of mortality to benthic resources. The Damage Assessment and Restoration Program of the FKNMS also performs detailed mapping, assessment, and monitoring of injured areas (usually related to vessel groundings) within the Sanctuary and uses these data to develop detailed coral restoration and rehabilitation programs. After restoration is complete, long-term (five-year) monitoring efforts are performed to determine the success and efficacy (or failure) of these restoration efforts. This office investigates 500-600 vessel groundings per year on coral reef and seagrass habitats within the FKNMS. The Coral 312 Program, consists of assessing damage to reefs by ships and providing technical information on adjudicated responsibility/liability against the person/company who damaged the benthic resources. This office also conducts emergency triage for damaged coral and on-site restoration, which is funded by proceeds from successful litigation related to the damage. The type of equipment/methods used to assess damage and rehabilitate corals are: visual, photo and video surveys and diver measurements of damage patterns. Damage patterns are quantified by divers and through using aerial imagery, surface (National Geodetic Survey's Shallow Water Positioning System) and underwater (CobraTac/AquaMap) GPS surveys. Video mosaics of the reef resources monitored have been developed using the commercial software RavenView. However, this product only creates strip mosaics with limited spatial accuracy. In the future, the restoration office wants to improve the efficiency of in-water surveys and the quality of the products produced for damage recovery and monitoring purposes.

NOAA Damage Assessment and Restoration - Mr. Bill Precht

Under the Marine Sanctuaries Act, NOAA's Florida Keys National Marine Sanctuary, collects data on the health of coral reefs and uses these data to support managerial and policy decisions on reef and fisheries conservation. Information on the Threatened *Acropora* species is of specific concern. The FKNMS currently partners with academia, other governmental entities, and NGO's, including but not limited to the University of North Carolina, Wilmington, RSMAS; other NOAA groups and sanctuary monitoring groups; Florida Marine Research Institute's Coral Reef Evaluation and Monitoring Project (FMRI CREMP); Mote Marine Laboratory, Dauphin Island Sea Lab/Florida Institute of Oceanography; and the Nature Conservancy. Monitoring techniques vary from group to group and program to program.

The UNCW rapid reef survey methodology consists of trained observers using stationary diver surveys to identify, count, and measure reef fish populations. In addition, trained divers survey the benthic community and mobile invertebrates using visual, photo, and video methods. The metrics collected include abundance, diversity, condition (partial mortality), and size of all benthic invertebrates and macroalgae, as well as reef rugosity. This program is based on a stratified random survey design and has conducted surveys at over 900 sites Sanctuary-wide within the last decade.

The CREMP reef survey protocol consists of collecting point-count data from permanent sites throughout the FKNMS and more recently Dade and Broward Counties. This project has been on-going since 1996. The biggest limitation of the CREMP effort is that field campaigns occur only once a year, limiting the ability to make interpretations on the impacts of acute disturbances such as bleaching events, hurricanes, and disease outbreaks.

Mote Marine Lab, in collaboration with the FKNMS, collects information on bleaching patterns using visual surveys and satellite information. Researchers from FIO/Dauphin Island Sea Lab conduct visual coral monitoring and develop population trend models in Sanctuary Preservation Areas (SPAs). The Nature Conservancy is currently using monitoring data as the basis for developing reef resilience strategies within the Sanctuary.

The Nature Conservancy- Mr. Chris Bergh

The Nature Conservancy coordinates the Florida Reef Resilience Program (FRRP) and the FRRP's Disturbance Response Monitoring (DRM) effort for shallow coral reefs of the Florida Keys and southeast Florida. TNC is concerned with the conservation of coral reefs and the impact of declining reef health on other natural and human communities. The focus is on resilience of the reefs to bleaching/disease events. Their work is facilitated through partnerships with collaborators such as NOAA, Florida Department of Environmental Protection, Florida Fish and Wildlife Conservation Commission, Universities (University of Miami, Nova Southeastern University, Florida Institute of Technology), Mote Marine Laboratory, and World Wildlife Fund. The FRRP methodology is based on a stratified random allocation of sampling sites in unique subregions and zones of the Florida reef ecosystem that are surveyed yearly at the peak of the summer high temperatures (August-September). Coral communities are surveyed by trained divers using visual methods (line and belt transects). The information collected includes coral cover, colony sizes, partial mortality, and prevalence of bleaching and diseases. The data collected are archived in an on-line database for report generation. The largest limitation that TNC has to contend with is that surveys need to be designed to respond to disturbances other than bleaching and disease (e.g., algal blooms, hurricanes and coldwater events). TNC is planning workshops in 2009 to address program shortcomings.

(Full presentations can be found in Appendix C.)

SERDP TECHNOLOGY DESCRIPTIONS AND DEMONSTRATIONS

Mosaicing- University of Miami - Dr. Pamela Reid, RSMAS

Efficient survey methodologies that provide comprehensive assessment of reef condition are fundamental to coral reef monitoring. Current state-of-the-art techniques in coral reef assessment rely on highly trained scientific divers to measure indices of reef health (e.g., substrate cover, species richness, coral size, coral mortality). First-generation video mosaics developed by Reid's team were an innovative survey technology that provided large-scale (up to 400 m^2), spatially accurate, high-resolution

images of the reef benthos without extensive survey times or a need for scientific divers. Despite these advances, the first-generation mosaic products were insufficient for species-level identification of many benthic taxa, thereby limiting the monitoring potential of the technique. Therefore, a second-generation mosaic survey technology was developed by Reid's team, integrating high-resolution still-image acquisition with high-definition video surveys of the reef benthos. The second-generation mosaic products have sub-millimeter benthic resolution, allowing for species identification of coral colonies as small as 3 cm, identification of macroalgal genera, and increased information on coral colony health and small scale competitive interactions. This advanced survey technology allows users to collect imagery on both a landscape and colony level over 100's of square meters in under an hour of in-water dive time. The resulting product has excellent archive potential and is a superior tool for tracking changes over time.

Mosaicing Demonstrations- University of Miami Team - Dr. Nuno Gracias

A fundamental building block of the mosaic creation process is image matching, which corresponds to detecting the same area of the benthos in two different images. Image matching allows for estimating the relative displacement of one image with respect to the other.

The mosaicing algorithm starts by performing image matching over the sequence of images in temporal order, since time consecutive images have maximum overlap. Next, an attempt is made to match images that are not sequential in time. Each successful image match provides a geometric constraint between two images. If enough constraints are found, then a set of images can be geometrically arranged to form a mosaic. The information from all image matches is used in a non-linear least square algorithm which finds the joint displacement of all images that best fits all the geometric constraints. Finally the images are blended to create a large composite view of the sea floor.

The current software uses the MATLAB computing environment, and can create mosaics of thousands of images with minimal user intervention and effort. User input is handled with easy-to-use graphical user interfaces. The software consists of the following modules:

1. Image extraction and correction – Allows for retrieving images from a video and correcting for lens and housing distortion.
2. Global matching – Performs image matching and estimates registration for all frames.
3. Manual inspection and correction – Allows for detailed inspection and additional user input on image registration for difficult images.
4. Image blending - Combines registered frames into a single mosaic.
5. Mosaic viewing - Allows point and click access to individual frames.

In addition to the basic mosaic creation capability, four enhanced capabilities have been created and demonstrated previously at a proof-of-concept level. These four capabilities have been streamlined and integrated into the mosaic software package:

1. Combining video with high resolution still photos - Increases spatial resolution of the mosaics, thereby improving taxonomic resolution;
2. Using additional positioning information – Improves geometric accuracy of the mosaics specially over high topography areas;
3. Improved blending – Reduces the visibility of the seams among neighboring images when rendering the final mosaics;
4. Removing refracted sunlight – Strongly attenuates or eliminates the disruptive patterns of refracted sunlight for very shallow water surveys.

The most practical approach for transitioning the mosaicing technology to end users is under consideration. One approach would be to publish the existing MATLAB code and user manuals. The limitation of this method is that there is no infrastructure in place to provide the pre-release software engineering (bug testing, error reports, unified GUI, installation scripts, etc.) or the customer service support that would be expected if this product were to become a fully developed commercial software package. A second approach would be to run a service bureau to produce mosaics for end users. Under this model, users would submit their imagery to a central facility and receive a mosaic in return; the software itself would not be released as a product. The limitation of this method is that a certain minimum demand for mosaics would be needed to sustain the facilities of a service bureau.

FIRe technology - Rutgers University - Dr. Max Gorbunov and Dr. Paul Falkowski

Development of advanced technologies for environmental monitoring and assessment of coral reef communities requires an understanding of how different environmental factors affect the key elements of the ecosystems and the selection of specific monitoring protocols that are most appropriate for the identification and quantification of particular stressors. The Rutgers team developed a Fluorescence Induction and Relaxation (FIRe) technique for assessing the health and viability of corals. The FIRe instrument illuminates an organic tissue with precisely controlled flashes of light and measures the amount of fluorescence response that comes back. The fluorescence levels can vary, based on environmental conditions and the presence or absence of a stressor(s), thus acting as an indicator of the health of the organism. The FIRe-retrieved physiological parameters include the quantum yields of fluorescence at the minimum and maximum levels (F_o and F_m, respectively), the efficiencies of photosynthetic energy conversion (F_v/F_m), the functional absorption cross section of Photosystem II, the rates of photosynthetic electron transport, photosynthetic turnover time, and coefficients of photochemical and non-photochemical quenching. Because the technique records an extensive suite of physiological parameters, there is a possibility to identify what stressor is involved and to distinguish between common natural stresses (e.g, thermal stress or photoinhibition) and anthropogenic stressors, such as metal toxicities. The measurements are sensitive, fast, non-destructive, can be done in real time and *in situ*.

The Rutgers team has designed and developed a set of FIRe instruments, including a bench-top FIRe fluorometer, diver-operated fluorometer, and moorable fluorometer. This instrumentation is used together with standard laboratory methods (lipid and protein analysis, molecular biology, microscopy, and fluorescence spectroscopy) and provides a comprehensive physiological diagnostic tool. The FIRe technology has been employed for basic research of the physiological responses of coral to natural stresses (thermal stress, photoinhibition, nutrient load) and to selected anthropogenic stressors such as metal toxicity. The research revealed that the developed diagnostics are very sensitive to changes in the coral physiology and records detrimental changes at early stages of the stress development - before any visible changes in coral coloration appear. On this background, algorithms are developed for identification of environmental stressors.

The photosynthetic efficiency (Fv/Fm) is the primary stress indicator. Healthy corals have Fv/Fm of about 0.50. Stressors usually lead to a decrease in Fv/Fm, with the exception of nutrient load that may increase Fv/Fm. Thermal stress is triggered by a 1-2 ^{o}C increase in temperature above its normal maximum and varies greatly between coral species. Research has revealed that the coral sensitivity to thermal stress is controlled by the lipid composition of photosynthetic membranes. Specifically, thermally resilient clones have a lower relative content of the major polyunsaturated fatty acid that simultaneously reduces the susceptibility of the membrane lipids to attack by Reactive Oxygen Species. The thermal stress leads to a characteristic decrease in both Fv/Fm and the rates of photosynthetic electron transport down Photosystem II (PSII). Photoinhibition also leads to a decrease in Fv/Fm ratio, but has no effect on the photosynthetic electron transport in PSII reaction centers. The target of thermal stress and photoinhibition is the primary photosynthetic reactions in PSII.

Metal toxicity analyses have shown that metals (copper, zinc, lead, and tin) inhibit growth rates but do not change the efficiency of the primary photosynthetic reactions at early stages of the stress development. Metals do, however, affect the photosynthetic turnover times and the maximum rates of photosynthetic electron transport. Therefore, secondary photosynthetic reactions are affected, but not the primary photosynthetic reactions, that is in striking contrast to common natural stressors. Metal poisoning also causes an increase in caspase activity (an indicator of program cell death) and tissue degenerations, thus suggesting damage to both coral host and algal symbionts.

FIRe Demonstration– Rutgers Team

The FIRe technology records the dynamics of fluorescence yields on the micro- to millisecond time scale, with the overall time of a single measurement of about 1 second. Because coral communities are non-uniform and show a high degree of spatial variability, even within a single colony, several readings on the same corals are taken, at different spots on a particular coral head. Acceptable repeatability is achieved with this technique. In the field, several readings on the same corals are taken, at different spots on a particular coral head. The prototype diver-operated system has a viewing screen so that the diver can determine in real-time if the fluorescence value is outside the normal range

of response. The diver then can take a sample for further analysis during that collection opportunity. For example, this technique can result in a reduction in cost when studying heavy metal contamination and impact and also can realize a reduction in the number of sites needing to be sampled.

The FIRe onboard computer conducts the measurements in fully automatic regime and performs initial data analysis in real time. The data are stored and downloaded after a dive. The dedicated data analysis software package fits the fluorescence profiles to a bio-physical model to retrieve physiological parameters of the organism. Rutgers has established a database of fluorescence response baseline data for corals from various locations in the Carribbean and Indo-Pacific regions. Also there is baseline data for a variety of stressors, such as copper, zinc, lead, and temperature. In the future, the Rutgers Team plans on writing algorithms to relate stress levels with the database of known stressors.

Integration of the Two Systems (FIRe and Mosaics)

One of the goals of this workshop was to gather information and identify how the two SERDP approaches/technologies might be complementary to each other and/or how they might be integrated to meet end-user needs. The challenge for the integration of the video mosaics and the FIRe technology is the different spatial scales at which these two systems presently work. The FIRe instrument collects physiological information at the cm-scale while the video mosaics, even with sub-mm pixel resolution, provide information at the plot scale (up to 500 m^2). Moreover, the data for the FIRe system are presently collected at short distance (< 5 cm from the surface of the target), while the video data required to build video mosaics are collected at 1.5 - 2 meters above the surface of the reefs. The future integration of these two systems will depend on the development of a FIRe instrument that is able to sample at larger distances from the surface of the reef and a system that synchronizes the collection of physiological and video data so that each fluorescence measurement is correlated spatially and visually with a position and organism within the landscape mosaic.

Although these technical challenges will remain in place until the technologies are further developed, the potential benefits of an integrated system were outlined in the workshop. <u>The added benefit of combining both methods in a single platform would be the identification of areas mosaiced within wide scale plots of reefs that are subject to declining coral health and may be moving toward future mortality or reduced growth.</u> This would help concentrate efforts on areas with higher risk of mortality and document resilient patches within communities. A joint platform would also enhance the ability to survey deeper reefs with reduced dive time.

GROUP DISCUSSION

Current Practices (Agency presentations)

Discussion based on Agency Presentations indicated a consensus that current monitoring and assessment strategies conducted by the agencies are, in general, adequate to meet present mandates with regards to coral reef monitoring. Desired capabilities that would expand present survey methodologies and specific challenges were also discussed. The issue of the high cost and safety related to field operations (e.g., boats, trained divers, deep diving) is of concern to all parties involved in coral monitoring. Therefore, development of streamlined and efficient survey methodologies that reduce dive and field time was recognized as a significant need. The need for techniques providing repeatable data acceptable to all agencies involved was also emphasized.

Limitations that constrain current monitoring as assessment efforts were discussed, and include the following:

1) Limited sampling frequency that precludes the assessment of cause and effect relationships of coral decline patterns
2) A lack of coordination and inconsistency of methodologies that precludes data from being fully shared by programs and agencies
3) Various agencies which are charged with the monitoring and protection of multiple habitats and jurisdictions, spreading the resources dedicated to coral reefs very thin.
4) A large degree of redundancy with several agencies surveying the same areas with limited communication.
5) A lack of uniform methods and sharing of resources leading to a general lack of efficiency.
6) A lack of explicit monitoring and assessment needs and a priori goals resulting in inadequate data being collected (data that do not answer the questions posed by the programs).
7) Monitoring and assessment requirements that have not been well-defined before the methods and the survey technology are chosen.
8) The idea that monitoring and assessment are two different topics and should not necessarily be considered unified efforts.
9) The need for a methodology that minimizes time-at-site while providing a wide range of detailed coral health metrics.
10) Different agencies have different goals/missions (drivers), therefore it would be difficult for one or even two technologies to fit all programs.
11) Science does not presently drive management policies with respect to coral reefs. A science-based approach is needed to address the optimal integration of survey methods and technologies.
12) A report card framework for coral reefs is needed, focusing overall ecosystem assessment, the role of reefs, and consequences of reef degradation.
13) The lack of forecasting tools, such as what might be addressed in part by the FIRe technology, also is a limitation of current practices. Development and implementation of technologies for assessing the physiological status of coral

with capabilities to detect detrimental change to the coral health at early stages should be an important component of coral monitoring programs.

Potential Utility of SERDP Technologies

Participants were in agreement regarding the potential usefulness of both the mosaicing and the FIRe technologies for advancing monitoring and assessment practices of the coral reef community. There was consensus that transition of both technologies to the end-user community would be valuable. Specific comments and suggestions included the following:

Mosaics offer unique opportunities for collecting and analyzing long-term monitoring data, developing new indicators of reef health, and contributing to other applications such as use in UXO munitions management and public outreach efforts. Future generations of still cameras will offer even higher-image capture rates that may enable mosaicing without the use of video cameras. One limitation of the mosaicing technology are that the cameras are downward looking, so objects under overhanging features will be obscured. In addition, the current resolution of the mosaics limits species identification to corals larger than 2 cm. However, there was general agreement that the mosaicing technology was ready for transition to the user community.

Participants were enthusiastic about the potential application of FIRe technology for identifying coral stressors. The suggestion was made that it would be useful to develop libraries to aid interpretation of the FIRe data, and to conduct lab work to determine inter and intra species variability, and diel fluctuations with the FIRe system. There was also interest in looking at the differences within a single colony based on the position of the light and probe. The participants were polled to find out what kind of stressors the users thought would be important to explore next. Coral diseases and petrochemicals were suggested, participants also pointed out a need to investigate signals from a combination of stressors. The intent was to focus on petrochemicals as the last specified Navy stressor of interest and (2) based on workshop participant input and concern about the synergistic/canceling effect of multiple stressors evaluate a mixed stressor signal (e.g, nutrient load in combination with thermal stress).

Consensus was also reached that the two technologies are indeed complementary and that integration could be implemented in the short term with existing (but separate platform) capabilities of the individual projects. Further joint development should be undertaken if system limitations relating to the differences in distance at which data is collected and spatial recording of the FIRe data within a mosaic can be overcome. Mr. Precht suggested conducting large-scale surveys with FIRe and mosaics aimed at detecting spatial stressor “hot spots”. It was noted that the FIRe technology would benefit from further field demonstration before it is put on a platform alongside the mosaicing cameras.

Technology Overlay and Potential Collaborations

The potential for the two SERDP technologies to augment and enhance the specific reef monitoring and assessment activities of the participating agencies was

discussed at length. Agency-specific input is outlined below and summarized in Table 1. Column 1 of Table 1 lists the Governmental and Non Governmental Agencies represented by workshop presenters and other participants. Columns 2, 3 and 4 are color coded to indicate potential contributions of mosaics (green), FIRe (yellow) or both technologies (purple) to augment or enhance monitoring of present metrics (Column 2), enable new desired capabilities (Column 3), or provide new opportunities for partnerships (Column 4). Text in Column 2 identifies indices of reef health presently monitored by each agency that could benefit from the use of mosaics and/or FIRe. Text in Column 3 identifies desired enhanced monitoring capabilities that could be accomplished using mosaics and/or FIRe. Column 4 summarizes potential collaborations using mosaics and/or FIRe. Appendix D contains the details of the information provided by presenters at the SERDP Coral Monitoring Workshop.

Table 1. Technology overlay and potential collaborations. See text for details.

Potential roles for: Mosaics FIRe Mosaics & FIRe

Agency	Current Metrics / Indicators	Desired Capabilities	Collaborations
Presentors			
US Navy	coral cover, colony size, disease & bleaching, mortality	increased sampling efficiency; multi-tier approach; safe, efficient, cost effective; digital technology;	
MMS	coral cover, diversity, coral growth		reef condition & coral growth, Flower Gardens
NPS	coral cover, disease & bleaching	detailed mapping; disease causes and infection research	coral monitoring program, Biscayne National Park
FWS	coral cover, diversity, disease & bleaching	improved monitoring techniques and technologies	monitoring at remote refuges, e.g. Palmyra Island
EPA	coral cover (2D & 3D), colony size, disease & bleaching, mortality, population structure	expand surveys to include additional benthic organisms and determine links to stressors	investigate sampling efficiency in different environments (mosaics); assess coral viability, identify stressors, and collect database of FIRe signatures
NOAA Fisheries	coral cover, colony size, disease & bleaching, mortality	reduce dive time and expand surveys to deep reefs (e.g., *Oculina* banks)	survey deep coral communities (i.e., *Oculina* banks) and thickets of thretened corals (i.e., *Acropora*)
NOAA Coastal Monitoring and Assessement	biomarkers, coral diseases	rapid, effective, non-destructive methods to evaluate coral physiological condition	use in-situ FIRe in combination with chemical, microbiological & biomarker sampling to assess coral response to mixed stressors.
NOAA Sanctuaries	injury & % cover at damaged and reference sites, recovery patterns	increase survey spped; reduce need for trained divers; improvements relative to strip mosaics	groundings assessment, FKNMS
NOAA Restoration	coral colony size, partial mortality, disease & bleaching % cover, urchin abundance	survey deep reefs (> 30 m)	survey coral reef and seagrass sites to compare survey methods, products, & cost effectiveness
TNC	coral cover, disease & bleaching, colony size, mortality	identify causes of changes in coral condition and demographics occurring between annual sampling events	continue ongoing collaboration with the Florida Reef Resilence Program
Other participants			
US Navy AUTEC			continue ongoing collaboration mosaicing permanent monitoring sites
NCRI			address pixel mixing problem in airborne remote sensing images
Florida Sea Grant			surveys at CREWS/ICON stations

POTENTIAL COLLABORATIONS WITH OTHER AGENCY PRESENTERS

Minerals Management Service: Mr. Sinclair expressed interest in the SERDP-funded video mosaicing technology because of its high resolution capability, the ability to survey deeper communities with reduced dive time, and the capability of providing a permanent visual record (i.e., high-resolution maps of the bottom). Potential collaboration to use video mosaics to evaluate coral reef condition and colony growth in the Flower Gardens was discussed.

National Park Service: Dr. Ruttenberg indicated that both the video mosaics (mapping, assessment) and fluorescence (disease and bleaching impacts) were potentially useful techniques that could be incorporated into a comprehensive coral reef monitoring program by NPS. Video mosaics were collected by the University of Miami team at St. Croix in collaboration with NPS staff in 2007. The potential for future integration of video mosaics in the coral monitoring program at Biscayne National Park was mentioned.

U.S. Fish & Wildlife Service: Dr. Wolfe indicated that FWS does not conduct its own monitoring and relies on partnerships with other agencies to fulfill its coral reef monitoring mandate. Potential collaborations with the SERDP-funded technologies would have to be conducted through FWS' partners (EPA, NOAA, USGS, etc.). Interest was expressed in conducting joint surveys incorporating mosaics and FIRe in remote refuges such as Palmyra Island.

U.S. Environmental Protection Agency: Dr. Fisher pointed out the potential for using mosaics to conduct statistical power analyses to determine sampling efficiency and change-detection levels in different environments. The University of Miami team has previously worked with Dr. Fisher and the EPA to conduct parallel surveys at one site surveyed regularly by a EPA coral disease research group to determine if the metrics obtained from both surveys were similar. Dr. Fisher also identified the FIRe technology as a potentially useful tool to develop early-warning indicators of reef degradation in watersheds affected by multiple stressors. He expressed interest in working with Rutgers to use FIRe for assessing coral viability and stressor identification and suggested monitoring rates of benthic primary production in lab experiments and in the field. Dr. Fisher also expressed interest to use FIRe technology for monitoring other organisms including macroalgae and phytoplankton.

NOAA Southeast Fisheries Science Center: Dr. Miller and other researchers from NOAA SEFSC have collaborated extensively with the University of Miami team, using video mosaics in the assessment of disturbance patterns to populations of the threatened coral *Acropora palmata* in the Florida Keys. Dr. Miller also recognized the potential for utilizing the FIRe method as an early warning indicator of coral diseases and bleaching impacts. Dr. Miller suggested further collaboration using mosaics for joint surveys of deep coral communities (i.e., *Oculina* banks), with a possible CRTF proposal.

NOAA Center for Coastal Monitoring & Assessment: Mr. Warner highlighted the potential benefits of including the FIRe technique in the assessment of chemical pollution and early impacts on exposed corals. He invited the Rutgers team to participate in a field campaign that involves fine-scale sampling of a well characterized coral reef ecosystem. Mr. Warner suggested using *in-situ* FIRe measurements in combination with chemical, microbiological and biomarker sampling to assess how corals respond to a mix of

environmental stressors, including thermal stress. Mr. Warner also discussed the potential benefits of incorporating video mosaics as a survey and mapping tool.

NOAA Marine Sanctuaries: Mr. Goodwin indicated that the University of Miami team has collaborated with NOAA on the survey of a vessel grounding scar in Biscayne National Park and that future joint assessments are planned to incorporate the video mosaic technique into the assessment of groundings within the FKNMS.

NOAA Damage Assessment and Restoration: Mr. Precht discussed the potential for using video mosaic capabilities for CREMP permanent sites and collaborating with the FKNMS in the monitoring the status and trends of threatened *Acropora* population using both video mosaics and FIRE techniques. He suggested performing a side-by-side comparison of survey methods, products, and cost effectiveness between NOAA and University of Miami groups.

The Nature Conservancy: In 2008, TNC established a collaboration with the University of Miami team to use video mosaics to monitor and map coral colonies within permanent sites. The data to be collected at these permanent sites will be used to quantify the impacts of bleaching and diseases on coral populations. Mr. Bergh and Dr. Kramer indicated interest in continuing collaboration between University of Miami and the Florida Reef Resilience Program.

Additional Workshop Participants

AUTEC: Mr. Tom Szlyk from the Navy's AUTEC Range indicated that The Atlantic and Gulf Rapid Reef Assessment (AGRRA) protocol has been used on a yearly basis in the recent past to assess the status and trends of coral reef communities at Andros Island. This methodology uses visual surveys conducted by trained divers to record cover of benthic organisms, colony sizes, partial mortality patterns, prevalence of bleaching and diseases, abundance of urchins, and surface topography. In the past several years, the SERDP-funded mosaic technology has been integrated into the reef survey protocol at Andros and mosaics have been used to map and monitor coral communities at more than twenty permanent sites around the AUTEC base. Mr. Szlyk indicated that due to the sampling interval (once a year) disturbance events such as disease outbreak and bleaching may be missed. The University of Miami team will continue ongoing collaboration at AUTEC with Mr. Szlyk and Mr. Marc Cimenello. Mr. Don Marx (NAVFAC ESC) brought up the importance of making sure that any data produced by the technologies developed under SERDP would be accepted by regulatory agencies.

NOVA Southeastern University/NCRI Center. Researchers from NOVA conduct regular assessment of reefs in Broward County Florida using a combination of visual surveys and remote sensing technologies (LIDAR, Multibeam, Satellite Imagery). Dr. Purkis identified the mosaic technology as a potential methodology for providing accurate ground-truthing of satellite imagery for the development of benthic habitat maps and to address the issue of within-pixel mixing of satellite imagery. A potential collaboration with the University of Miami group was discussed within the context of surveying dense patches of the threatened coral *Acropora cervicornis* in Broward County.

Florida SeaGrant. Ms. Fletcher indicated that the mosaicing and FIRe technologies are both potentially beneficial for assessing the status and trends of deep coral reefs and cultural resources (e.g., coral communities on ship wrecks, archeological digs). As a science outreach coordinator, Ms. Fletcher also recognized the tremendous potential of using landscape video mosaics as display and education tools. Potential collaborations using mosaics and FIRe were suggested for sites in Florida and La Parguera, PR where CREWS/ICON stations are deployed.

SUMMARY and RESULTS

The workshop defined the DoD client perspective on coral reef assessment and monitoring needs. Federal policy mandates that DoD characterize, assess, and monitor underwater benthic communities at Air Force, Army, and Navy facilites and ranges in order to document compliance with national policy and to ensure that DoD operations do not lead to natural resource degradation, particularly with respect to coral reefs. As a participant in the U.S. Coral Reef Task Force (CRTF), DoD is interested in developing efficient survey methodologies that provide a comprehensive assessment of reef conditions. Specifically, the Navy is looking for technologies and methodologies that will enable the collection of data with less dive time, reproduce data collection transects reliably year after year, and retain flexibility to be modified based on expert evaluation of site conditions at the time of the survey. DoD is need of a rapid deployment capability to document coral reef groundings. DoD is also interested in exploring how emerging technologies may foster new opportunities to develop productive partnerships between the Department of the Navy and other organizations.

The workshop also examined methodologies and needs of other agencies with mandates for coral reef monitoring and assessment. Participants were in agreement that current monitoring and assessment strategies conducted by the agencies are, in general, currently adequate to meet present mandates. There was broad interest from all agencies in developing methodologies that reduce dive time, increase cost efficiency and provide repeatable data. Specific challenges and enhanced capabilities that would expand present methodologies were also discussed, especially a projected need to expand coral reefing monitoring to the ecosystem level, highlighting detailed mapping with improved accuracy compared to strip (1D) mosaics, monitoring deep reefs, assessing cause and infection patterns of coral disease, providing non destructive methods for determining coral physiology and support for preemptive risk evaluation of coral reef health.

The two recently developed techniques for coral reef monitoring, landscape mosaics and fluorescence induction relaxation techniques (FIRe), were introduced to project participants. Presentations and demonstrations outlined the capabilities of these techniques, and the potential integration of the two technologies. Workshop participants were in agreement regarding the potential usefulness of both technologies for advancing monitoring and assessment practices of the coral reef community. <u>In particular, consensus was reached that both techniques offer potential for more efficient methods of monitoring coral cover, colony size, mortality, bleaching and disease, population structure, extent of injury and recovery patterns, and documentation of coral reef ecosystem metrics. There was also consensus that transition of both technologies to the end-user community would be valuable.</u>

Participants expressed opinions that mosaics offer unique opportunities for collecting and analyzing long-term monitoring data and for developing new indicators of coral reef health. The mosaics were considered superior tools for damage assessment and public outreach efforts. It was also suggested that the mosaicing could play an important role in the issue of shallow water munitions management for unexploded ordnance. There was also general agreement that the mosaicing technology is ready for transition to the user community and paths for commercialization were discussed. One strategy under consideration is to license the technology to a commercial software company such that individuals could buy software to produce their own mosaics. An alternative plan would be to commercialize a service under which mosaics would be produced on a fee-per-mosaic basis. Participants generally seemed to favor Option 2, but recognized that an informed decision would require a cost benefit analysis.

Participants were enthusiastic about the potential application of FIRe technology for identifying coral stresses. Suggestions were made regarding the need to develop libraries to aid in the interpretation of the FIRe data and to conduct lab work to determine inter and intra species variability, diel fluctuations and looking at the differences within a single colony based on position of the probe and light when using the FIRe technology. It has also been suggested that the FIRe technology could be employed and validated at non-DoD test sites with a known stressor environment, e.g., at a NOAA sites in Puerto Rico. Follow-on work for the FIRe technology will consist of focusing on petrochemicals as the last specified Navy stressor of interest and investigating the synergistic/canceling effect of multiple stressors, e.g. nutrient load and thermal stress.

Participants also indicated that regulatory stakeholder agencies would have to agree that this technology possesses the potential to become a mutually acceptable component of their surveys, as both technologies are different from what is currently being accepted as the standard. Coinciding with that challenge is the matter of making technologies as user-friendly as possible or at least providing a practical ability for general field marine ecologists to learn and operate the system(s). Regulatory acceptance could be addressed through the ESTCP Program by involving regulators in field demonstrations.

The overall consensus was that the two technologies are complementary, but not necessarily synergistic, to each other. Integration of the two technologies onto a single platform could be useful in the future to some in the user community, but, in the short term, integration would not be necessary to benefit from the capabilities of the separate technologies when deployed separately. Future integration efforts would benefit from additional lab/field work to develop libraries to aid the interpretation of the FIRe data. There was commentary that separate system development may be as useful as integrated system development.

A matrix was developed based on workshop presentations and discussion illustrating how user-defined coral reef monitoring and assessment needs can be met by the two SERDP-developed technologies (Appendix D). This matrix indicates the potential contributions of mosaics, FIRe, or both technologies to facilitate or improve present monitoring methodologies, enable new capabilities, and provide opportunities for new partnerships.

APPENDICES

Appendix A- List of Participants

Name	Title	Affiliation	Address	City/State	Phone	E-mail
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Dr. Margaret Miller	Ecologist	NOAA- Fisheries	NOAA-Fisheries, Southeast Fisheries Science Center 75 Virginia Beach Dr.	Miami, FL	305-361-4561 x 561	margaret.w.miller@noaa.gov
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Mr. Humberto Guarin	Marine Operations	Rosenstiel School of Marine and Atmospheric Science, University of Miami	223 SLAB	Virginia Key , FL	305-361-4716	hguarin@rsmas.miami.edu
Mr. Don Marx	Marine Ecologist	NFESC	6506 Hampton Blvd	Norfolk, VA	757-322-4376	donald.marx@navy.mil
Mr. John Noles	Environmental Planner	Naval Facilities Engineering Service Center, Atlantic	6506 Hampton Blvd	Norfolk, VA	757-322-4891	john.noles@navy.mil

Name	Title	Affiliation	Address	City/State	Phone	E-mail
Mr. Thomas Szlyk	Staff Environmental Engineer	Naval Undersea Warfare Center Detachment AUTEC	801 Clematis Street	West Palm Beach, FL	(561) 832-8566, Ext. 7249	thomas.szlyk@autec.navy.mil
Mr. Marc Ciminello	Staff Environmental Engineer	Naval Undersea Warfare Center Detachment AUTEC	801 Clematis Street	West Palm Beach, FL	561-832-8566	marc.ciminello@autec.navy.mil
Mr. Bill Wild	Environmental Scientist	Space & Naval Warfare Systems Center - Pacific	71750 (PL-BS) SPAWARSYSCEN - Pacific 53475 Strothe Rd	San Diego, CA	619-553-2781	bill.wild@navy.mil
Dr. Ken Richter	Oceanographer	Space & Naval Warfare Systems Center - Pacific	KENNETH E. RICHTER 71750 (PL-BS) SPAWARSYSCEN 53560 HULL STREET	San Diego, CA	619-553-2780	ken.richter@navy.mil
Ms. Cheryl Kurtz	Marine Ecologist	Space & Naval Warfare Systems Center San Diego	CHERYL A. KURTZ 71750 (PL-BS) SPAWARSYSCEN 53560 HULL STREET	San Diego, CA	619-553-5313	cheryl.kurtz@navy.mil
Ms. Kristen Lau	OSD Staff to Dr. Hall	HydroGeoLogic Inc.	11107 Sunset Hills Road, Suite 400	Reston, VA	703-326-7830	klau@hgl.com
Dr. John Hall	Sustainable Infrastructure Program Manager	DOD - SERDP/ESTCP	901 N. Stuart St., Suite 303	Arlington, VA 22203-1853	703-696-2125	john.hall@osd.mil

Appendix B- Final Agendas

SERDP Coral Reef Monitoring & Assessment Workshop

Day 1- Tuesday November 18, 2008

Agenda

Time	Description	Presenter
8:30 – 8:45 AM	Welcome & Introductions	Bill Wild, Navy: SPAWAR Pacific
8:45 – 9:00 AM	SERDP/ESTCP: Program Overview and Sponsor Role	Dr. John Hall, OSD: SERDP/ESTCP
9:00 – 9:30 AM	DoD Client Perspective	Ms. Lorri Schwartz (for Mr. Tom Egeland), Office Assist. Sec. of Navy for Installations & Environment
9:30 – 9:40 AM	Agency/Organization Perspectives	Mr. James Sinclair, Minerals Management Service
9:40 -9:50 AM		Dr. Matt Patterson, National Parks Service
9:50 – 10:00 AM		Bret Wolfe, Fish & Wildlife Service
10:00 – 10:10 AM		Dr. William Fisher, Environmental Protection Agency
10:00 – 10:20 AM	Q/A for previous 5 speakers	All
10:20 – 10:35 AM	**Break**	
10:35 – 10:45 AM	NOAA Perspectives	Dr. Margaret Miller, NOAA SE Fisheries
10:45 – 10:55 AM		Rob Warner, NOAA Center for Coastal Monitoring & Assessment (CCMA) Biogeography Branch
10:55 – 11:05 AM		Bill Goodwin, NOAA Marine Sanctuaries
11:05 – 11:15 AM		Dr. Bill Precht, NOAA Damage Assessment and Restoration
11:15 – 11:35 AM	Q/A for previous 4 speakers	All
11:35 – 12:35 AM	Group discussion 1: Monitoring/Assessment Needs	Bill Wild, Navy: SPAWAR Pacific
12:35 – 1:35 PM	**Working Lunch (continued discussion)**	
1:35 – 2:20 PM	University of Miami Research (includes 10–15 minutes Q/A)	Dr. Pamela Reid, Univ. Miami
2:20 – 3:05 PM	Rutgers University Research (includes 10–15 minutes Q/A)	Dr. Max Gorbunov and Dr. Paul Falkowski, Rutgers University
3:05 – 3:15 PM	Integration of UM/Rutgers Technologies	Dr. Diego Lirman, Miami Dr. Max Gorbunov, Rutgers
3:15 – 3:30 PM	**Break**	
3:30 – 5:00 PM	Group discussion 2: Overlay of Monitoring/Assessment Needs with Miami/Rutgers Technologies	Bill Wild, Navy: SPAWAR Pacific
5:00 PM	**Adjourn**	
7:30 PM	**Group dinner at Jaguar Ceviche Spoon & Latam Grill**	

SERDP Coral Reef Monitoring & Assessment Workshop

Day 2- Wednesday November 19, 2008

Agenda

Time/ Location	Description	Presenter
9:00 – 9:10 Library	Agency/Organization Perspectives	Chris Bergh, The Nature Conservancy (Florida Reef Resilience Program)
9:15 – 10:15 AM/ Library	Lab Demonstrations	Rutgers and Miami Teams
10:15 – 10:30/ Library	**Break**	
10:30 AM – 12:30 PM/ NORTH GROSVENOR- 3rd Floor	Hands-on processing and applications	Rutgers and Miami Teams
12:30 PM – 1:30 PM	**Lunch**	
1:30 – 3:15 PM/ Library	Synthesis and Collaboration: Discussion/Overlay of SERDP Technologies	Wild/Reid/Gorbunov
3:15 – 3:30 PM/ Library	Closing Remarks	Dr. John Hall, OSD: SERDP/ESTCP
3:30 PM	**Adjourn**	

SERDP Coral Reef Monitoring & Assessment Workshop

Day 2- Wednesday November 19, 2008

Agenda

Time/ Location	Description	Presenter
9:00 – 9:10 Library	Agency/Organization Perspectives	Chris Bergh, The Nature Conservancy (Florida Reef Resilience Program)
9:15 – 10:15 AM/ Library	Lab Demonstrations	Rutgers and Miami Teams
10:15 – 10:30/ Library	**Break**	
10:30 AM – 12:30 PM/ NORTH GROSVENOR- 3rd Floor	Hands-on processing and applications	Rutgers and Miami Teams
12:30 PM – 1:30 PM	**Lunch**	
1:30 – 3:15 PM/ Library	Synthesis and Collaboration: Discussion/Overlay of SERDP Technologies	Wild/Reid/Gorbunov
3:15 – 3:30 PM/ Library	Closing Remarks	Dr. John Hall, OSD: SERDP/ESTCP
3:30 PM	**Adjourn**	

Appendix C - Agency Perspective Presentations

Strategic Environmental Research and Development Program

Coral Reef Workshop
University of Miami
RSMAS
18-19 Nov, 2008

Systems Center PACIFIC

Two New Developmental Efforts

- University of Miami
  - High Resolution Landscape Mosaics for Coral Reef Mapping and Monitoring
- Rutgers University
  - Analysis of Biophysical, Optical and Genetic Diversity of Coral Reef Communities using Advanced Fluorescence and Molecular Biology Techniques

Group Discussion #1
Monitoring/Assessment Needs

- Overlaps and gaps
  - What are the methodologies and technologies currently being used by the speakers and what technology gaps/limitations do they face?
- Summarizing and Prioritizing the assessment needs
  - How do these organizations foresee themselves conducting coral reef monitoring in the future?

Group Discussion #2
Overlay of Monitoring/Assessment Needs with Technologies

- Summary spread sheet of agency input
  - data collected: geo Location; spatial extent; depth range; temporal frequency; metrics / indicators; gaps / limitations.
- Overlay of SERDP technologies
  - shows how the mosaics, fluorescence, and the integration of these two will help fulfill the user needs.
- Application of integrated mosaic-fluorescence data
  - where can the integrated technology be used?
  - potential demonstration sites

GROUP DISCUSSION #3 Synthesis and Collaboration:

- Understanding the DoD client perspective on assessment and monitoring needs
- Understanding other potential user perspectives (i.e., in addition to DoD) on what their coral reef monitoring and assessment needs are and how these two SERDP-developed technologies may help address those needs
- Identifying how the two approaches/technologies are complementary to each other and how they can be integrated to meet end-user needs.

SERDP/ESTCP
Program Overview and Sponsor Role

Dr. John A. Hall
Sustainable Infrastructure Program Manager
SERDP/ESTCP

SERDP Coral Reef Monitoring & Assessment Workshop
November 18, 2008

Strategic Environmental Research and Development Program (SERDP)

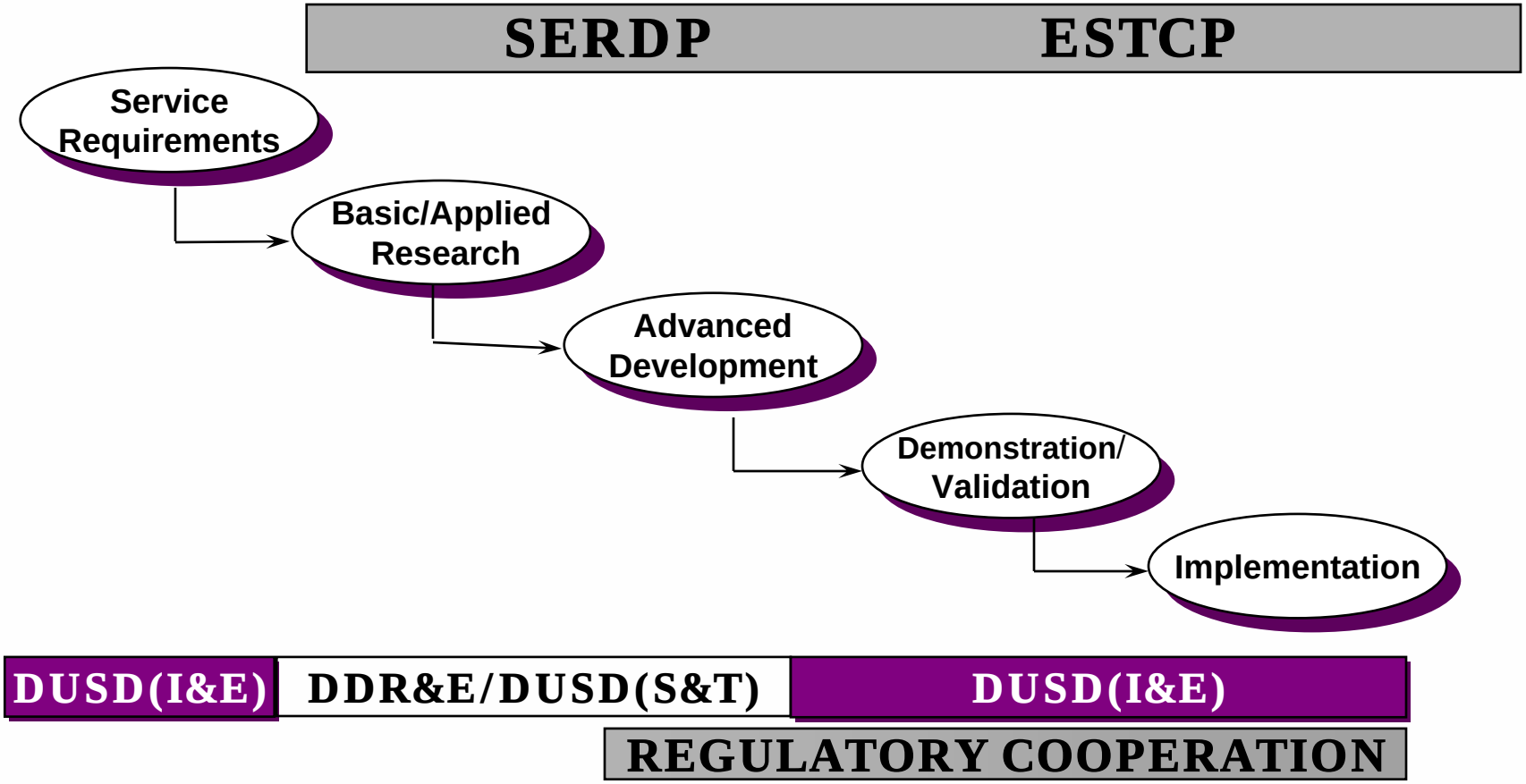
- Established by FY 1991 Defense Authorization Act
  - DoD, DOE, and EPA partnership

- SERDP is a requirements driven program that:
  - Responds directly to user requirements generated by the Services
  - Identifies high-priority, DoD environmental science and technology needs or investment opportunities that address these requirements

Environmental Security Technology Certification Program (ESTCP)

- Established in 1995
- Demonstrate innovative and cost-effective environmental methodologies and technologies
  - Capitalize on past investments
  - Transition methods and technology out of the lab and field
  - Validate operational cost and performance
- Promote implementation
  - Identify DoD user community
  - Satisfy users by direct application at a DoD facility/site
  - Gain regulatory acceptance
  - May lead to technology transfer outside of DoD

Environmental Science and Technology Development Process



Focus Area Management Structure

Munitions Management

Weapons Systems & Platforms

Environmental Restoration

Sustainable Infrastructure

Sustainable Infrastructure (SI)

- **Natural Resources**
- Cultural Resources
- Facilities
- Energy

Natural Resources Sub-Focus Area

- Future Areas of Emphasis/Initiatives
  - Ecological Forestry
  - Arid Lands Ecology and Management
  - Pacific Island Ecology and Management
  - Coastal/Estuarine Ecology and Management
  - Living Marine Resources Ecology and Management
  - Species Ecology and Management
    - TER-S
    - Invasive Species
  - Watershed Processes and Management
  - Climate Change Impacts and Adaptation

Living Marine Resources Ecology and Management

- Marine mammal population and habitat modeling
- Effects of naval sound on marine mammals
- Coral reef monitoring and assessment

SERDP Coral Reef Projects

- SI-1333 High Resolution Landscape Mosaics for Coral Reef Mapping and Monitoring (Universit of Miami)
- SI-1334 Analysis of Biophysical, Optical, and Genetic Diversity of Coral Reef Communities Using Advanced Flourescence and Molecular Biology (Rutgers University)

SERDP Objectives for the Workshop

- Understand the DoD client perspective on coral reef assessment and monitoring needs.
- Understand other potential user perspectives (beyond DoD) on needs and how the two currently funded SERDP projects (SI-1333 and SI-1334) may help address those needs.
- Identify how the two project approaches/ technologies are complementary to each other and how they can be integrated to meet end-user needs.

SERDP Solicitation Process

- Annual Solicitations to Meet DoD Needs
  - Two Solicitations (Core and SEED)
  - Open to All: Government, Academia, Industry
- Competitive Award
  - External Peer Review
  - Internal and Scientific Advisory Board Review
- Transition to Demonstration/Validation

ESTCP Solicitation Process

- Annual Solicitations
  - Topic areas (BAA) for non-DoD leads
  - Mature methodologies and technologies for DoD leads
  - Natural resource and energy topic areas started in FY08
  - Identify DoD liaison for BAA proposals
- Competitive Process
  - Pre-proposal
  - Full proposal
  - Oral presentation
  - Program Office and ESTCP Technical Committee review/down-selects throughout

General Solicitation Timelines

- SERDP
  - Annual Solicitation - November
  - "SEED" Solicitation – November
  - Selection in June/July
  - SAB Reviews in September/October

- ESTCP
  - Annual Solicitation - January
  - Selection in September

Getting the Details

- SERDP: www.serdp.org
- ESTCP: www.estcp.org
- Online Library: http://docs.serdp-estcp.org/
  - Final Reports
  - Fact Sheets
  - Cost and Performance Reports
- TER-S Regional Workshops
  - www.serdp.org/tes

SERDP Coral Reef Monitoring and Assessment Workshop

DoD Client Perspective

Mr. Tom Egeland
ODASN (E)

November 18 and 19, 2008

DoD Mission & Policy

Mission: To provide the military forces needed to deter war and to protect the security of our country.

Policy: Sustain healthy natural resources for future generations while fulfilling the mission.

DoD Authorities

- Authorized to manage natural resources on property under its control.
- Major drivers are Sikes Act, Clean Water Act, Clean Air Act, Marine Mammal Protection Act, Endangered Species Act, and various Executive Orders including EO 13089 for Coral Reef Protection.
- Coral reefs resources given special protection in internal policy, directive and instruction.

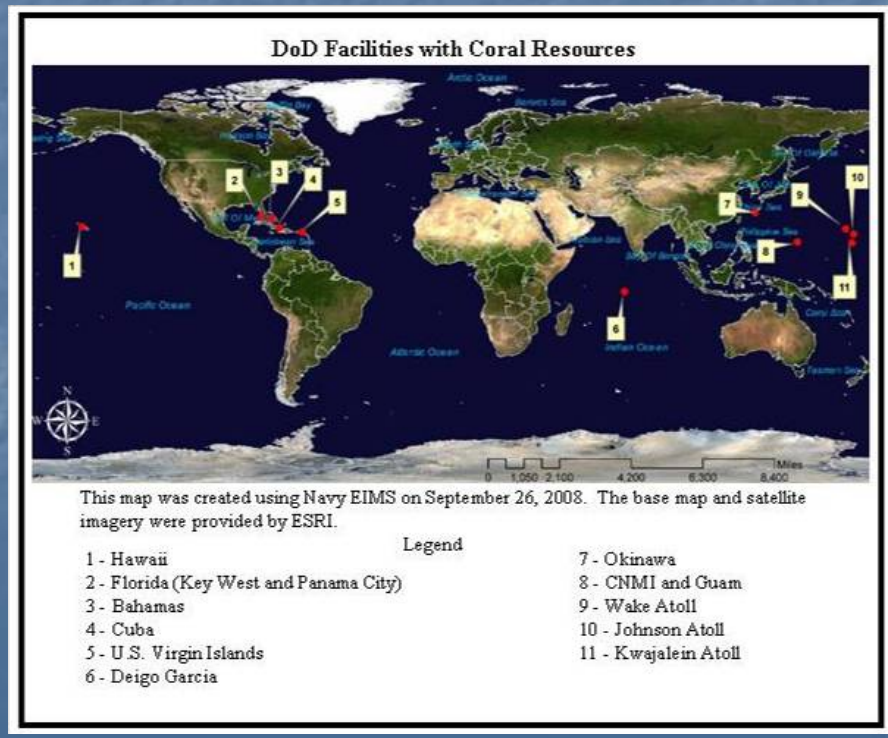
DoD Conservation Instruction 4715.3

- Sustain access for military training and testing at DoD facilities while ensuring that the natural and cultural resources are preserved for future generations.

DoD Resource Stewardship

- DoD physical plant consists of more than 571,200 facilities (buildings, structures and utilities) located on more than 3,700 sites, on nearly 30 million acres.

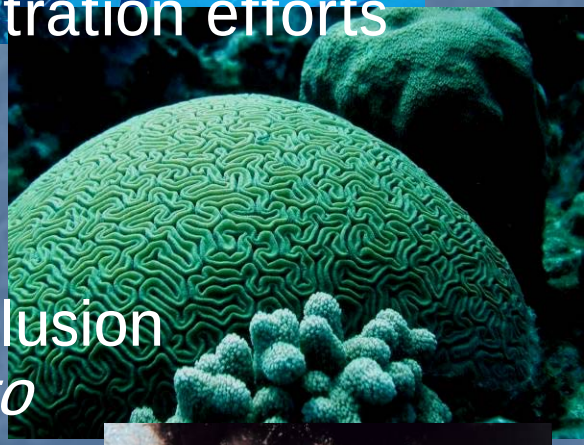
- Locations with coral resources include:
  - Commonwealth of Northern Marianas Islands
  - Wake Island
  - Johnston Island
  - Kwajalein Atoll
  - Guam
  - Hawaii
  - Okinawa
  - Diego Garcia
  - Andros Island, Bahamas
  - Cuba
  - U.S. Virgin Islands
  - Key West and Panama City, FL



DoD Programs & Projects

Natural Resources Conservation Programs

- Resource management and protection integrated into all aspects of DoD operations
- Compliance Programs
- Pollution Prevention Programs
  - P-2 Afloat (Navy)
  - Plastics Removal in Marine Environment (Navy)
- Programs to fund research and demonstration efforts
  - SERDP
  - NESDI
  - Legacy
- Positive resource management plus exclusion of other resource users leads to *de facto* preserves at DoD facilities
  - Vieques Island, Kingman Reef and Palmyra Atoll now managed as marine sanctuaries



DoD Programs & Projects

Marine Resource Protection Projects

- Reference database for Resource Managers containing scientific literature about DoD coral reef sites
- Sinking the retired aircraft carrier ex-ORISKANY for an artificial reef
- Clean up of tires from failed artificial Osborne Reef
- Beach clean-up efforts in Hawaii and other locations
- Active coral reef ecosystem protection through NEPA, assessment and monitoring
- Active development of research/demonstration projects related to coral reefs

DoD Statement of Need for SERDP Technologies

- Efficient assessment of benthic habitats to support routine activity planning
  - Reduced time and expense for data collection
  - Reasonable operator experience and dive time requirements
  - Experts spend more time in lab analyzing data than in field collecting data

- Data quality to support compliance requirements now and near-future
  - Support Habitat Equivalency and NEPA analyses
  - Coral Reef Protection Act reauthorization
  - Broadly accepted methodology for mapping, assessment and i*n-situ* coral reef health monitoring
  - Data/image archival capability, data compatibility with existing software

DoD Coral Reef Assessment and Monitoring

- Rapid survey/assessment
  - Reduce cost, dive time for each agency
  - Retain key strengths of a diver-based approach
  - Overcome the limitations of diver-based or photo-quadrat/video transect methods
- Example DoD projects with potential benefit
  - Fort Kamehameha Outfall
  - Kilo Wharf Extension
  - Guam expansion
- Other regulatory needs
  - Section 404/401 permits
  - Standard assessment methodology

DoD Cooperation and Partnerships

- Data and methods should facilitate interoperability between DoD components and cooperation with other Federal and State agencies
  - Widely accepted assessment model
  - Trusted QA/QC procedures
  - Military digital data requirements
- New technologies should facilitate partnerships for research and development
  - Mutual benefit to use same tools
  - Low cost, high benefit
  - Potential to leverage research needs

DoD Client Perspective

U.S. Department of the Interior
Minerals Management Service

MMS Securing Ocean Energy & Economic Value for America

Protection and Monitoring of Reef Communities in the Gulf of Mexico

James Sinclair, Marine Biologist, MMS Gulf of Mexico

Environmental Mission of the MMS

- As a part of Department of Interior, Minerals Management Service is committed to ensuring a safe environment.
- Oversees the safe and environmentally sound exploration and production of our Nation's offshore mineral resources.
- To manage the mineral resources on the outer continental shelf in an environmentally sound and safe manner.

History of Protection

- No Activity Zone: March 1974
  - 100 m isobath
  - No oil and gas activity
- 1-Mile Zone: 1975
  - Shunting all drilling muds and cuttings to within 10 m of the bottom
  - Monitoring the effects of operations on biota of the banks
- 3-Mile Zone: 1975 – shunting required
- 4-Mile Zone: by 1983 – shunting required
- Long-Term Monitoring replaced industry monitoring in 1988

MMS Securing Ocean Energy & Economic Value for America

The MMS Role in Protecting Reefs

- Regulation of oil and gas activities on the outer continental shelf
- Federal waters
- Connected infrastructure and support

Regulations to Protect Reefs

- Live Bottoms
  - Low Relief
  - Pinnacle Trend
- Potentially Sensitive Biological Features (PSBF's)
- Topographic Features
- Chemosynthetic Communities

MMS Securing Ocean Energy & Economic Value for America

Flower Garden Banks Monitoring

- Random video transects (16)
- Repetitive quadrat photos (8 m2) (40)
- Lateral growth photos (*Diploria strigosa*) (60)
- Perimeter video (200 m)
- Urchin and lobster surveys. (200 m)
- Continuously recording water quality instrumentation (temperature, salinity, pH, turbidity). Water sampling and water column profile measurements. Nutrient analyses.
- Fish surveys (radius of 7.5 m each) (24)

MMS Securing Ocean Energy & Economic Value for America

Monitoring Needs

- Mapping to identify habitats
- Characterize habitats
- Updated baseline data

MMS Securing Ocean Energy & Economic Value for America

U.S. Department of the Interior
Minerals Management Service

MMS Securing Ocean Energy & Economic Value for America

?

James.Sinclair@mms.gov

South Florida and Caribbean Perspectives to Coral Reef Monitoring and Assessment

Matt Patterson, NPS SFCN Network Coordinator
Dr. Benjamin Ruttenberg, SFCN Marine Ecologist

Outline

1. Identify your agency/organization’s role and objectives with respect to reef monitoring and assessment.

2. Summarize methodologies/technologies used by your agency for coral reef monitoring and assessment.

3. Describe how your current monitoring and assessment approaches meet or do not meet your needs.

4. Identify and prioritize unmet monitoring and assessment needs.

5. Identify any plans your agency/organization has to improve its approaches to monitoring and assessment.

Vital Signs Monitoring Networks

South Florida/Caribbean Network

Florida/Caribbean Office (FLACO)

SFCN Vital Signs Monitoring Plan

National Park Service
U.S. Department of the Interior

Natural Resource Program Center

South Florida / Caribbean Network
Vital Signs Monitoring Plan – Phase 3
DRAFT

Natural Resource Report NPS/SER/SFCN/NRR—2007/001

Big Cypress National Preserve
Biscayne National Park
Buck Island Reef National Monument
Dry Tortugas National Park

Everglades National Park
Salt River Bay National Historical Park and Ecological Preserve
Virgin Islands National Park

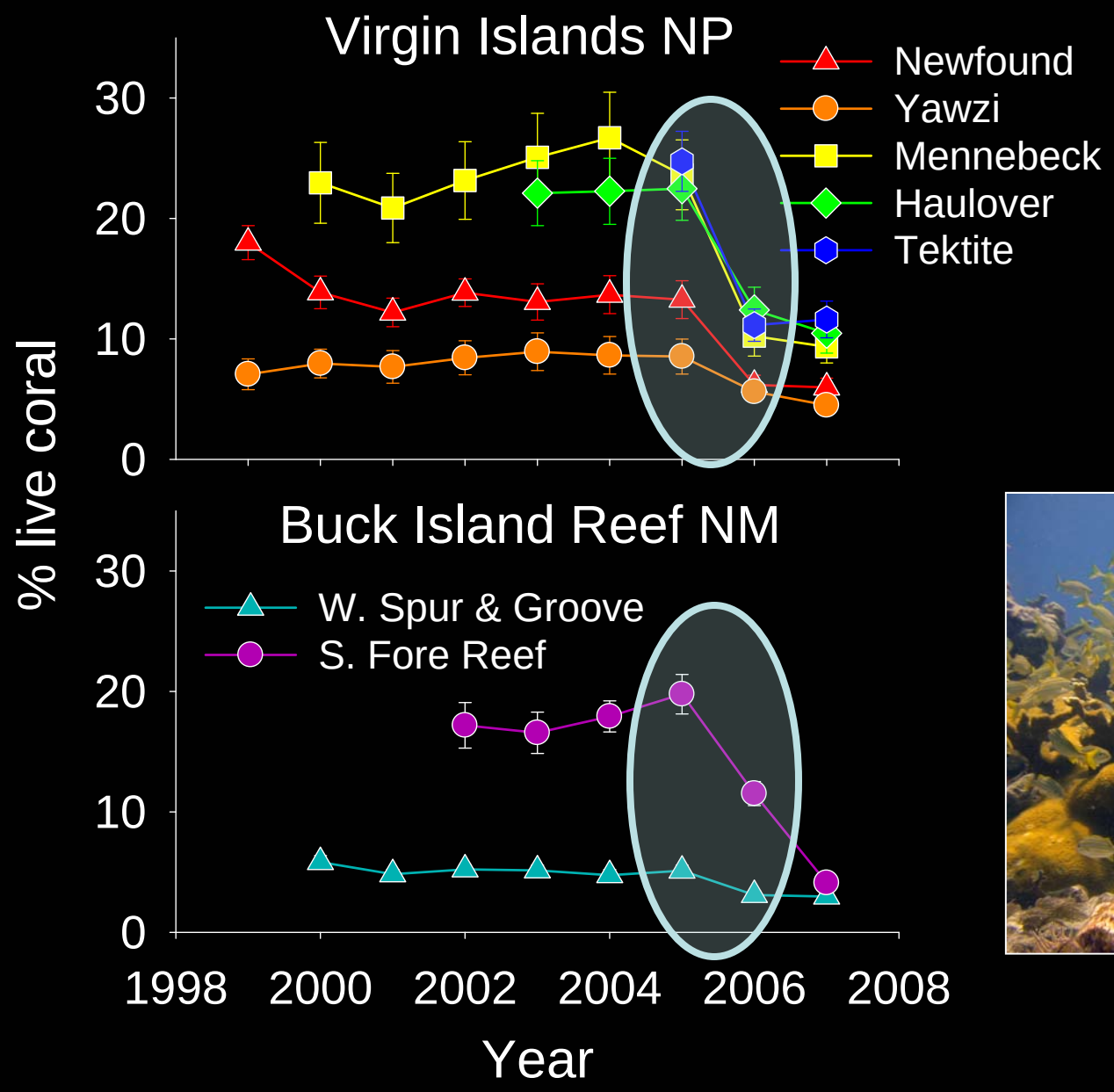
Group	SFCN Core Vital Signs
Marine	Marine Benthic Communities
	Marine Fish Communities
	Marine Exploited Invertebrates
Inter-tidal and above	Colonial Nesting Birds
	Wetland Ecotones and Community Structure
	Forest Ecotones and Community Structure
	Mangrove-Marsh Ecotone
	Freshwater fish and large macro-invertebrates
	Amphibians

Marine Benthic Communities (=coral reefs)

- Most previous work in USVI
- Annual monitoring of coral reef communities
- Expanded to include specific sites in all 4 parks

2. Summarize the methodologies and technologies currently used by your agency/organization for coral reef monitoring and assessment.

- Annual video transect surveys
- Grab and analyze still images
- Data: % cover of benthic functional groups, *Diadema,* T^{o} and coral disease
- Index sites
  - 20 10m permanent transects
  - 5 in STJ, 2 each in Buck Island, Dry Tortugas and Biscayne
- Extensive sites
  - 4 10m permanent transects per site
  - 18 sites in DRTO


Virgin Islands NP
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Yawzi
Mennebeck
Haulover
Tektite
Buck Island Reef NM
W. Spur & Groove
S. Fore Reef
% live coral
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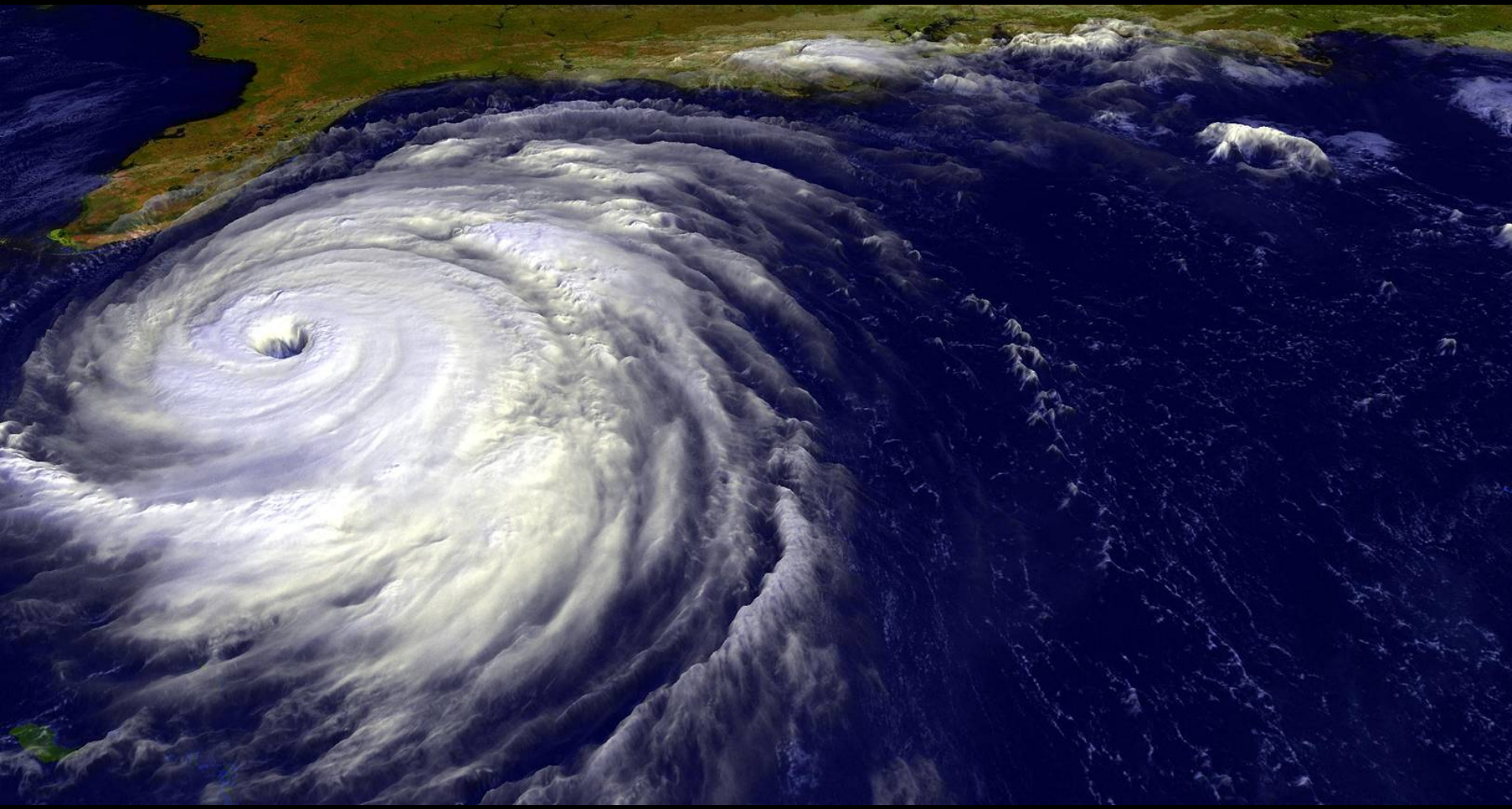

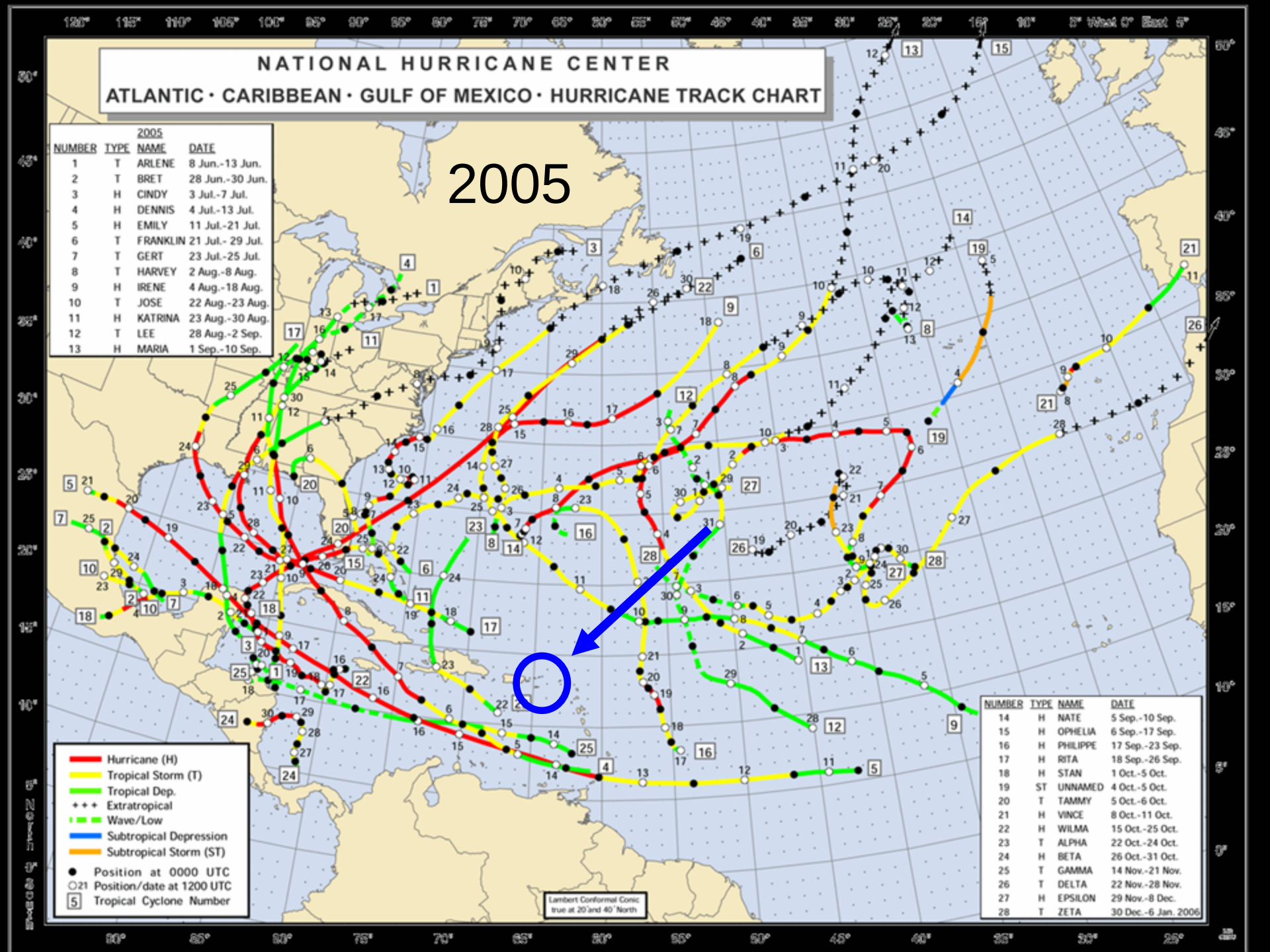
NATIONAL HURRICANE CENTER
ATLANTIC · CARIBBEAN · GULF OF MEXICO · HURRICANE TRACK CHART
2005
2005
NUMBER TYPE NAME DATE
1 T ARLENE 8 Jun.-13 Jun.
2 T BRET 28 Jun.-30 Jun.
3 H CINDY 3 Jul.-7 Jul.
4 H DENNIS 4 Jul.-13 Jul.
5 H EMILY 11 Jul.-21 Jul.
6 T FRANKLIN 21 Jul.- 29 Jul.
7 T GERT 23 Jul.-25 Jul.
8 T HARVEY 2 Aug.-8 Aug.
9 H IRENE 4 Aug.-18 Aug.
10 T JOSE 22 Aug.-23 Aug.
11 H KATRINA 23 Aug.-30 Aug.
12 T LEE 28 Aug.-2 Sep.
13 H MARIA 1 Sep.-10 Sep.
NUMBER TYPE NAME DATE
14 H NATE 5 Sep.-10 Sep.
15 H OPHELIA 6 Sep.-17 Sep.
16 H PHILIPPE 17 Sep.-23 Sep.
17 H RITA 18 Sep.-26 Sep.
18 H STAN 1 Oct.-5 Oct.
19 ST UNNAMED 4 Oct.-5 Oct.
20 T TAMMY 5 Oct.-6 Oct.
21 H VINCE 8 Oct.-11 Oct.
22 H WILMA 15 Oct.-25 Oct.
23 T ALPHA 22 Oct.-24 Oct.
24 H BETA 26 Oct.-31 Oct.
25 T GAMMA 14 Nov.-21 Nov.
26 T DELTA 22 Nov.-28 Nov.
27 H EPSILON 29 Nov.-8 Dec.
28 T ZETA 30 Dec.-6 Jan. 2006
Hurricane (H)
Tropical Storm (T)
Tropical Dep.
Extratropical
Wave/Low
Subtropical Depression
Subtropical Storm (ST)
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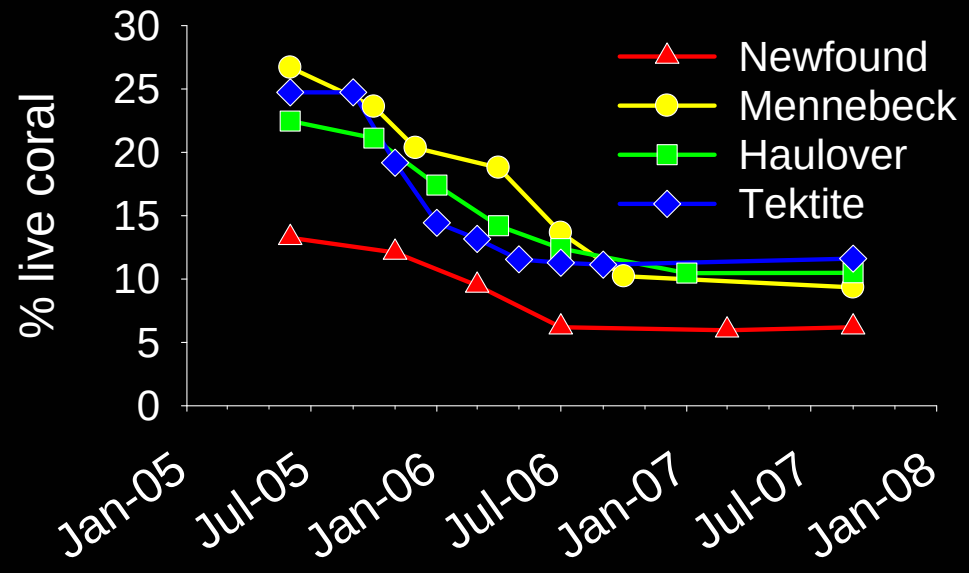
21 Position/date at 1200 UTC
5 Tropical Cyclone Number
Lambert Conformal Conic
true at 20°and 40° North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Jan-08
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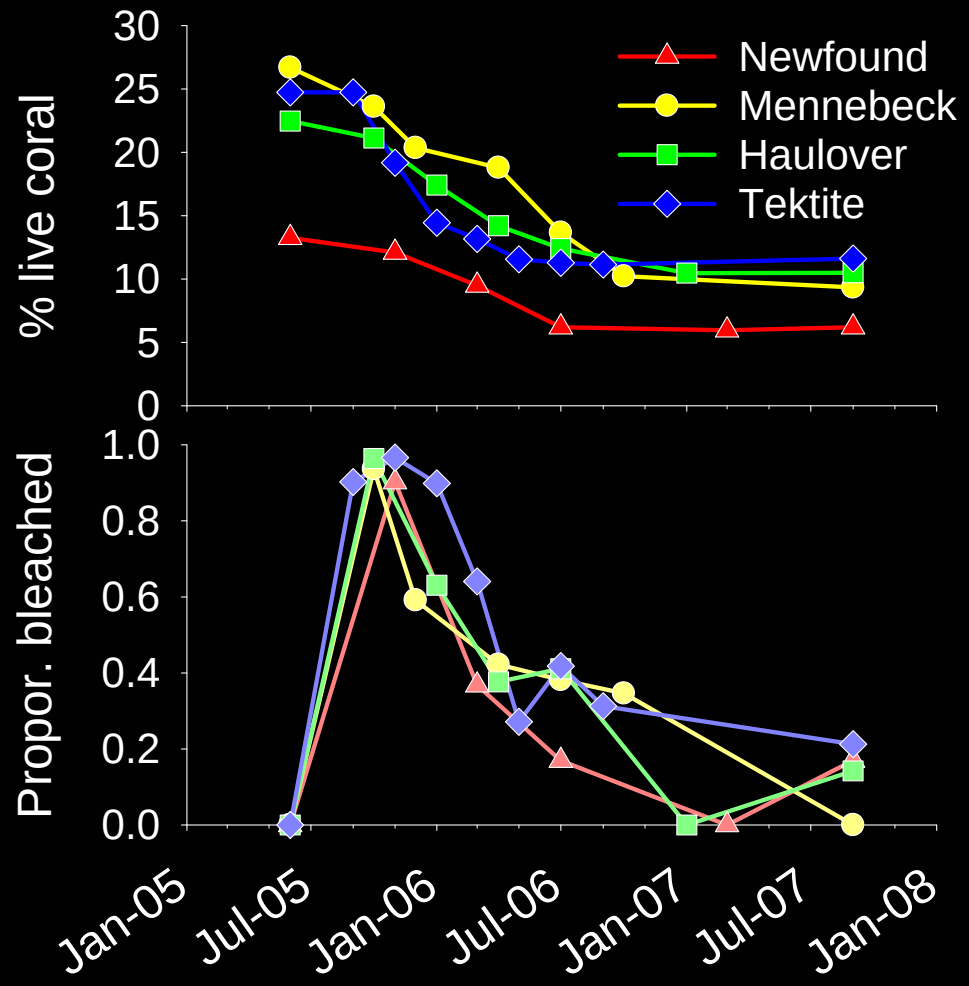
Mennebeck
Haulover
Tektite

St. John

Tektite Reef
Haulover Reef
Mennebeck Reef
Newfound Reef

St. John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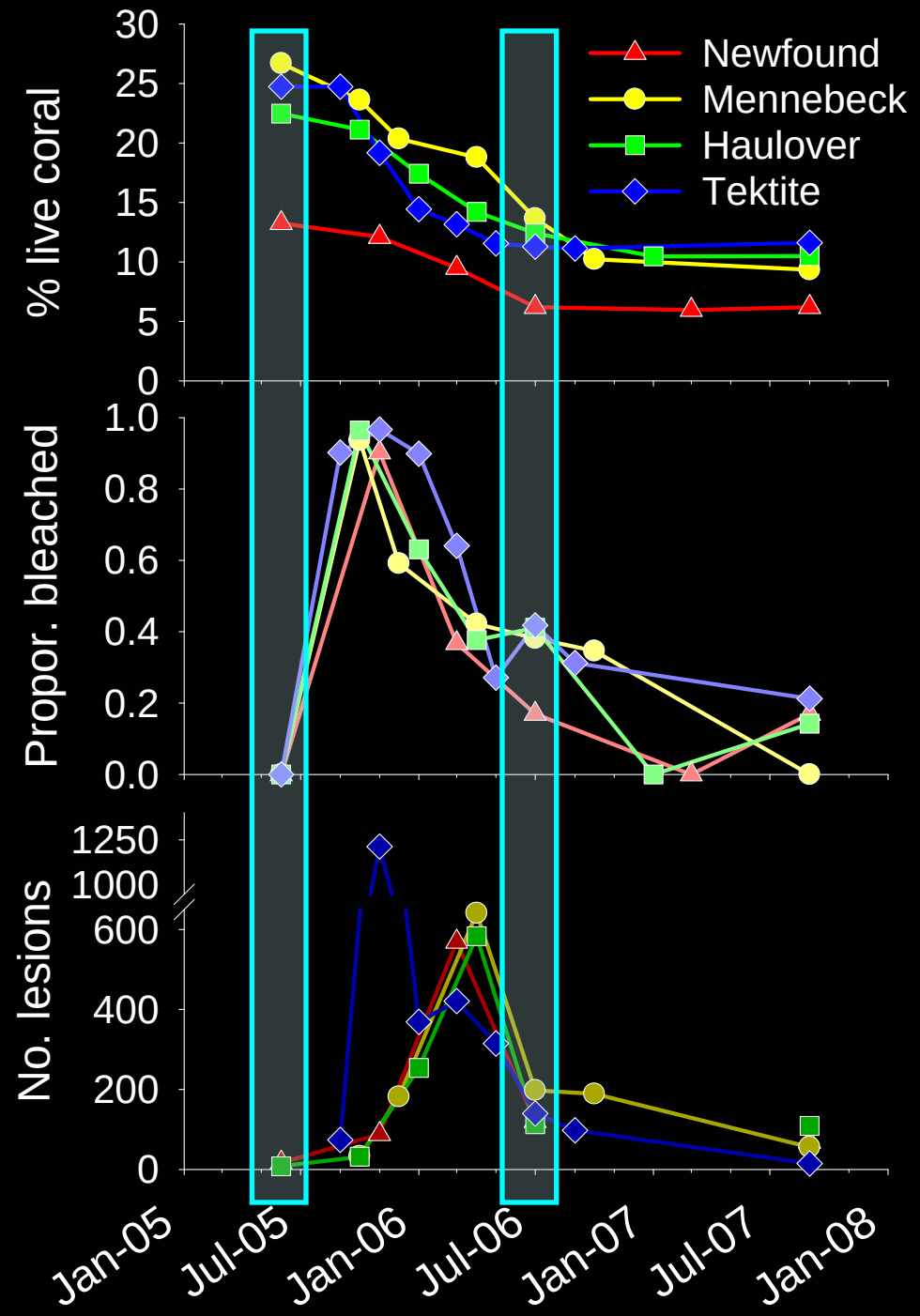
Jan-08

Tektite Reef
Haulover Reef
Mennebeck Reef
Newfound Reef

St. John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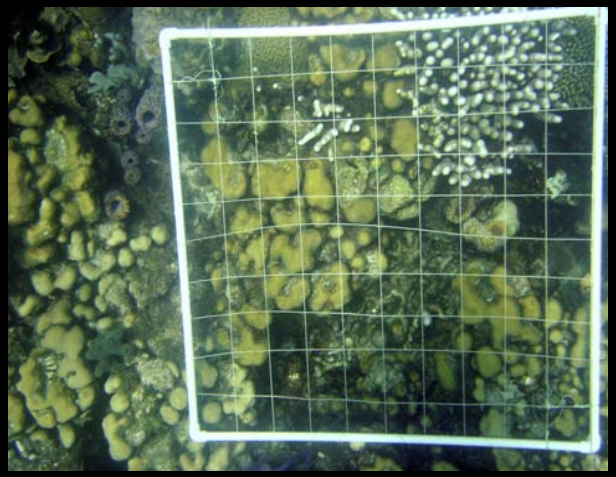
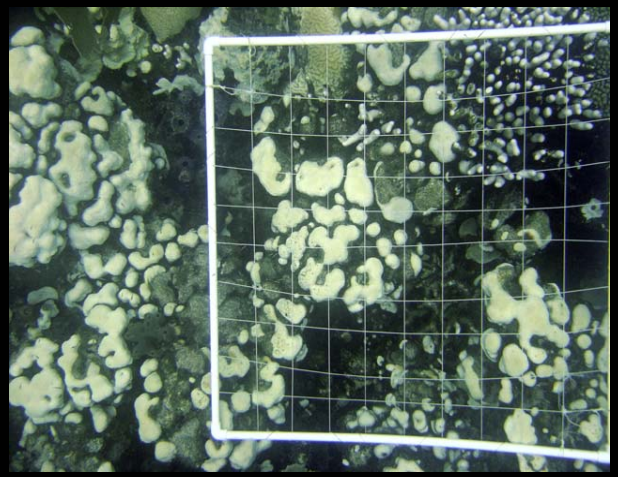
Jan-08

Tektite Reef
Haulover Reef
Mennebeck Reef
Newfound Reef



August 2005



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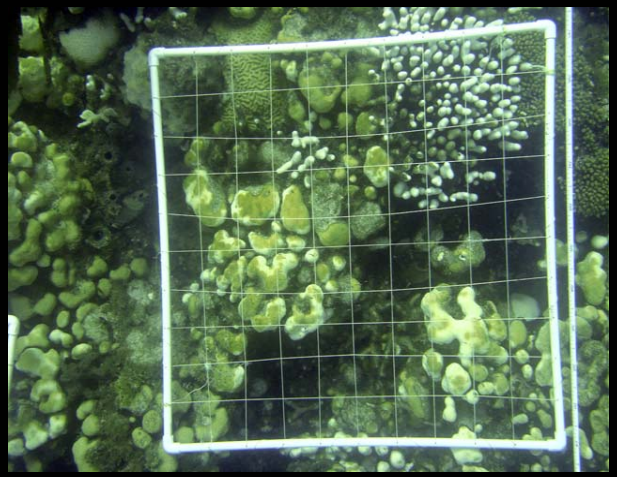
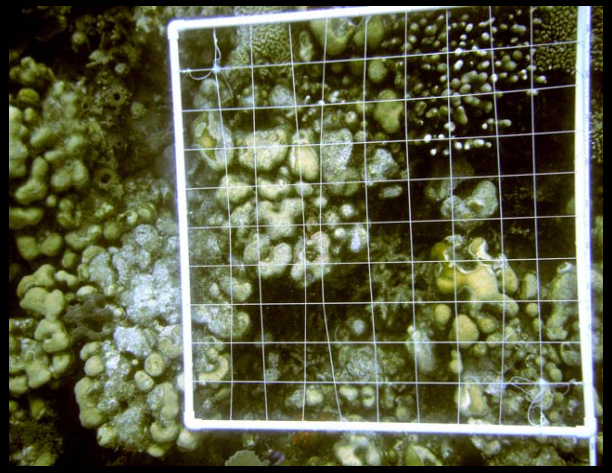
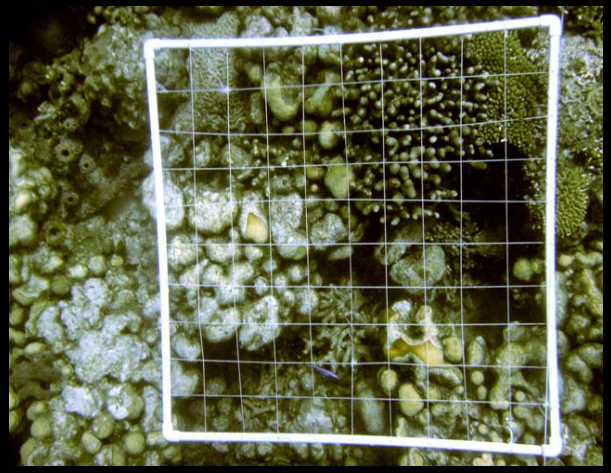


October 2005

November 2005



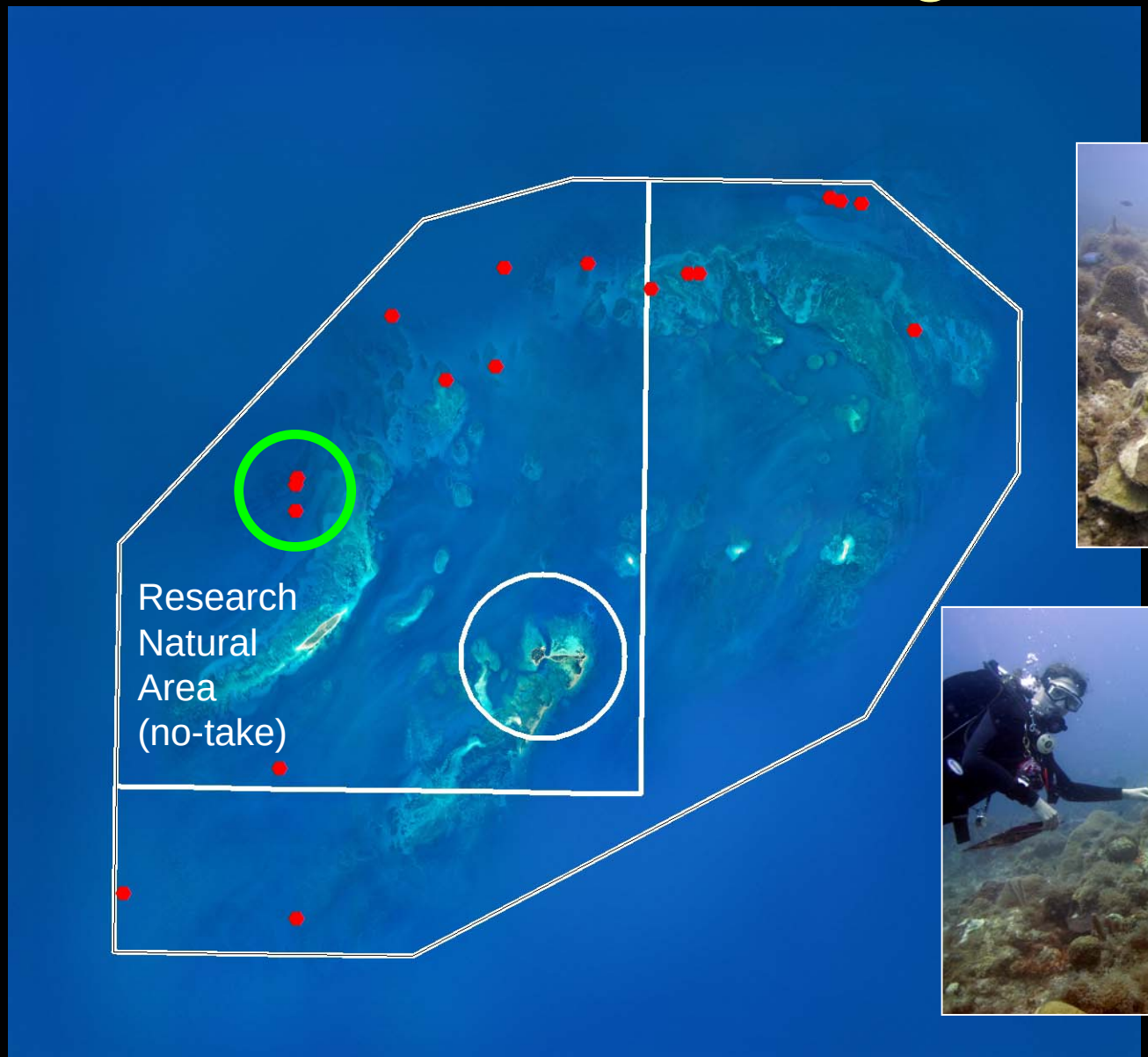
December 2005



January 2006

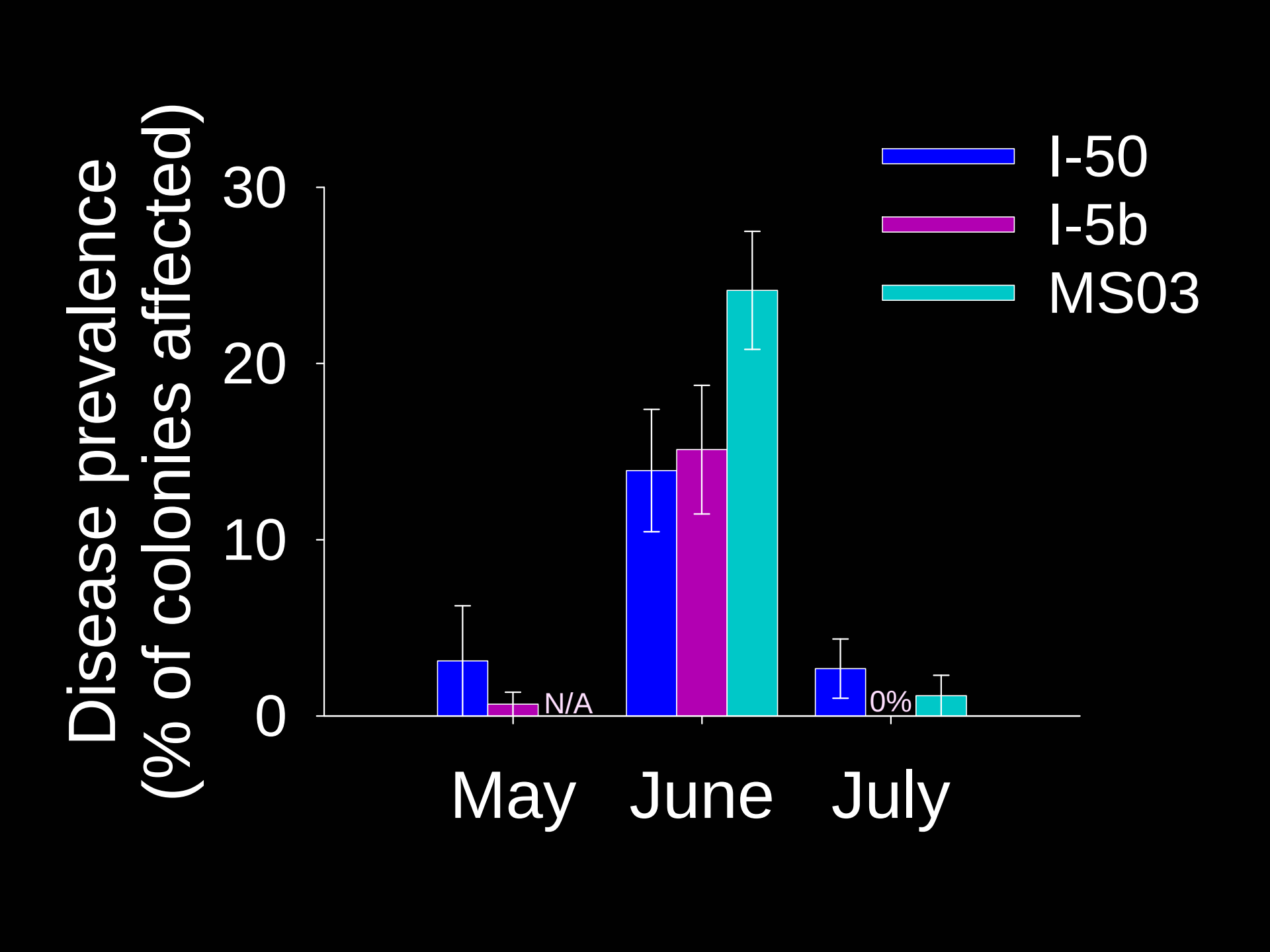
Photo by NPS

Dry Tortugas National Park
Coral Reef Monitoring Extensive Sites



Rapid Response to Disease Event

1. June 19, 2008 - Contact CDHC (Cheryl Woodley) describing the outbreak
2. June 20-22 – Initial Response: photos, prevalence and spatial extent of outbreak.
3. July 10, 2008 - International Coral Reef Symposium meeting to plan for a rapid response cruise to DRTO the following week
4. July 16-18, 2008 - NPS provided logistical support to George Mason Univ. (Drs. Bob Jonas, Geoff Cook). Collected samples of diseased corals. CDHC provides support for analysis of samples collected (biomarkers, histology, and bacteria culture).


Disease prevalence
(% of colonies affected)
30
20
10
0
N/A
0%
May
June
July
I-50
I-5b
MS03

3. Briefly describe to what degree your current monitoring and assessment approaches either meet or do not meet your needs.

- Methods are repeatable, testable, and have the statistical power to detect change.
- Mapping products have known accuracy by attribute
- Deep water work will require modified methods, mixed gas, ROV, or other technologies

4. If you have unmet monitoring and assessment needs, identify what these are and the priority you assign to them.

1. Detailed mapping of areas around NPS units (habitat and bathymetry)
2. Circulation models with larval transport
3. Coral disease causation and infection research
4. Ocean acidification
5. Lionfish eradication research

5. Identify any plans your agency/organization has to improve its approaches to monitoring and assessment.

- Near completion of detailed coral monitoring protocol.
- Improved cross calibration testing of data analysts.
- Upgrade to High Definition videography
- Use of interferometric sonar and LIDAR for mapping
- Use of drop camera for deep water evaluation of habitat
- Expanded monitoring for marine fish communities, seagrass, lobster and conch, recruitment.

Acknowledgements

NATIONAL PARK SERVICE

CORAL DISEASE & HEALTH CONSORTIUM
SOLUTIONS TODAY FOR REEFS TOMORROW.

GEORGE MASON UNIVERSITY

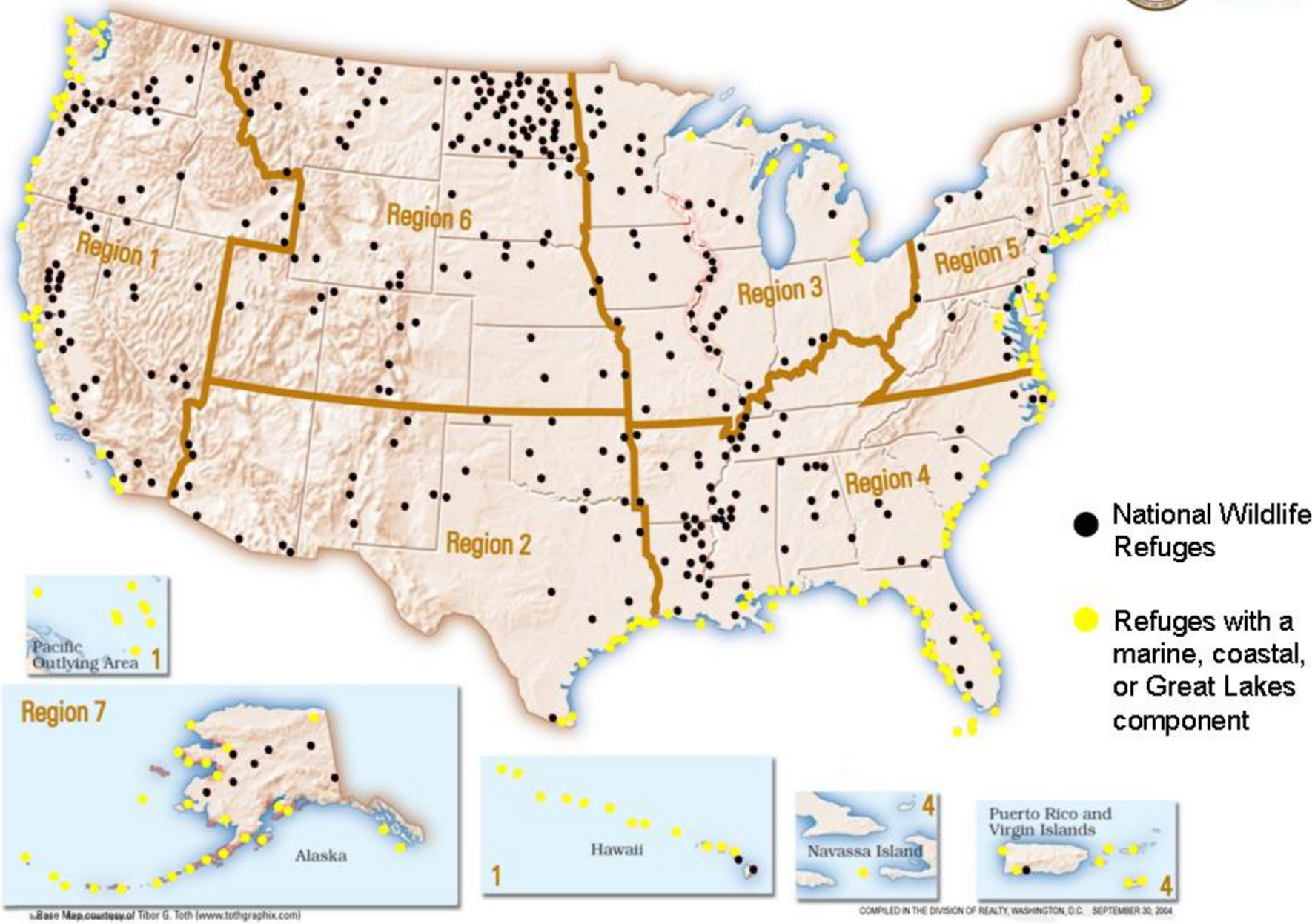
National Wildlife Refuge System: Protecting America's Coral Reefs

Presentation at SERDP Coral Reef Monitoring and Assessment Workshop

18 November 2008

Bret Wolfe
U.S. Fish and Wildlife Service
National Wildlife Refuge System, Marine Program
Arlington, Virginia

National Wildlife Refuge System



Great White Heron NWR
Key West NWR
36 km
Image NASA
Image © 2008 TerraMetrics
© USFWS
Google

Navassa Island NWR
79 km
Image NASA
Image © 2008 DigitalGlobe
© USFWS
Image © 2008 TerraMetrics
Google

SERDP Coral Reef Monitoring & Assessment Workshop

Day 1- Tuesday November 18, 2008

Agenda

Time	Description	Presenter
8:30 – 8:45 AM	Welcome & Introductions	Bill Wild, Navy: SPAWAR Pacific
8:45 – 9:00 AM	SERDP/ESTCP: Program Overview and Sponsor Role	Dr. John Hall, OSD: SERDP/ESTCP
9:00 – 9:30 AM	DoD Client Perspective	Ms. Lorri Schwartz (for Mr. Tom Egeland), Office Assist. Sec. of Navy for Installations & Environment
9:30 – 9:40 AM	Agency/Organization Perspectives	Mr. James Sinclair, Minerals Management Service
9:40 -9:50 AM		Dr. Matt Patterson, National Parks Service
9:50 – 10:00 AM		Bret Wolfe, Fish & Wildlife Service
10:00 – 10:10 AM		Dr. William Fisher, Environmental Protection Agency
10:00 – 10:20 AM	Q/A for previous 5 speakers	All
10:20 – 10:35 AM	**Break**	
10:35 – 10:45 AM	NOAA Perspectives	Dr. Margaret Miller, NOAA SE Fisheries
10:45 – 10:55 AM		Rob Warner, NOAA Center for Coastal Monitoring & Assessment (CCMA) Biogeography Branch
10:55 – 11:05 AM		Bill Goodwin, NOAA Marine Sanctuaries
11:05 – 11:15 AM		Dr. Bill Precht, NOAA Damage Assessment and Restoration
11:15 – 11:35 AM	Q/A for previous 4 speakers	All
11:35 – 12:35 AM	Group discussion 1: Monitoring/Assessment Needs	Bill Wild, Navy: SPAWAR Pacific
12:35 – 1:35 PM	**Working Lunch (continued discussion)**	
1:35 – 2:20 PM	University of Miami Research (includes 10–15 minutes Q/A)	Dr. Pamela Reid, Univ. Miami
2:20 – 3:05 PM	Rutgers University Research (includes 10–15 minutes Q/A)	Dr. Max Gorbunov and Dr. Paul Falkowski, Rutgers University
3:05 – 3:15 PM	Integration of UM/Rutgers Technologies	Dr. Diego Lirman, Miami Dr. Max Gorbunov, Rutgers
3:15 – 3:30 PM	**Break**	
3:30 – 5:00 PM	Group discussion 2: Overlay of Monitoring/Assessment Needs with Miami/Rutgers Technologies	Bill Wild, Navy: SPAWAR Pacific
5:00 PM	**Adjourn**	
7:30 PM	**Group dinner at Jaguar Ceviche Spoon & Latam Grill**	

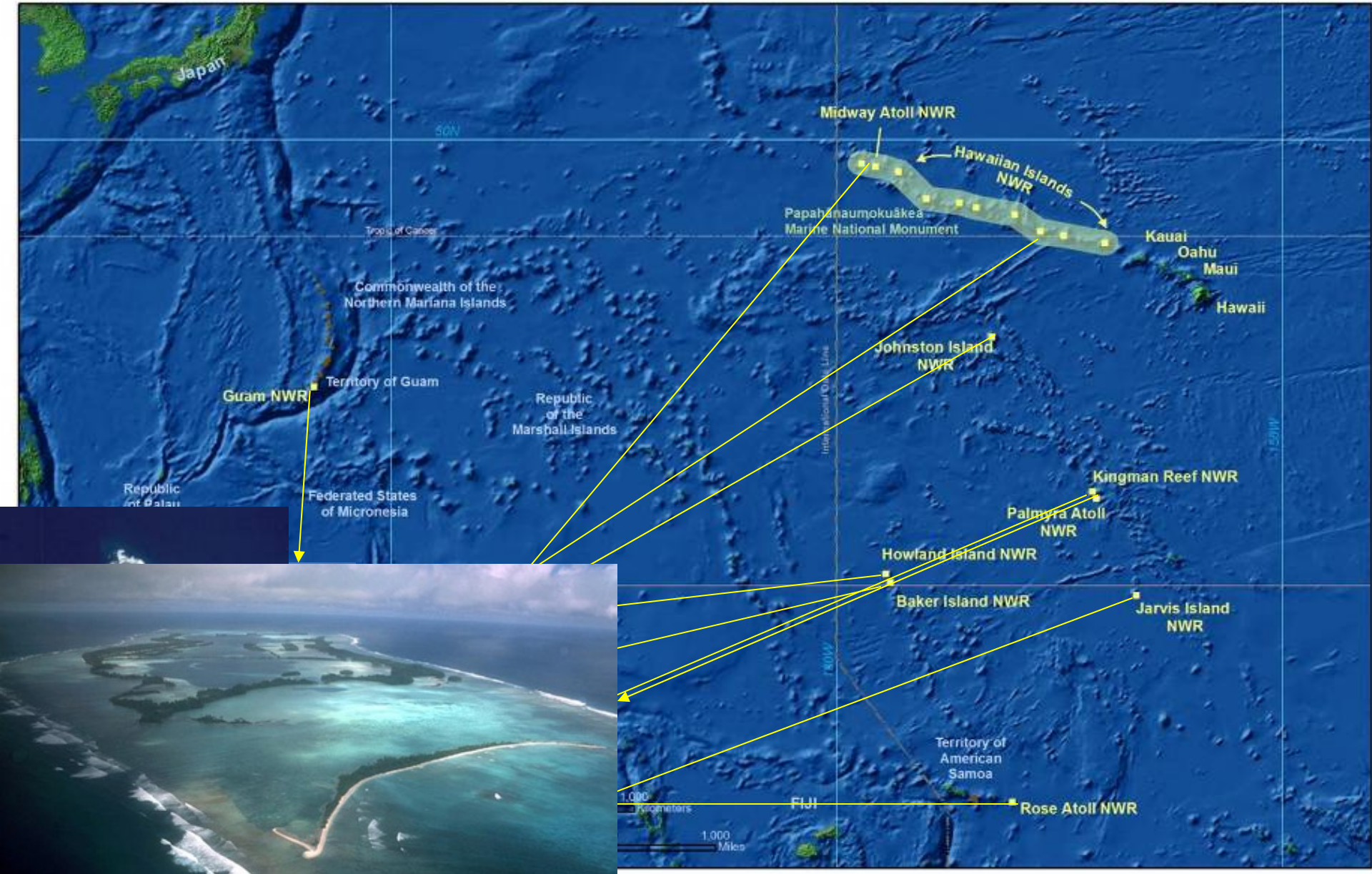
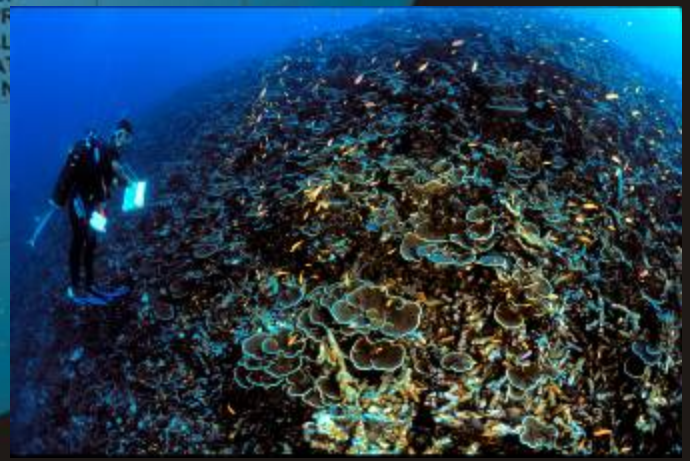


Photo by Rob Shallenberger/USFWS

onal Wildlife Refuges in the Pacific



MIDWAY ATOLL NWR
HAWAIIAN ISLANDS NWR COMPLEX
KAUAI NWR
OAHU NW
MAUAI
BIG ISL
JOHNSTON ATOLL NWR
KINGMAN REEF NWR
JARVIS ISLAND NWR
HOWLAND ISLAND NWR
BAKER ISLAND NWR
ROSE ATOLL NWR

Monitoring

Florida Keys Refuge Complex

- Interagency agreement with EPA
- Water Quality Protection Program for Florida Keys NMS
- FWS intends to continue to support the long-term partnership

Monitoring

Navassa Island NWR

- No pre-1998 baseline data
- Research cruises approximately every 2 years
- Overfishing is major threat
- Difficult to enforce regulations
- Remote enforcement methods

Monitoring

Hawaiian Islands and Midway Atoll NWRs

- Coral monitoring at permanent sites since 2000
- Northwestern Hawaiian Island Reef Assessment and Monitoring Program
- Numerous fisheries monitoring programs
- Permitted research by UC-Santa Cruz

Monitoring

Remote Pacific Refuge Complex

- Annual and biennial research cruises
- Towed diver surveys (2 km in length)
- REAs covering between 1000-5000 m^2
- Photo-quadrat/video surveys at permanently marked 50-100 m transects
- Recruitment studies

Palmyra Atoll Research Consortium (PARC)

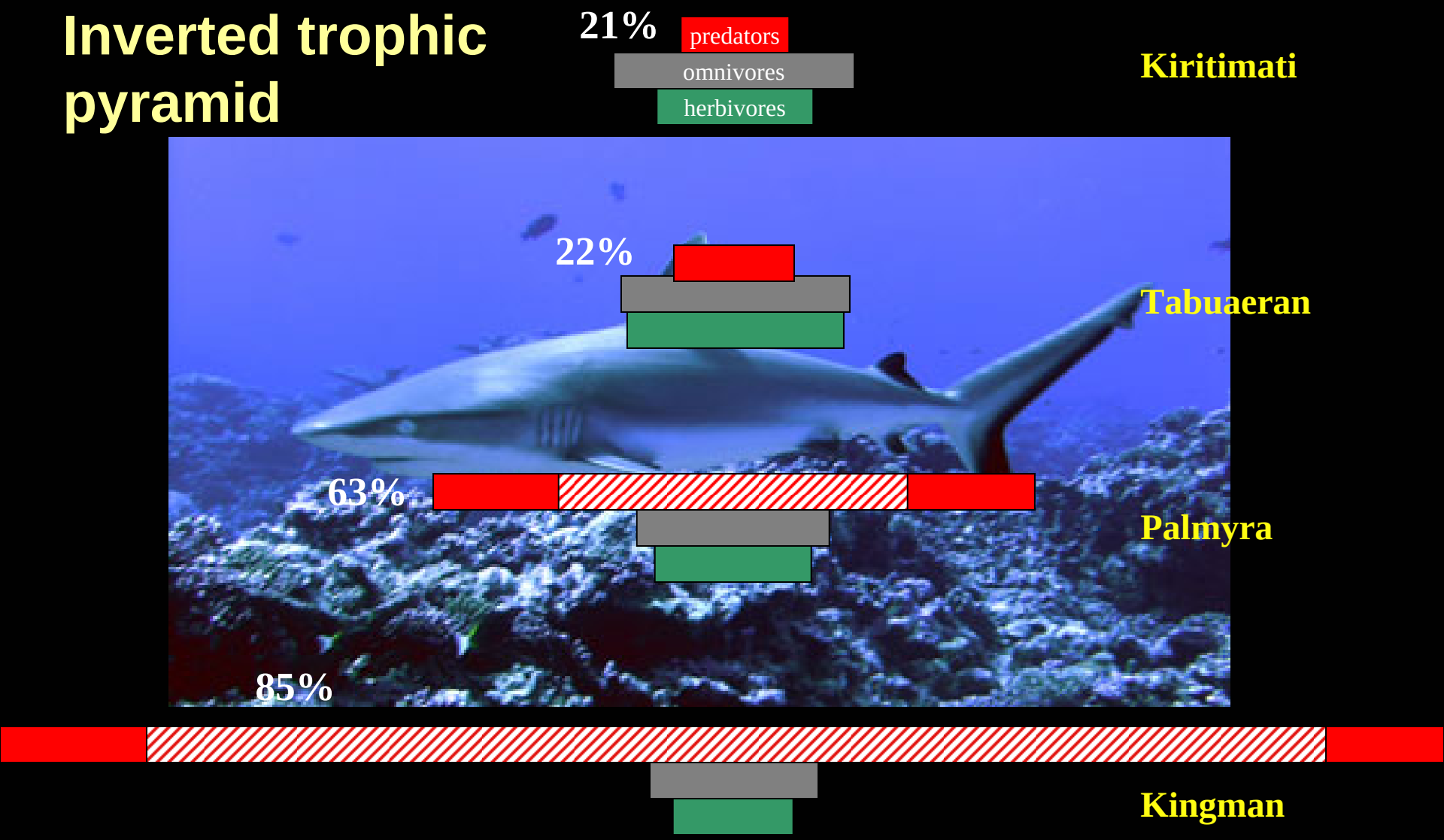
- founded in 2004
- $1.5 million donation from Moore foundation
- Supported by US FWS and TNC
- Research focuses
  1) Biodiversity of Palmyra
  2) Terrestrial/Marine Interface
  3) Marine Biology, Climate Change, and Biogeochemical Structure

Palmyra Atoll NWR

Kingman Reef NWR

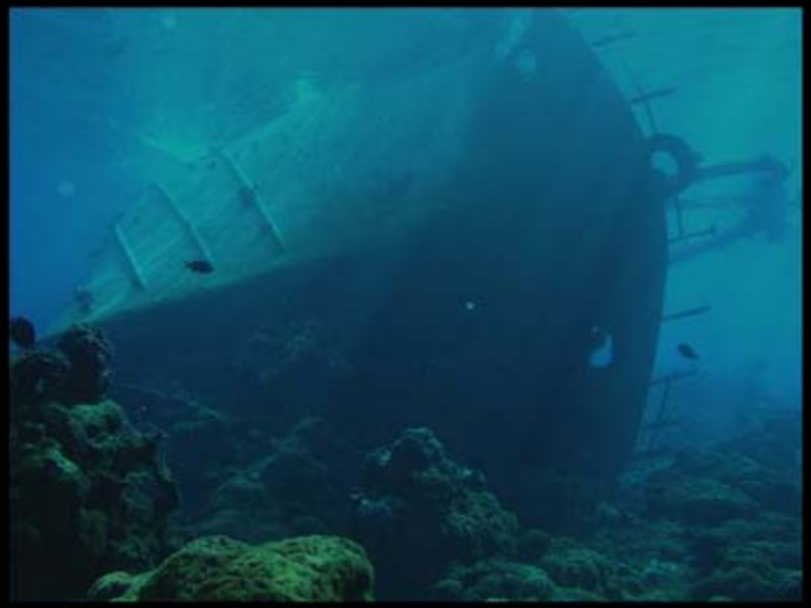
http://www.palmyraresearch.org

Inverted trophic pyramid



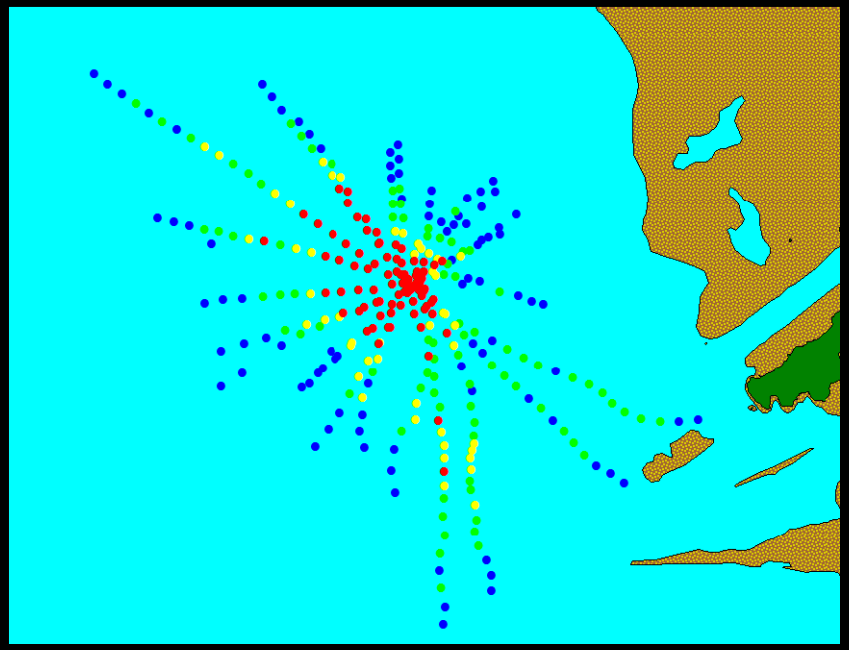
Healy 2008; Sandin *et al 2008*

Unique Challenges



Large area of cyanobacteria growth
March 2008

Corallimorph Infestation at Palmyra Wreck



Color codes correspond to estimated benthic cover of corallimorphs: red=high, yellow=medium, green=light, blue=no visible corallimorphs.
(Work and Aeby 2007)

Coral Reef Monitoring and Assessment Needs

- Enforcement of fishing regulations and other illegal activities
- Additional partnerships
- Additional transects at remote refuges
- Improved monitoring techniques and technologies
- Improved understanding of invasive species

Contact Info:

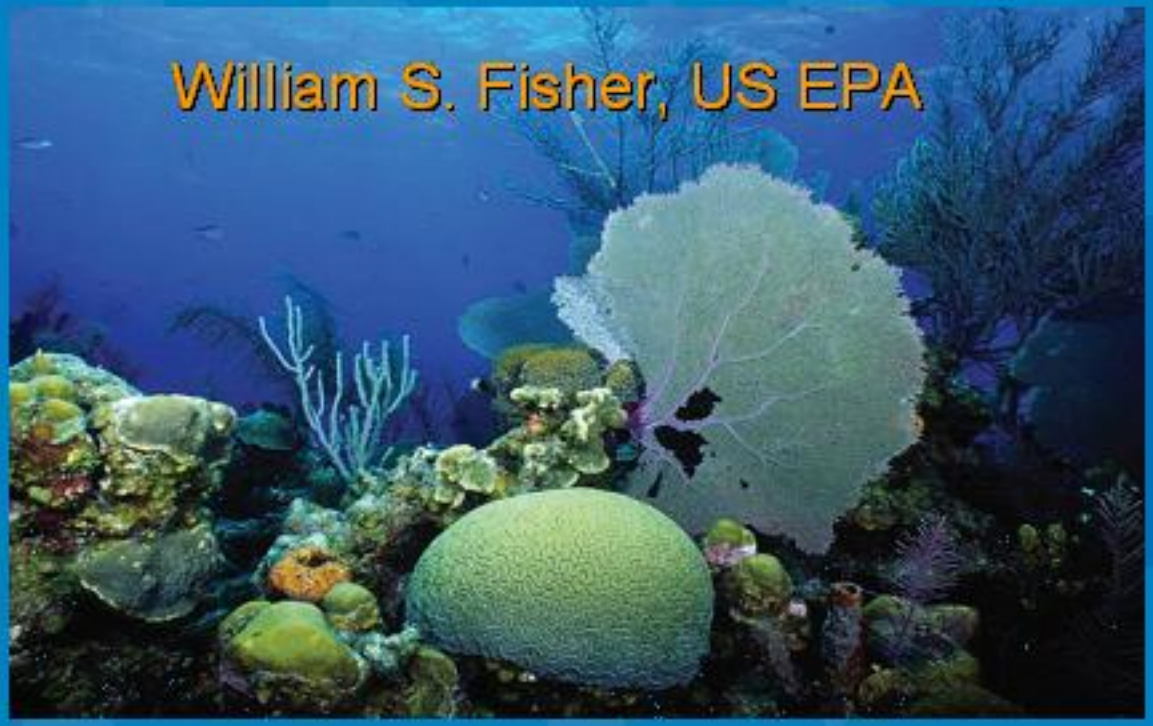
Bret Wolfe
National Wildlife Refuge System, Marine Program
bret_wolfe@fws.gov

Jim Maragos, Ph.D. Coral Reef Biologist
Pacific/Remote Islands National Wildlife Refuge Complex
jim_maragos@fws.gov

Coral gardens at Palmyra Atoll NWR

United States
Environmental Protection
Agency

U.S. EPA CORAL REEF PROGRAMS: BIOCRITERIA DEVELOPMENT and ECOSYSTEM SERVICES RESEARCH



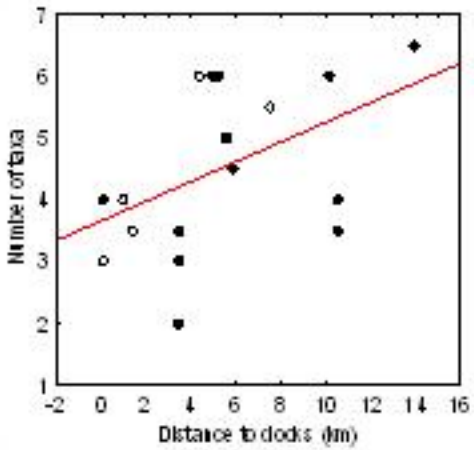
William S. Fisher, US EPA

Office of Research and Development
National Health and Environmental Effects Research Laboratory, Gulf Ecology Division

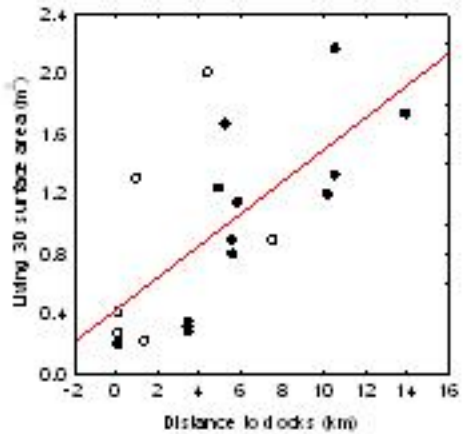
April 14, 2009

United States Environmental Protection Agency

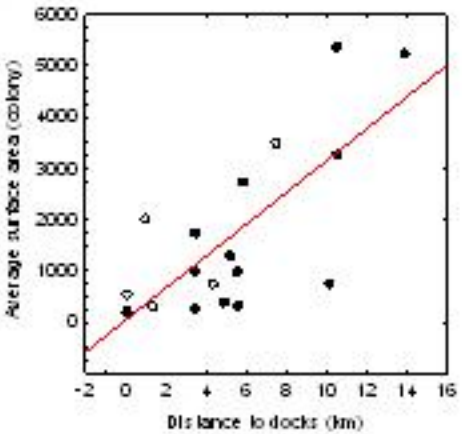
Taxa



Live SA (3D)



Colony SA



>>>> distance from disturbance >>>>

Ecosystem Services

Stony Corals

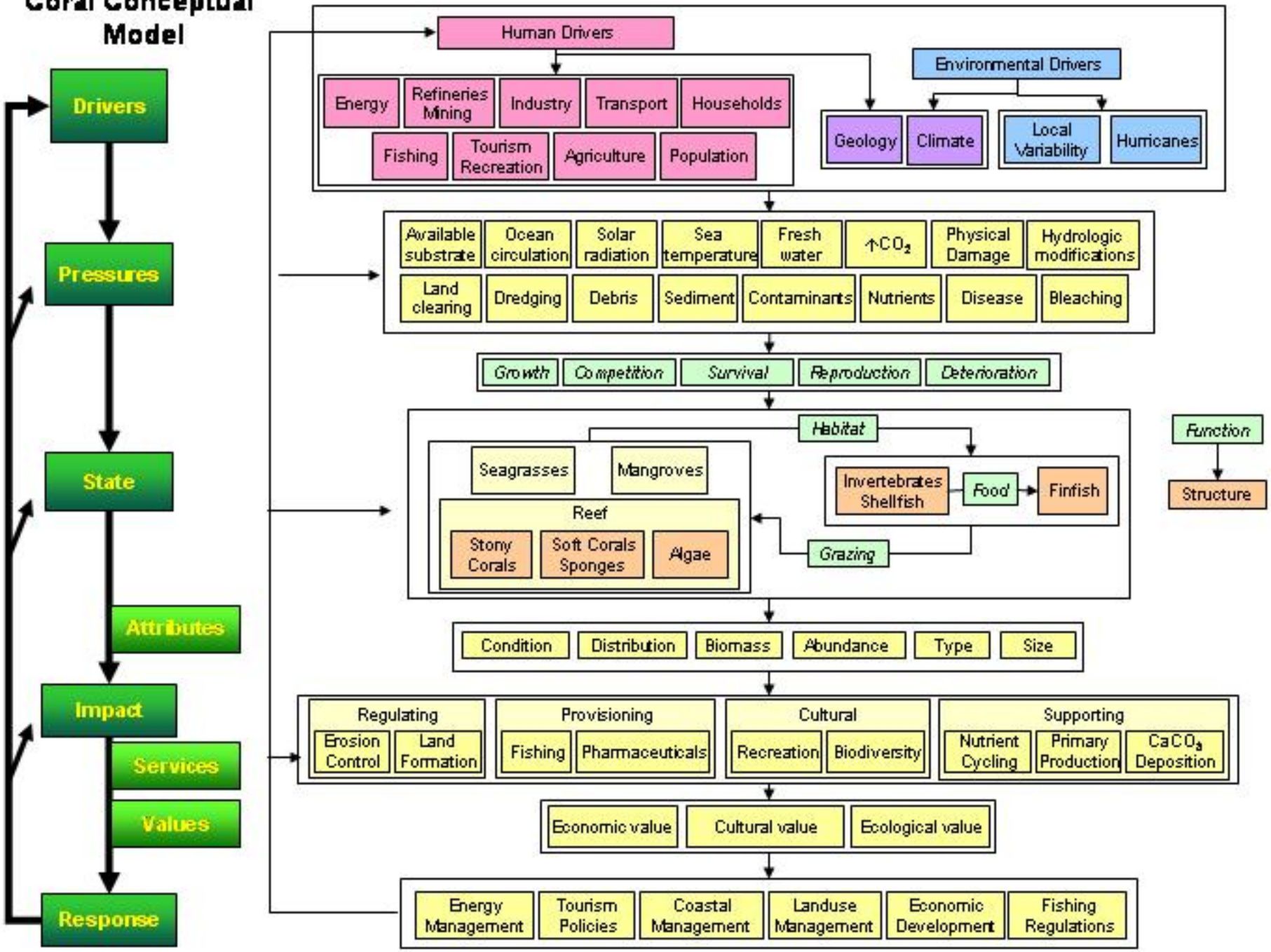
Ecological value

- Habitat for fish and invertebrates
- Biodiversity
- Primary Production

Economic value

- Tourism
- Fishing
- Shoreline protection
- Bio-mining (drugs)

Coral Conceptual Model



Do M&A approaches meet our needs?

- Rapid, easily measured, easily transferred
- Transparent, easily interpreted
- Information relevant to ecosystem management
- Indicators responsive to human disturbance

What needs are not met?

- Only one assemblage, stony coral
- Unknown connection to stressor identification
- Unknown connection to ES and values

Unmet monitoring needs

1. Long-term monitoring programs
   - Standard core procedures
   - Local implementation
   - Linkage to regulatory action
2. Assessment integration
   - Indices from multiple assemblages
   - Specific links to stressor identification
   - Links to chemical and physical criteria
3. Indicators and programs to measure ecosystem services

Plans to improve our program

- Additional assemblages
  - simultaneous surveys of stony corals, soft corals, sponges, macro-invertebrates and fish
- Stressor identification
  - cumulative associations
  - specific methods (after finding of impairment)
- Monitoring strategies that are jurisdiction-specific
- Power of assessment tools
- Ecosystem service indicators

fisher.william@epa.gov

United States Environmental Protection Agency

Following are slides to address potential questions

Historical Approaches to Stony Coral Bioassessment

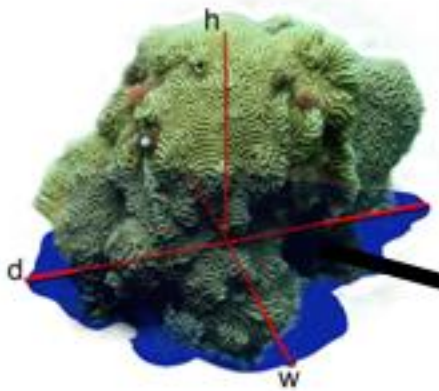
Colony-based protocols

focus on number but not size or surface area

Surface area protocols

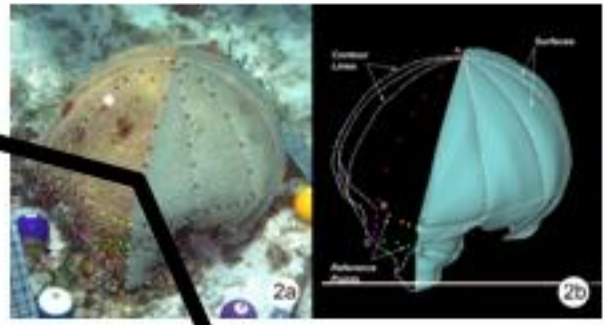
focus on amount of live coral tissue but not number or size of individual colonies

Estimating 3D Colony Surface Area



Colony measurements

Conversion factors derived from 3D photographic reconstruction



Colony surface area (3D)

Courtney et al. (2007) JEMBE 351:234-242

Management Options

Marine Protected Areas

- fishing, boating, tourism restrictions

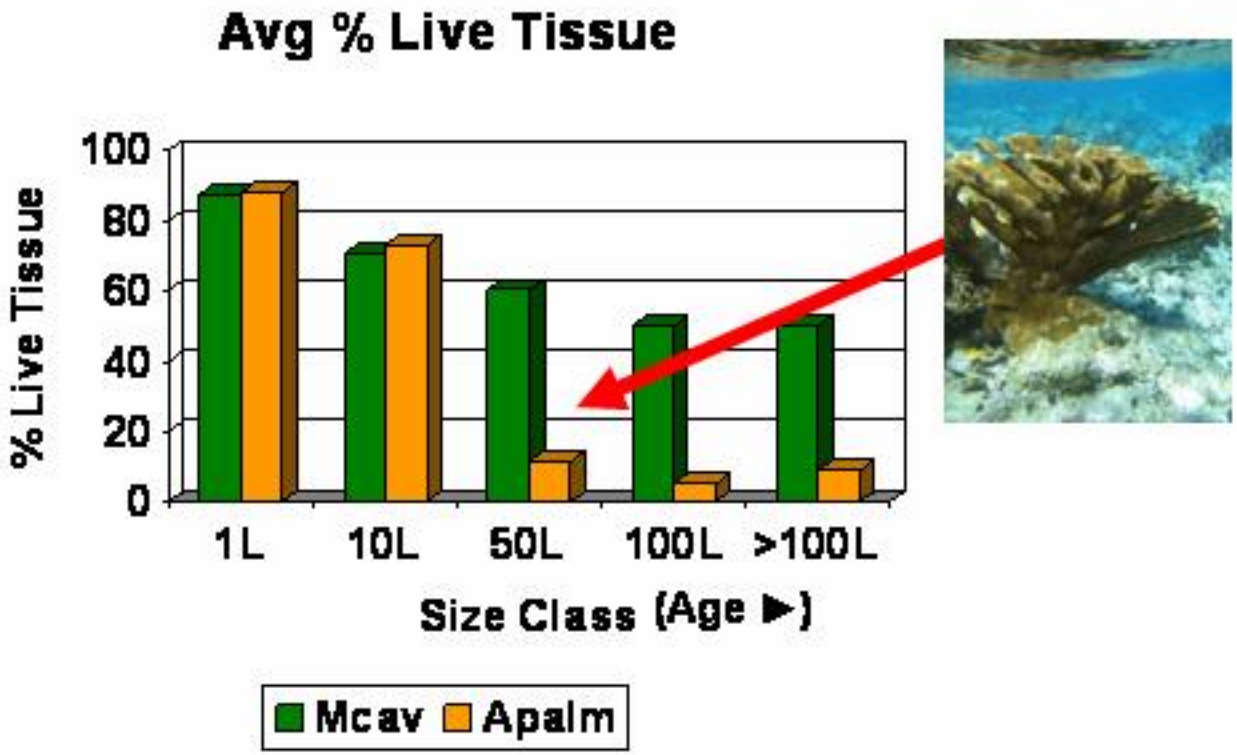
Managing for 'resilience'

- resilient populations and habitats

Biocriteria and other Clean Water Act regulations

- watershed and waterbody pollutants
- mandate to identify waterbody impairment

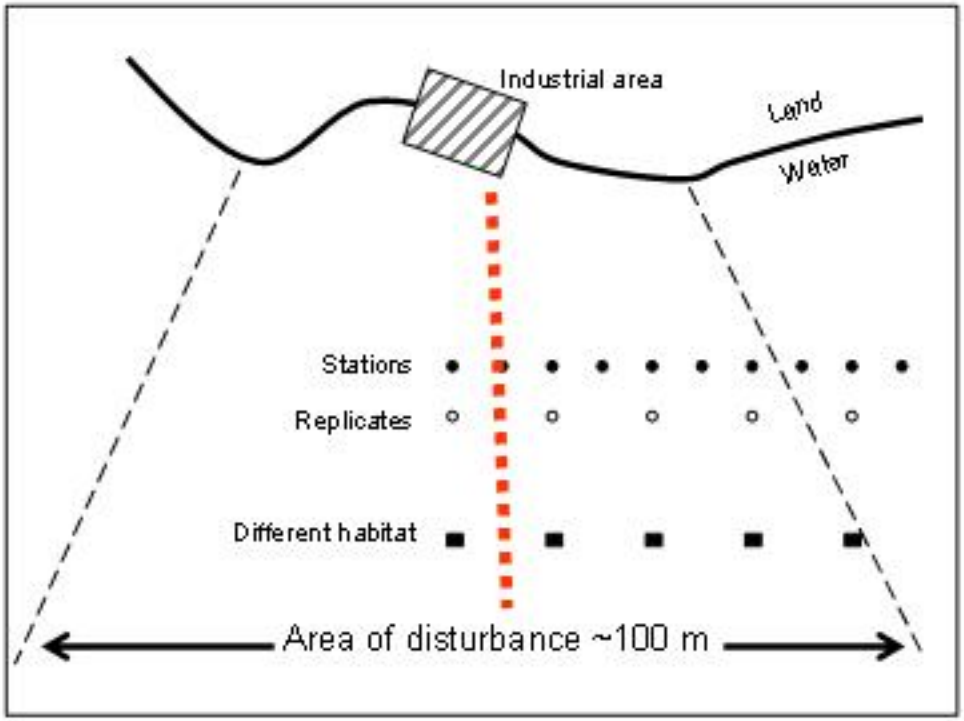
Size-Related Condition



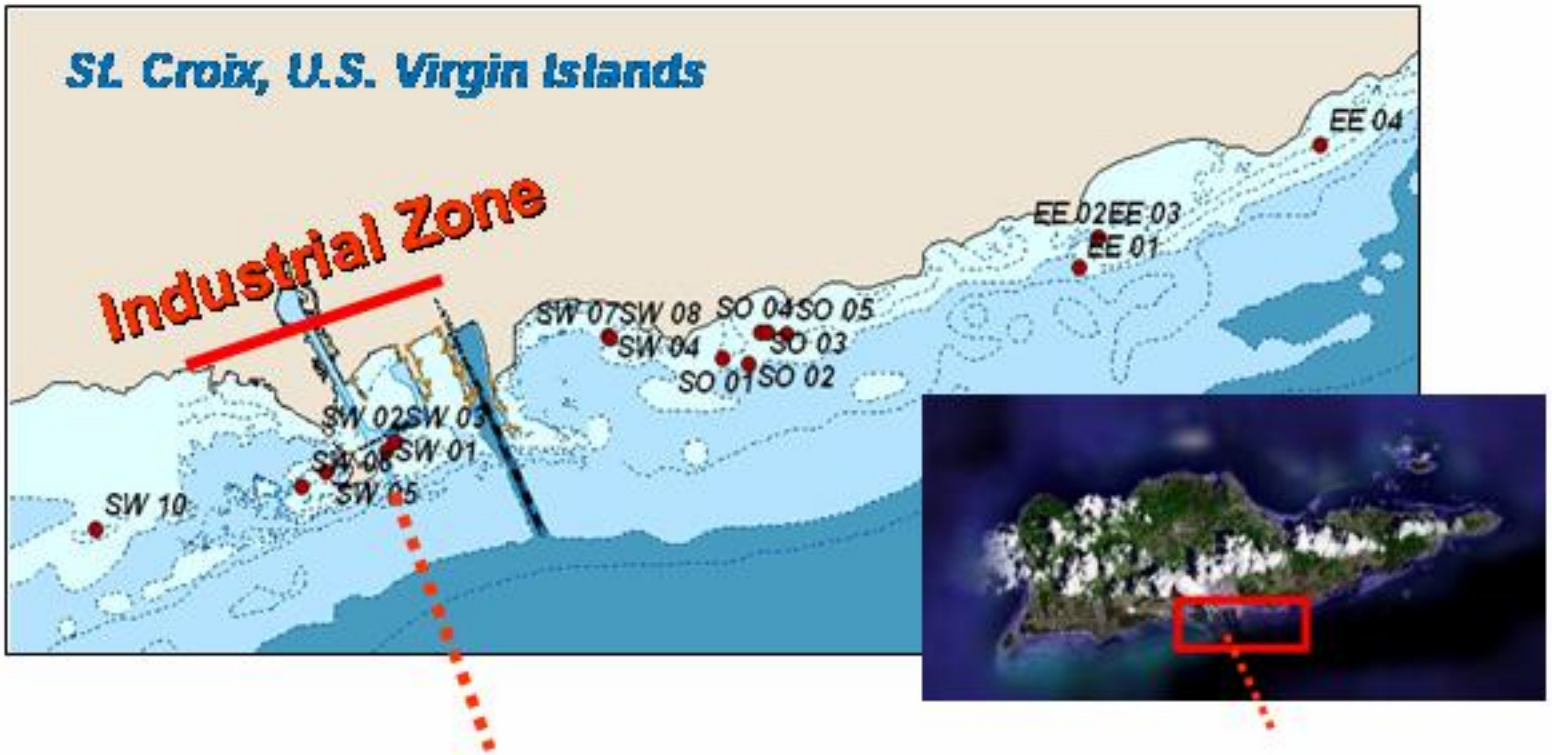
Data from Florida Keys

Screening for Metrics

Metrics
indicators that respond to human disturbance over natural variation



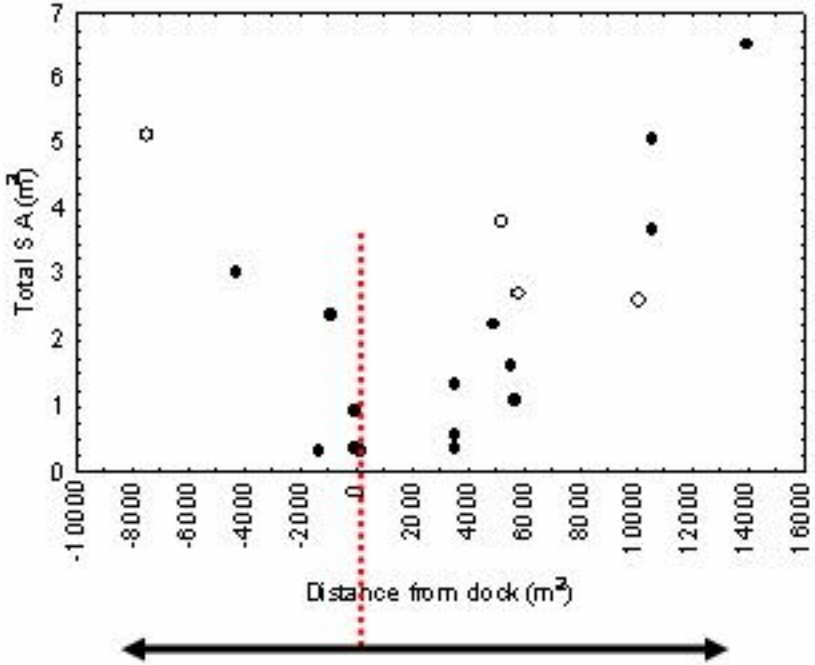
Selecting Metrics



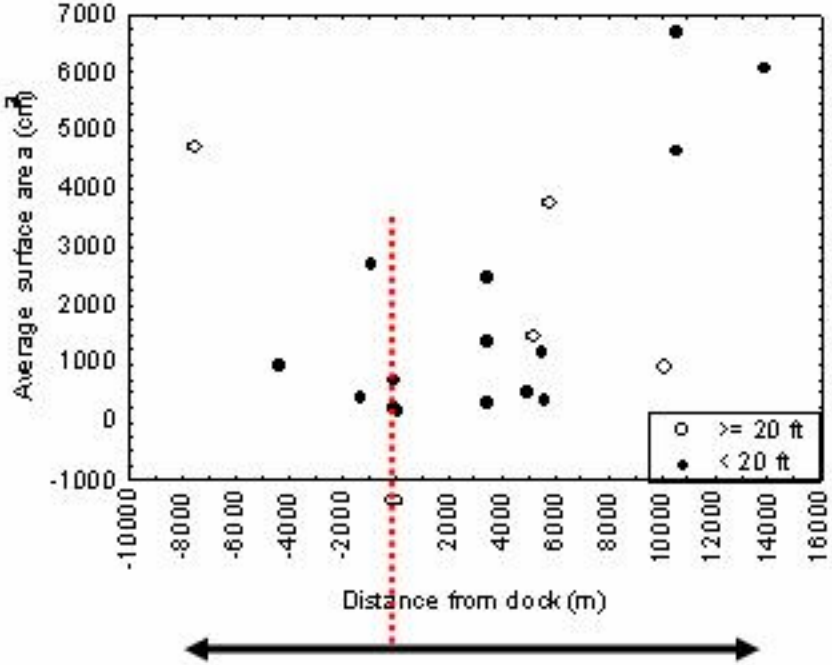
Center of human disturbance

Candidate Metrics

Total 3D SA



Average Colony Size



distance from disturbance

Potential for Coral Reef Protection using CWA

- Coral knowledge, monitoring and bioassessment expertise
- Authority of the CWA

- Proven process to establish regulatory programs using bioassessments
- Desire of jurisdictions to embrace CWA and improve management options

Ecosystem Services Research Program

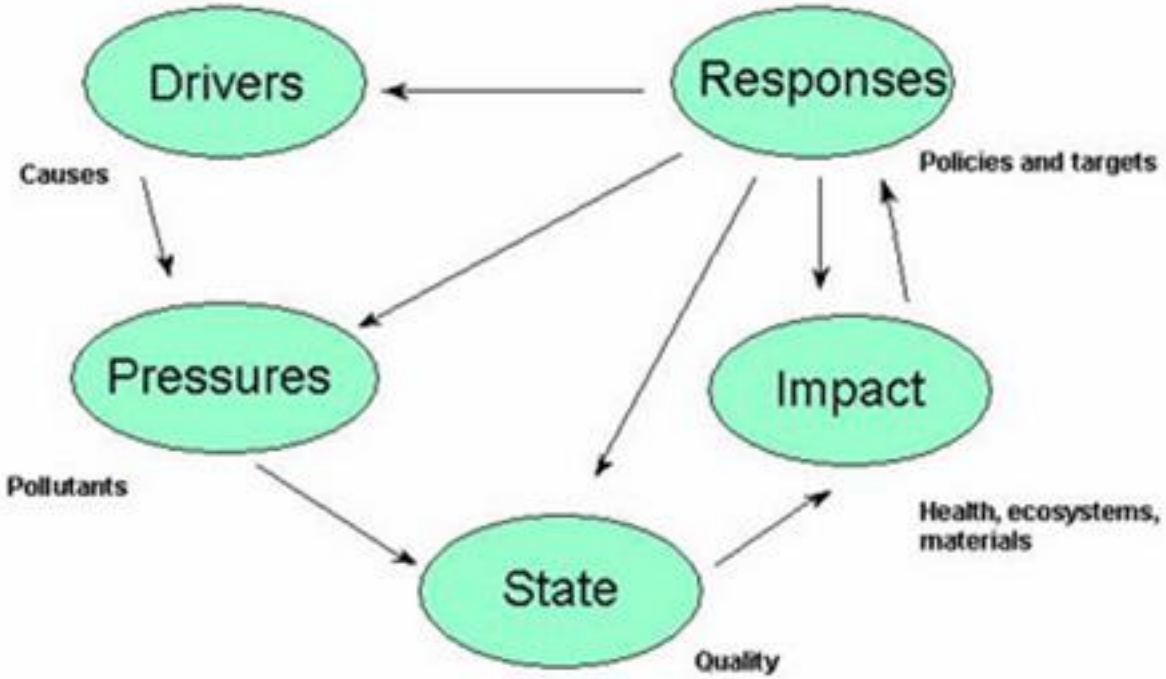


Figure 1. The DPSIR assessment framework

Biocriteria

Numeric values or narrative descriptions (thresholds) established ***to protect the biological condition*** of aquatic life inhabiting waters of a given designated use.

Require defensible bioassessment procedures to establish and enforce regulatory thresholds

Biological Assessments

- Applied in many CWA programs*
- Indicators relevant to management
- Metrics distinguish anthropogenic stressors
- Thresholds (criteria) describe expected or desired condition
- Monitoring results support regulatory decisions

*
Biocriteria
404 permits & mitigation plans
401 certifications
Dredging and ocean dumping permits
NPDES permits & 301(h) decisions
Permits for coastal development
Others….

United States Environmental Protection Agency

EPA Bioassessment Protocol for Stony Corals

Assessment procedures for biocriteria and other CWA regulatory activities

http://www.epa.gov.bioindicators/coral/coral_biocriteria.html

Stony Coral RBP

Colony Observations

Colony ID
Colony Size
% Live Tissue

Surface Area

3D SA calculated from colony size

Multiple Relevant Indicators

Multiple Indicators

Colony Identification

Taxa Richness

Community Composition

Relative Spp Abundance

Species Diversity

Abundance

Density

Population Structure

Community Structure

Colony Size

Surface Area

Total 3D SA

Total 3D Coral Cover

Live 3D SA

Live 3D Coral Cover

Size-Related Condition

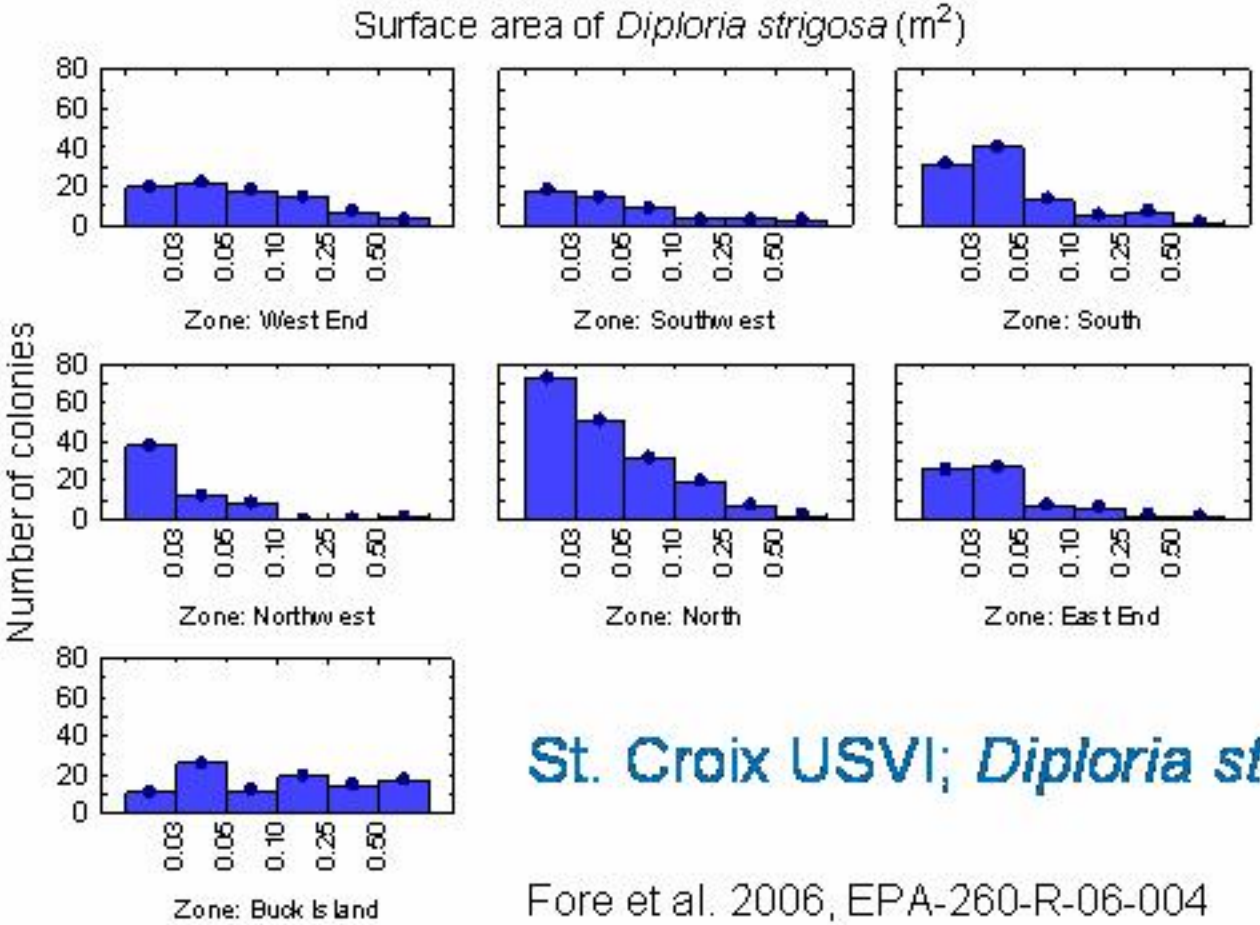
% Live Tissue

Colony %LT

Avg %LT

% Live Coral

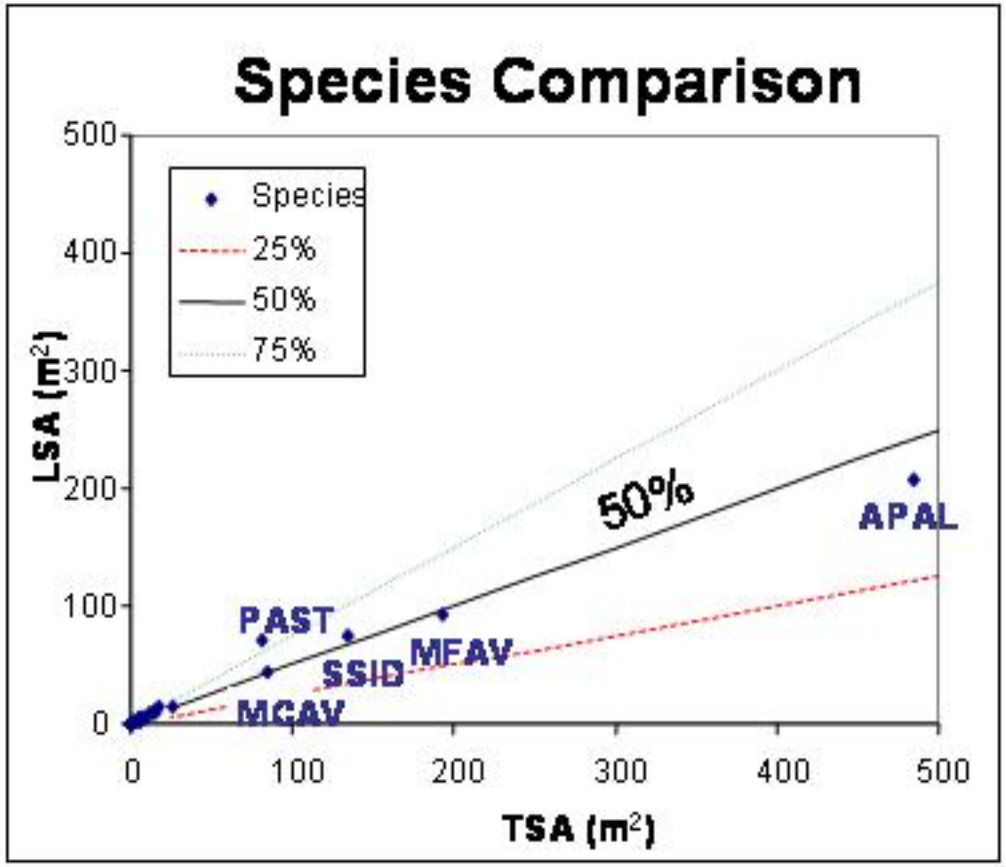
Population Structure



St. Croix USVI; *Diploria strigosa*

Fore et al. 2006, EPA-260-R-06-004
http://www.epa.gov.bioindicators/coral/coral_biocriteria.html

Percent Live Coral



Key West Stations

Science, Service, Stewardship

Coral and Coral Reef Monitoring

Southeast Fisheries Science Center

Margaret Miller

NOAA FISHERIES SERVICE

NOAA
FISHERIES
SERVICE

General areas of interest

- Fishery Independent Monitoring of fishes and inverts
- Coral/Habitat status
  - —Habitat value/associations for fishes (EFH)
  - —Coral condition, population dynamics
- Protected Species Monitoring
  - —*Acropora* spp. corals

- Restoration / Evaluation

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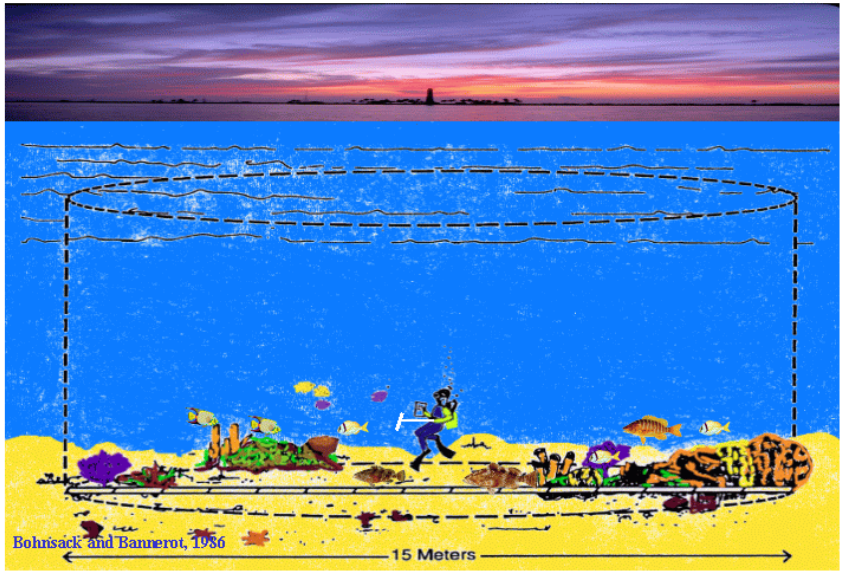
SERVICE

Fishery Independent Monitoring (Reefish Visual Census) Bohnsack/Ault et al.

- Stationary plot method
- Multispecies, size and abundance
- Applied in optimized habitat-stratified approach to achieve efficient regional ‘stock assessments’
- Being applied in several regions

- In-water intensive activity



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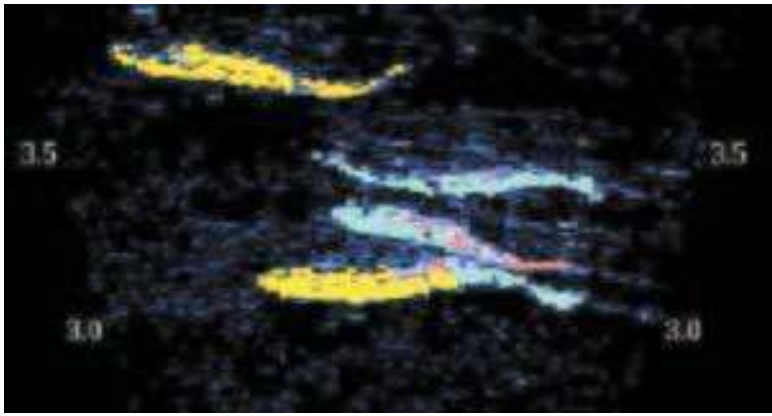
SERVICE

Fish/Habitat association

- Grouper/Reef association (Kellison, Gleason, Rivera)
  - Reef characteristics (mostly architectural) related to dstribution of
    - Juvenile
    - adult
    - SPAG
  - Acoustic techniques being applied
  - Florida Keys and Puerto Rico



- Mangrove association
  - Visual surveys of limited application
  - Developing DIDSON sonar system (Serafy & Kellison)

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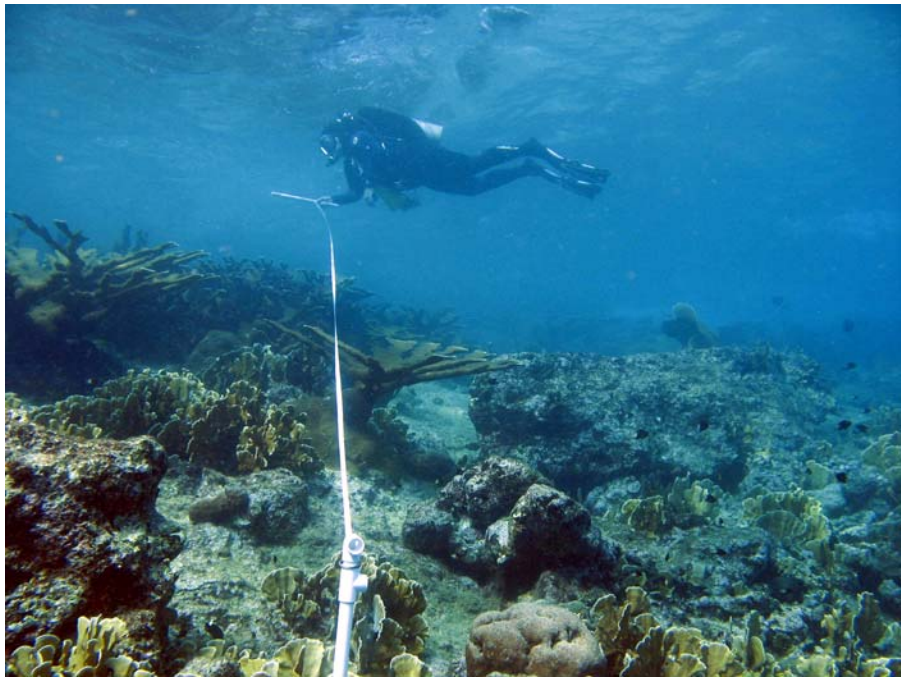
SERVICE

Coral/Benthic Status

- Classical approaches
  - —In situ transects
  - —Photo- or video- sampling
    - % cover

- Coral condition
  - —Size
  - —Bleaching
  - —Disease states



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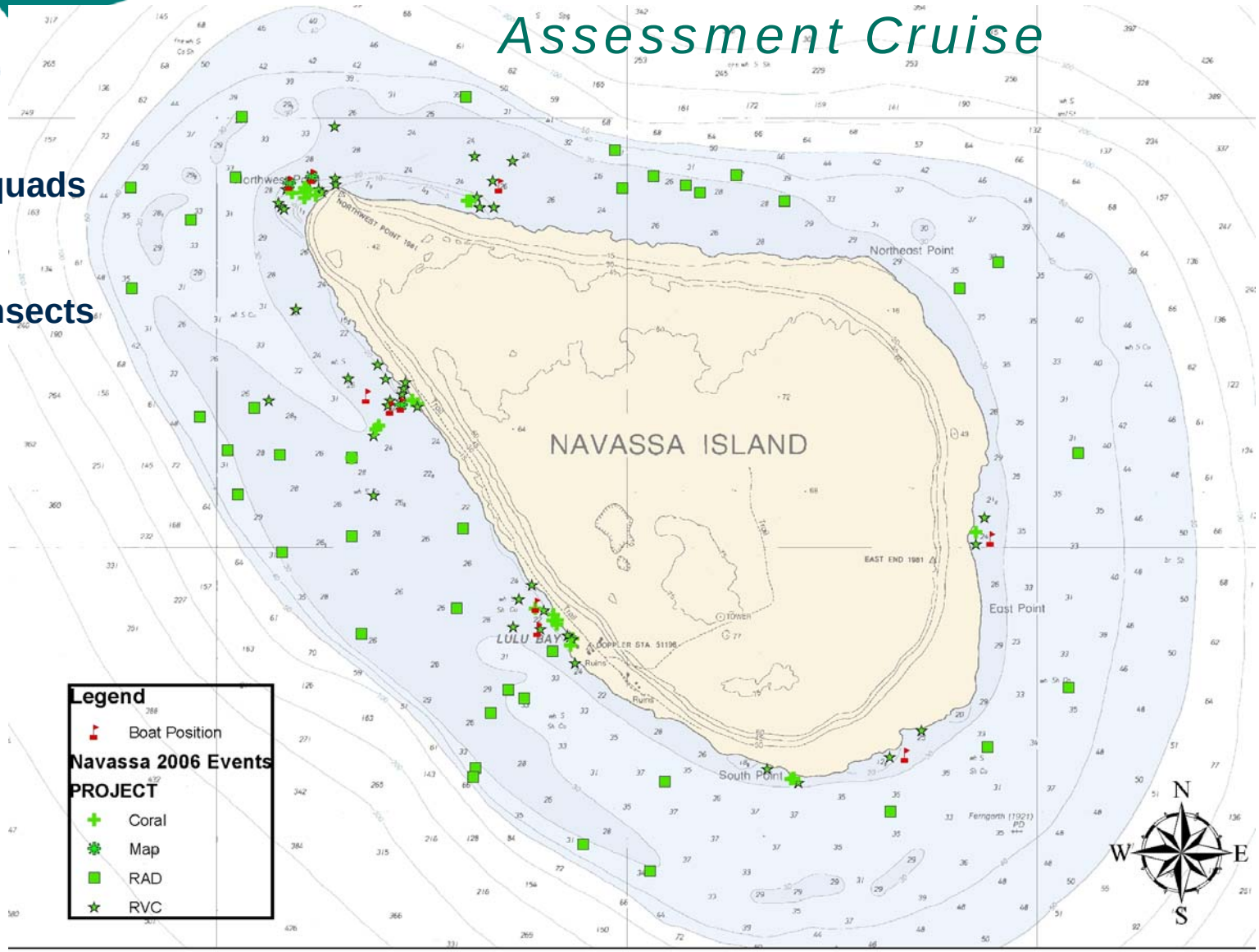
Navassa 2006 Reef Assessment Cruise

- RVC/Photoquads
- RVC only
- Benthic transects only



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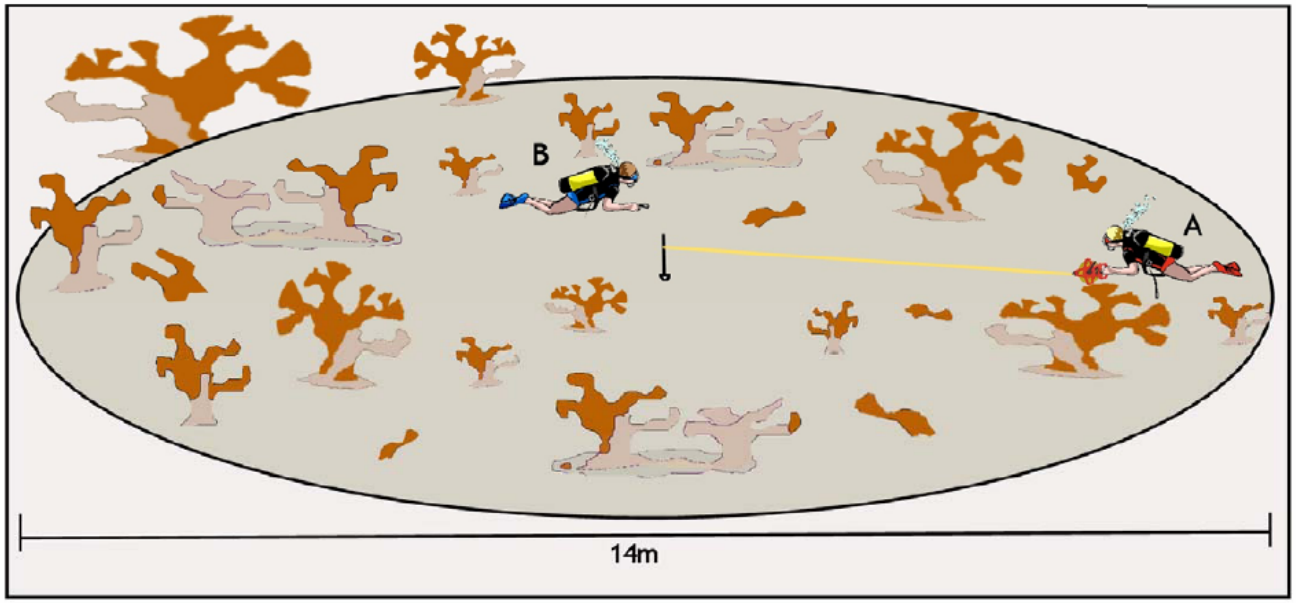
SERVICE

Acropora spp. Monitoring

- Targeted monitoring effort required
  - —Confined habitat (A.palmata particularly)
- Remnant populations often spatially patchy and very low population density

NOAA FISHERIES SERVICE

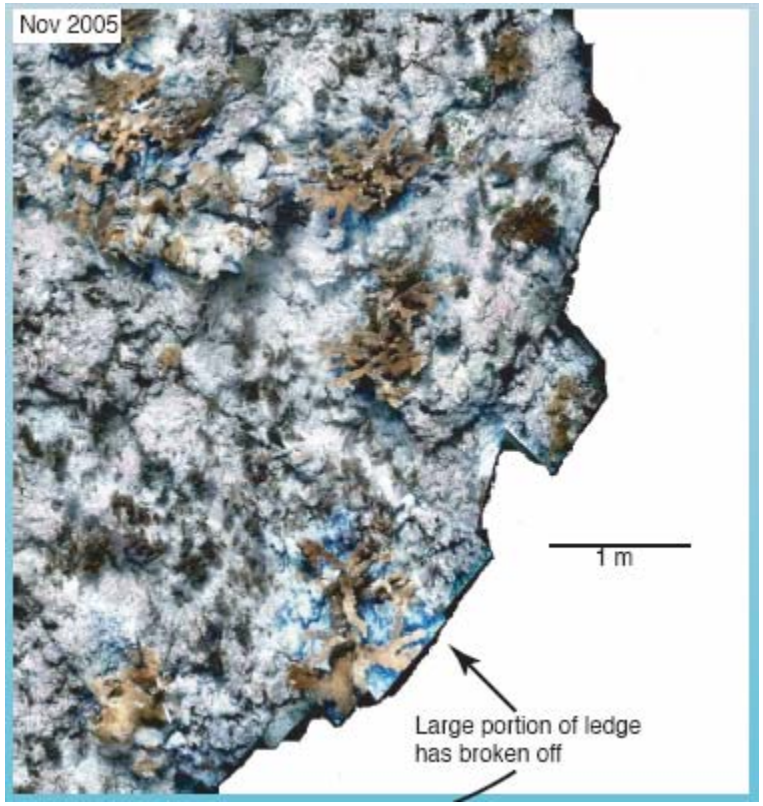
Demographic Monitoring Protocol



- Incorporates fixed plots (7m radius)
  - —Allows some assessment of recruitment via scrutiny of actual ‘real estate’
- Tagged colonies
  - —some assessment of growth
  - —Relative prevalence of ‘threats’
  - —Mortaltiy
    - To some extent attributable to individual ‘threats’

NOAA
FISHERIES
SERVICE

SERDP Mosaics have been applied

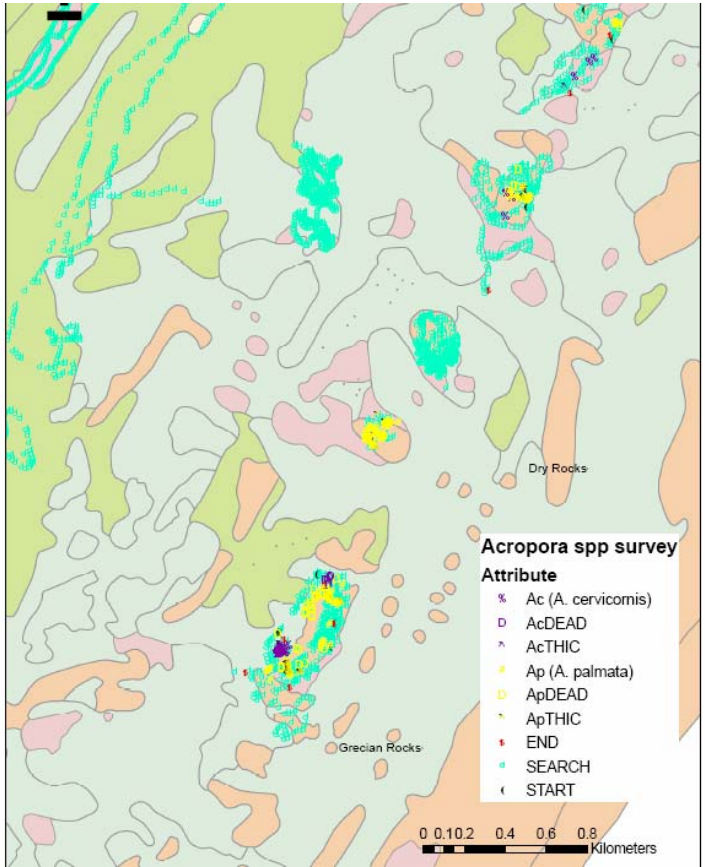


NOAA FISHERIES SERVICE

In-water distribution mapping

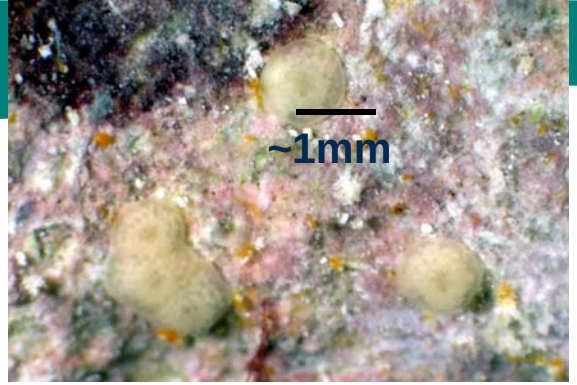
- Snorkellers w/GPS (boat tow or scooters) survey targeted habitats

Fig. 5: Aerial photo of Navassa Island with the GPS survey tracks (blue) and

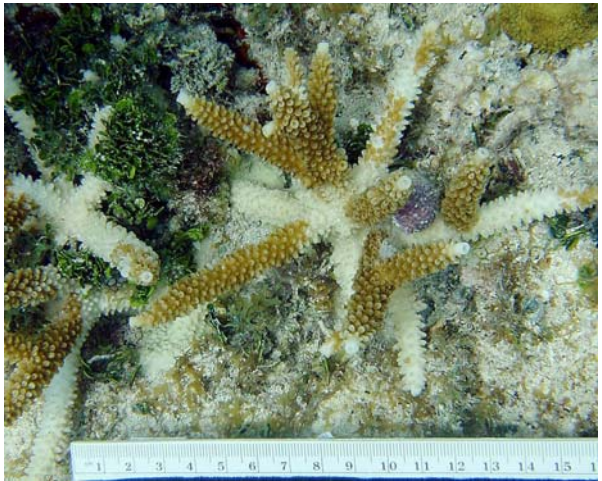


NOAA
FISHERIES
SERVICE

Dilemmas



- Tiny coral recruits
  - —Several years of ‘black box’ at post-settlement stage about which we know nothing due to inapparency
    - No ‘natural’ expectation to judge performance of culture efforts
- Coral disease diagnosis
  - —Gross visual signs are inadequate to understand health status of corals
- Efficiency/Cost



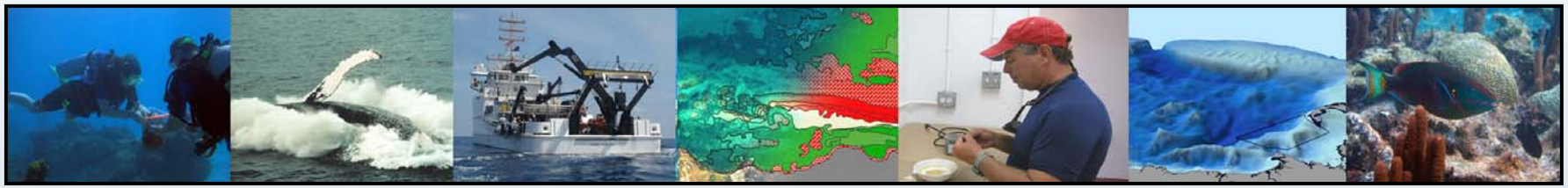
CCMA MISSION

To assess and forecast coastal and marine ecosystem conditions through research and monitoring

http://ccma.nos.noaa.gov/

Organizational Structure

- **Biogeography Branch**



- **Coastal and Oceanographic Assessment, Status & Trends Branch (COAST)**

- **Research Coordination and Administrative Support Branch (RCAS)**

Coral Reef Monitoring and Assessment Workshop

REGION	**shallow water reefs** KM2
U.S. Virgin Islands	344
Puerto Rico	2,302
Navassa	3
Florida	30,801
Flower Garden Banks	0
Main Hawaiian Islands	1,231
Northwestern Hawaiian Islands	1,595
American Samoa	55
Pacific Remote Island Areas	252
Marshall Islands	13,456
Federated States of Micronesia	14,517
Northern Mariana Islands	124
Guam	108
Palau	2,529

Deep water reefs (30 – 200 m) on going in multiple regions (30 – 1000 m)

1999 – 2001 photogrammetry and imaging spectroscopy, (Warner 1997 to 2001 Caribbean - photos sharpen HSI + other source)

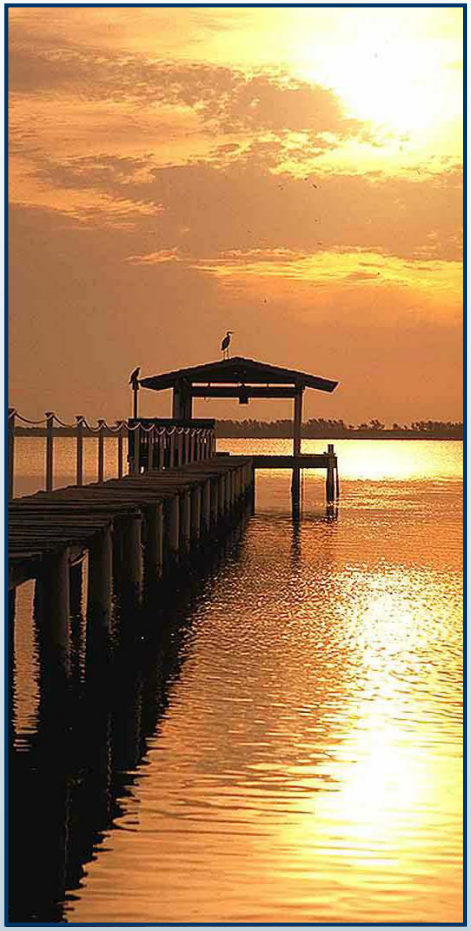
2001 – 2004 IKONOS imagery

2005 – 2008 multibeam and LiDAR (camera system on drop frame and ROV)

Guiding Principles

- **Address National Issues with Local Approach**
- **Support Diverse, Collaborative Partnerships**
- **Provide Science & Research to Directly Support Management and Policy Decisions**
- **Integrate Research Across Scientific Disciplines**
- **Serve as a Link Between Science Conducted in Academia and Specific Needs of Coastal Decision-Makers**

Center Capability Highlights



- **Assessments**
  - ✓ *Coral Reef Ecosystems*
  - ✓ *Marine Protected Areas*
  - ✓ *Eutrophication & Nutrients*
- **Integrated Ecosystem Modeling & Mapping**
- **Coastal Contamination, Status & Trends**
- **Oceanographic Remote Sensing**
- **Harmful Algal Bloom Detection & Forecasting**
- **Biogeography & Spatial Ecology**

CCMA scientists conduct field observations on regional and national scales to provide the best available scientific information for resource managers and researchers, and to provide technical advice, and accessibility to data.

Strength Through Partnership

Enhancing Cooperative Research Partnerships

- Federal, State, Regional, Local Governments
- Academic Institutions
- Non-Governmental Organizations
- Tribes

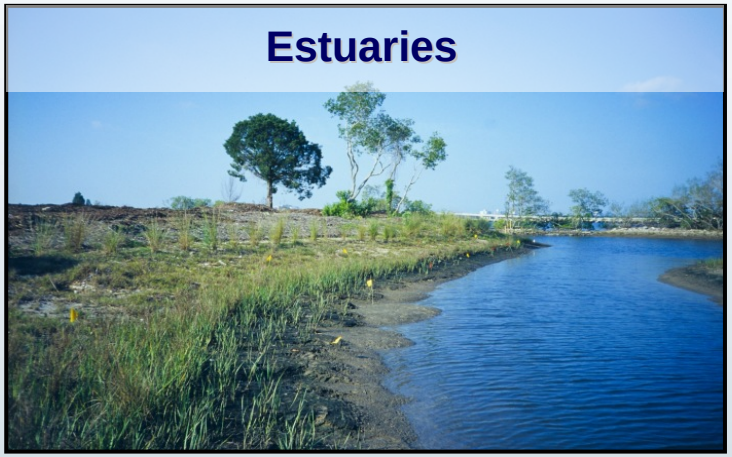
Roles of Partners

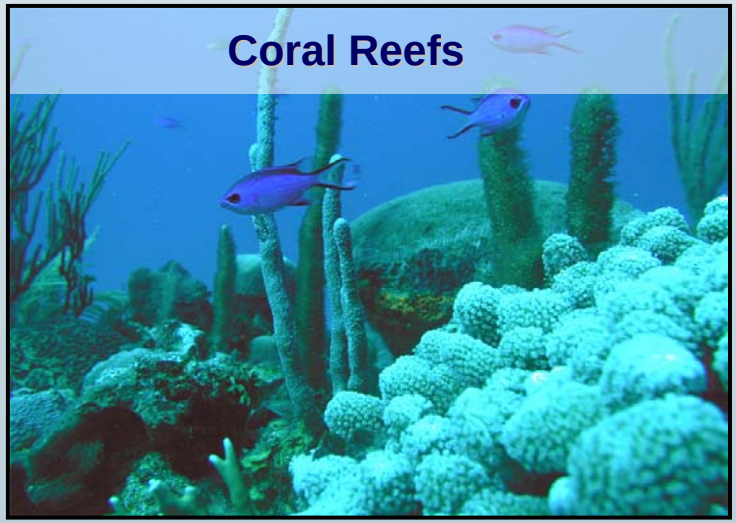
- Collaborative Work with CCMA
- Project Planning, Execution, and Product Development
- Technical expertise
- Local Knowledge

"The shortage of human and logistical infrastructure in Southwest Alaska makes field work here challenging and expensive. Partnering with NOAA's Center for Coastal Monitoring and Assessment makes vital water quality monitoring feasible here that would be difficult if not impossible otherwise."

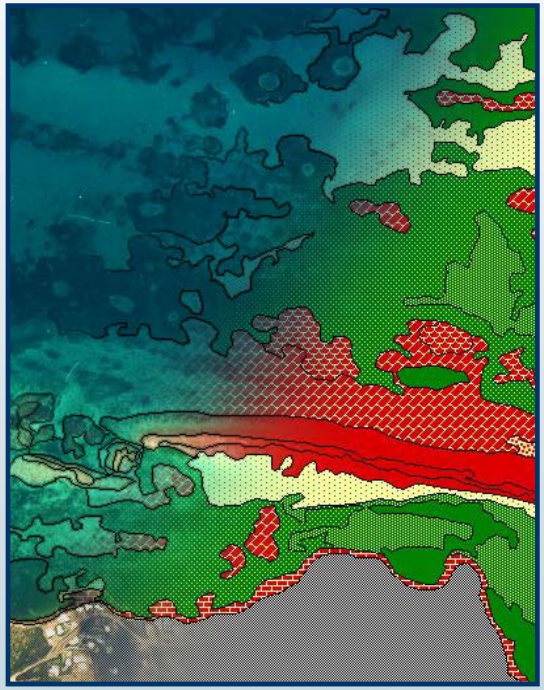
- Bristol Bay Native Association

Ecosystem-based Research

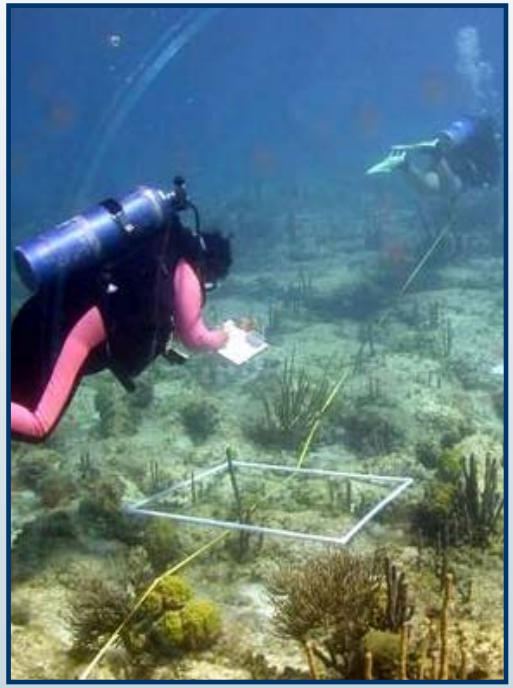




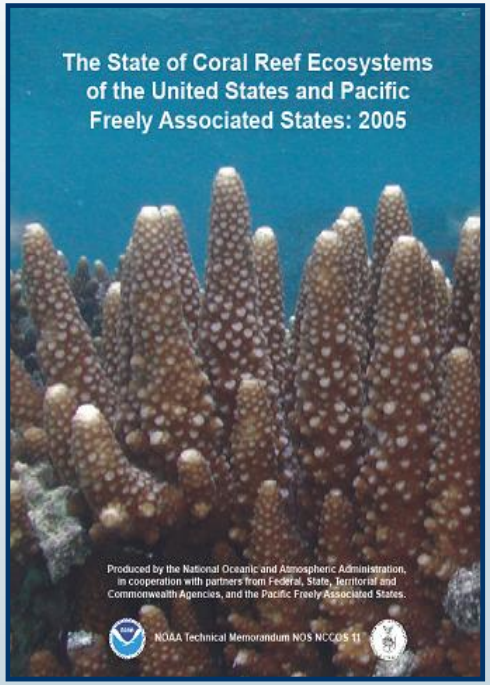
Biogeography Branch: Coral Reef Ecosystem Assessment Mapping & Monitoring



Mapping



Monitoring



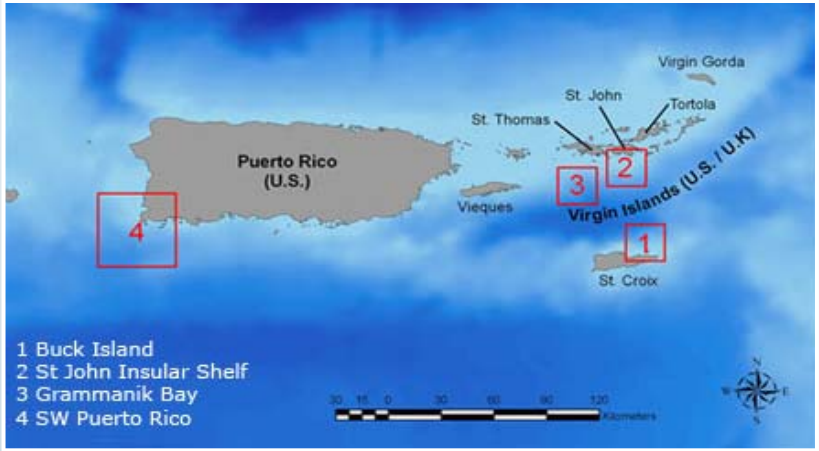
Assessment Products

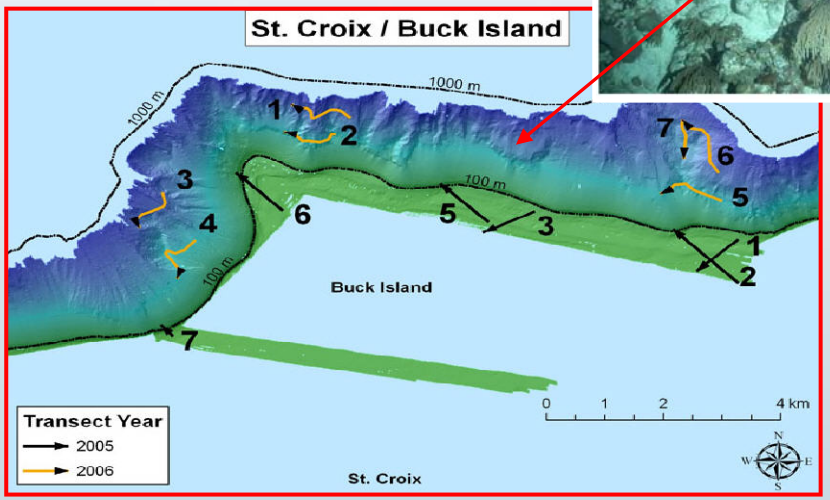
"The fish monitoring and tracking work that NOAA's Center for Coastal Monitoring and Assessment does in the VI National Park and VI Coral Reef National Monument is of vital importance in determining the status of fish populations in our waters. Tracking of fish will enable ecological linkages to be established between the park, monument, and adjacent habitats. All of this work will enable effectiveness of various degrees of marine protected areas to be assessed. This work could not be accomplished with current levels of NPS funding and resources."

- V.I. National Park/V.I. Coral Reef National Monument

Biogeography Branch: Tool Development/Technical Consultation

Example: Benthic Habitat Viewer Browser Tool



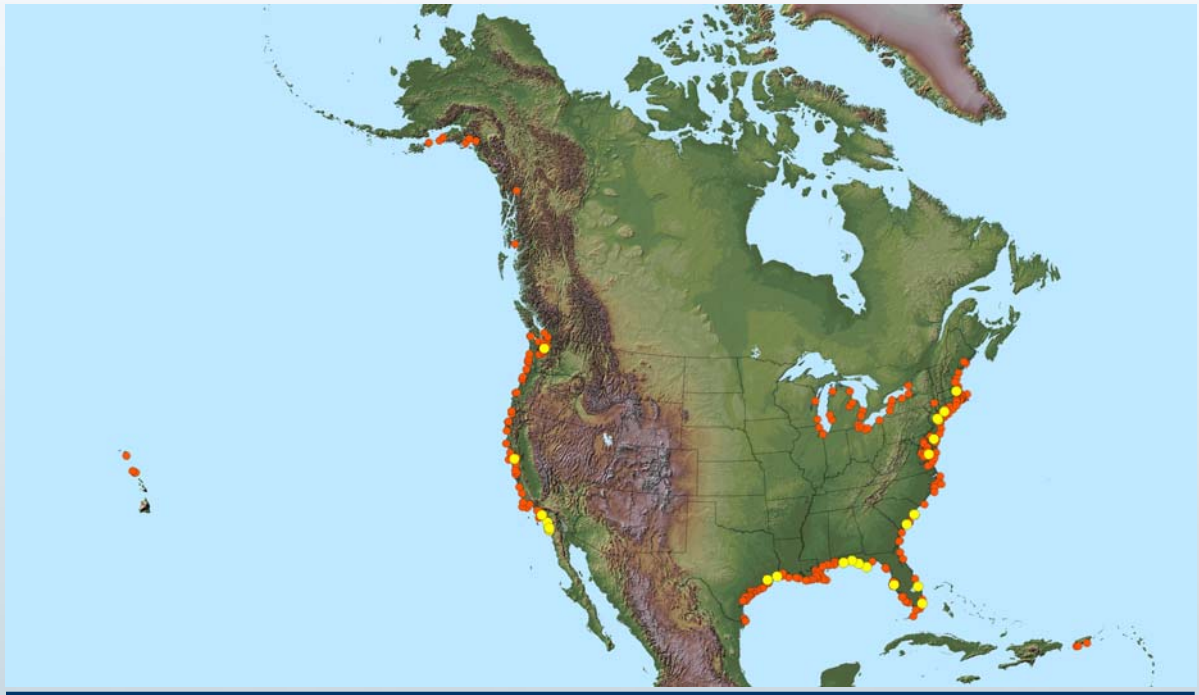


Services: ArcGIS Tools, Browser Tools, Consultation

"The GIS tool created and operated for us during the Sanctuary Advisory Council and Research Area Working Group meetings has been invaluable for helping us look at possible Research Areas that will mazimize achieving our science and management objectives in a wide range of habitats and for a wide range of species, with minimum impact on user groups."

- Gray's Reef National Marine Sanctuary

COAST Branch: Coastal Pollution



Mussel Watch Sites | Bioeffects Assessment Locations

- **National Status & Trends:**
  Mussel Watch &
  Bioeffects Programs
- **280 sites nationwide monitored annually for 120 contaminants**
- **Nation's longest running coastal contaminant monitoring program**
- **Comprehensive assessments of environmental contamination, toxicity, and biological community condition in bays and estuaries**

"Collaborating with NOAA's Mussel Watch Program benefits the Southern California Coastal Water Resources Project and other organizations such as the Multi-Agency Rocky Intertidal Monitoring network (MARINe) by increasing the spatial coverage of coastal environmental monitoring to include areas of special biological significance and putting chemical contaminant levels along our coastline into a 'national perspective'."

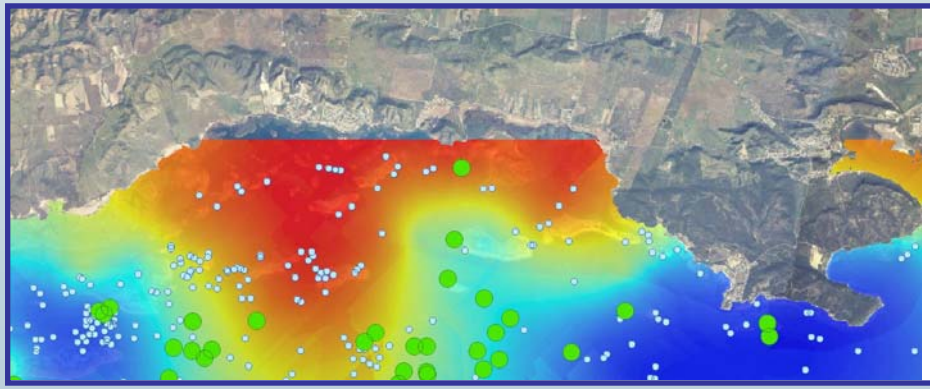
- Southern California Coastal Water Research Project

COAST Branch: Contaminant Distributions in Caribbean Ecosystems

Project Objectives

- To assess chemical contaminant levels in water, sediments, and coral tissues
- Identify and quantify biomarkers and identify pathogens in coral tissues
- Develop and test hypotheses relating contaminant burdens to measures of coral health
- Link Results of these exercises to ongoing regional coral reef ecosystem monitoring – including coral health and diversity; reef fish distribution, abundance, and diversity; phycology, and land use practices
- Evaluate application of the analytical construct to other areas in the US Caribbean and Pacific basins

PAH Plume: Strong Negative Correlation with Coral Species Richness



Nonparametric: Spearman's Rho

Variable	by Variable	Spearman Rho	Prob>|Rho|	-.8 -.6 -.4 -.2 0 .2 .4 .6 .8
Mean(r0)	CORAL_RICH	-0.7939	0.0061	

Green dots indicate locations where coral species richness was within the top 25th percentile for brain, branching, pillar, encrusting, mound and boulder corals. Blue dots are remaining locations at reef sites.

Interviews with individuals involved with mapping and monitoring

- May map and characterize at finer scales
- New characterization tools (automation)
- Underwater positioning system
- Fine scale oceanic dynamics for larval dispersion
- Acoustic monitoring and identification of fish
- Listening systems to monitor fish spawning
- Possible future instruments – hyperspectral; perhaps fused with other sensors
- AUV platform

NOAA's Coral Reef Conservation Program: past, present and future

*1998 US Coral Reef Task Force (CRTF)

*2000 NOAA's Coral Reef Conservation Program (CRCP) "lead national efforts to better understand and conserve coral reefs, reef species, and the human communities that depend on them…"

*2001 CRCP projects integrated into Coral Reef Ecosystem Integrated Observing System (CREIOS), compatible with Integrated Ocean Observing System (IOOS).

*2007 "Roadmap" for future CRCP endeavors; three top priorities

- Impacts of fishing
- Impacts of land-based sources of pollution and
- Impacts of climate change

*2008 and 2009 - CRCP "redefining the scope of its national program activities, including a reassessment of mapping and monitoring activities in CREIOS".

CREIOS – composed of four NOAA Line Offices and program offices.

Source: NOAA Coral Reef Conservation Program, National Program for Mapping, Monitoring, and Data – White Paper (draft)

CREIOS Pacific Workshop, week of November 17, 2008

Coral Reef Monitoring and Assessment Workshop

*** In September 2007 NOAA's Coral Reef Conservation Program underwent an external review and subsequently developed a Roadmap for implementation of the results of the review. The Program has narrowed its focus to three threats to coral reefs 1) Climate change 2) Land-based sources of pollution 3) Impacts from fishing and has created working groups for each threat to determine goals and objectives for each.**

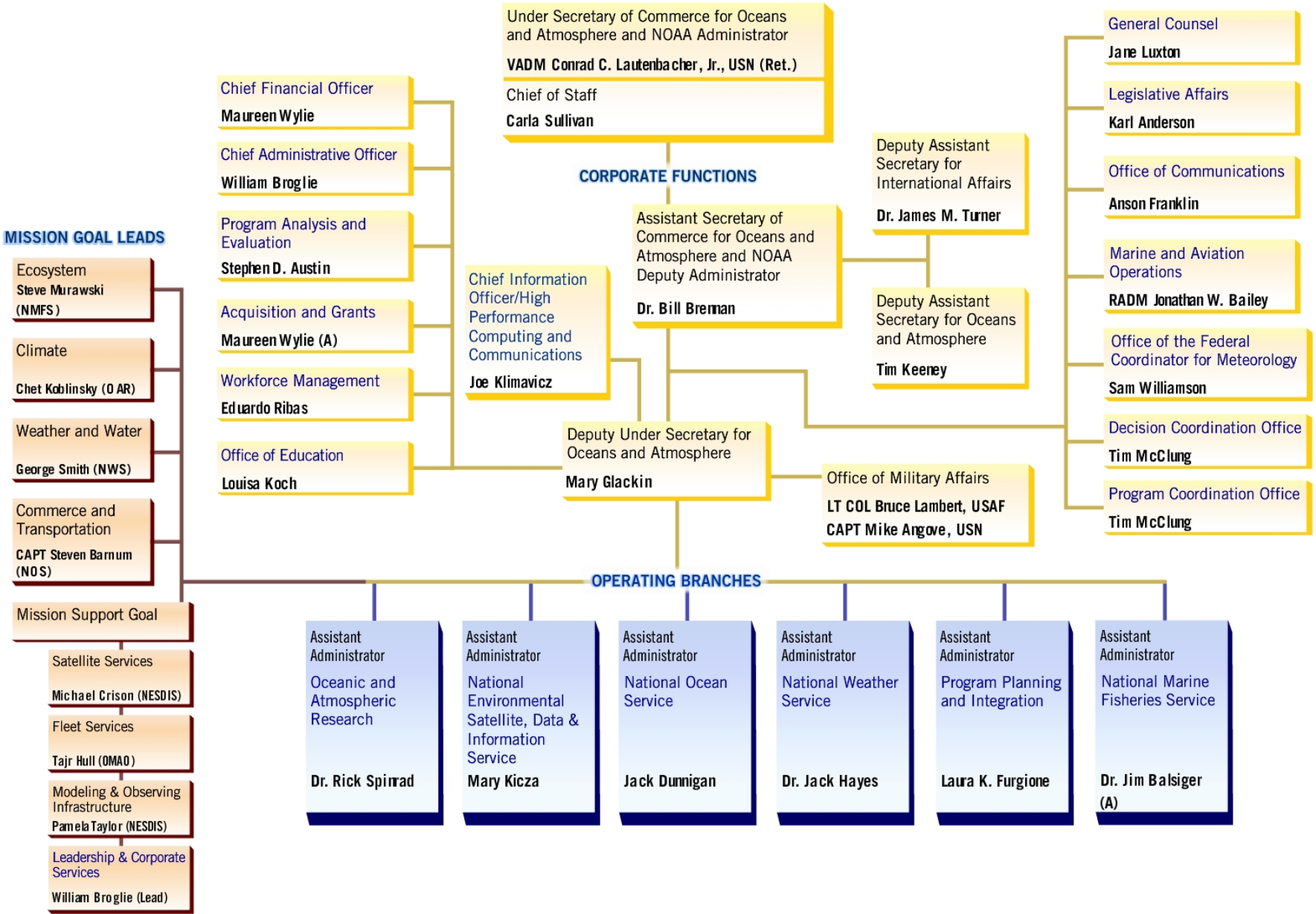
*** Also as part of the roadmap implementation we are reviewing and potentially revising long-term plans for our monitoring and mapping activities, collectively known as the Coral Reef Ecosystem Integrated Observing System (CREIOS), to ensure they are cost-effective, aligned with management needs, and allow for the timely delivery of required products and services to all essential users, given funding constraints.**

*** As a first step, this month the CRCP will bring together Pacific coral reef ecosystem managers and CRCP scientists at a three-day workshop in Honolulu, Hawaii. A subsequent workshop will be held in the Caribbean next year.**

*** As mentioned, this process may bring about some changes in direction and we look forward to partnering with DOD as we move forward with our monitoring program.**

Shannon Simpson CRCP 2008.11.14

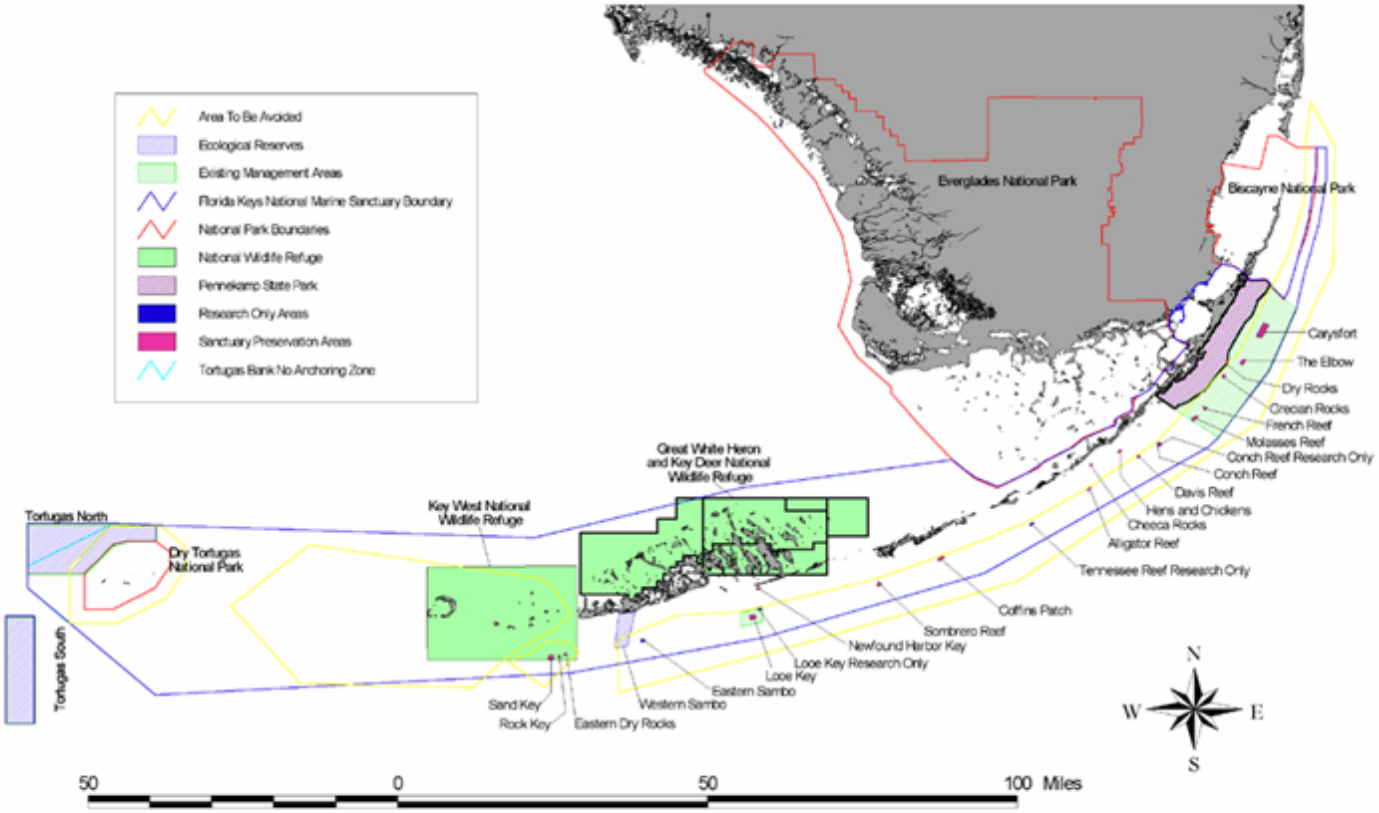
NOAA ORGANIZATION



Injury Assessment and Restoration Monitoring on Coral Reefs within the Florida Keys National Marine Sanctuary

Bill Goodwin
Sanctuary Resources Manager
FKNMS

Florida Keys National Marine Sanctuary



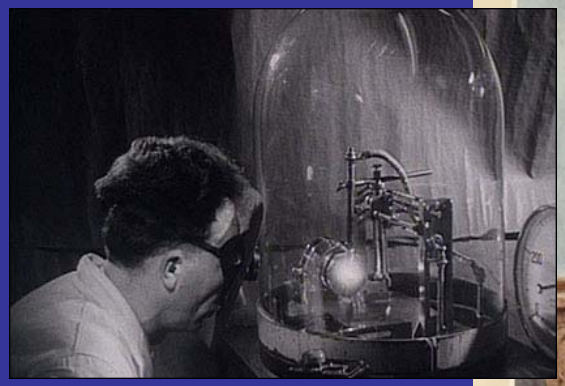
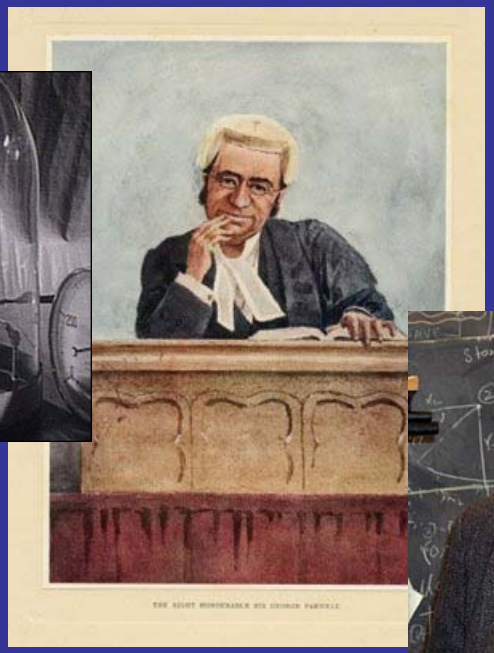
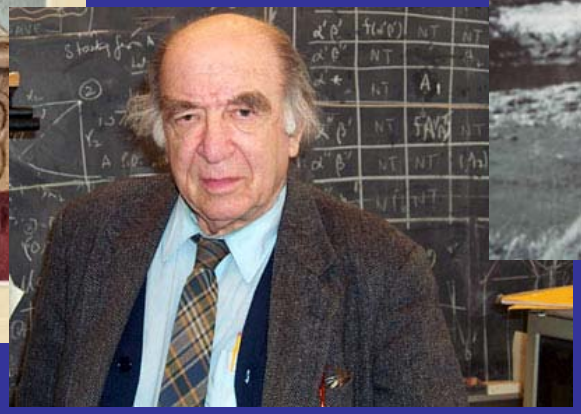
2900 square nautical miles of Sanctuary
500-600 vessel groundings annually
Approximately 15% occur on coral reef habitat

Statute 312

Whenever a grounding occurs within a national marine sanctuary, NOAA can seek damages to cover response, injury and damage assessment, restoration and replacement of the damaged habitat or acquisition of equivalent habitat, and compensation of the public for the value of the damaged resources until full recovery.

Primary goal of the Sanctuary's Coral 312 program:

To prepare rapid, cost-effective, litigation-quality claims for injuries to coral resources resulting from vessel groundings and other mechanical injuries, and to implement the restoration and monitoring of coral reef ecosystem injuries

The Coral 312 Program uses an interdisciplinary team of biologists, economists, lawyers, and resource managers to assess and recover natural resource damages from the vessel owner/operator who cause these injuries. The funds collected are then used to implement the restoration of and monitor restored coral reef ecosystems

Elements of a 312 Case
(in the order in which they usually occur)

- *Initial response*
- *Assessment*
  - *Documentation*
  - *Quantification*
  - *Location*
  - *Description*

- *Emergency triage/restabilization (if necessary)*
- *Primary restoration*
- *Monitoring*
- *Compensatory projects*

Basic Assessment Tools

Boat

Waterproof paper or slate

Dive/snorkeling gear

Coral Injury Assessment Field Data for Vessel Groundings

Vessel/Site Name: **Assessors:**

Date:

Time:

Location: **Tide State:**

Water Visibility: **Water Depth:**

Current: **Site Marked:** Y N

Sea State: **With:** Float Stake

GPS Position:

Length/Heading of Track(s):

Habitat Type: Patch Reef Bank Reef Coral Rubble Hard Bottom

Coral/Other Species Impacted:

Notes/Site Sketch: Use to describe keel grooves, trenching, fractured colonies, broken branches, dislodged/overturned colonies, scarified/parking lot, berms of coral rubble, bottom paint skid marks, striations, prop scars, or nicks in colonies.

Basic Assessment Tools

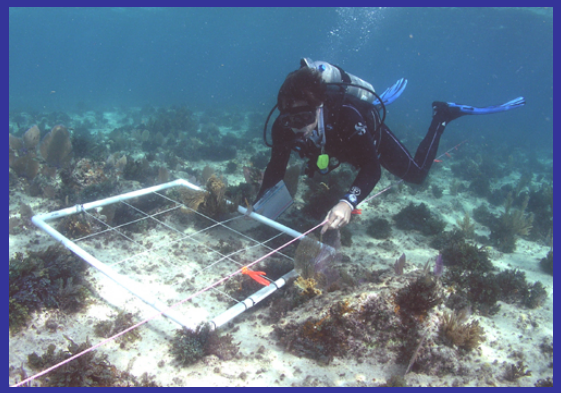
•Compass

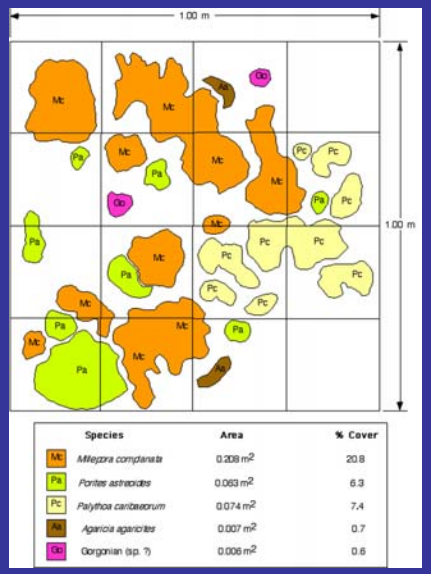
•GPS

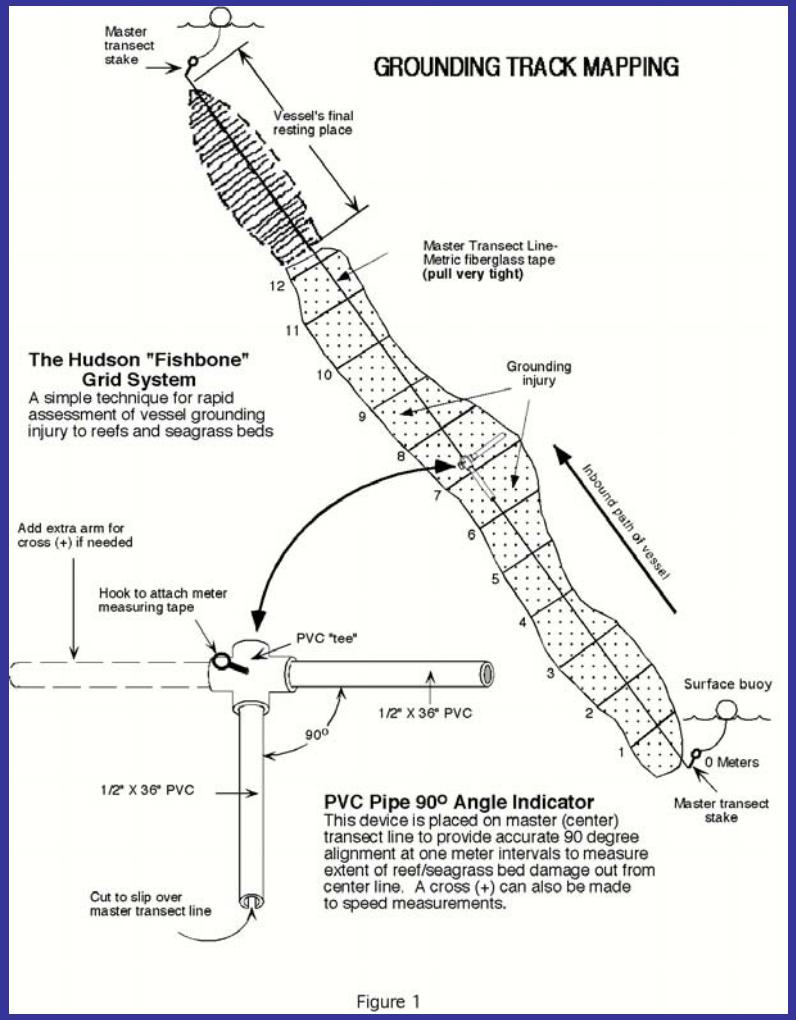
•U/W Photo/Video

•Meter Tape

•Quadrat







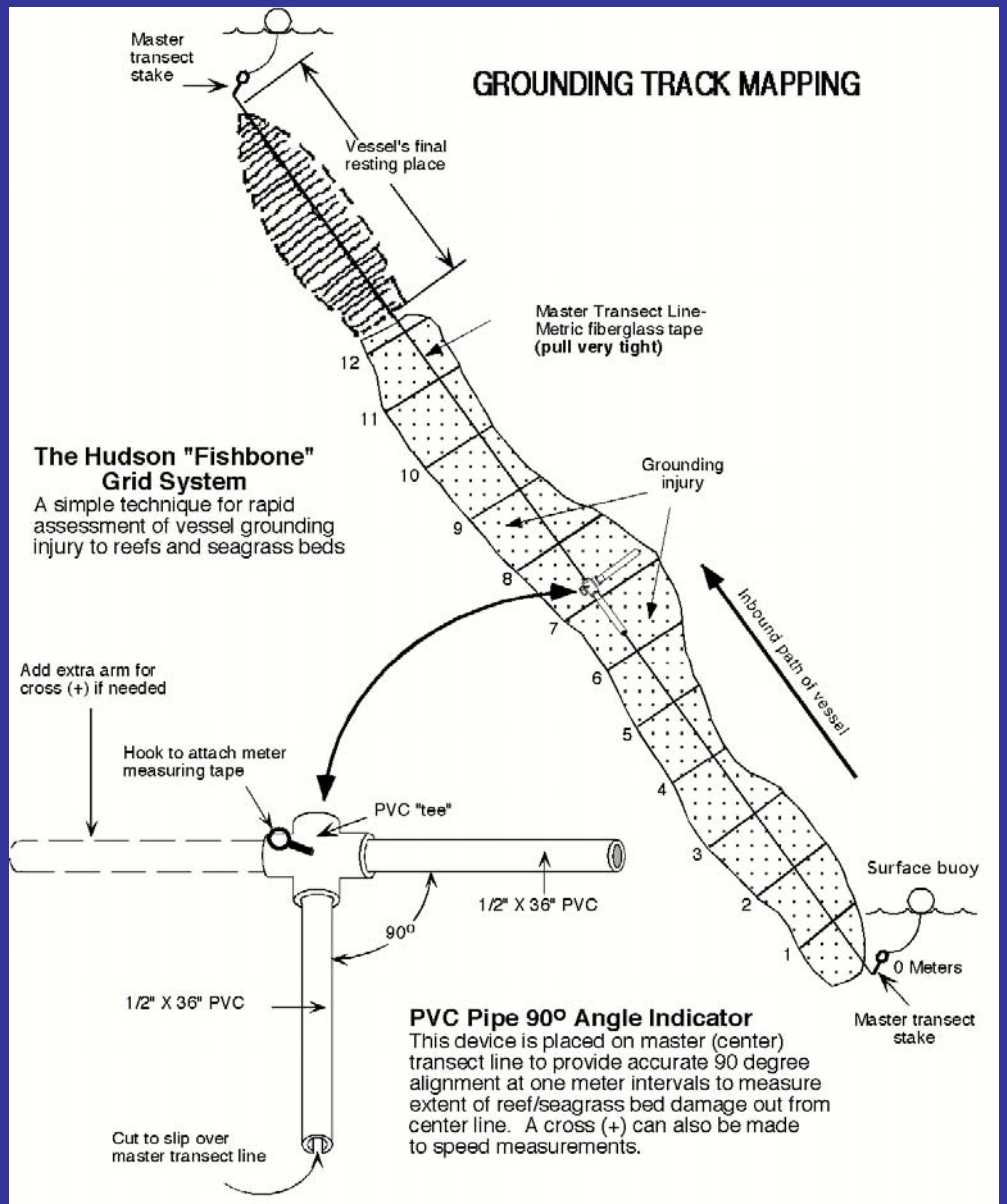
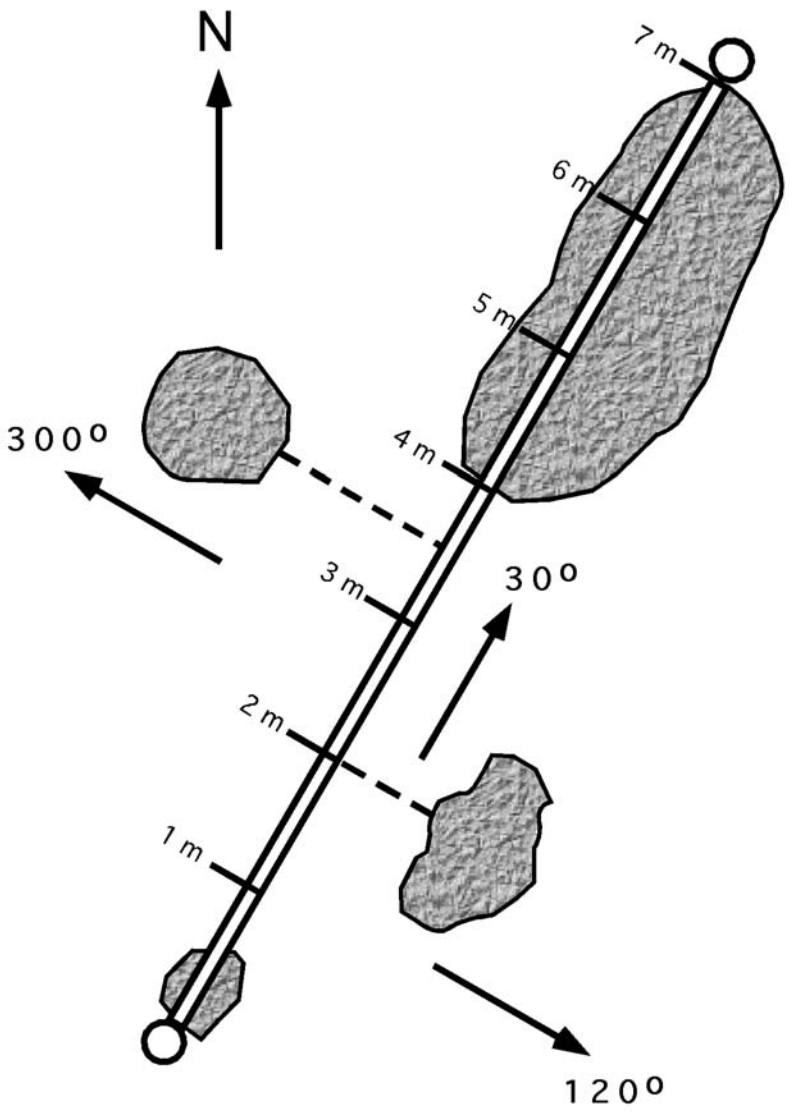

GROUNDING TRACK MAPPING
Master transect stake
Vessel's final resting place
Master Transect Line-
Metric fiberglass tape
(pull very tight)
12
11
10
9
8
7
6
5
4
3
2
1
Grounding injury
Inbound path of vessel
The Hudson "Fishbone" Grid System
A simple technique for rapid assessment of vessel grounding injury to reefs and seagrass beds
Add extra arm for cross (+) if needed
Hook to attach meter measuring tape
PVC "tee"
1/2" X 36" PVC
90°
1/2" X 36" PVC
Cut to slip over master transect line
Surface buoy
0 Meters
Master transect stake
PVC Pipe 90° Angle Indicator
This device is placed on master (center) transect line to provide accurate 90 degree alignment at one meter intervals to measure extent of reef/seagrass bed damage out from center line. A cross (+) can also be made to speed measurements.

Figure 1

Measurement/Mapping of Intermittent Coral Injury



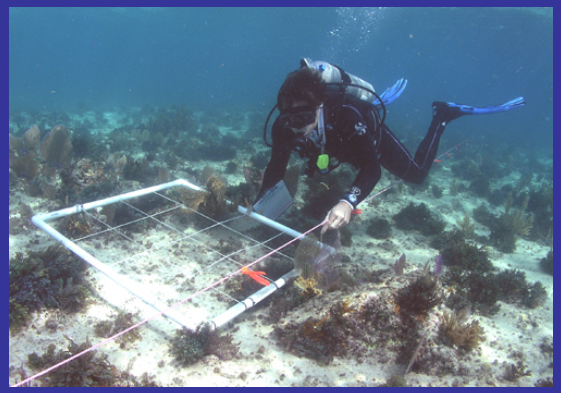
Basic Assessment Tools

•Compass

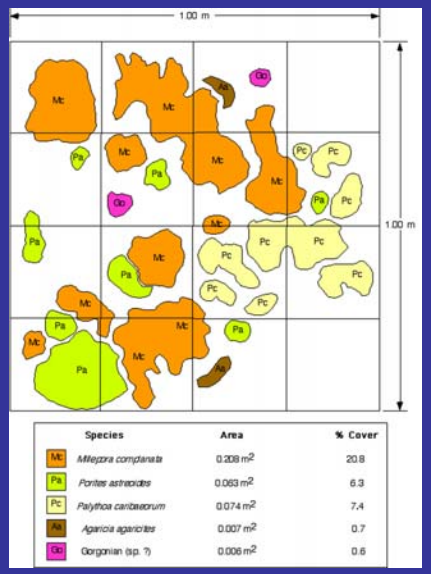
•U/W Photo/Video

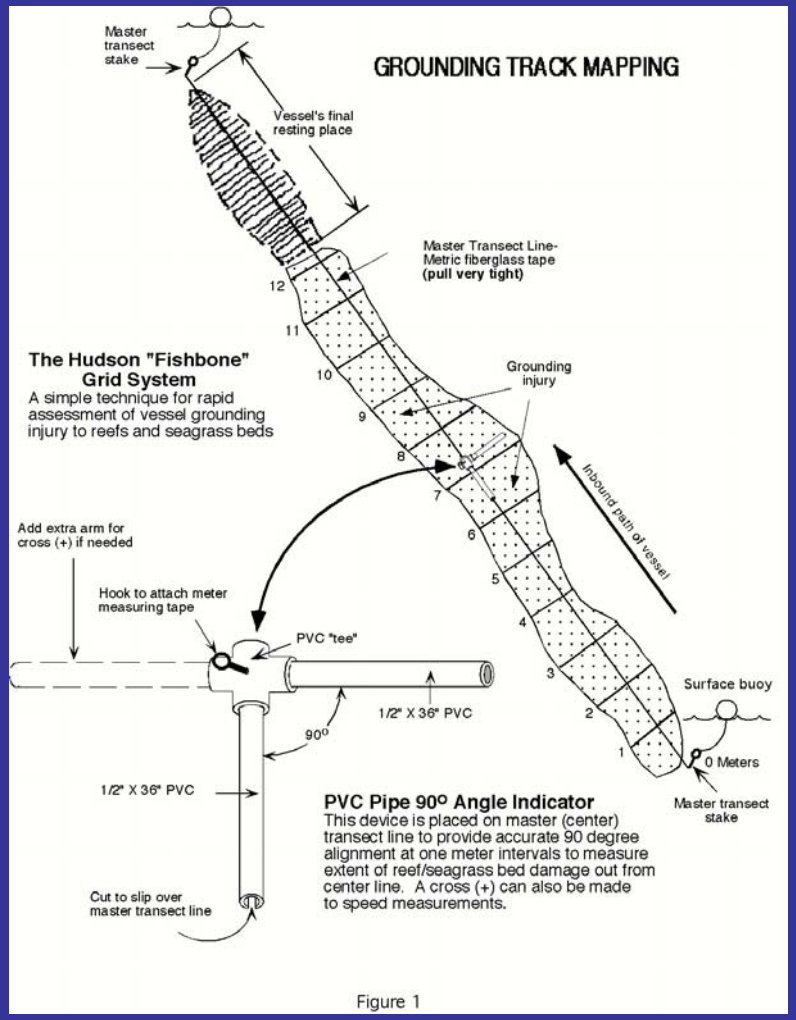
•Meter Tape

•GPS

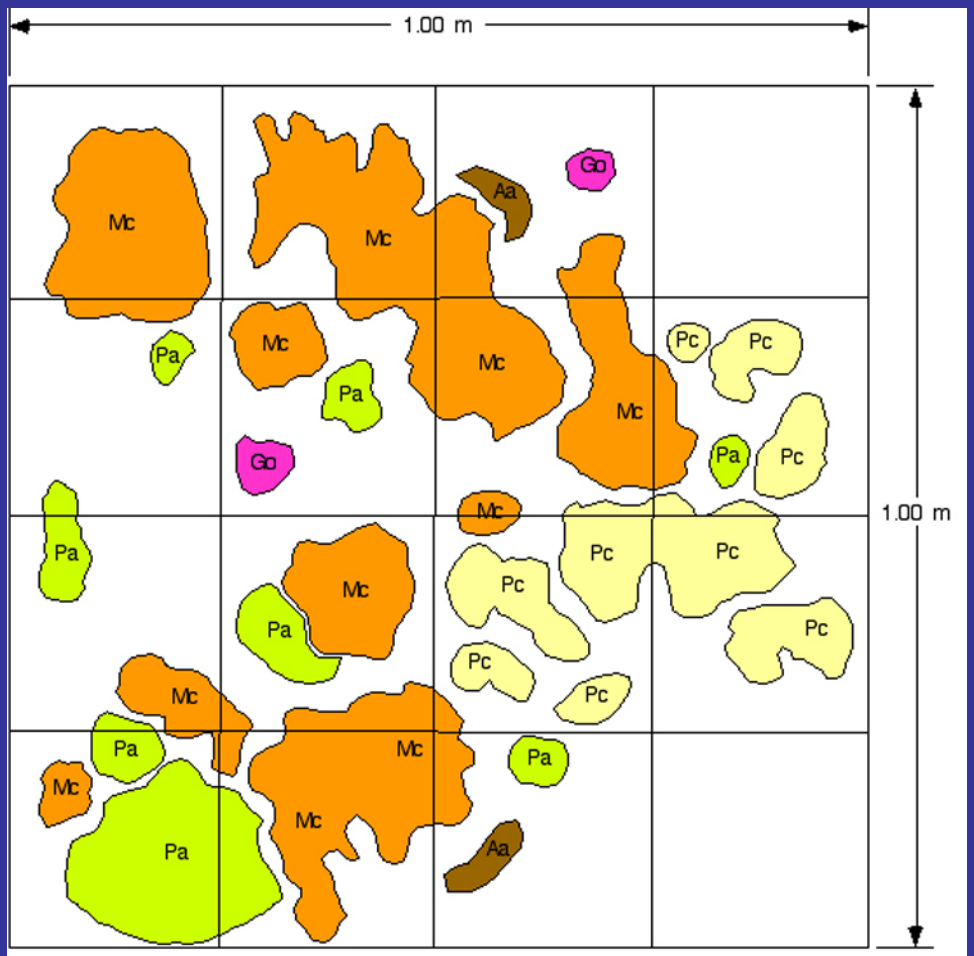


•Quadrat





Digitized benthic map of unimpacted reef crest adjacent to injury; generated from photo quadrat data



	Species	Area	% Cover
Mc	*Millepora complanata*	0.208 m^2	20.8
Pa	*Porites astreoides*	0.063 m^2	6.3
Pc	*Palythoa caribaeorum*	0.074 m^2	7.4
Aa	*Agaricia agaricites*	0.007 m^2	0.7
Go	Gorgonian (sp. ?)	0.006 m^2	0.6

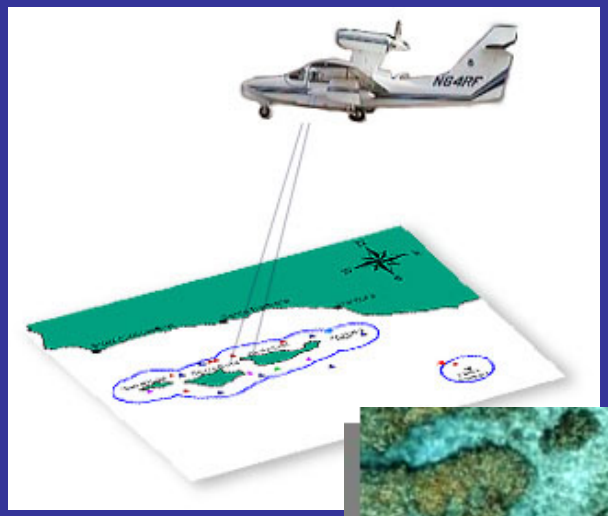
Total living coral cover = 35%

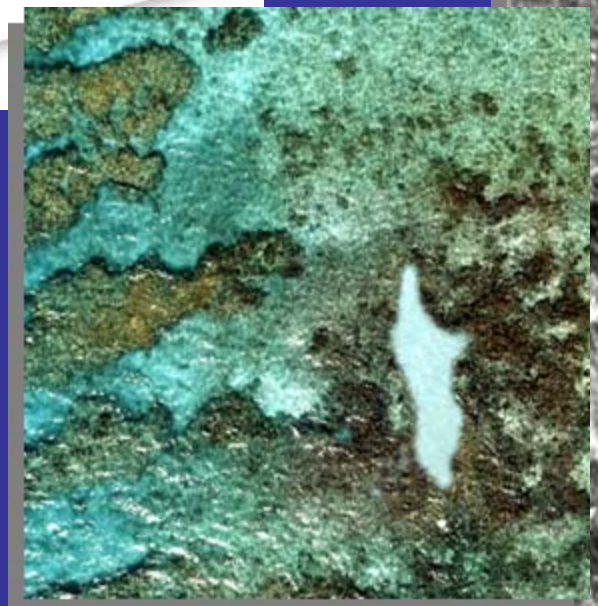
Coral Cover loss in A-A′= 5.1 m^2

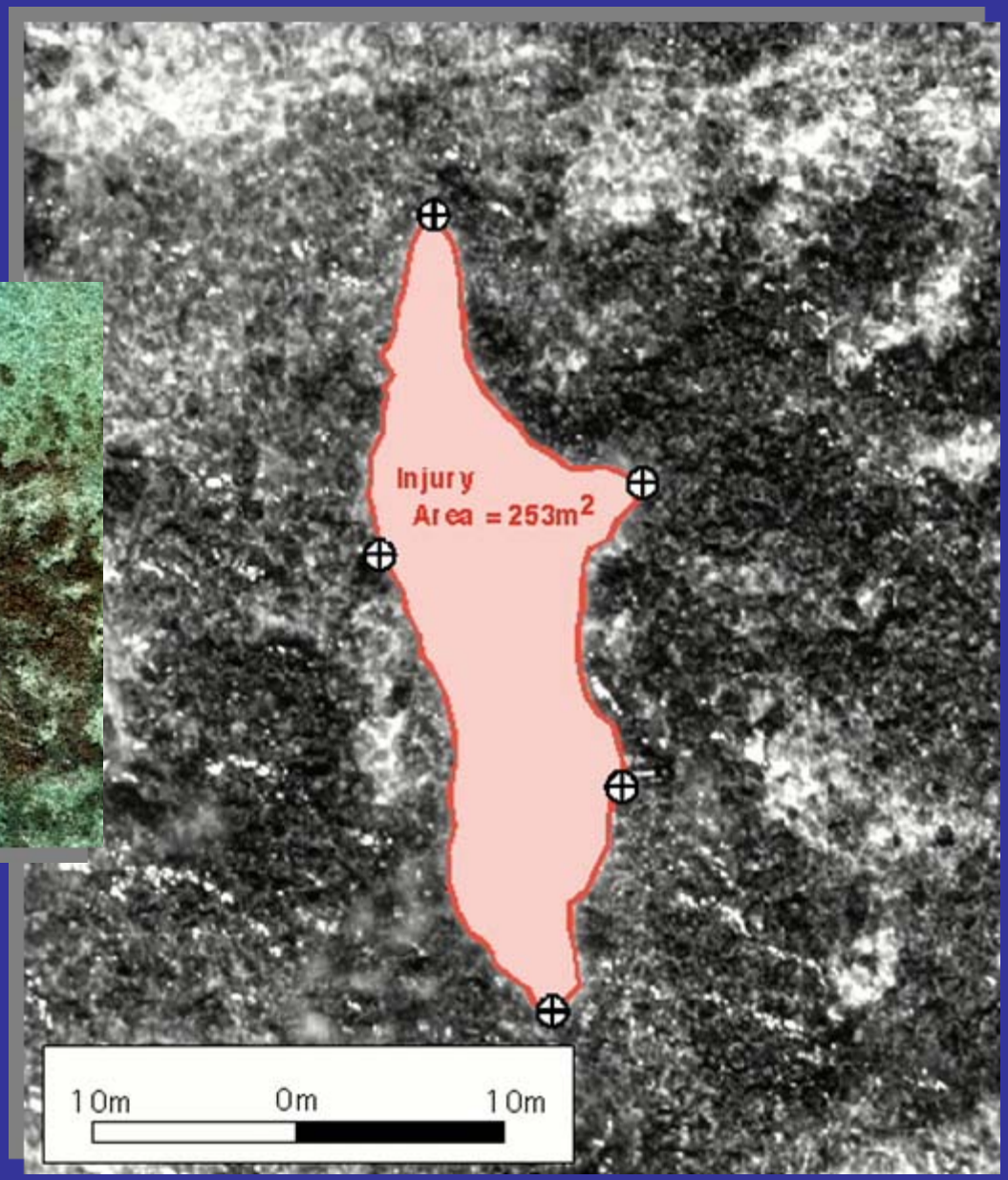
Basic Assessment Tools

Buoys and Stakes for Temporary, Long Term and Permanent Site Marking

Advanced Techniques





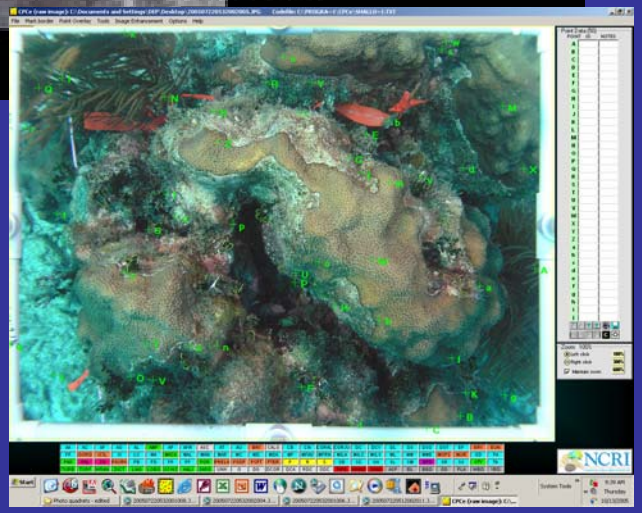


Aerial photo analysis

Advanced Techniques

Video Transects

Linear video image collage
- RavenView
- Snap DV



Point count analysis of single frame from video

Advanced Techniques

Underwater mapping systems

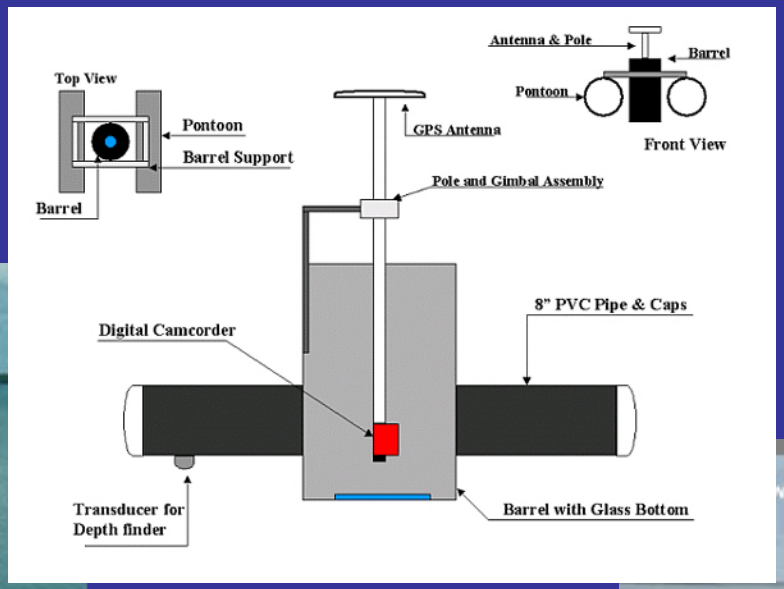
M/V Casitas - NW Hawaiian Islands

•AquaMap

•CobraTac

Advanced Techniques

NOAA/NGS
Shallow Water Positioning System (SWaPS)



Remote control unit

Skiff-mounted unit

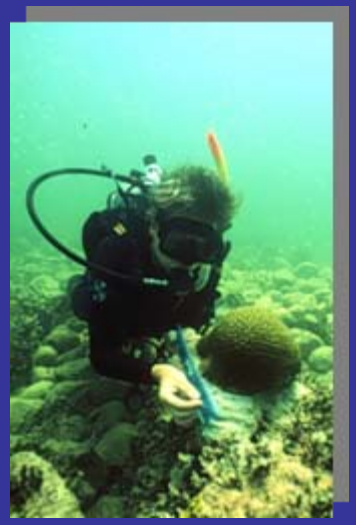
Single frame from SWaPS video transect with corresponding positioning data displayed at bottom of frame

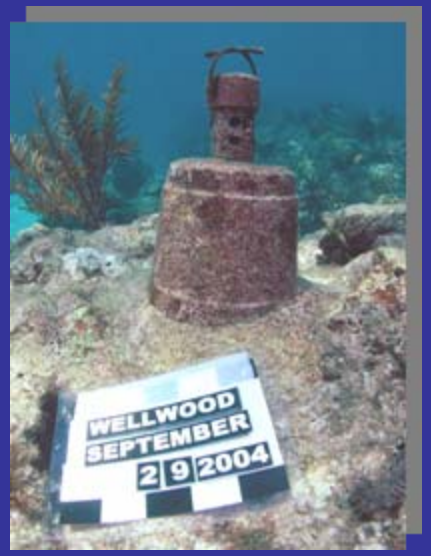
M/V Adaro site, Grecian Rocks Reef

Assessment phase

Restoration completed

Monitoring is an essential component of any major coral reef restoration effort…



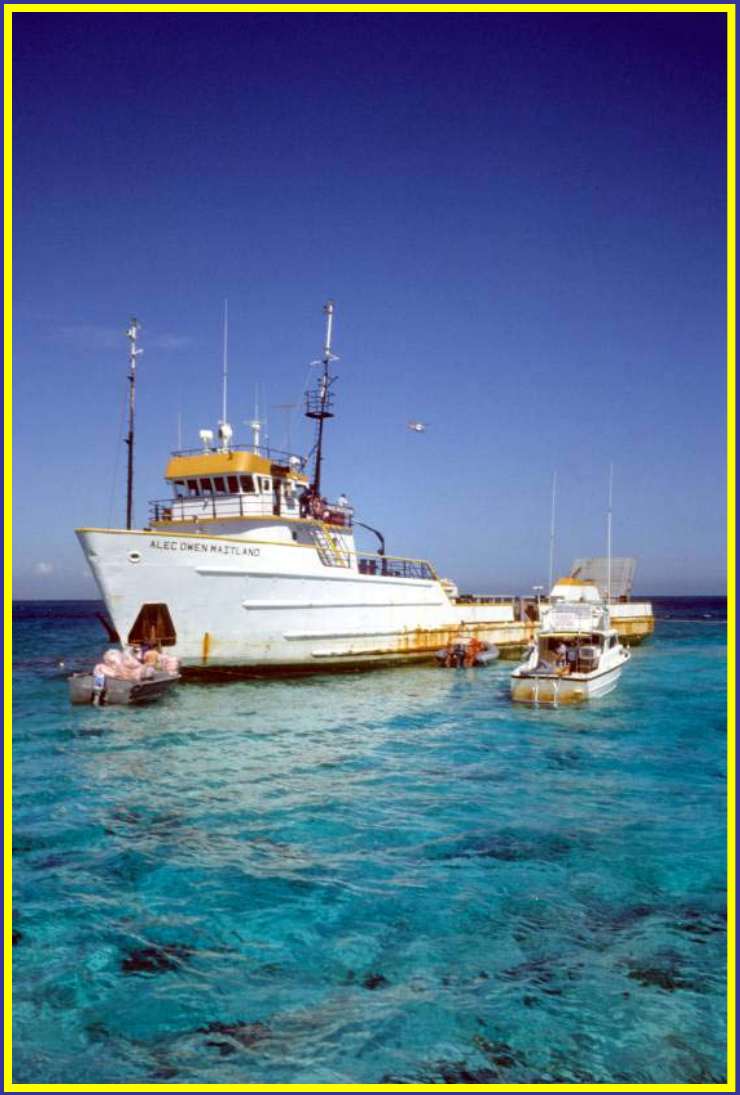


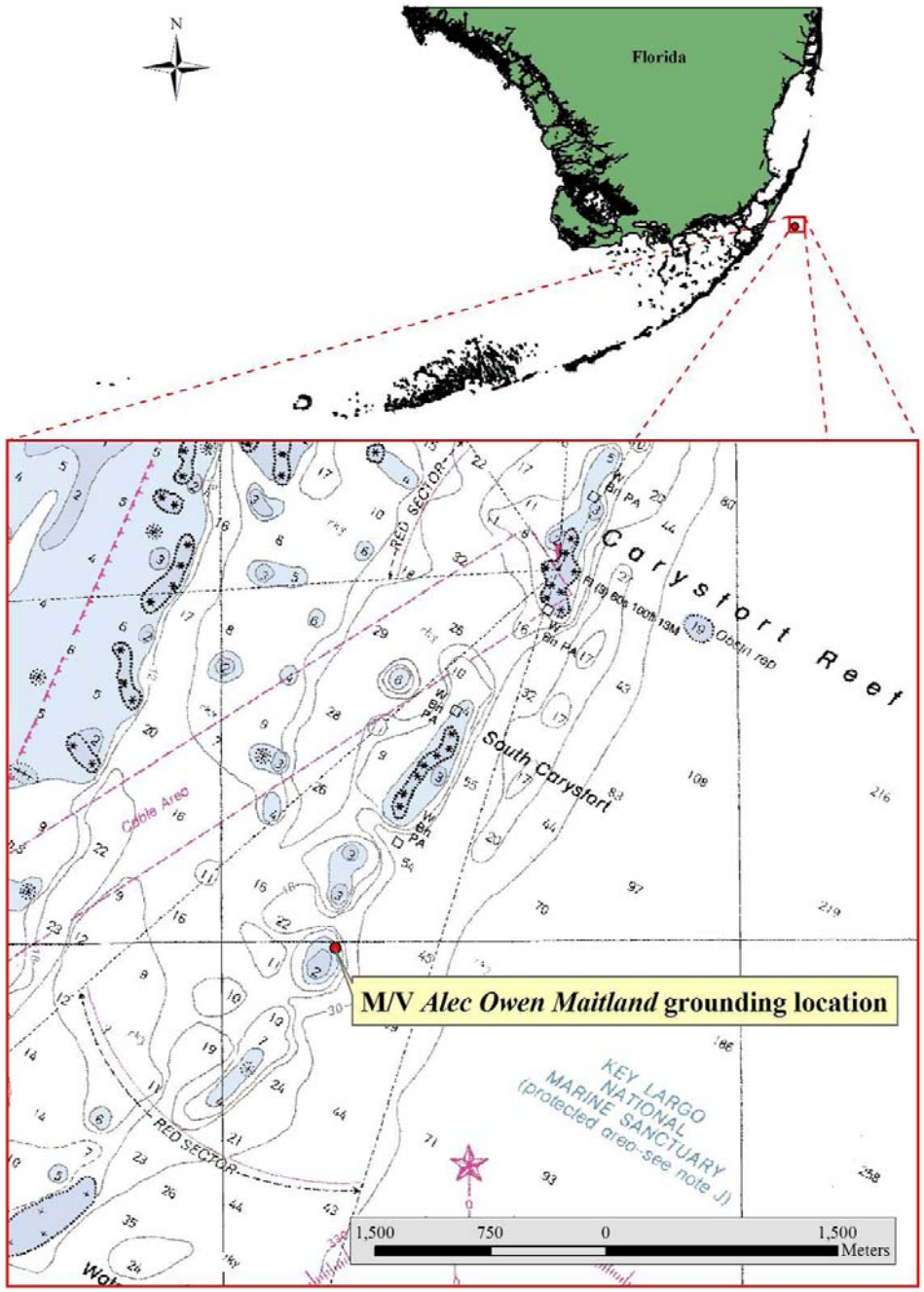
Learn from Past Projects

Monitoring Efforts

- Currently monitoring 33 restoration sites within the FKNMS (both seagrass and coral)
- Determine Success and Efficacy (or Failure) of Past Efforts
- Understand what works – what doesn’t – and why?
- Implement Adaptive Management Program

Restoration Case Study
M/V Alec Owen Maitland




N
Florida
Carysfort Reef
South Carysfort
RED SECTOR
Cable Area
M/V *Alec Owen Maitland* grounding location
KEY LARGO NATIONAL MARINE SANCTUARY (protected area--see note J)
1,500 750 0 1,500
Meters

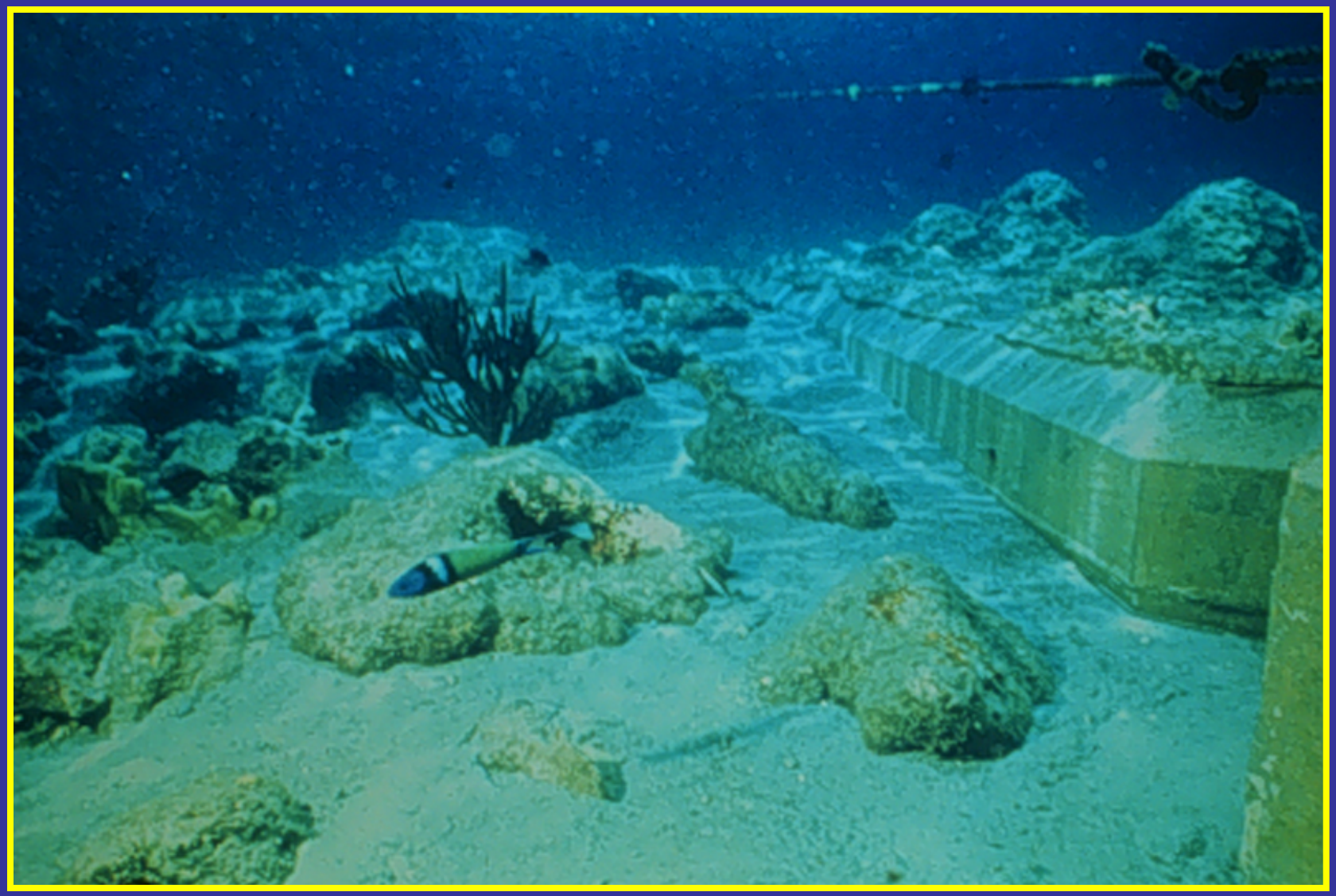
Significant injuries to coral reef resources resulted from crushing effect of vessel's hull

However, the most serious injury occurred when the captain attempted to “power off” of the reef, causing an enormous “blowhole”, or prop-dredged excavation

Sidewall view of prop-wash crater

Blueprint for restoration of prop-wash excavation crater

Deployment of modules from work barge

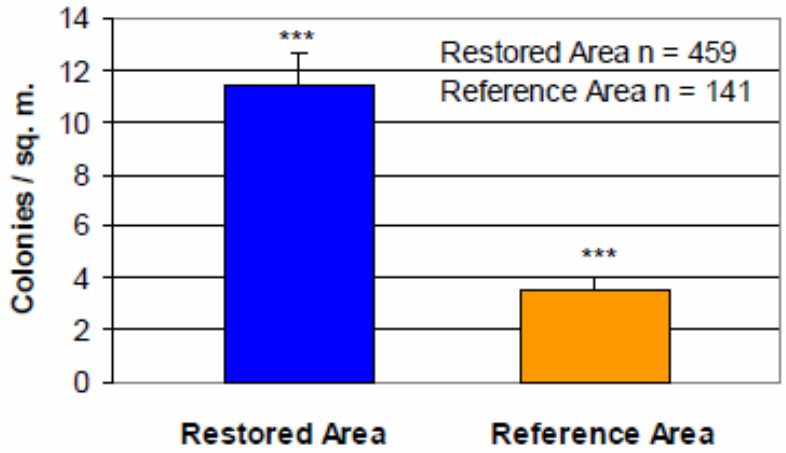


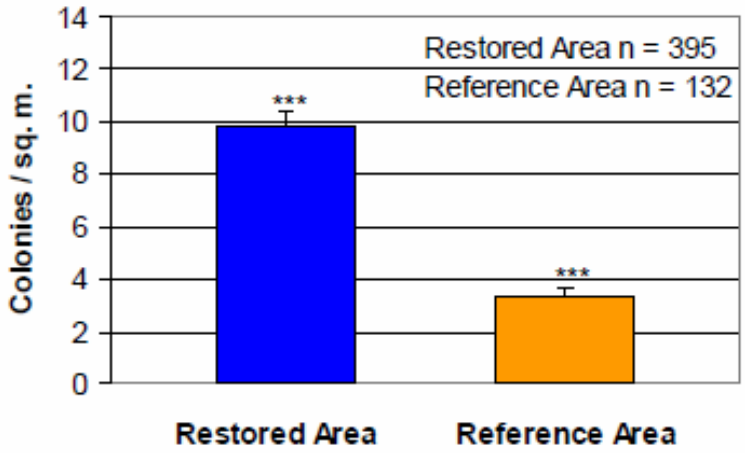
The finished product

Ten years later…

Diver conducting survey in restoration area (left) and reference area (right)

Representative benthic organisms surveyed on the *Maitland* restoration armor units. Starting from top left: *Diploria* sp., *Siderastrea siderea*, *Stypopodium zonale* next to *Halimeda* sp., and *Porites astreoides* next to *Gorgonia ventalina*





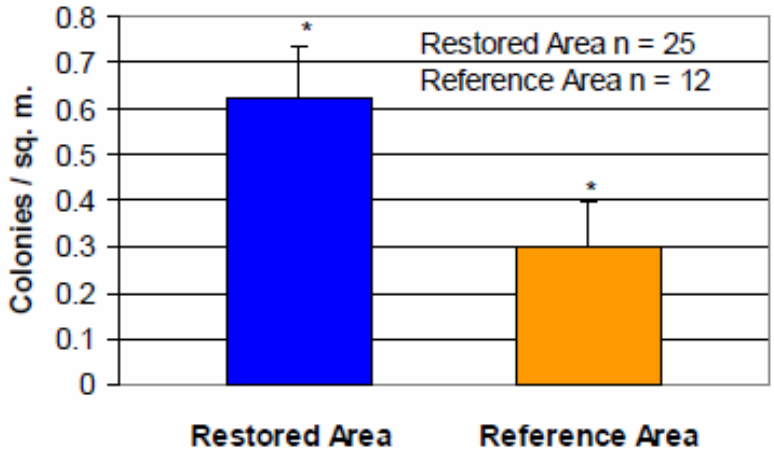
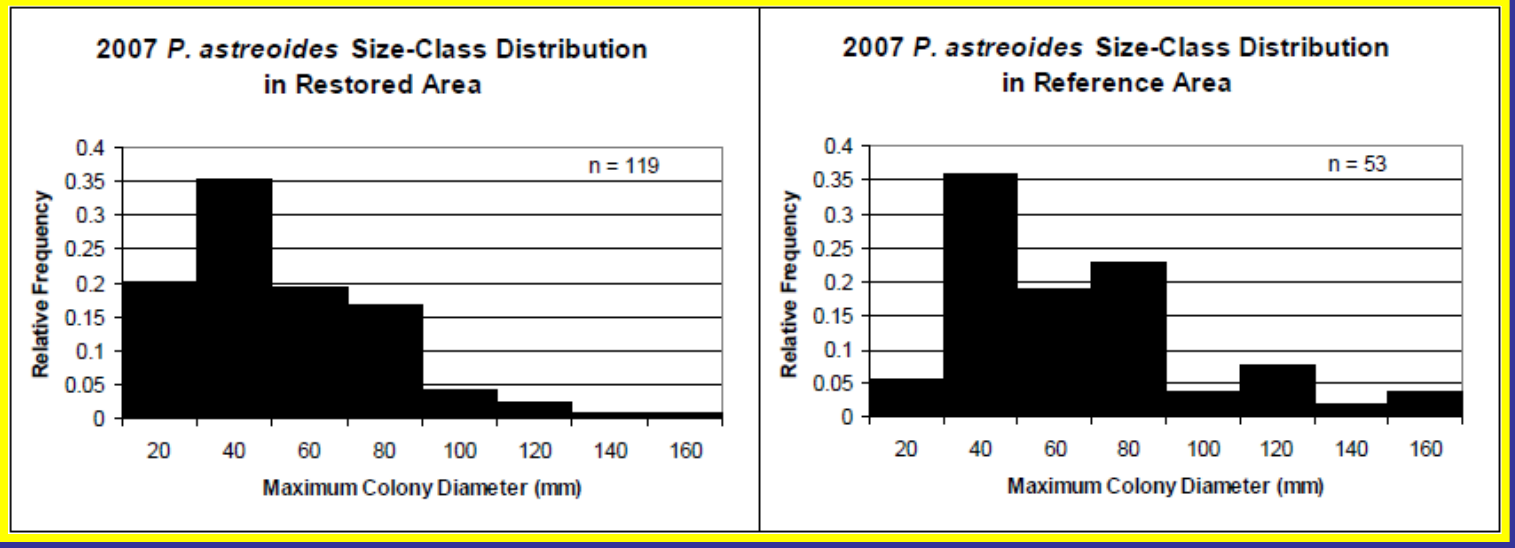


Figure 11. 2004 densities of all 3 groups of corals (Note differing scales used for *Millepora*). Error bars = Standard Error; *** notation indicates highly significant difference ($p < 0.0001$), * notation indicates significant difference.

Number of Scleractinian colonies, by species, surveyed in 2007 at the *Maitland* restoration site.

Species	Restored area	Reference area
Agaricia spp.	0	2
Diploria spp.	2	0
Favia fragum	0	1
Montastraea cavernosa	0	0
Porites astreoides	119	53
Porites porites	17	9
Siderastrea radians	11	3
Siderastrea siderea	19	4
Total	**168**	**72**



The rapid convergence rates observed in this study were influenced by the life-history characteristics of *Porites astreoides*, the dominant coral on both the reference habitat and the restoration structures.

***Porites astreoides* is an opportunistic coral with a relatively small adult colony size, and recruitment and survivorship rates among the highest in the region (Miller et al. 2000; Kojis & Quinn 2001; Tougas & Porter 2002).**

In contrast, where reference communities are dominated by corals with limited sexual recruitment and very large adult colony size like *Montastraea* spp. and *Acropora* spp (Szmant 1986), convergence rates can be expected to be significantly slower.

What have we learned?
(or, what to do, what not to do, and why)

- Most reef restoration efforts have been set *ad hoc*
- Most efforts have not been founded on scientific data
- Ecosystem function has been absent in the decision-making process
- *Surprise* – community structure of restored reefs are converging on natural reefs in spite of our efforts

SERDP Coral Reef Monitoring and Assessment Workshop

Surveys of coral reef and hard-bottom habitats

FKNMS

William F. Precht
NOAA – Florida Keys National Marine Sanctuary

Several monitoring activities that are ongoing in the FKNMS have been modified slightly to become part of the three-level FKNMS Zone Monitoring Program. The FKNMS Zone Monitoring Program began in 1997.

Rapid assessment and monitoring of coral reef habitats in the Florida Keys National Marine Sanctuary

Principal Investigator:

Steven L. Miller, Center for Marine Science, University of North Carolina at Wilmington (UNCW)

Project Team:

Mark Chiappone, Center for Marine Science, University of North Carolina at Wilmington

Leanne M. Rutten, Center for Marine Science, University of North Carolina at Wilmington

Dione W. Swanson, Division of Marine Biology and Fisheries, Rosenstiel School of Marine and Atmospheric Science, University of Miami

Rapid Assessment Methods

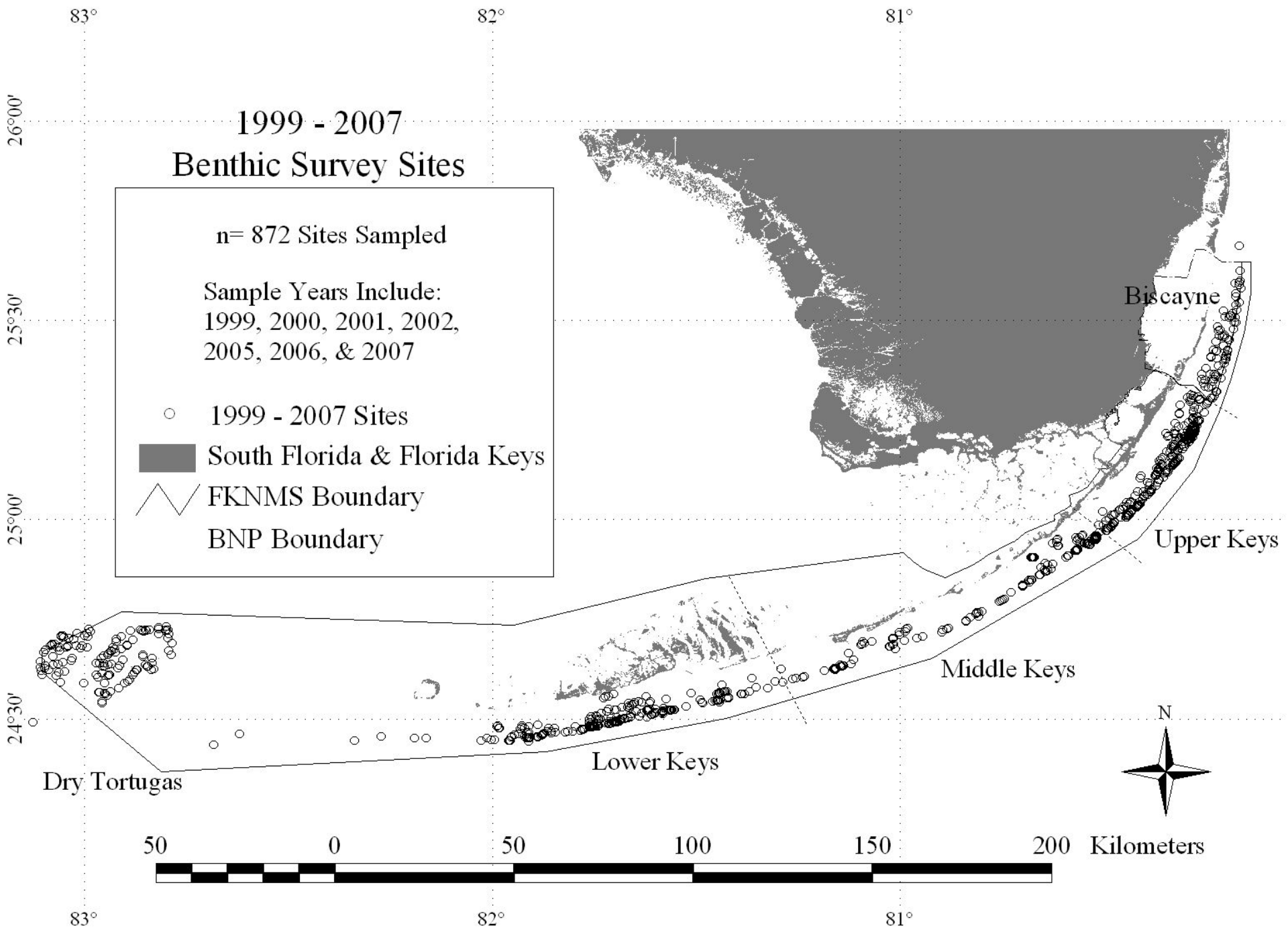
people.uncw.edu/millers

Program Objectives

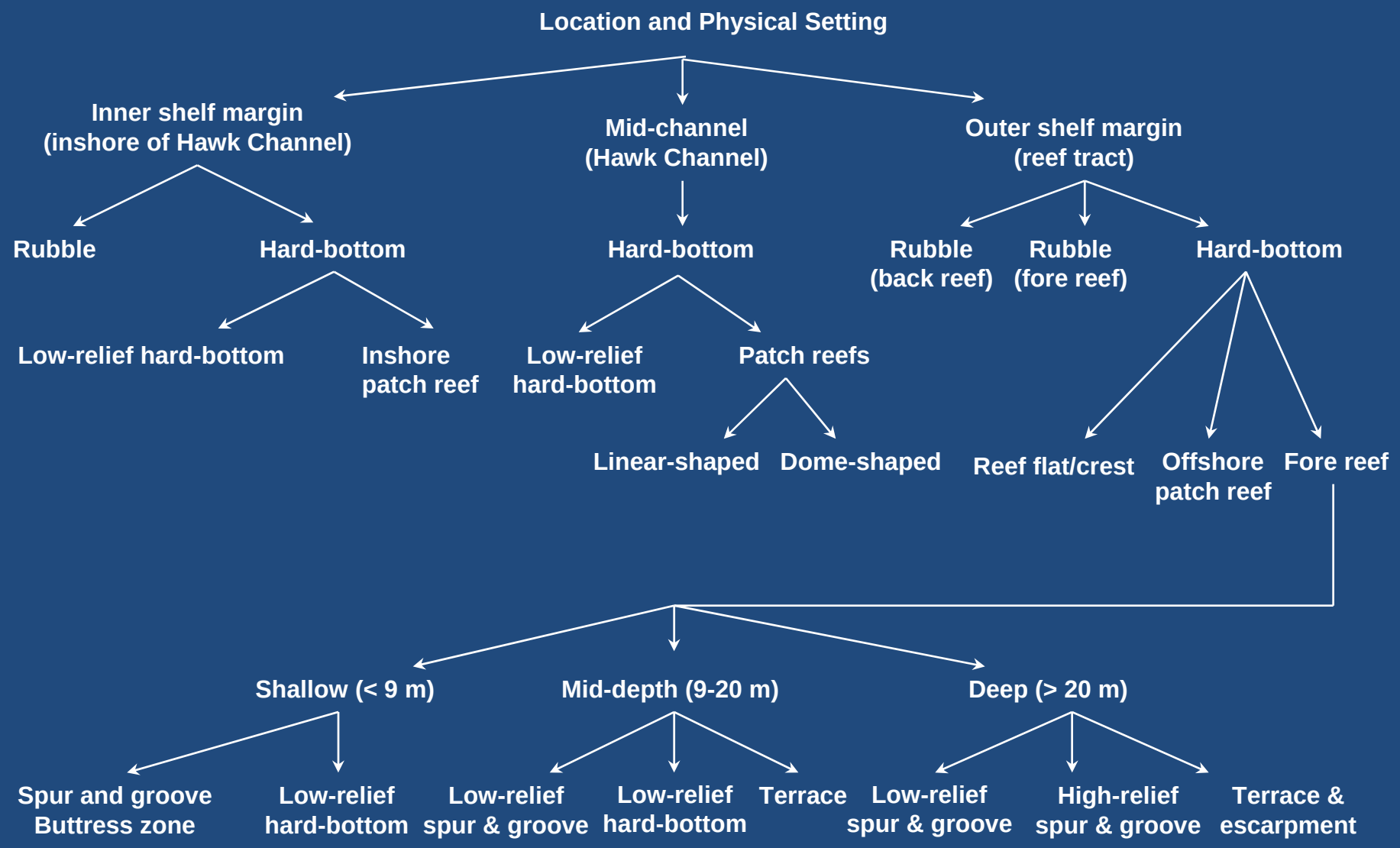
- **Rapid assessment of coral reef and hard-bottom communities**
  - ✓ **Keys-wide, nearshore to offshore**
  - ✓ **Multiple habitat types (and depths)**
  - ✓ **<u>No-take zones (23) vs. reference sites (500+)</u>**
  - ✓ **Multidisciplinary approach linked with reef fish assessments**

Rapid Assessment Methods

- 25-m transects for benthic cover
  - point-intercept
  - video and photo archives
- 25-m x 0.4-m belt transects
  - Species richness (coral, sponge, gorgonian)
  - Gorgonian abundance and height
  - Juvenile coral abundance and size
  - Adult coral abundance, size and condition
  - Urchin density and size
  - Marine ornamental species density
  - Substratum topography (vertical relief, slope, depth)
  - Density, length and impacts of fishing gear

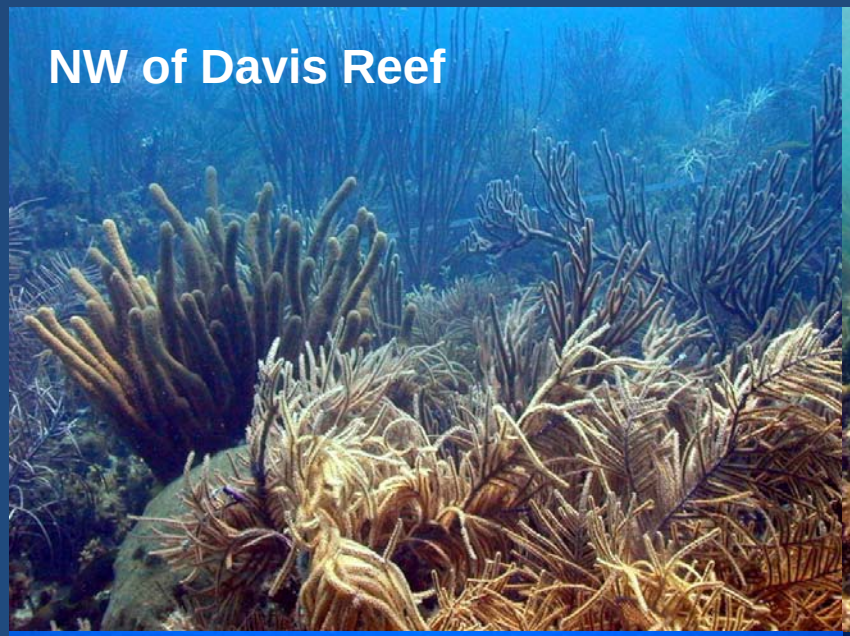

1999 - 2007
Benthic Survey Sites
n= 872 Sites Sampled
Sample Years Include:
1999, 2000, 2001, 2002,
2005, 2006, & 2007
1999 - 2007 Sites
South Florida & Florida Keys
FKNMS Boundary
BNP Boundary
83°
82°
81°
26°00'
25°30'
25°00'
24°30'
Biscayne
Upper Keys
Middle Keys
Lower Keys
Dry Tortugas
N
50
0
50
100
150
200
Kilometers

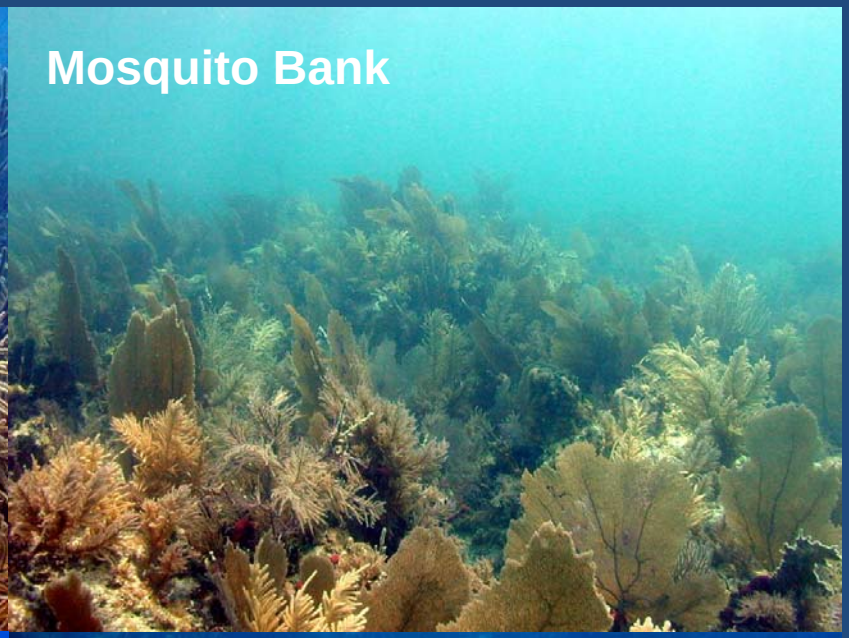
Structural Classification of Florida Keys Hard-bottom Habitats



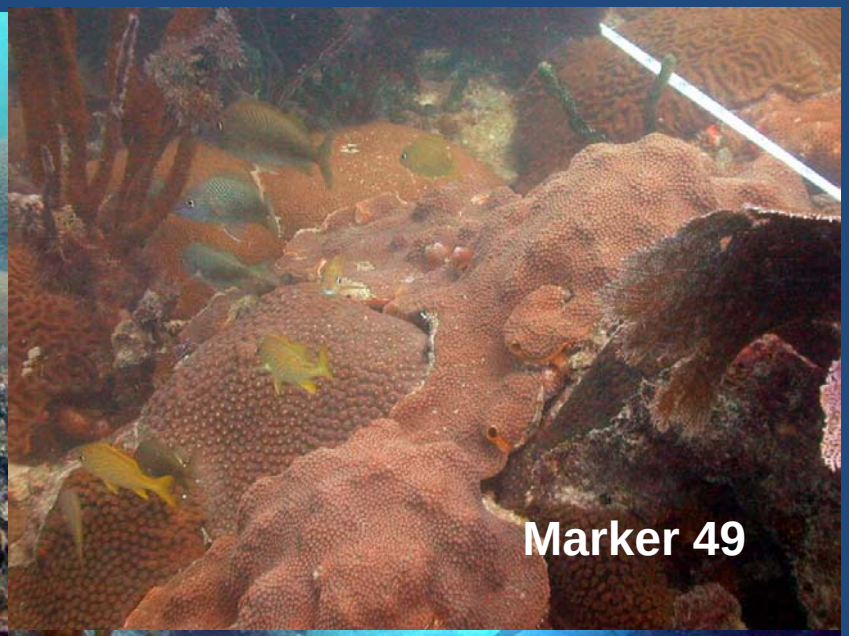
Upper Keys Spur and Groove

Offshore Patch Reefs

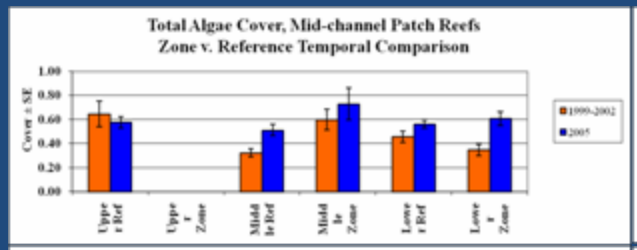


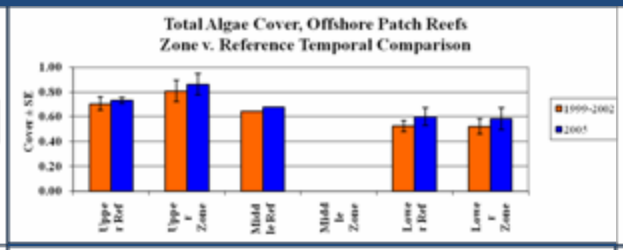


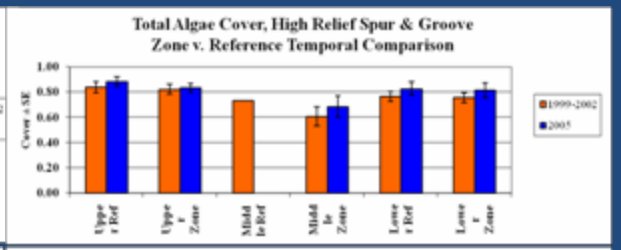
Mid-channel Patch Reefs

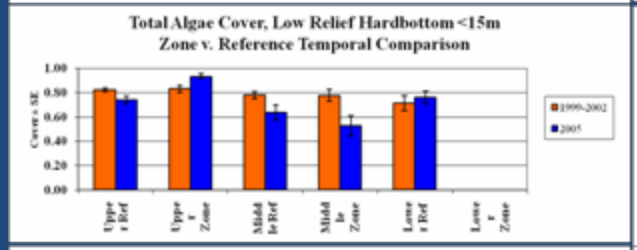


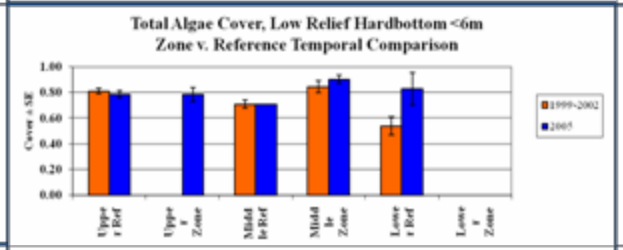
Coral Reef Assessment Results

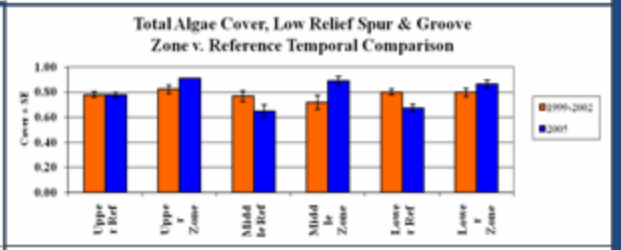


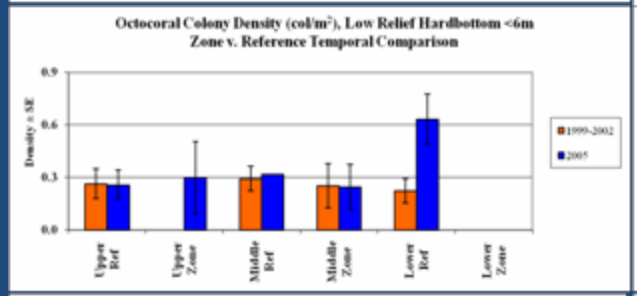


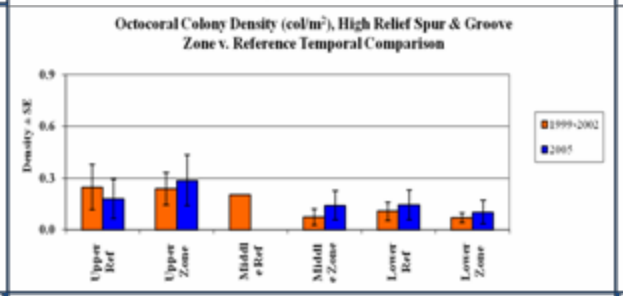


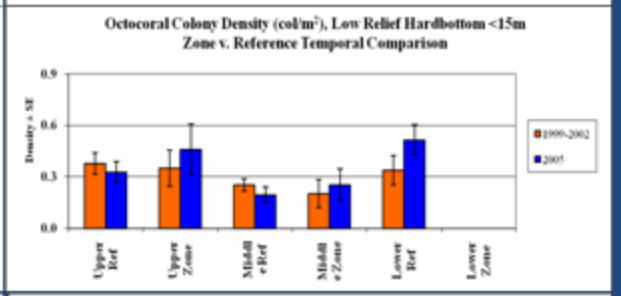


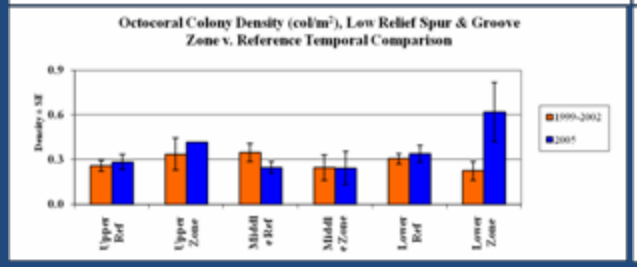


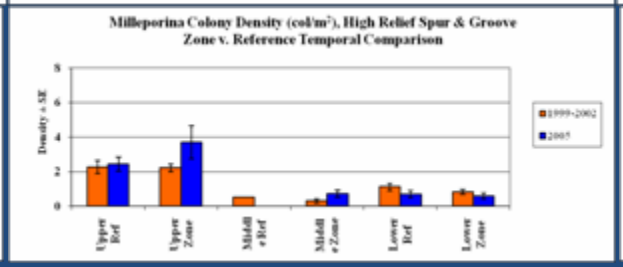


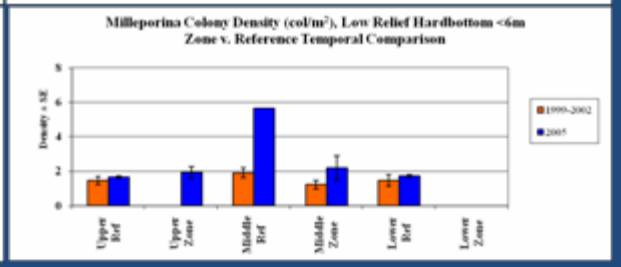




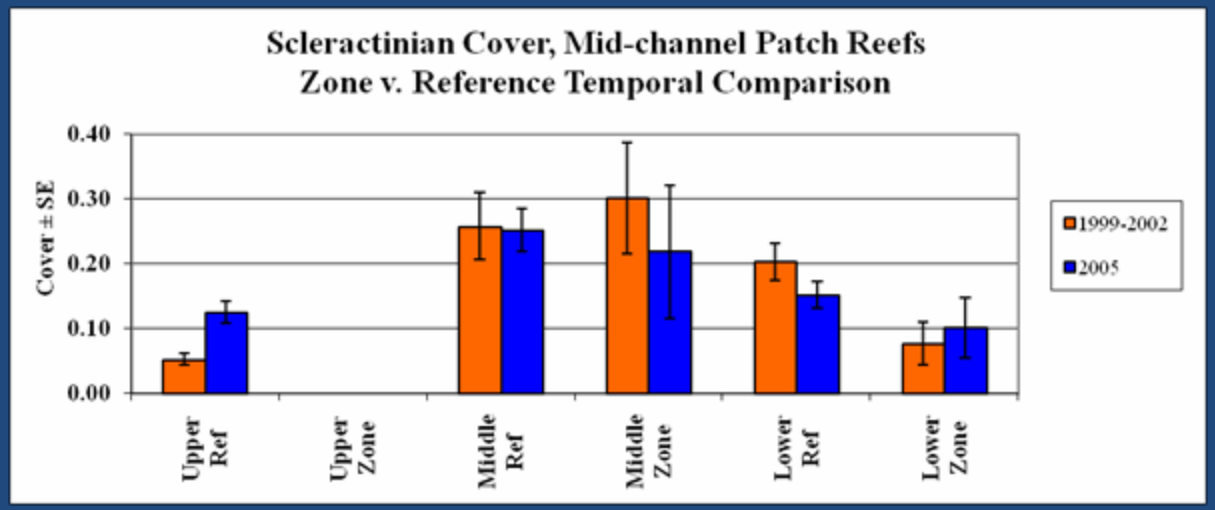


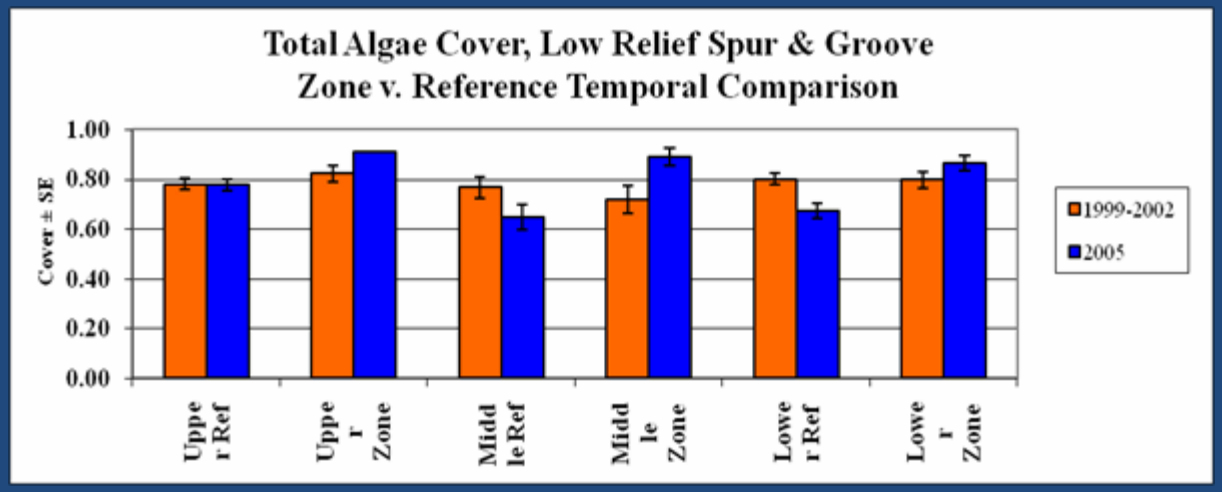




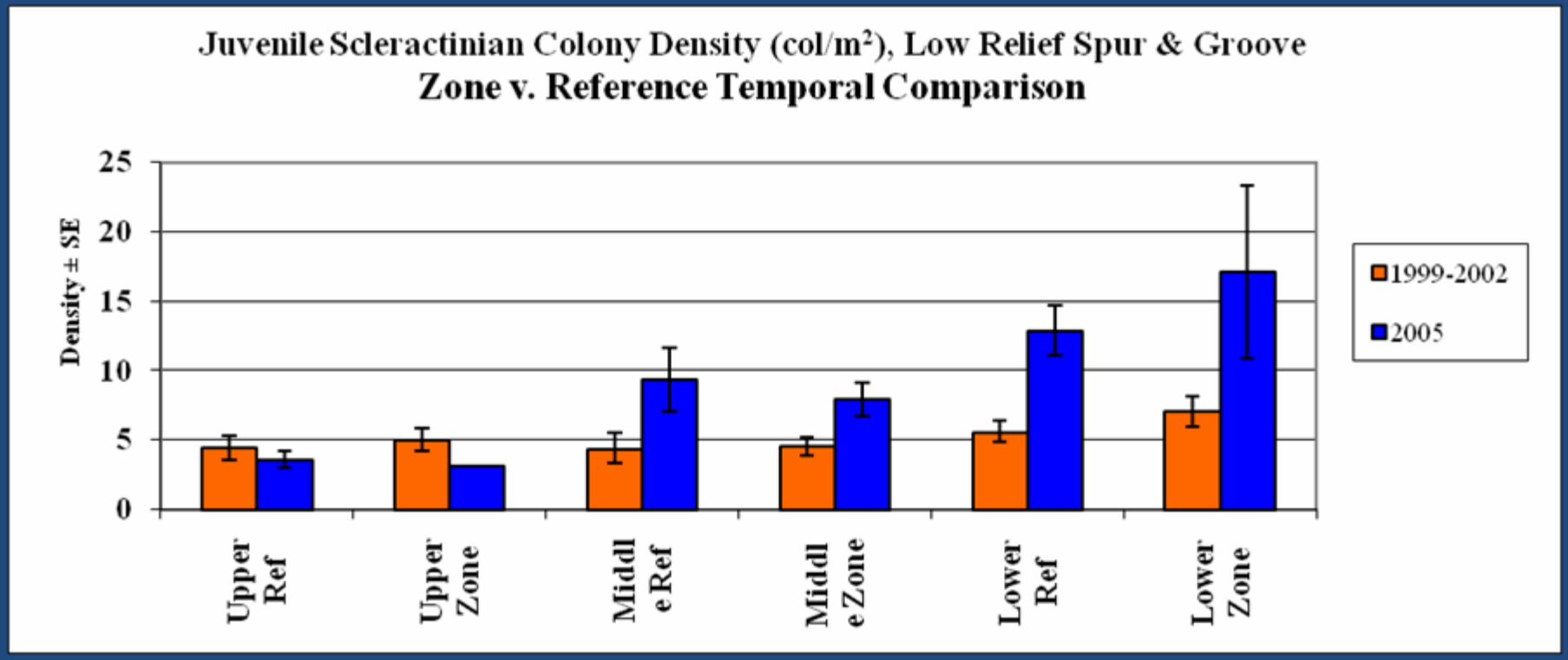


Coral and Seaweed Cover

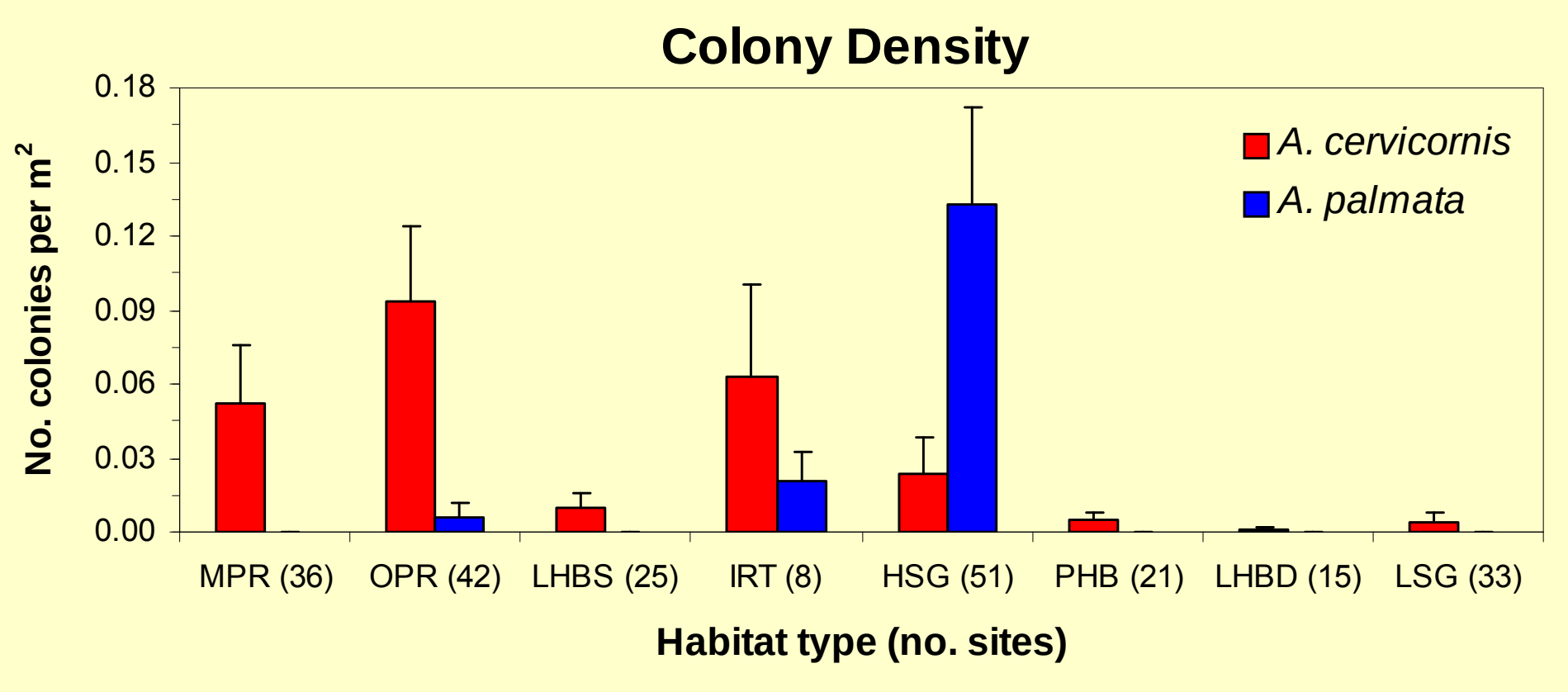




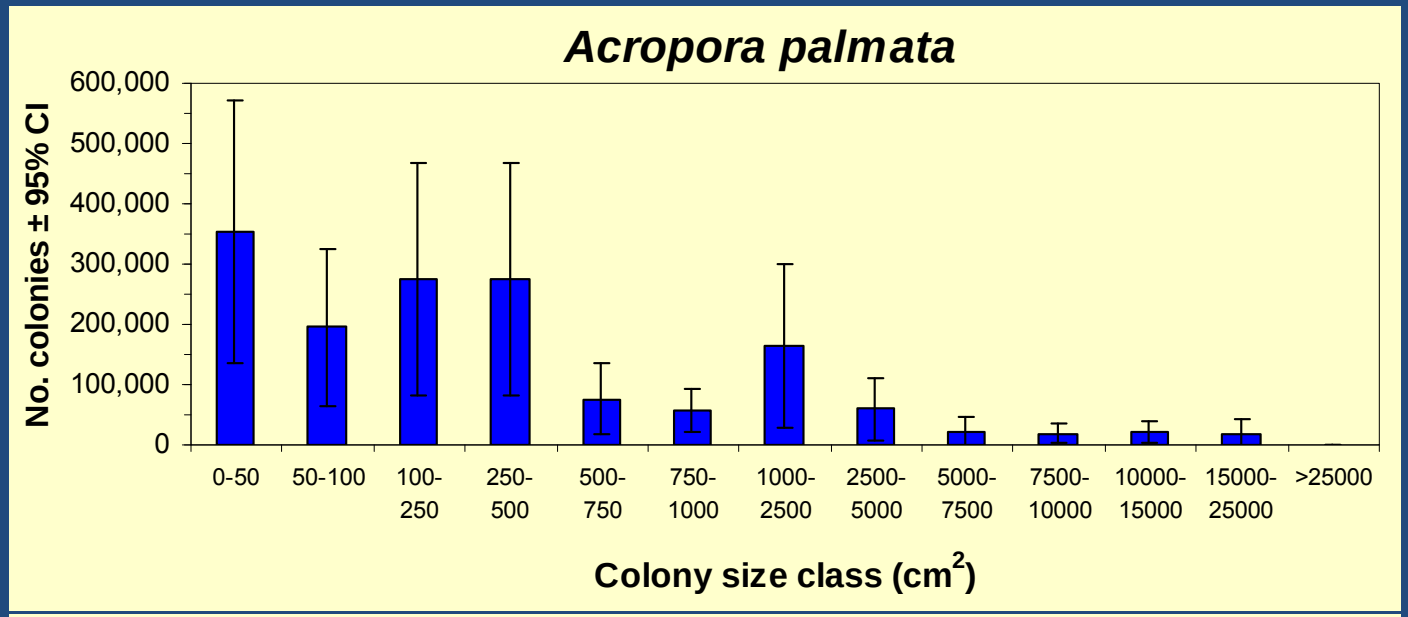
Juvenile Coral Density

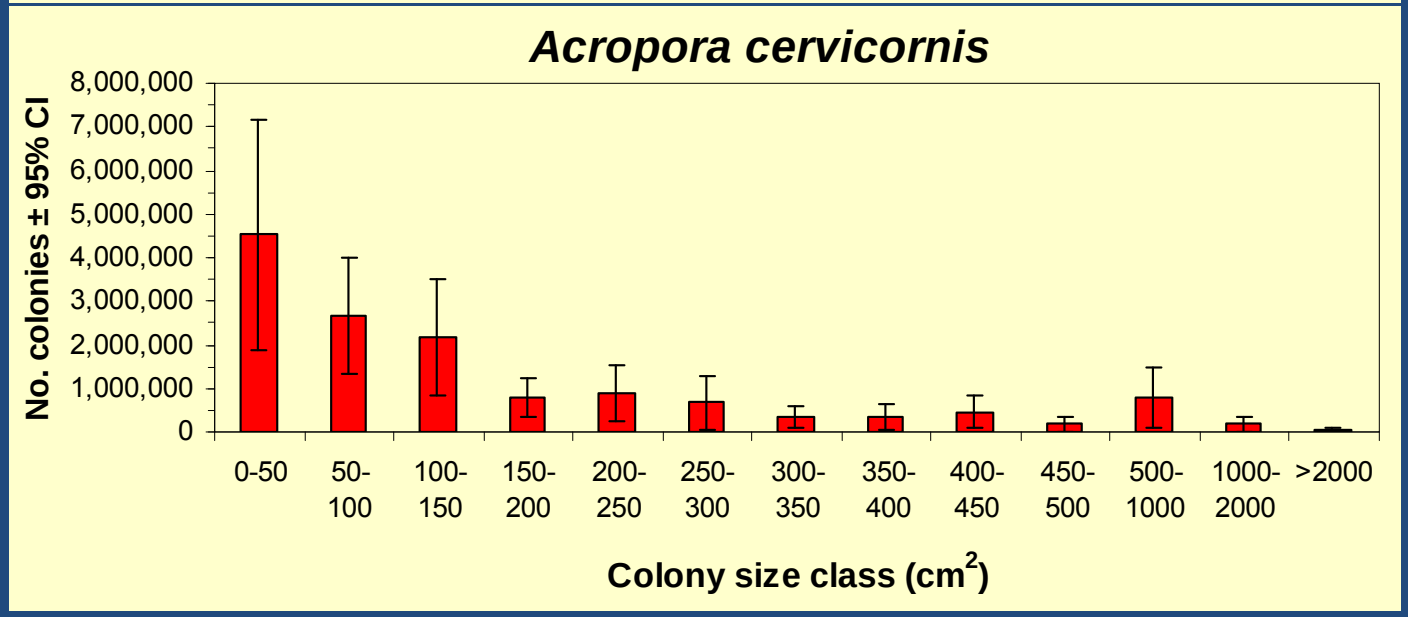


Acropora Colony Abundance 2007

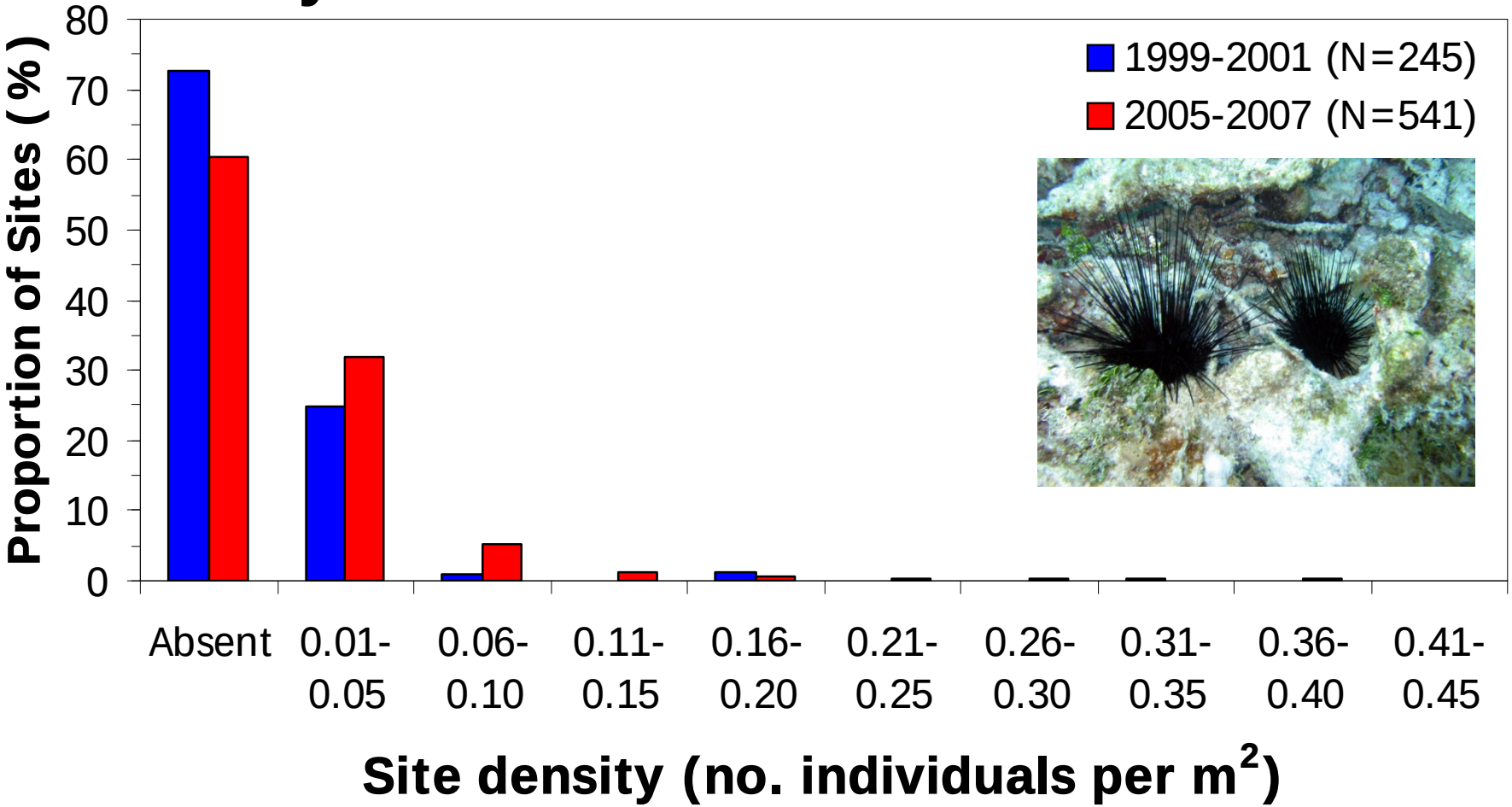


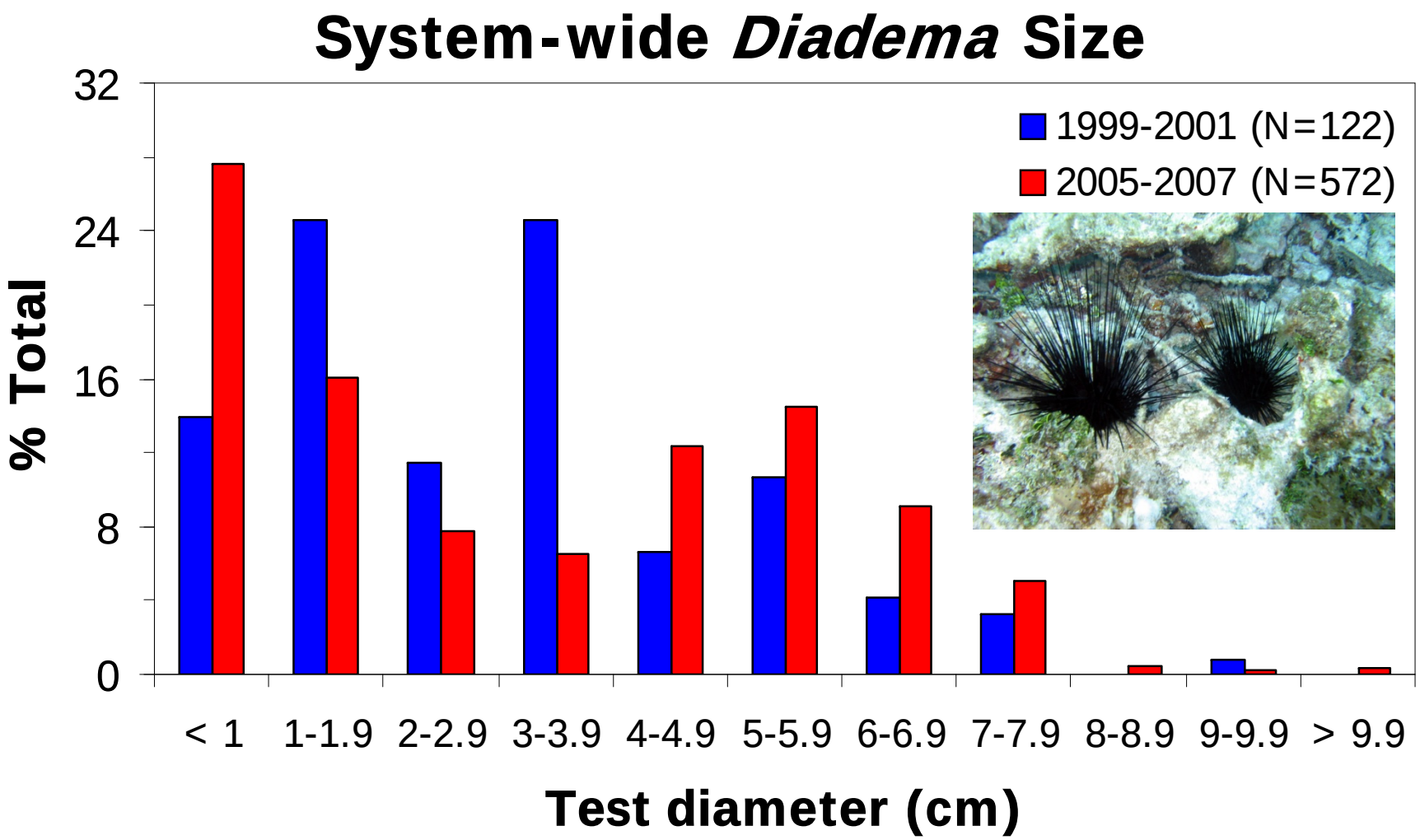
Acropora Population Abundance by Colony Size Class 2007

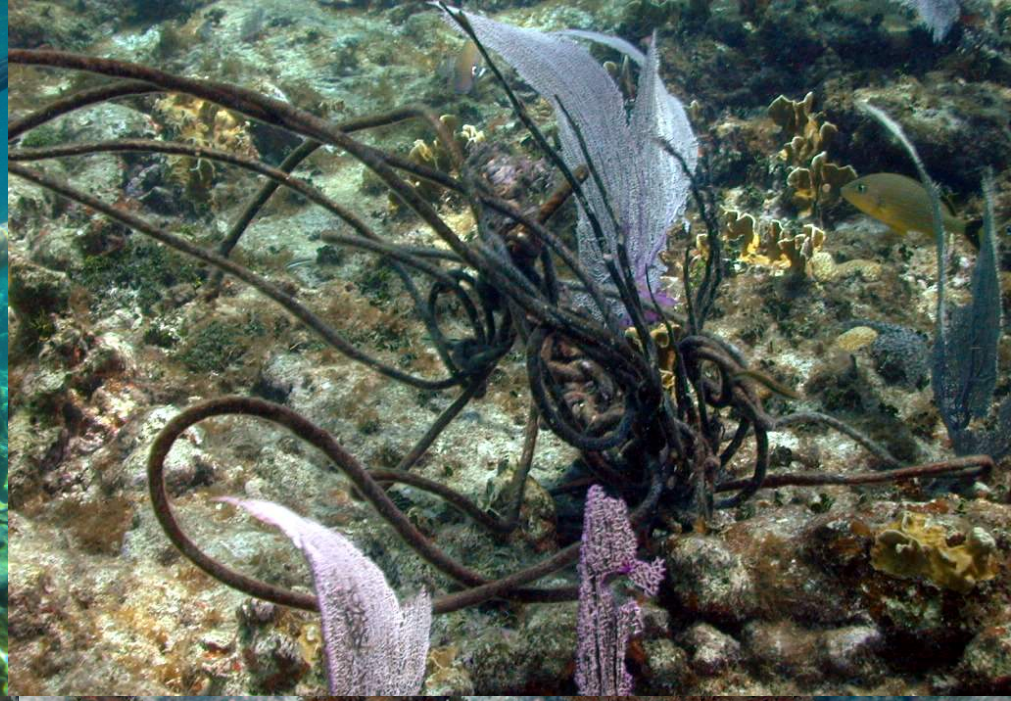




System-wide *Diadema* Densities




System-wide *Diadema* Size
% Total
32
24
16
8
0
1999-2001 (N=122)
2005-2007 (N=572)
< 1
1-1.9
2-2.9
3-3.9
4-4.9
5-5.9
6-6.9
7-7.9
8-8.9
9-9.9
> 9.9
Test diameter (cm)



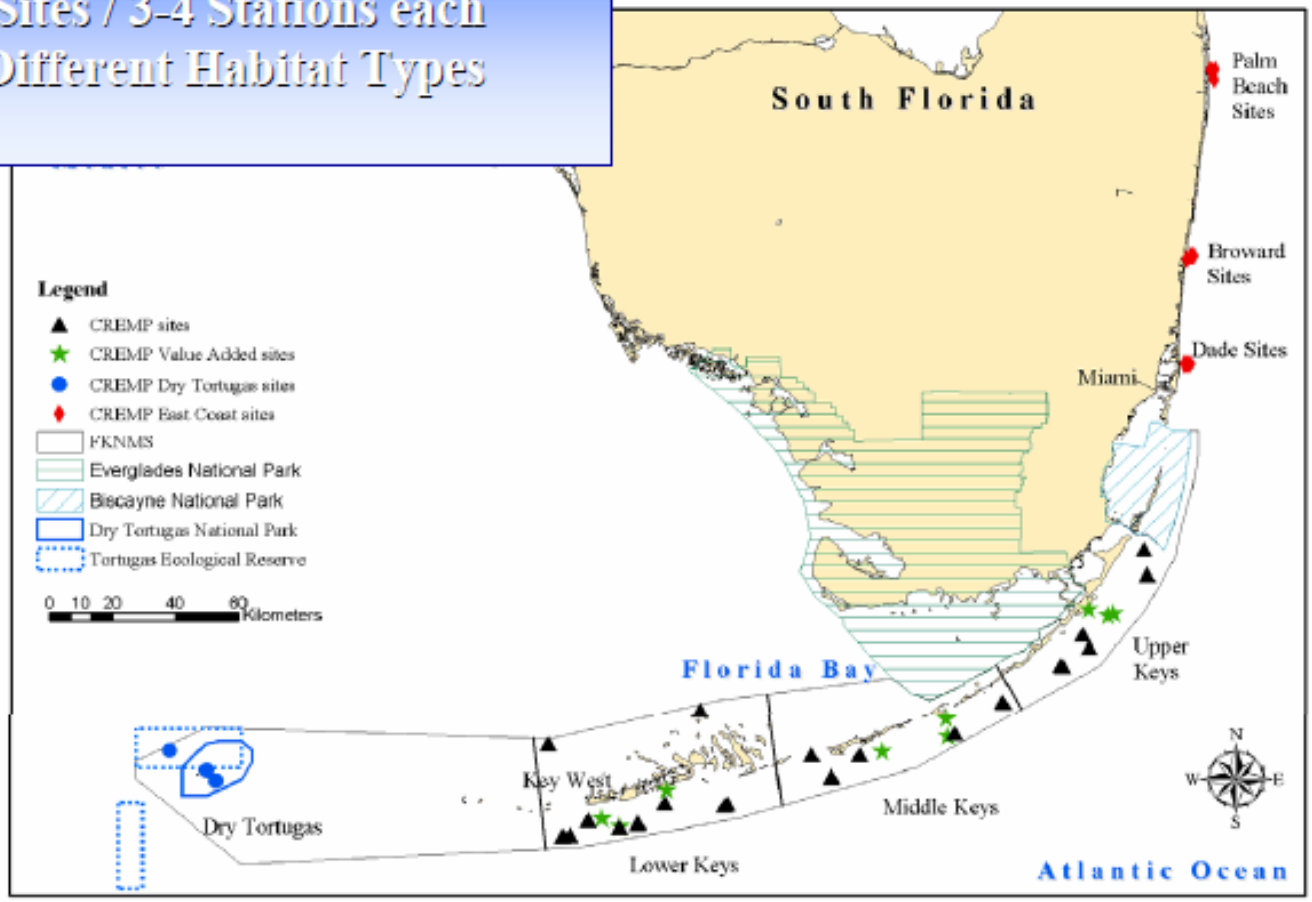
Florida Keys National Marine Sanctuary Coral Reef Evaluation and Monitoring Project

The Florida Keys National Marine Sanctuary Coral Reef Evaluation and Monitoring Project (FKNMS CREMP) sampling sites and stations were selected and installed in 1995. Originally 40 sites and 160 stations were selected for monitoring. The original 40 CREMP sampling sites were selected using a stratified, or layered, random sampling procedure based on the U.S. Environmental Protection Agency's (EPA) Environmental Mapping and Assessment Program (EMAP). Stratification, or the arrangement of the layers, was based on habitat type, with four main habitat types defined: nearshore hardbottom, patch reefs, offshore shallow reefs (roughly 10 to 20 feet of depth), and offshore deep reefs (about 30 or 50 feet deep). While sampling sites were selected in a random matter, stations were installed with the intention of monitoring specific aspects of the selected habitats. In 1999 three sites totaling 12 stations were installed and sampled in the Dry Tortugas as part of the FKNMS CREMP monitoring, for a total of 43 sites and 172 stations.

Coral Reef Evaluation and Monitoring Project (CREMP)

FWC Coral Biologists

- 43 Sites / 3-4 Stations each
- 4 Different Habitat Types



METHODS

Each site consists of two to four monitoring stations delineated by permanent markers. Stations are approximately 2 x 22 meters and are generally perpendicular to the reef crest. Within each station, field sampling consists of a station species inventory (SSI), video transects (three transects per station) and a bio-eroding sponge survey (Figure 2). Nine sites (3 in each of the geographical areas) have been designated Value Added Sites. In addition to SSI, video transects, and bio-eroding sponge surveys, sampling at these sites includes diseased coral surveys, stony coral abundance surveys, and temperature surveys.

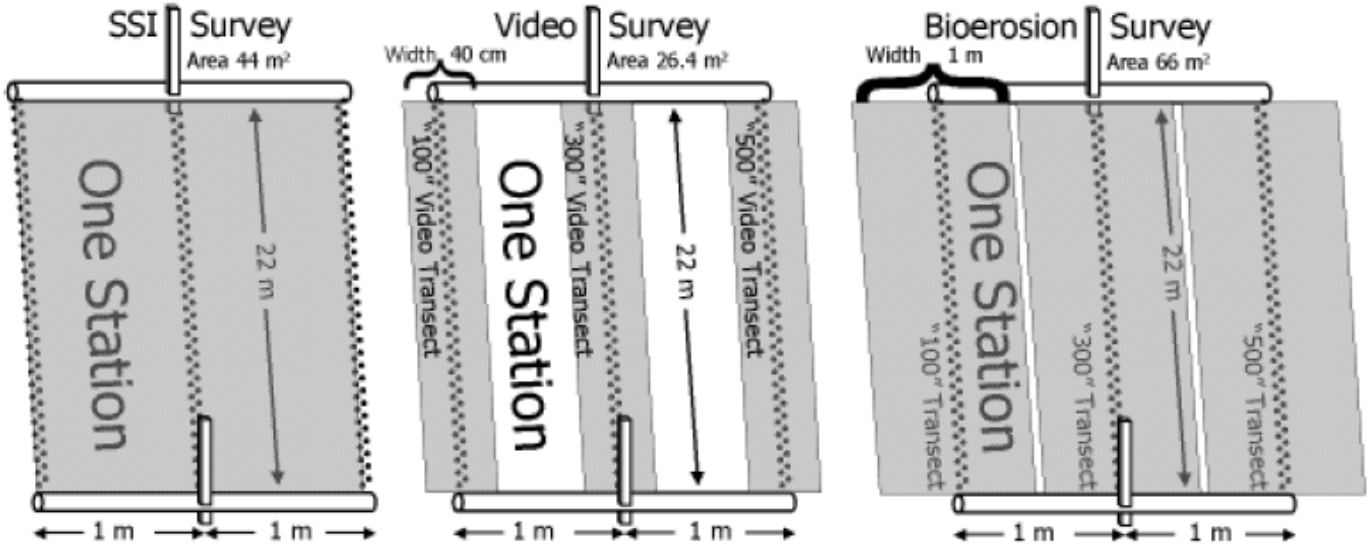
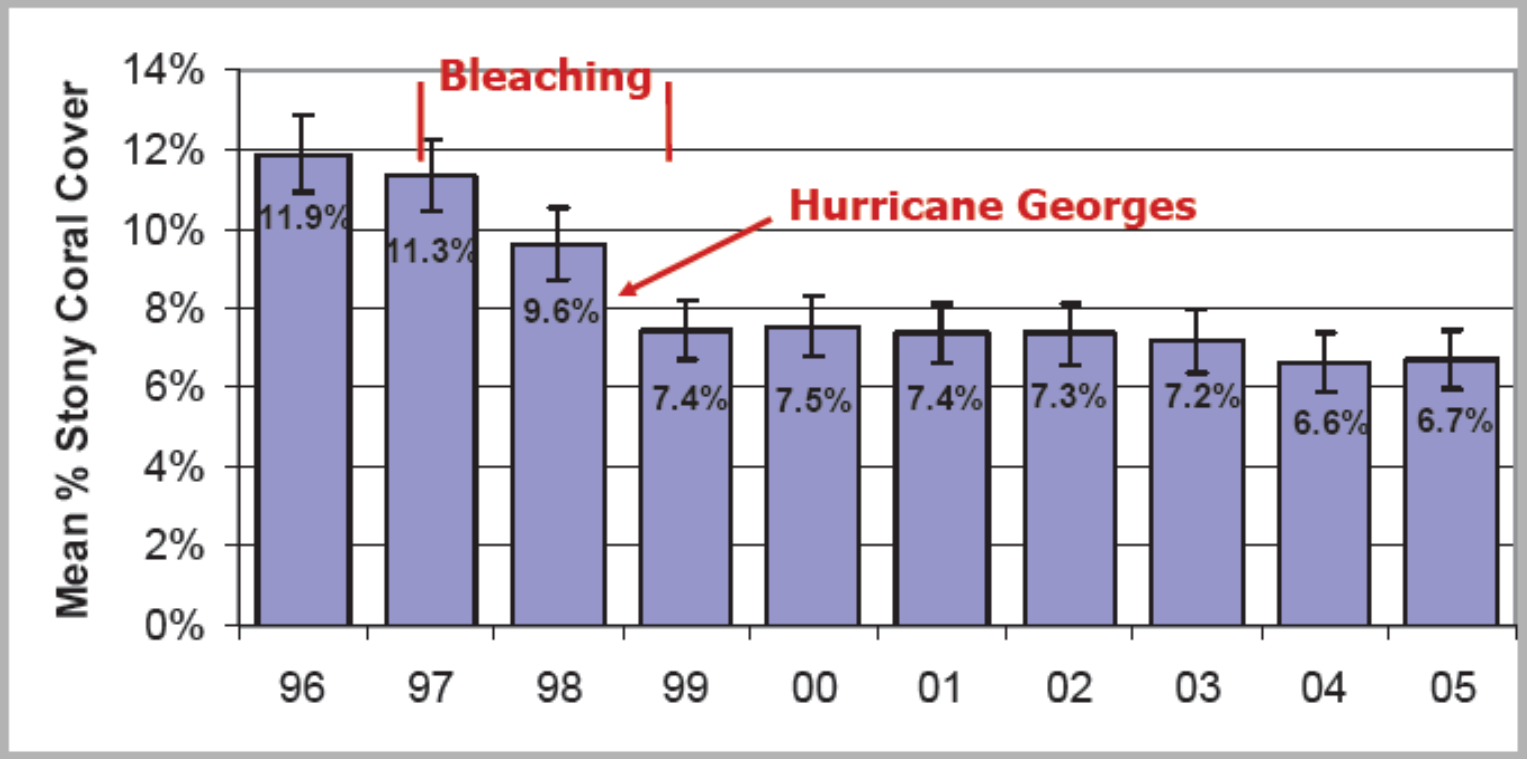


Figure 2. Layout of CREMP stations and area sampled by each method.

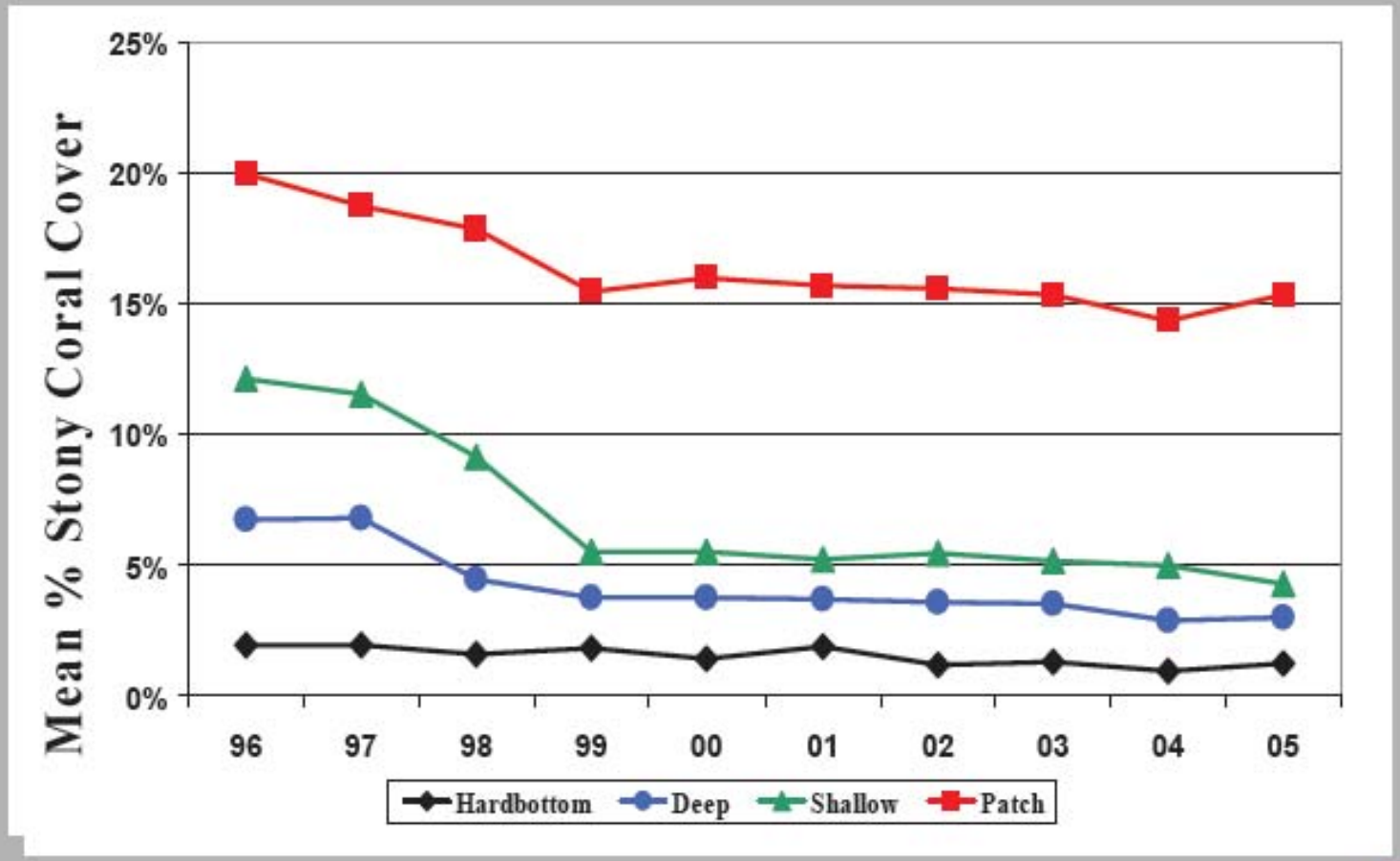
Stony Coral Cover Sanctuary-wide 1996-2005



A decrease in stony coral cover was observed sanctuary-wide for each year from 1997 through 1999. Mean percent stony coral cover in 2005 did not change significantly (alpha = 0.05). Sanctuary-wide during 2005, mean stony coral cover was 6.7%.

http://www.floridamarine.org

Stony Coral Cover by Habitat Type, 1996-2005



During 2005, stony coral cover increased at patch reef habitats, decreased at shallow reef habitats and remained unchanged at hard bottom & deep reef habitats.

Florida Keys Coral Bleaching Early Warning Network

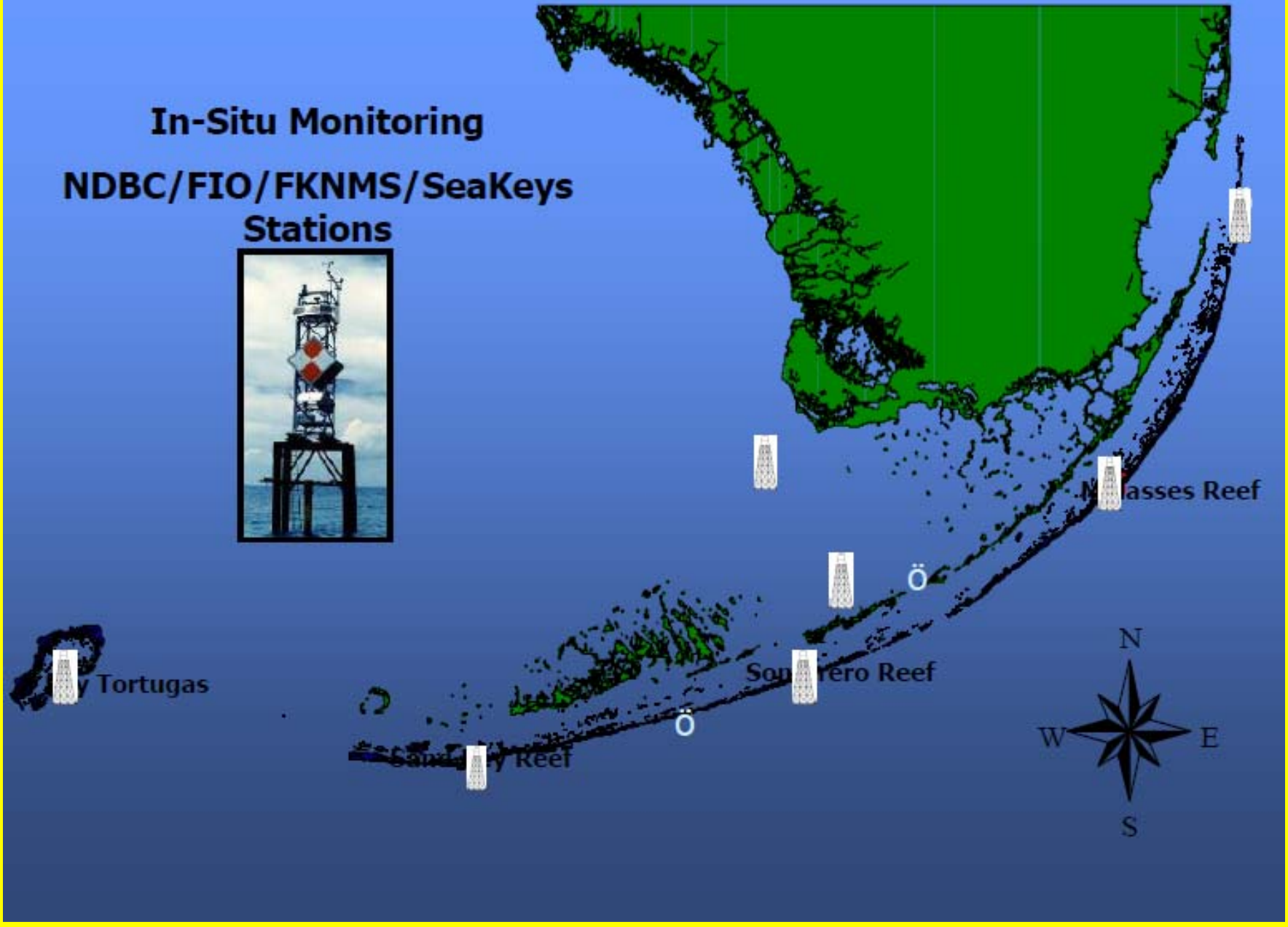
"BleachWatch"

Erich Bartels

Cory Walter

In-Situ Monitoring

NDBC/FIO/FKNMS/SeaKeys Stations



NOAA Coral Reef Watch Program

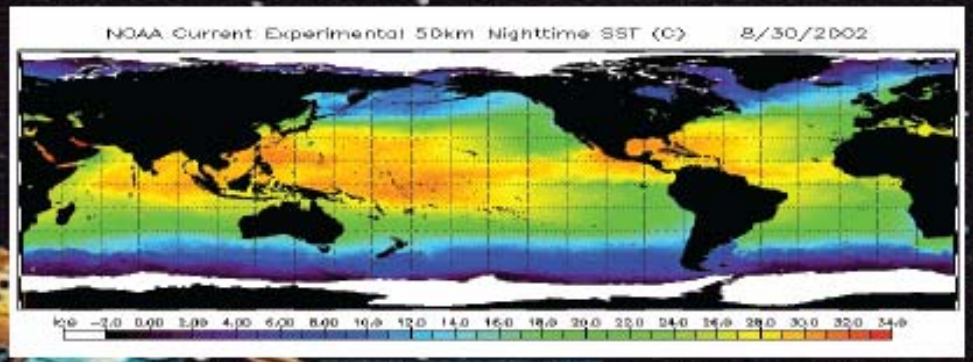
Satellite Near Real-Time Coral Bleaching HotSpot Products

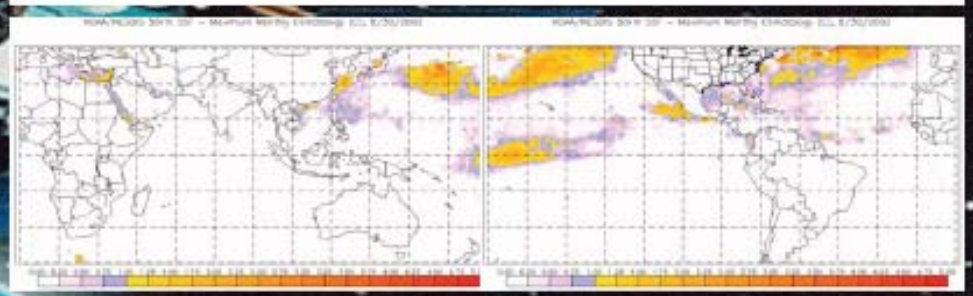
(Twice-weekly at 50km resolution)

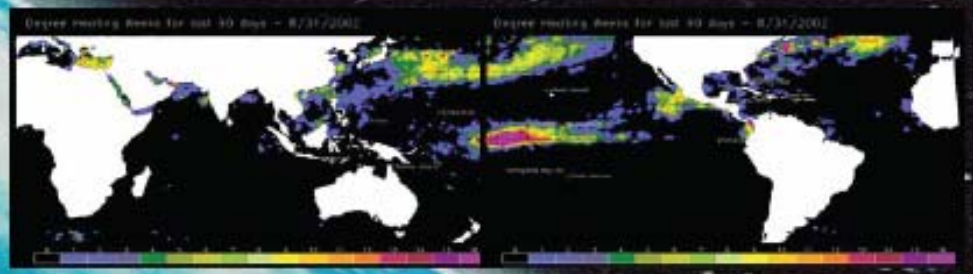
Sea Surface Temperature
(Night-time only)

Bleaching HotSpots

Degree Heating Weeks (DHW)







http://coralreefwatch.noaa.gov

Bleaching Observations

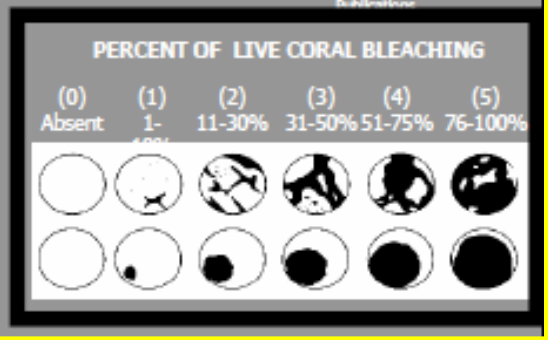
- Severity of Bleaching

- Types of Corals Bleached

Coral ID Table

- Percent of Bleached Corals

Percent Cover/Bleaching Table



Coral Diseases of the Lower Florida Keys

Lauri MacLaughlin, Resource Manager, FKNMS
Debbie Santavy, EPA, Gulf Breeze Laboratory
Kim Ritchie, Mote Marine Laboratory

The EPA has conducted research cruises each summer since 1997, sampling 36 sites on the reefs between Key West and the Dry Tortugas. The study utilizes a circular (10 meter radial arc) transect method developed by Edmunds in 1991. Coral counts, diseased coral counts, and bleached coral counts are recorded to determine the distribution and frequency of disease.

Data reveals 11 disease conditions affecting 18 species of stony corals and sea fans. According to research completed in 1998, the greatest incidence of disease and bleaching was found on the Key West reefs, where approximately 22% of the corals were diseased and 26% were bleached. The focus of the sanctuary's involvement in coral disease work includes assessment, treatment, and ongoing monitoring. For example, sanctuary staff supports the EPA disease cruises each summer, and the sanctuary funded the development and implementation of the Marine Ecosystem Events Response Assessment (MEERA) rapid response program at Mote Marine Lab.

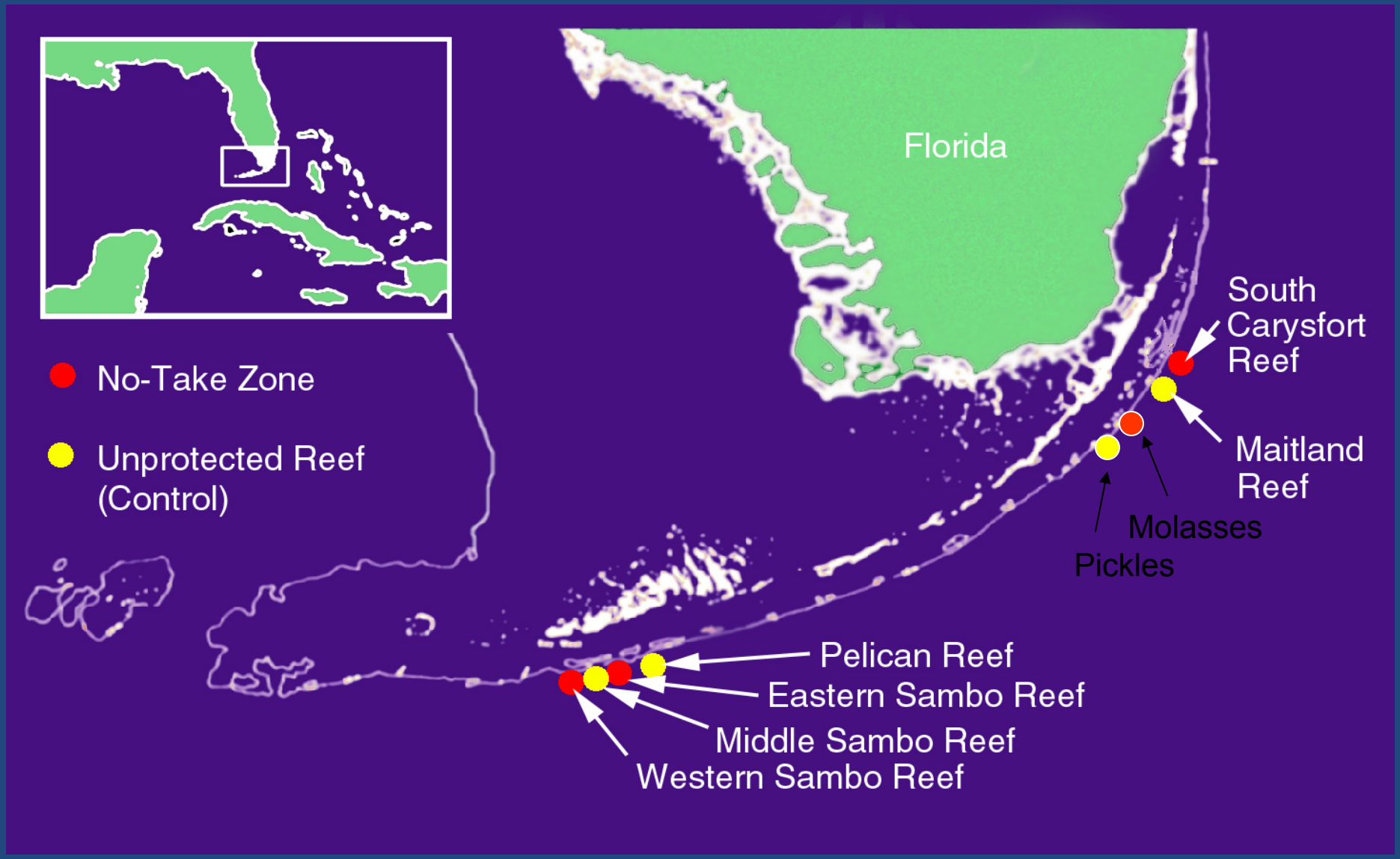
Resilience of coral reef benthic communities in the Fully Protected Zones of the Florida Keys

Struan R. Smith
Biology Dept.
Georgia State University
Atlanta GA

Richard B. Aronson
Dauphin Island Sea Lab
Dauphin Island AL

John C. Ogden
Florida Institute of Oceanography
St. Petersburg, FL

Florida Keys FPZ and Reference area study sites



Study Design

- Three pairs of Fully Protected Zones (FPZ) and Reference (REF) areas were established in 1997-98:

  South Carysfort and Maitland in the Upper Keys.

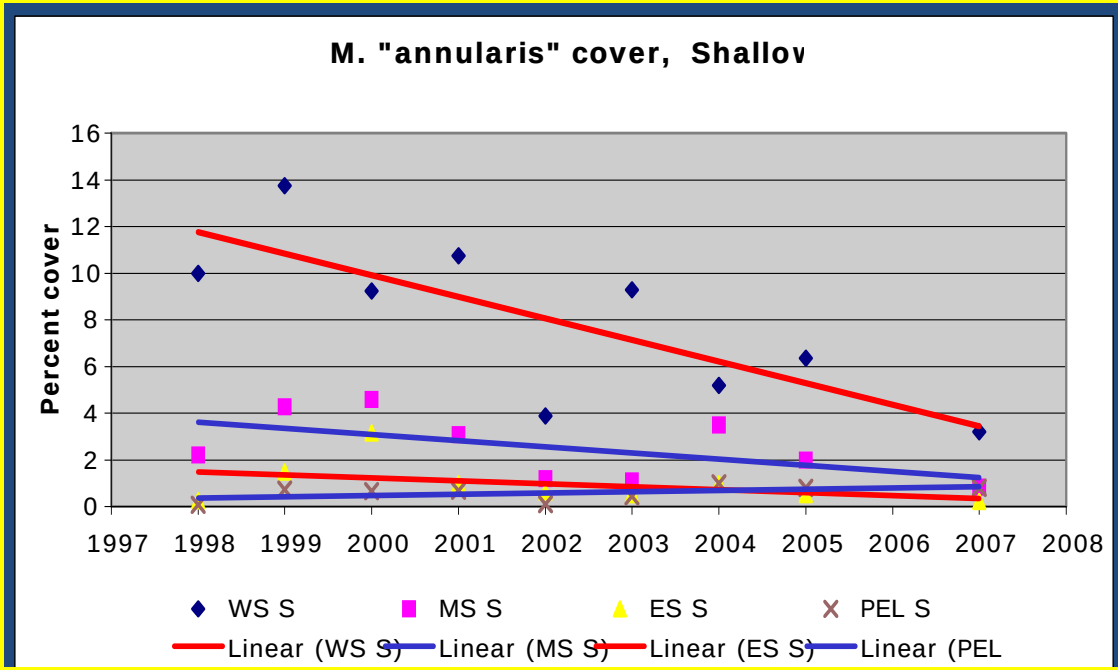
  Western Sambo and Middle Sambo in the Lower Keys

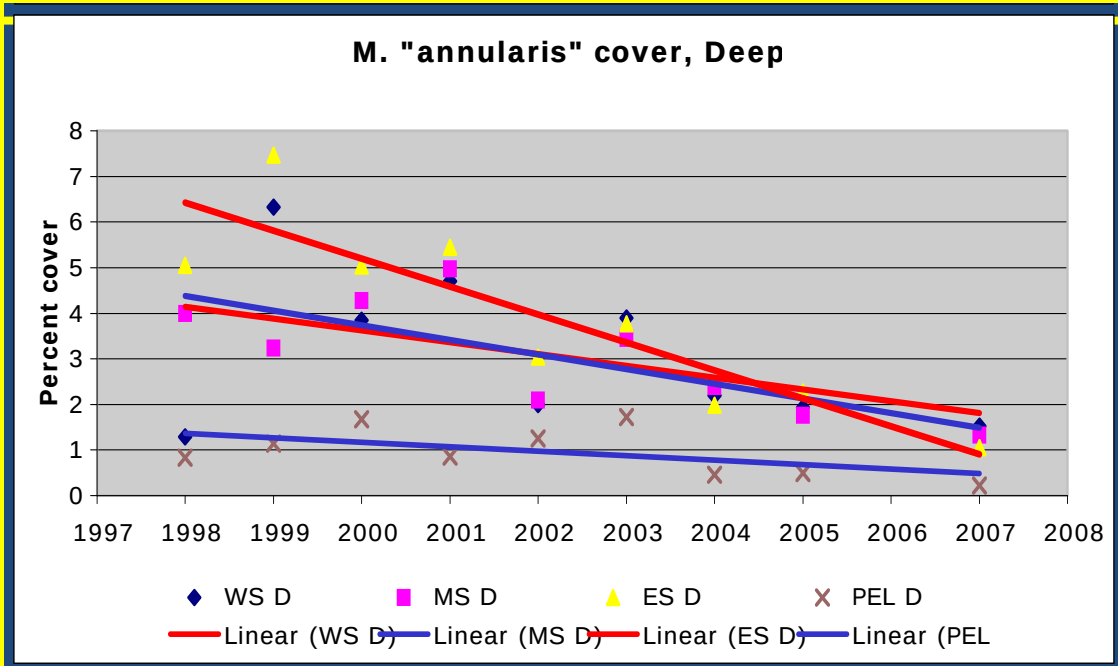
  Eastern Sambo and Pelican Shoal in the Lower Keys.

- Two depths (8-11m and 14-18m) studied in each FPZ and reference area.
- Sites established in Fall 1997, with data collection from 1998 to 2007.
- No data in 2006 due to loss of funding.
- Utilized permanent quadrats to assess patterns of coral recruitment, growth and survival
- Random video transects used to asses coral cover
- Sampling was done annually in early summer.

Trends in *M. "annularis"* cover in FPZ and reference sites in the lower Keys

- Derived from 10 random 25 m video tape transects down slope
- 10 random dots x 50 frames per transect.
- Overall coral overage has declined at nearly all sites since 2000.
- Both *M. "annularis"* and *M. cavernosa* show declines at all sites and depths.
- FPZ: WS and ES; REF: MS, PEL

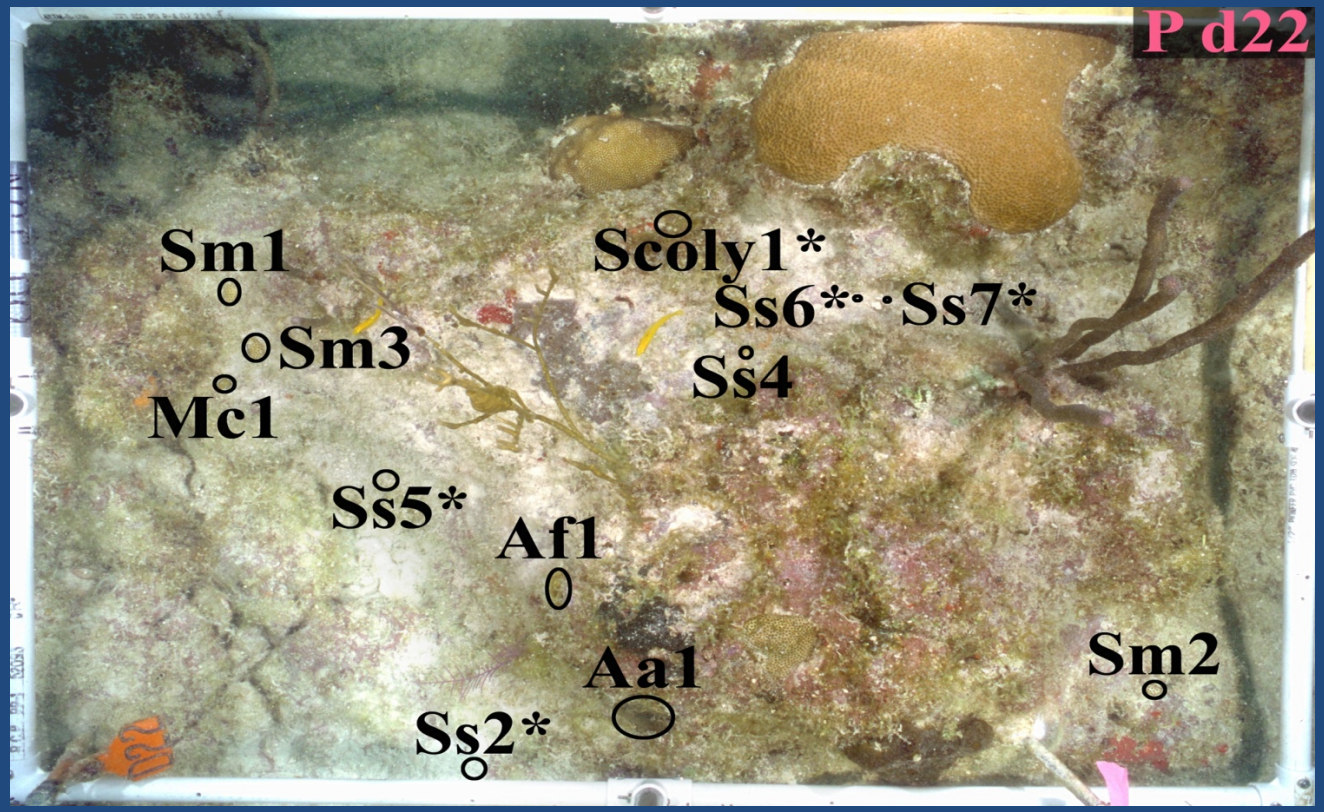




Coral recruitment and juvenile coral sampling method

At each depth in study area:

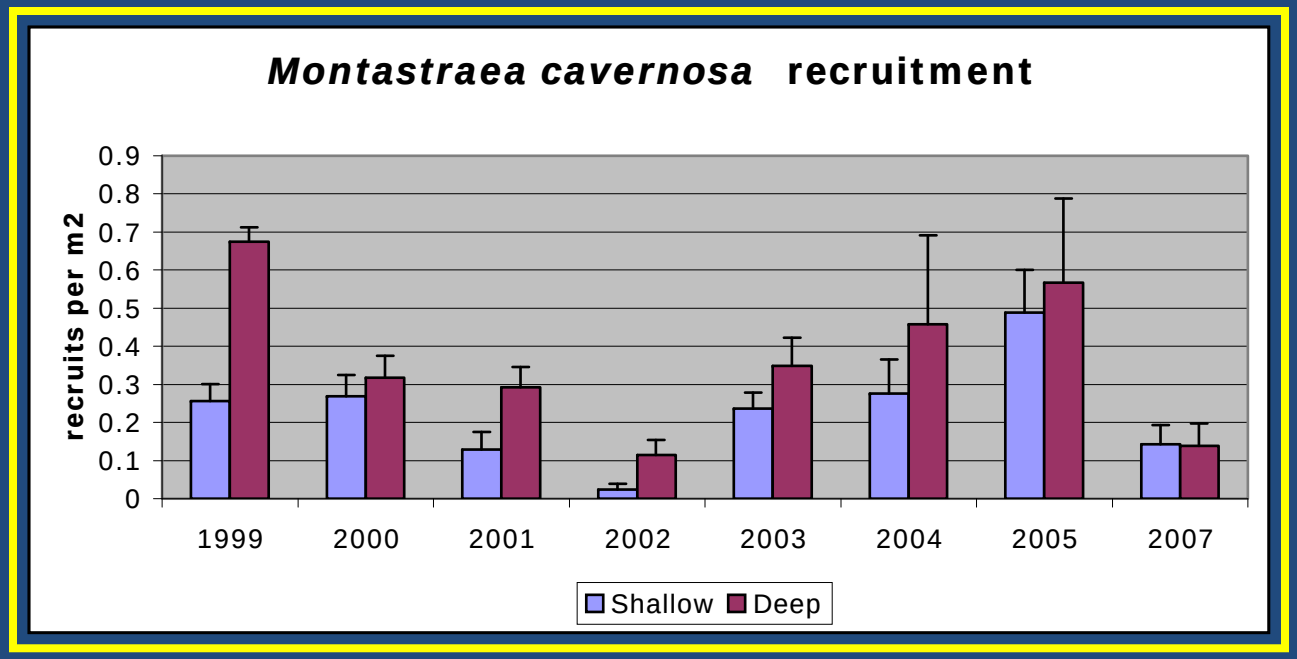
- Thirty two permanent 0.62 m^2 quadrats for visual census of coral recruitment and survival of juvenile corals (≤ 5 cm diameter).
- Annotated image of each quadrat is used *in situ* for rapid and accurate re-surveys each year.



Patterns of *Montastraea* sp. recruitment

Recruitment from visual census in the permanent quadrats:

- *M. cavernosa:* between 8 to 25 new colonies per year per depth
- *M. "annularis":* only 1 to 6 per year, not consistent by depth or year



Population models of key reef building corals *Montastraea* " annularis" and *M. cavernosa*

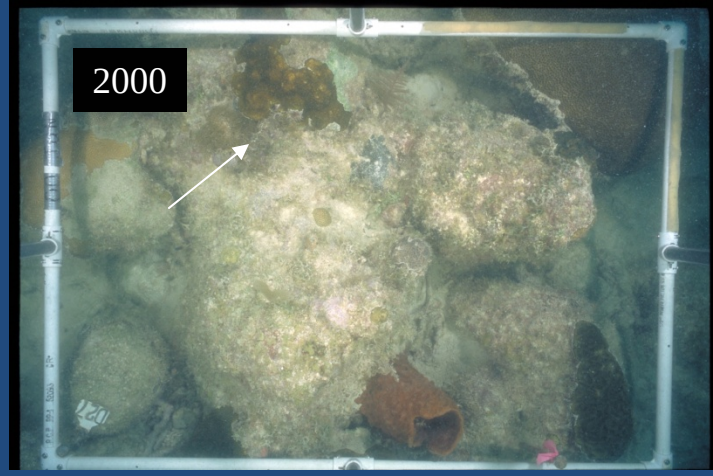
- Evaluate patterns of survival and growth of larger colonies in quadrats (~15 cm^2 to ~400 cm^2) and the adjacent areas (200 - 3000 cm^2).
- Integrate with recruitment and juvenile mortality quadrat data.
- Use transition matrix models to predict future population growth

M. faveolata and *M. franksii*

M. cavernosa

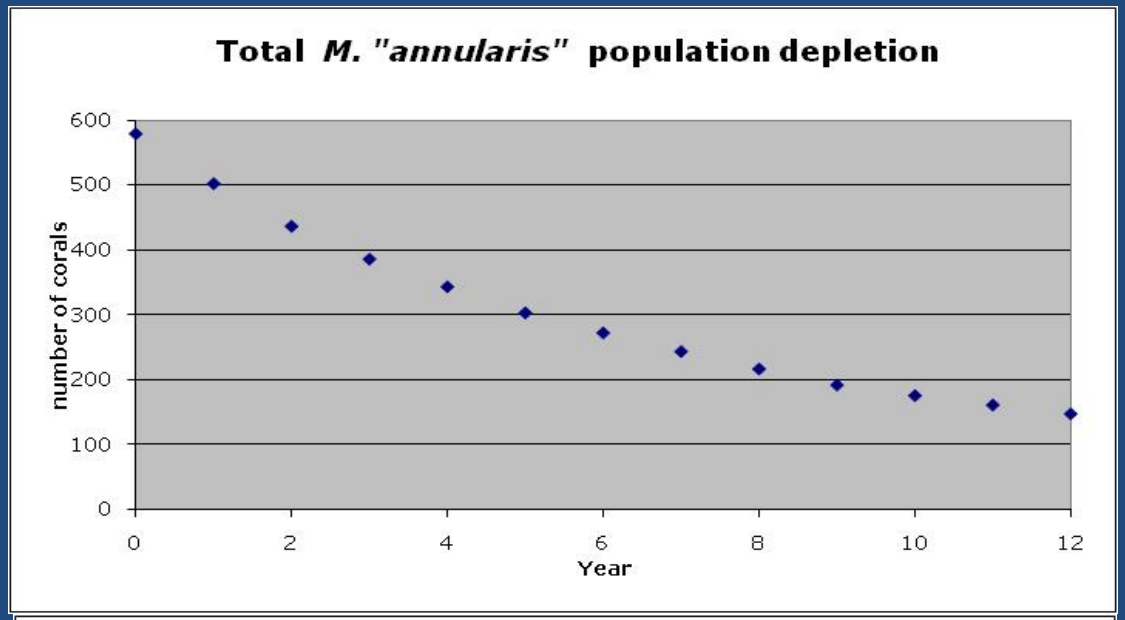
Montastraea spp. recruitment, growth and survival from 1998 to 2007

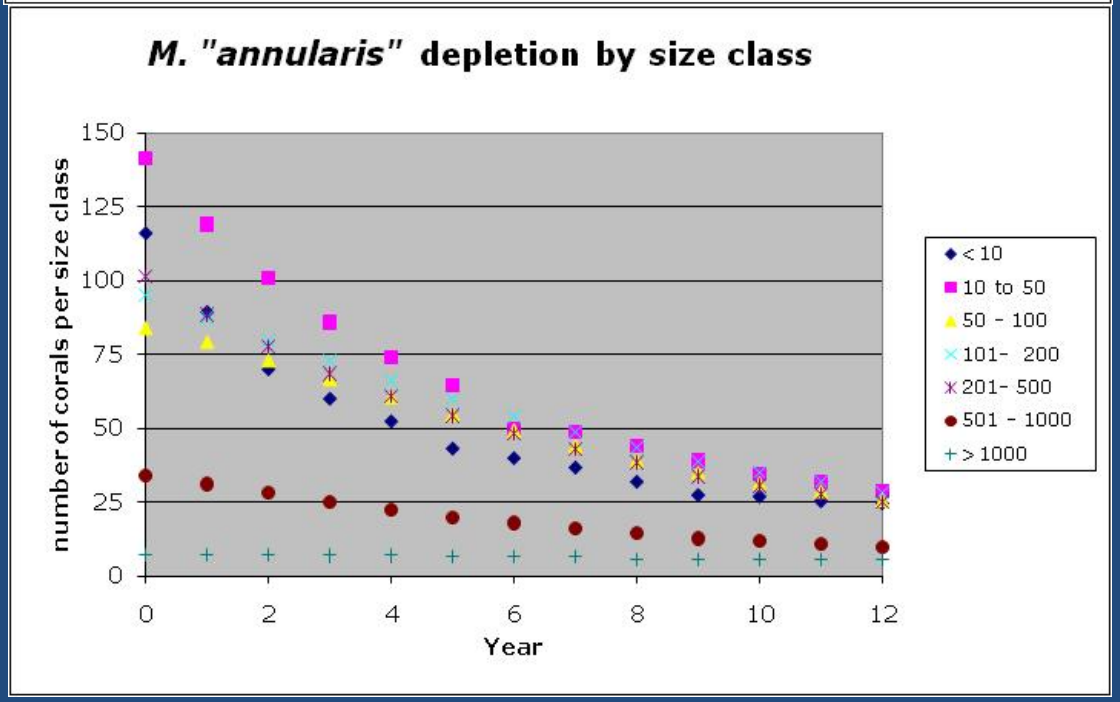
- Individual *Montastraea* colonies in the deep and shallow quadrats in the Lower Keys were assessed for growth, shrinkage and mortality in each year from 1998 to 2007
- Adjacent larger colonies (200- 3000 cm^2) were included, starting in 2003.
- Colony area determined by digital planimetry, 3 measurements per colony.
- Patterns of change (growth, shrinkage, mortality) categorized in 7 size classes:
  - < 10, 11-50, 51 -100, 101- 200. 201-500, 501-1000, >1000 cm^2
- Data pooled across sites and depths, due to small sample sizes per depth and site.



Population model: *M. "annularis"*

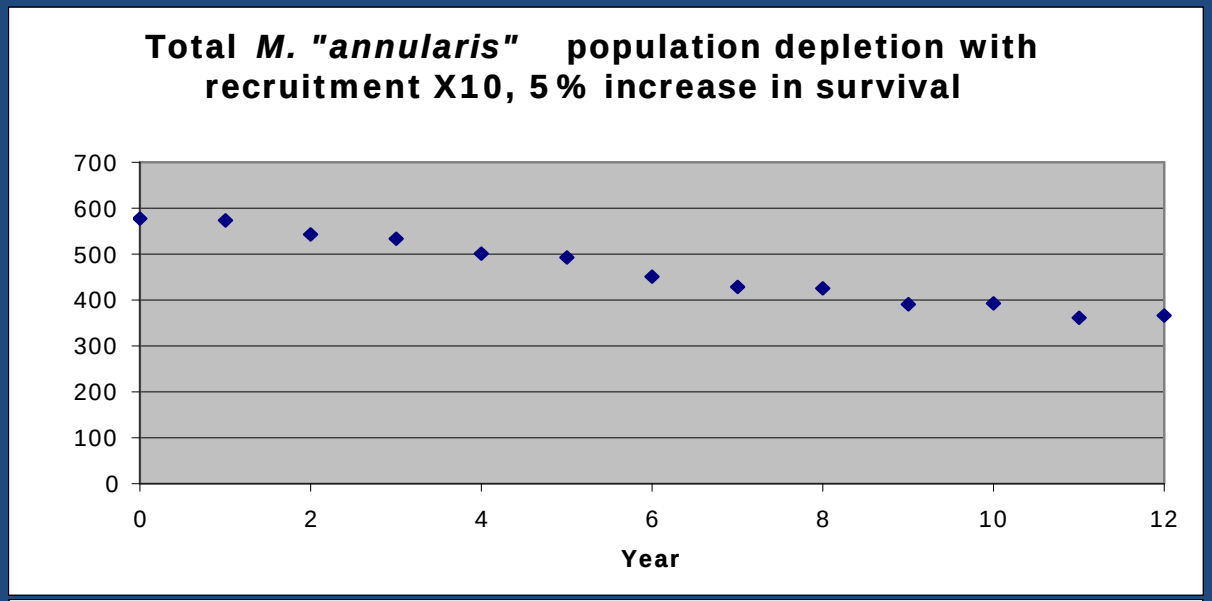
- The model was run for 12 years.
- At the start of each model year a random number of recruit (3, 4 or 6) were added to the smallest size class, based on our empirical measurements.
- Population is reduced by 50% in only 6 years.
- Low recruitment rates and high shrinkage and mortality rates in the smaller size classes never allow the population to increase.

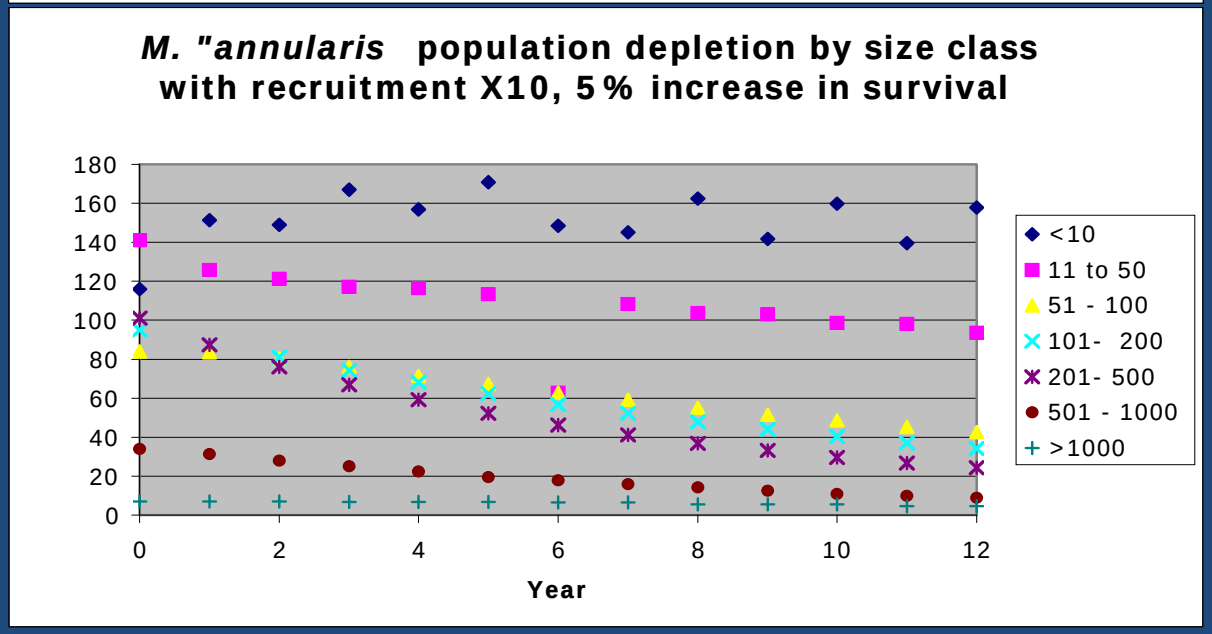




Population model: *M. "annularis"* with recruitment X10 and 5% increase in survival in three smallest size classes

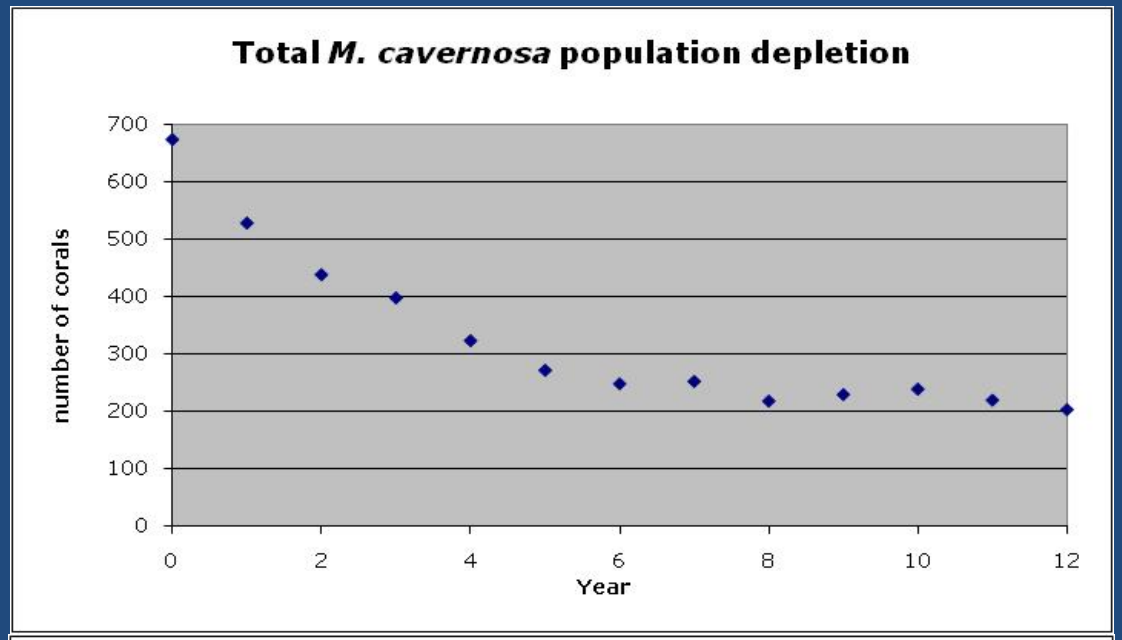
- Random recruitment of 30, 40 or 60 colonies per year.
- Survival increased by 5% in ≤10, 11-50 and 51-100 cm^2 size classes
- Population decline is slowed
- Shrinkage and mortality in larger size classes are significant in preventing population growth.

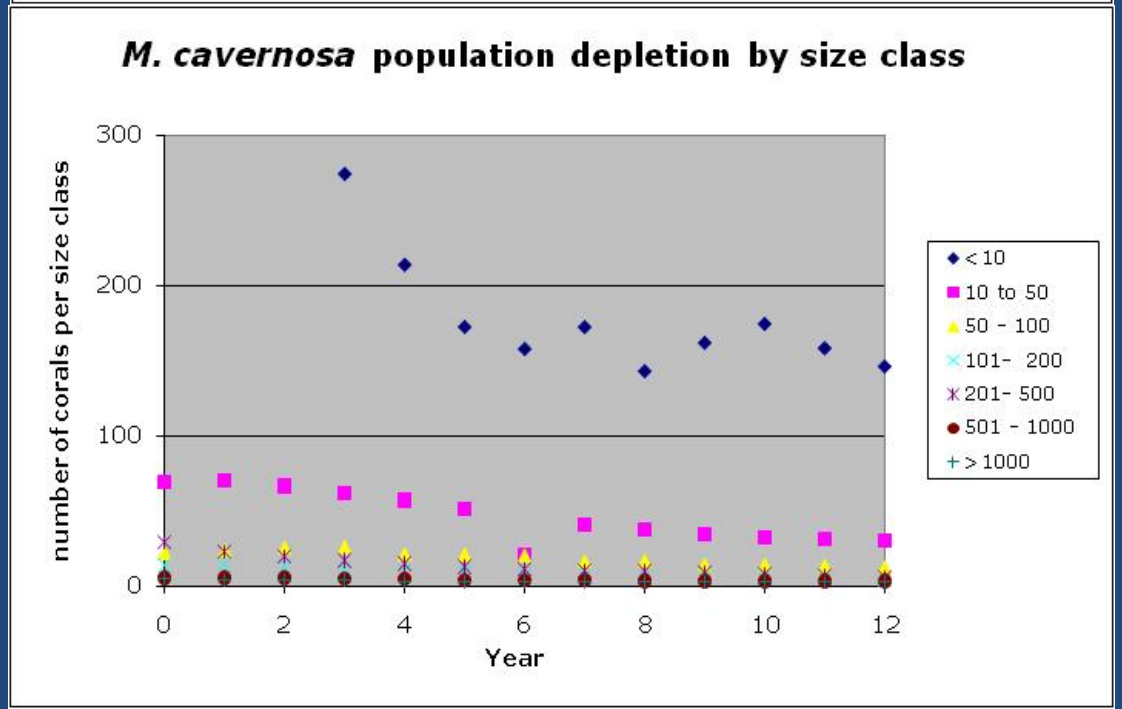




Population model: *M. cavernosa*

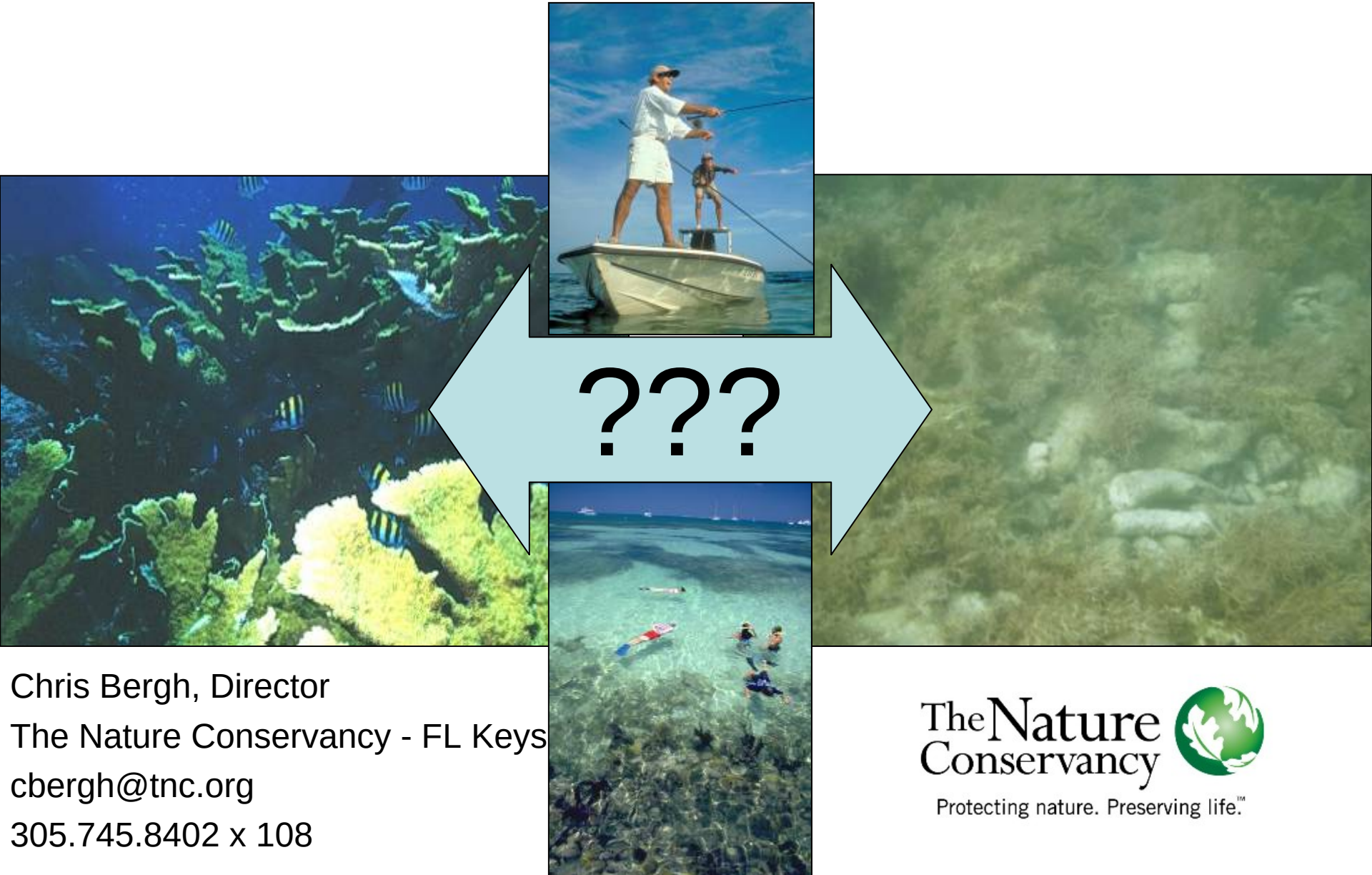
- Maintained a steady population of smallest colonies due recruitment of 20 to 60 colonies per study areas per year.
- Juvenile mortality limits successful transition of recruits to larger classes
- Mortality and shrinkage patterns in larger size classes limit population growth patterns.





Applicant Name	Applicant Affiliation	Project Title	SPAs	SUAs	WSER	TNER	TSER
Dr. Christopher Gledhill	NOAA Fisheries	SEAMAP Reef Fish Survey				X	X
Dr. Christopher Lobban	University of Guam	A search for ciliate-Symbiodinium symbioses in the Florida Keys and Dry Tortugas	Looe, Sand			X	
Mr. Matthew Long	University of Virginia	Total ecosystem fluxes of calcium and oxygen over coral reefs using the new eddy correlation technique	Cary, Grec, Mol, Con, All, Cof, Looe	Con, Ten, Looe			
Dr. C. Drew Harvell	Cornell University	Keys Wide Monitoring of Disease and Immune Responses of Sea Fan Corals	Cary, Mol, Con, All, Looe	Con, Ten, Looe			
Mr. Gabriel Delgado	Florida Fish and Wildlife Conservation Commission	Queen Conch Monitoring in the Florida Keys	X	X	X		
Mr. Margaret Miller	NOAA/NMFS/SEFSC	Ecological Restoration in the FKNMS	Elb, DrRx, Grec, Fren, Mol				
Mr. Marc Galloway	Marine Resources Development Foundation/MarineLab	Support of MarineLab Environmental Education Programs: Collecting of Biological Specimens and Performing Reef Check Benthic Surveys	X				
Dr. Ilze Berzins	The Florida Aquarium	Culture and Propagation of Hard Corals for Research and Restoration Activities			X		
Dr. Sidney Pierce	University of South Florida	Phylogenetic analysis of Florida populations of the sea slug, Elysia crispata	Looe				
Dr. Struan Smith	Georgia State University	Benthic community responses to ten years of management in the FKNMS	Cary, Mol	ES	X		
Dr. David Eggleston	North Carolina State University	Passive Underwater Acoustic Studies: an Emerging Ecological Tool for Florida Reefs and other Coastal Environments	EDR, Rock, Sand		X		
Dr. Niels Lindquist	UNC Chapel Hill	Role of sponges in N cycling and total respiration in coral-reef ecosystems		Con			
Dr. Mark Fonseca	NOAA/NOS	Comparative analysis of the function of disturbed and undisturbed coral reef and non-coral ecosystems in the Tortugas: Phase II-measuring the refugia effect of establishing a reserve				X	X
Mr. Ken Nedimyer	Healthy Reefs, Inc.	Staghorn Coral (Acropora cervicornis) restoration using corals grown in an offshore coral nursery	Mol				
Dr. Steven Miller	University of North Carolina at Wilmington	Coral Reef Rapid Assessment, Monitoring, and Modeling in the Florida Keys National Marine Sanctuary	X	X	X	X	X
Ms. Lyza Johnston	University of Miami / RSMAS	Reproductive biology and early life-history of the corallivorous gastropod, Coralliophila abbreviata	Cary, Elb, DrRx, Grec, Fren, Mol				
Dr. Mark Butler	Old Dominion University	Characterization of Hardbottom & Patch Reef Community Dynamics with a Focus on Lobster, Sponges, Urchins and Octopus			X		
Dr. Kimberly Ritchie	Mote Marine Laboratory	Multi-Disciplinary Multi-Institutional Coral Studies in the Dry Tortugas				X	
Dr. Margaret Miller	NOAA Fisheries SEFSC	Development of health/disease screening tools for Acropora spp.	Cary, Elb, DrRx, Fren, Mol				
Dr. Su Sponaugle	University of Miami/RSMAS	Age, Growth, and Recruitment of Reef Fish in the Upper Florida Keys	Fren, Mol				
Dr. Laurie Richardson	Florida International University	Microbial Community Organization and Development of a Pathogenic Consortium, funded by NIH	Cary, DrRx, Grec, Fren, Mol, Con, Dav				
CDR James Verlaque	NOAA Ship Nancy Foster	Permit for the NOAA Ship Nancy Foster to enter the Area To Be Avoided (ATBA) to support FKNMS and EPA research on coral condition		X		X	X
Dr. Pamela Hallock Muller	University of South Florida	Application of Bioindicators to Study Ecological Function of Florida Keys Reefs	Cary, Grec, Mol, Con, All, Cof, Som, Looe	Con, Ten, Looe	X		
Dr. Mark Warner	University of Delaware	Evaluating the Impact of Zooxanthellae Physiology and Diversity on the Loss and Recovery of Coral-algal Symbioses: Developing a Predictive Physiological Model for Coral Bleaching	Con, Cheec				
Dr. Pamela Reid	University of Miami/RSMAS	Single Beam Acoustic Seabed Classification in Coral Reef Environments	Cary, Elb, Dav				
Dr. Alejandro Acosta	Florida Fish & Wildlife Conservation Commission/FWRI	Performance Evaluation of Marine Zoning in the Florida Keys National Marine Sanctuary	Looe	ES	X		
Dr. Thomas Cuba	Delta Seven Inc.	Secondary succession on damaged coral reefs: What happens after the disease and A preliminary investigation into factors affecting the patterns of open space development (disease)	Looe				
Dr. Elizabeth Johns	NOAA's Atlantic Oceanographic and Meteorological Lab	Interdisciplinary Coastal Oceanographic Observations	X	X	X	X	X
Dr. James Leichter	University of California at San Diego	Longterm Temperature Variability on the Florida Keys Reef Tract	Cary, Elb	Con			
Dr. Erin Lipp	University of Georgia	Pathogenicity of select Serratia marcescens isolates against elkhorn coral, Acropora palmata	EDR, Rock, Sand		X		
Mr. David Palandro	University of South Florida	Coral reef habitat change and water clarity assessment (1984-2002) for the Florida Keys National Marine Sanctuary using Landsat satellite data	Mol, Som, Looe				
Dr. John Lamkin	NOAA-Fisheries Southeast Fisheries Science Center	Juvenile Snapper Micro-Acoustic Tagging (JSMAT)			X		
Ms. Meaghan Johnson	The Nature Conservancy	The Effects of Major Disturbances on the Resilience of Corals in Florida	X	X	X	X	X
Dr. Sandra Brooke	Fish and Wildlife Research Institute	Growth and survival of Montastrea annularis juveniles in the Florida National Marine Sanctuary, Middle Keys Region	Looe				
Dr. Erin Lipp	University of Georgia	Prevalance and diversity of Serratia marcescens as a pathogen of elkhorn coral, Acropora palmata	EDR, Rock, Sand		X		
Dr. James Fourqurean	Florida International University	Long Term Seagrass Monitoring in the Florida National Marine Sanctuary	X	X	X	X	X
Dr. Steven Rohmann	NOAA/NOS/MB	Measuring light reflectance of seafloor habitat features with a spectrometer in the Western Sambo Ecological Reserve	Looe	Looe	X		
Dr. David Zawada	U.S. Geological Survey	USGS ATRIS Mapping in the Lower Florida Keys		Con, Looe,			
Ms. Nikki Poulos	Florida International University	Restoring predatory fish populations within marine reserves: Does this result in trophic cascades which influence the effectiveness of protected areas on coral reef habitats?	Mol	Con, ES	X		
Dr. Jessica Ward	University of California, San Diego	Role of bacteria in coral disease resistance	Con	Con			
Dr. Alina Szmant	University of North Carolina at Wilmington	Recruitment of the reef building coral Montastraea faveolata: Larval dispersal, settlement and post-settlement survivorship	DrRx				
Dr. Mark Hay	Georgia Institute of Technology	Herbivore Diversity as a Biological Fulcrum to Restore Coral Reefs	Fren, Mol				
Mr. Michael Burton	NOAA/National Marine Fisheries Service	Assess/Monitor effects of MPA status on reef fish populations and spawning aggregations in the Tortugas Ecological Reserves				X	X
Dr. Joseph Pawlik	University of North Carolina Wilmington	Ecology of Sponges on Florida Reefs: Demography and Bleaching		Con			
Dr. Alina Szmant	University of North Carolina - Wilmington	(1) Ecological Restoration in the FKNMS, and (2) Coral Reef Genomics: A genome wide approach to the study of cnidarian symbiosis	Cary, Elb, DrRx, Grec, Fren, Mol				
Dr. Kimberly Ritchie	Mote Marine Laboratory	Molecular Interactions between Coral and Associated Bacteria	Looe				
Dr. Pamela Reid	University of Miami / RSMAS	Application of ROV-based video technology to complement coral reef resource mapping and monitoring	Mol				
Dr. Dana Williams	University of Miami and NOAA/SEFSC	Restoration of Acropora palmata using naturally occurring fragments	Cary, Elb, DrRx, Fren, Mol				
Ms. Adrienne Romanski	University of Miami/RSMAS and Columbia University	Symbiodinium communities associated with diseased scleractinian corals in the Florida Keys	Fren				
Dr. Dana Williams	University of Miami RSMAS & SEFSC	Assessment of Threats to Acropora spp. in the Florida Keys: Proximal Causes and their Relative Importance to Remnant Populations	Cary, Elb, DrRx, Grec, Fren, Mol				
Dr. Kimberly Ritchie	Mote Marine Laboratory	Coral Health and Disease in the Florida Keys and Antibiotic Properties of Coral Eggs, Sperm, Eggs/sperm Bundles, and Planulae	X	X	X	X	
Dr. Jerald Ault	University of Miami/RSMAS	Acoustic Telemetry Tracking of Reef Fish to Determine Population Flux Rates in Open and Fully-Protected Marine Reserves				X	
Mr. Don DeMaria	Sea Samples	Collection and Taxonomy of Shallow Water Marine Organisms				X	X
Dr. Robert Halley	U. S. Geological Survey	Integrated Geologic Studies of Coral Reefs	Cary				
Dr. Dana Williams	University of Miami RSMAS & SEFSC	Genotypic Assessment of Threatened Acropora palmata in the Upper FKNMS	Cary, Elb, DrRx, Grec, Fren, Mol				
Ms. Megan Lowenberg	Florida State University	Characterization of Bacterial Flora of Spiny Lobster, Panulirus argus, and the Etiology of Shell Disease in Spiny Lobster	X		X		
Dr. James Leichter	University of California at San Diego	A Multiscale Investigation of High Frequency Upwelling on the Florida Keys Reef Tract	Cary, Elb, Con, Looe, Sand	Con, Ten			
Dr. C. Drew Harvell	Cornell University	Resistance Measures in Diseased and Bleached Scleractinian Corals During Thermal Events	Looe				
Dr. Felicia Coleman	Florida State University	Population Density, Demographics, and Predation Effects of Adult Goliath Grouper	X	X	X	X	X
Dr. Tonya Shearer	Georgia Institute of Technology	Fine-scale Genetic Structure of Scleractinian Corals and Implications for Reef Restoration	Con	Con			
Dr. Deborah Santavy	U. S. Environmental Protection Agency	Assessing the Health of Coral Reefs in the Florida Keys	X	X	X	X	X
Dr. William Fisher	U. S. Environmental Protection Agency	In Situ Growth Rates of Stony Corals using Nondestructive Photographic Methods	X	X	X	X	X
Dr. Nerida Wilson	Auburn University	Recent or relic distribution? The nudibranch Glossodoris sedna in the Caribbean	Mol				
Dr. James Lindholm	Pfleger Institute of Environmental Research	Movement Behavior of Fishes: The Role of Scale and No-Take Protection in the Conch Reef SPA/RO	Con, Dav	Con			
Ms. Nicole Fogarty	Florida State University	The Significance of Hybridization in Depauperate Caribbean Acroporid Coral Populations	Looe		X		
Dr. Greg Piniak	NOAA/NOS/NCCOS	Field Calibration of a Spatial Recovery Model for Injured Coral Reefs	X	X	X		
Dr. Stephen Monismith	Stanford University	The Role of Hydrodynamics in Determining Nutrient Fluxes to Conch Reef		Conch			
Dr. Eberhard Gischler	Johann Wolfgang Goethe-University	Partial Mortality of Massive Reef-building Corals: An Index of Patch Reef Condition, Florida Reef Tract (Re-visit)	Cary, DrRx, Cheec, NFH		X		
Mr. Gustavo Paredes	University of California San Diego	The Impact of Fishing on Caribbean Coral Reef Food Webs and Mechanisms of Recovery	Cary, Fren, Mol, Looe, Sand		X	X	X
Dr. Robert Ginsburg	RSMAS/University of Miami	Characterization of the Tortugas Bank Ecosystem and Its Condition				X	
Dr. Paula Mikkelsen	American Museum of Natural History	Qualitative Survey of Molluscan Diversity in the Florida Keys	X	X	X		
Dr. Laurie Richardson	Florida International University	Distribution and Etiology of Two Coral Diseases in the Florida Keys National Marine Sanctuary: Black Band Disease and White Plague Type II	Cary, Elb, DrRx, Grec, Fren, Mol, Con, Dav, H&C, Cheec, All				
Dr. Diego Lirman	University of Miami	Coral Size-Frequency Distributions as Indicators of Reef Health: Monitoring and Modeling Approaches	Cheec, Cof, H&C				
Dr. Rodney Bertelsen	Florida Fish and Wildlife Conservation Commission	Evaluation of Potential Spillover of Lobsters from the Dry Tortugas National Park to the Tortugas Ecological Reserve (North)				X	
Dr. Pamela Hallock Muller	University of South Florida	Larger Foraminifera as Bioindicators of Coral Reef Health	Cary, Grec, Con, All, Cof, Som, Looe,	Con, Ten, Looe	X		
Dr. Judith Lang		Experimental Reduction of Predatory Snails from Low-Density Stands of Elkhorn Corals in the Upper Keys	Cary, Elb, Grec, Mol, Dav, H&C, Cheec, All				
Mr. Kevin Rademacher	National Marine Fisheries Svc.	Southeast Area Monitoring and Assessment Program (SEAMAP)	X			X	X

Florida Reef Resilience Program



Chris Bergh, Director
The Nature Conservancy - FL Keys
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305.745.8402 x 108

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The Florida Reef Resilience Program Steering Committee

Department of Environmental Protection FLORIDA

National Oceanic and Atmospheric Administration NOAA U.S. Department of Commerce

Australian Government
Great Barrier Reef Marine Park Authority

Florida Fish and Wildlife Conservation Commission

University of Miami RSMAS Rosenstiel School of Marine & Atmospheric Science

WWF

National Park Service

Mote Marine Laboratory

Florida Institute of Technology
High Tech with a Human Touch

USF University of South Florida College of Marine Science

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The Florida Reef Resilience Program spans the reefs from the Dry Tortugas to St. Lucie Inlet

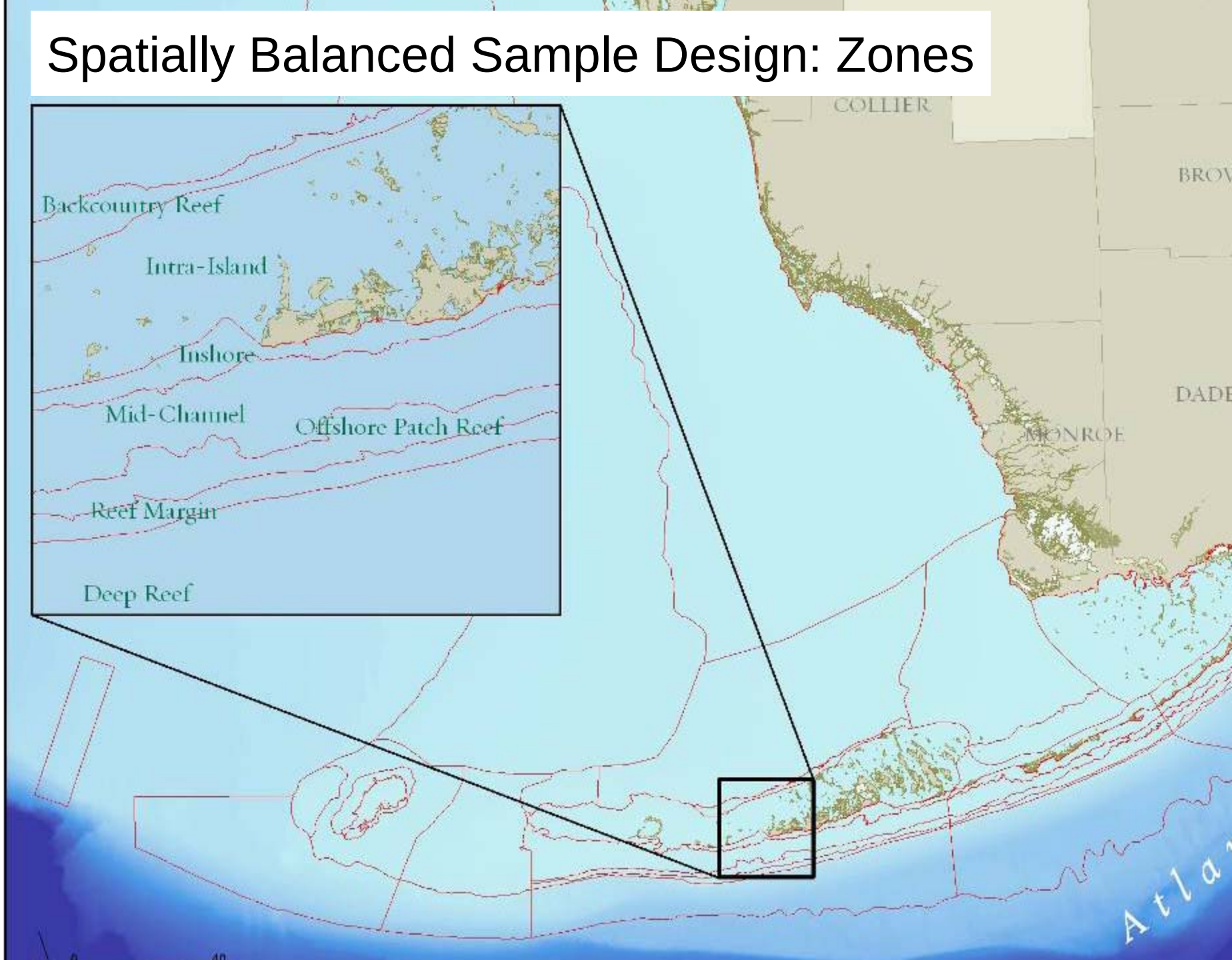
Which Reefs are Resilient?
Disturbance Response Monitoring

- Monitor coral condition during and after disturbance, with bleaching as a focus

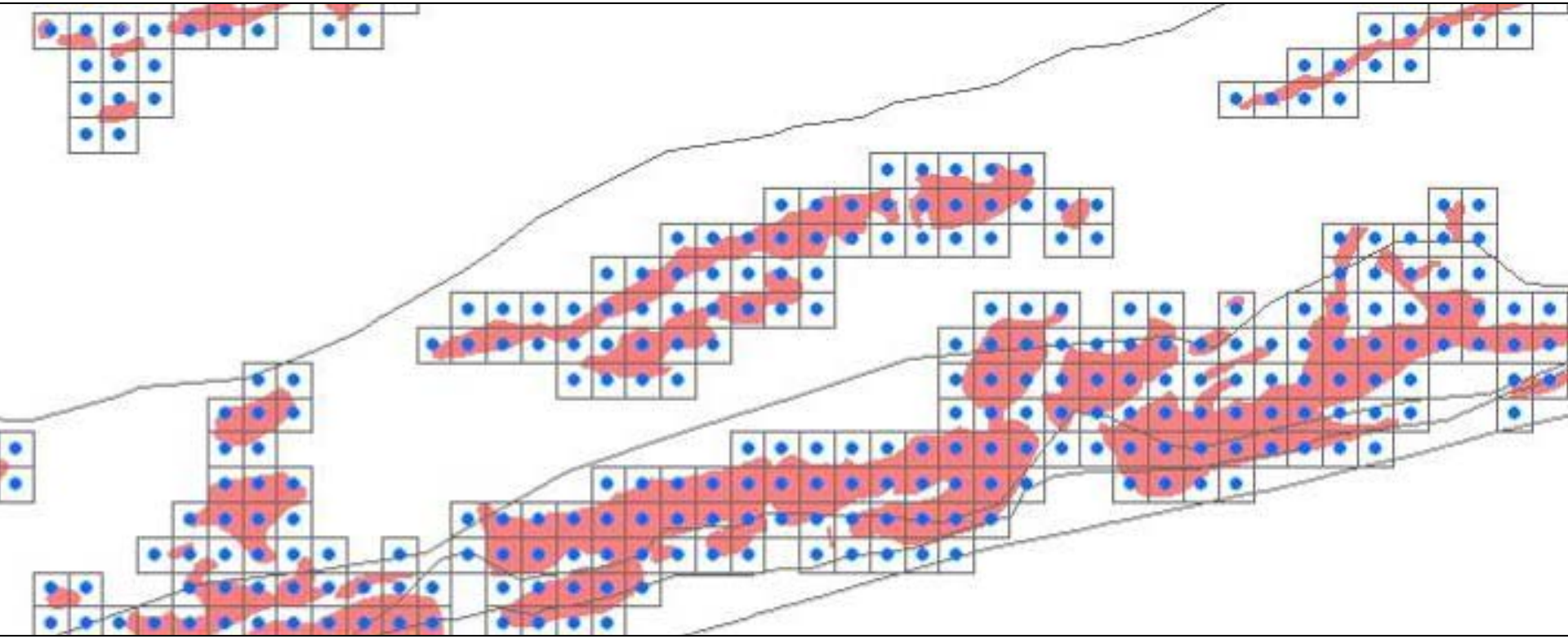
- Scientists surveyed Florida's shallow coral reefs during peak annual temperatures of 2005, 2006 and 2007 (and 2008)

Spatially Balanced Sample Design: Subregions

Spatially Balanced Sample Design: Zones



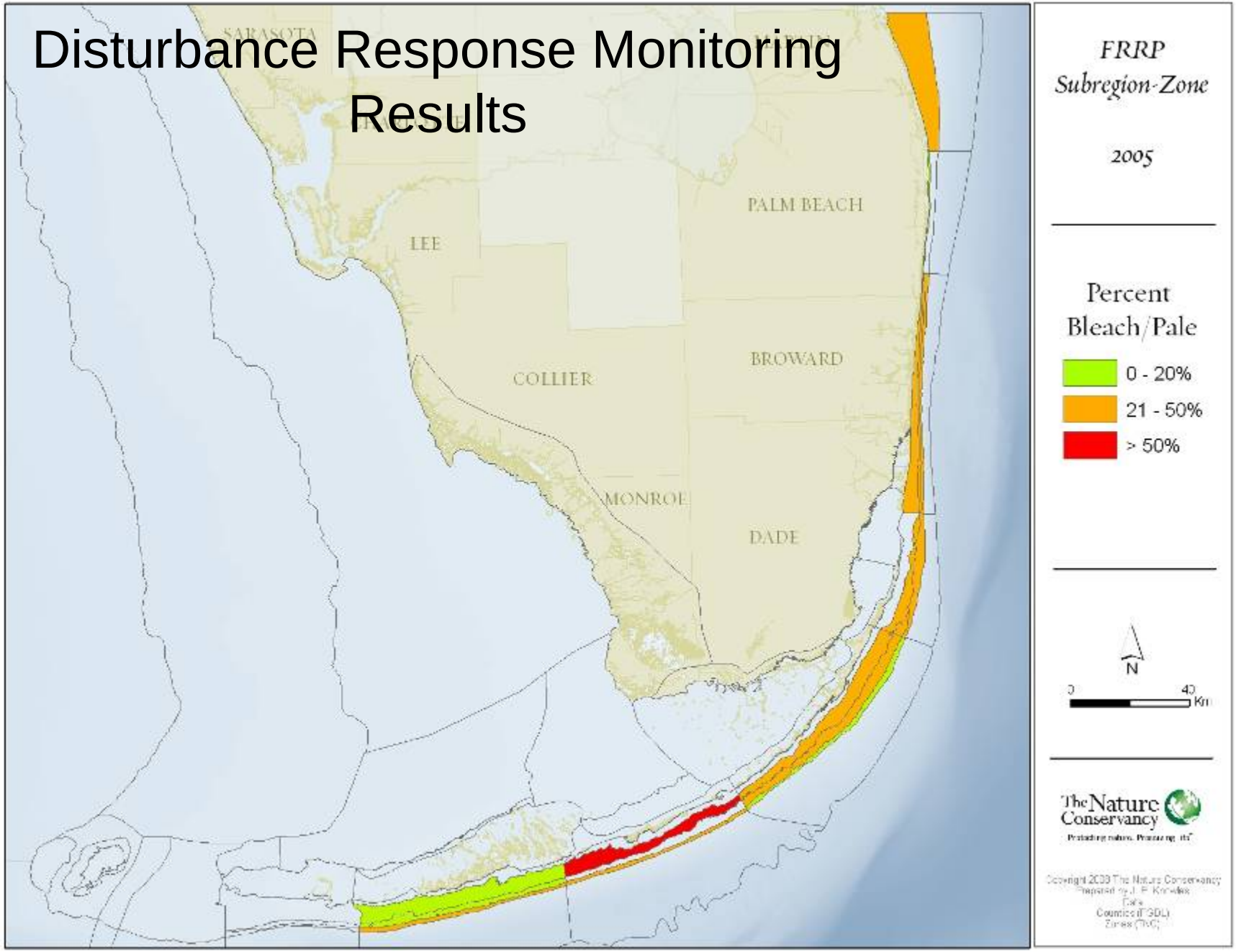
Spatially Balanced Sample Design: Sites



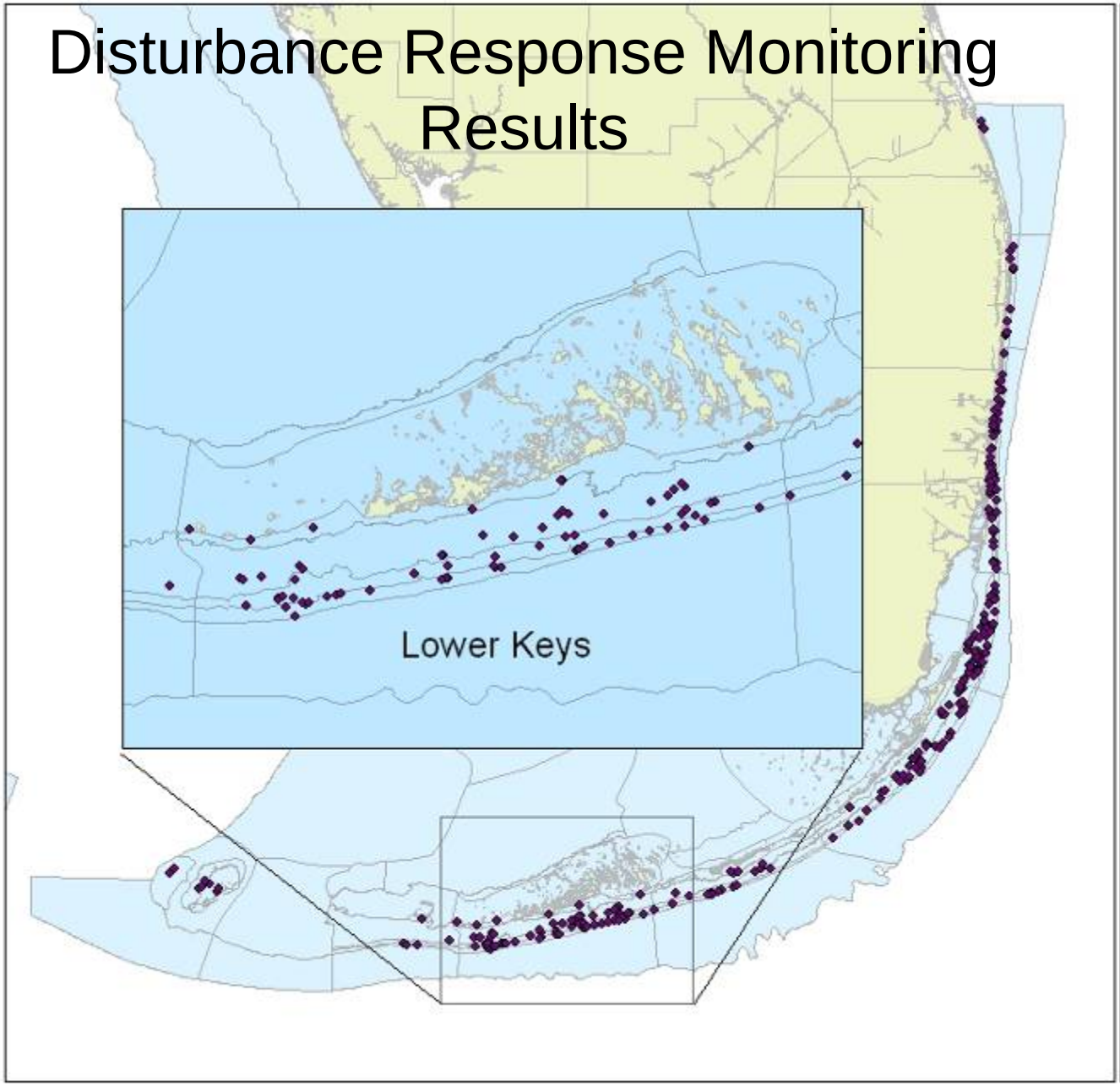
Disturbance Response Monitoring
Field Methods

- Random sites generated and assigned to teams
- 1 x 10m belt transects (2/site)
- Measure/assess all corals (>=4 cm)
- Species level identification
- Bleaching and disease (visually)
- Data entered online
- Database queried for results

Disturbance Response Monitoring Results



Disturbance Response Monitoring Results



FRRP
Sub-region-Zone

Lower Keys

2005-2007

Top Quartile
Total Area
Broadcast Spawners

- Large corals

N

0 80 Km

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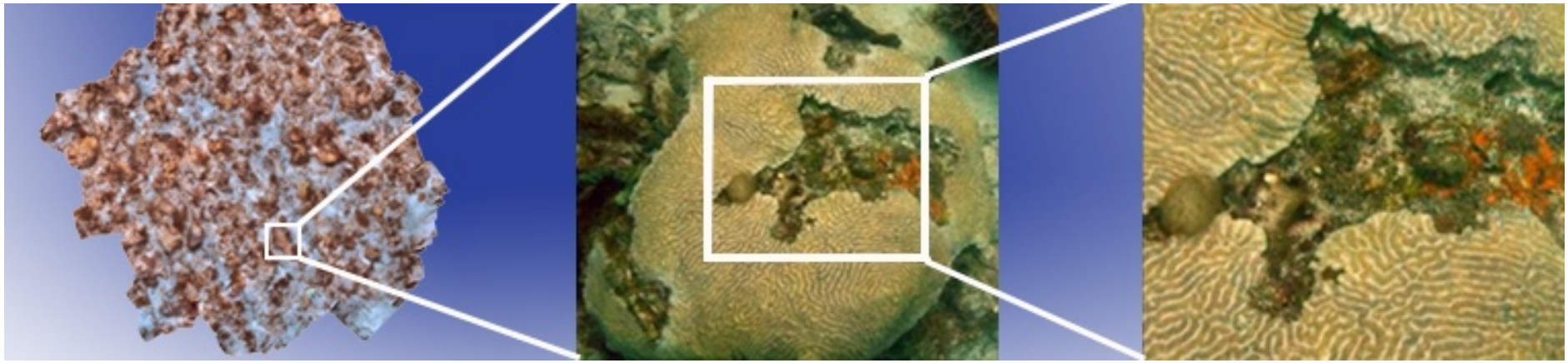
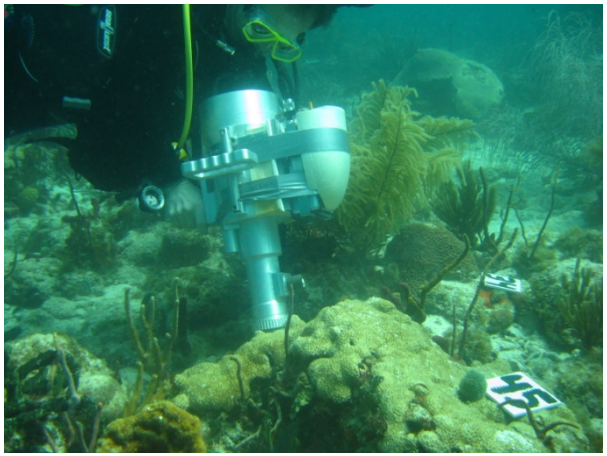
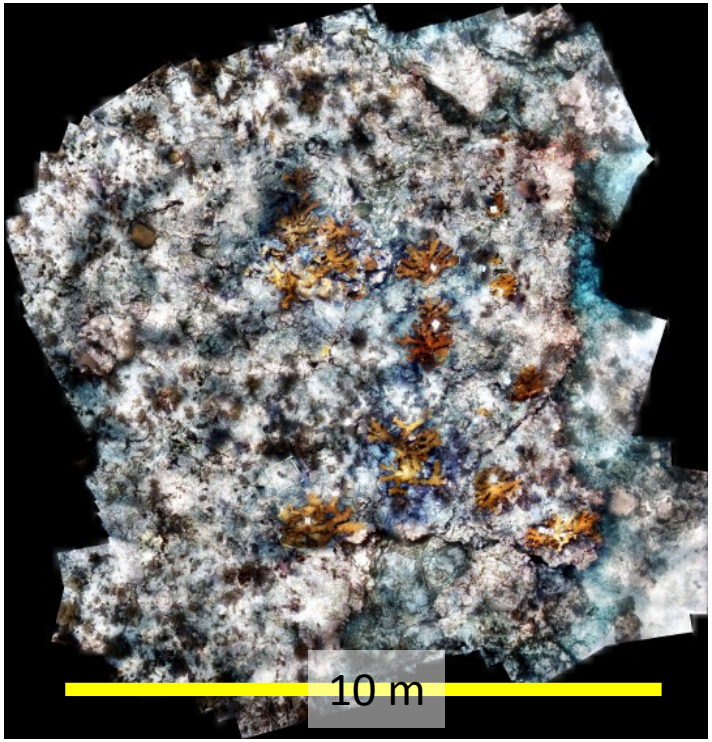
Landscape Video Mosaic & Fluorescence Induction and Relaxation System Integration

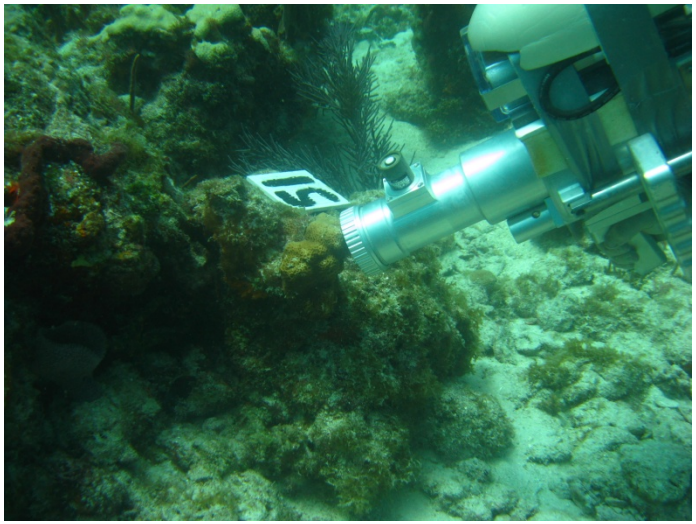




Dr. Diego Lirman, Dr. Pam Reid, Dr. Maxim Gorbunov and Dr. Paul Falkowski

Background





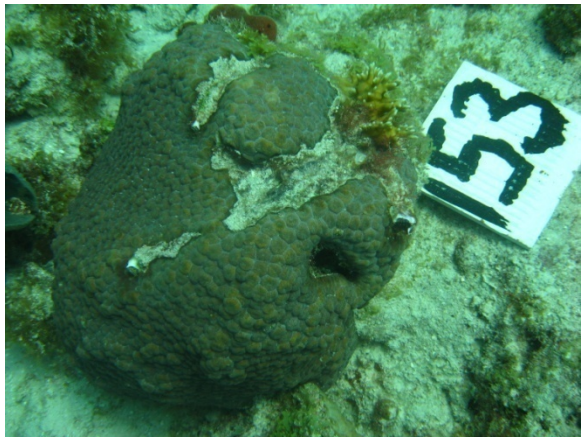
Simultaneous development of 2 technologies

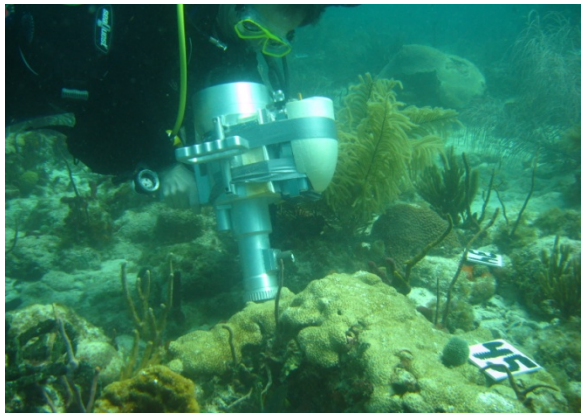
- Cover different spatial scales
- Assess different indicators of reef health

Integration Project

- Can these two be integrated?
- Benefits of a combined technology
- Prototype Technology

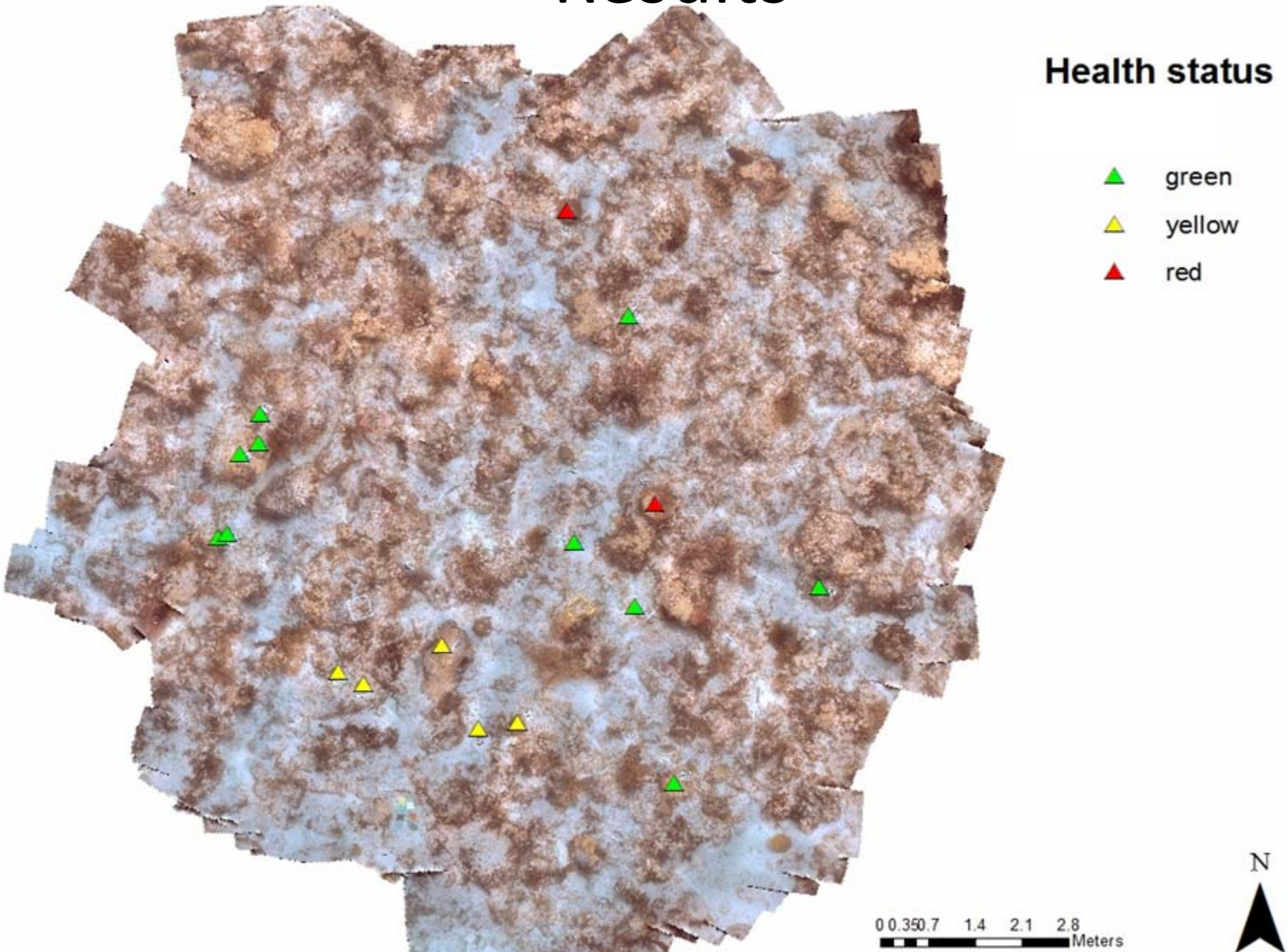
Field Integration



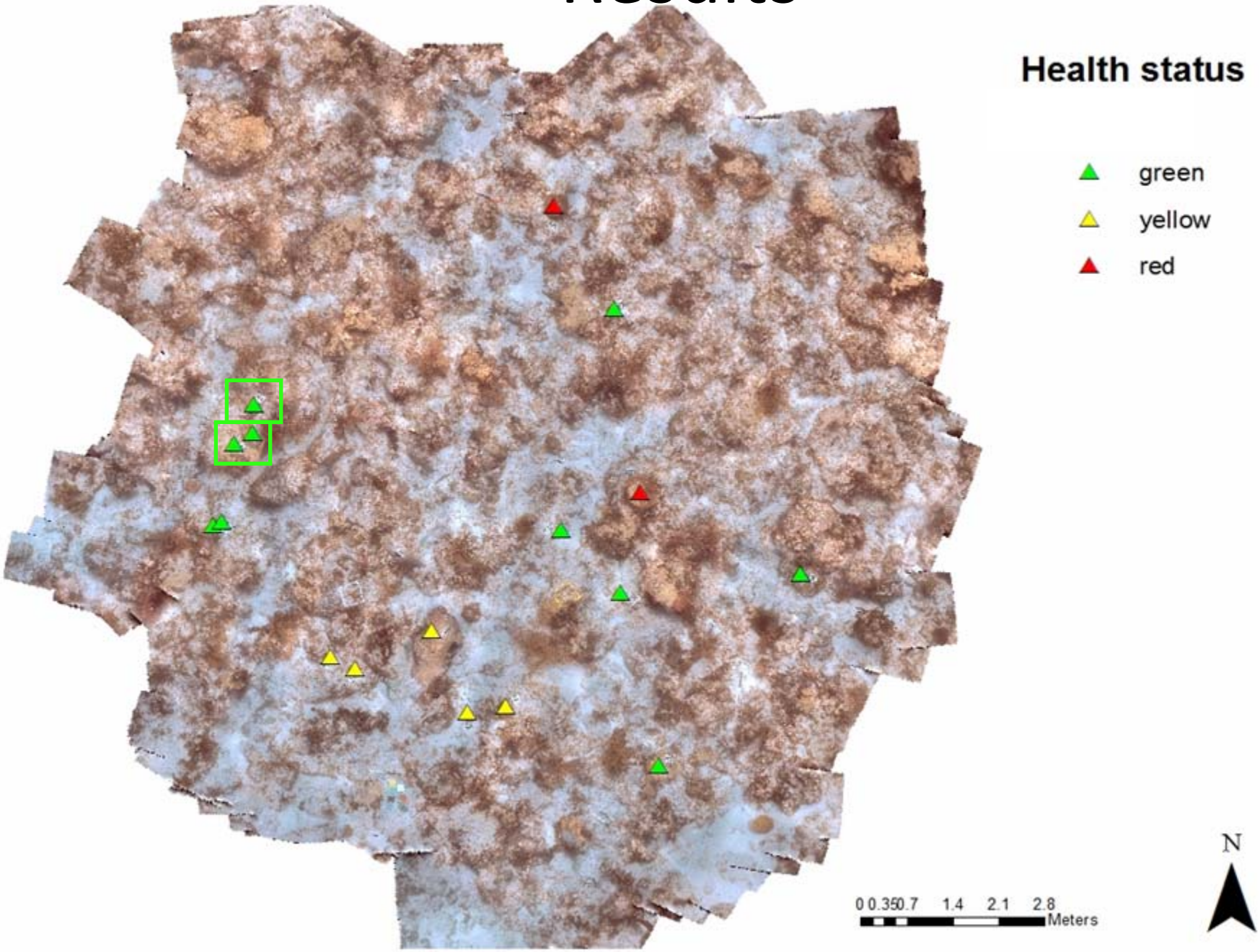


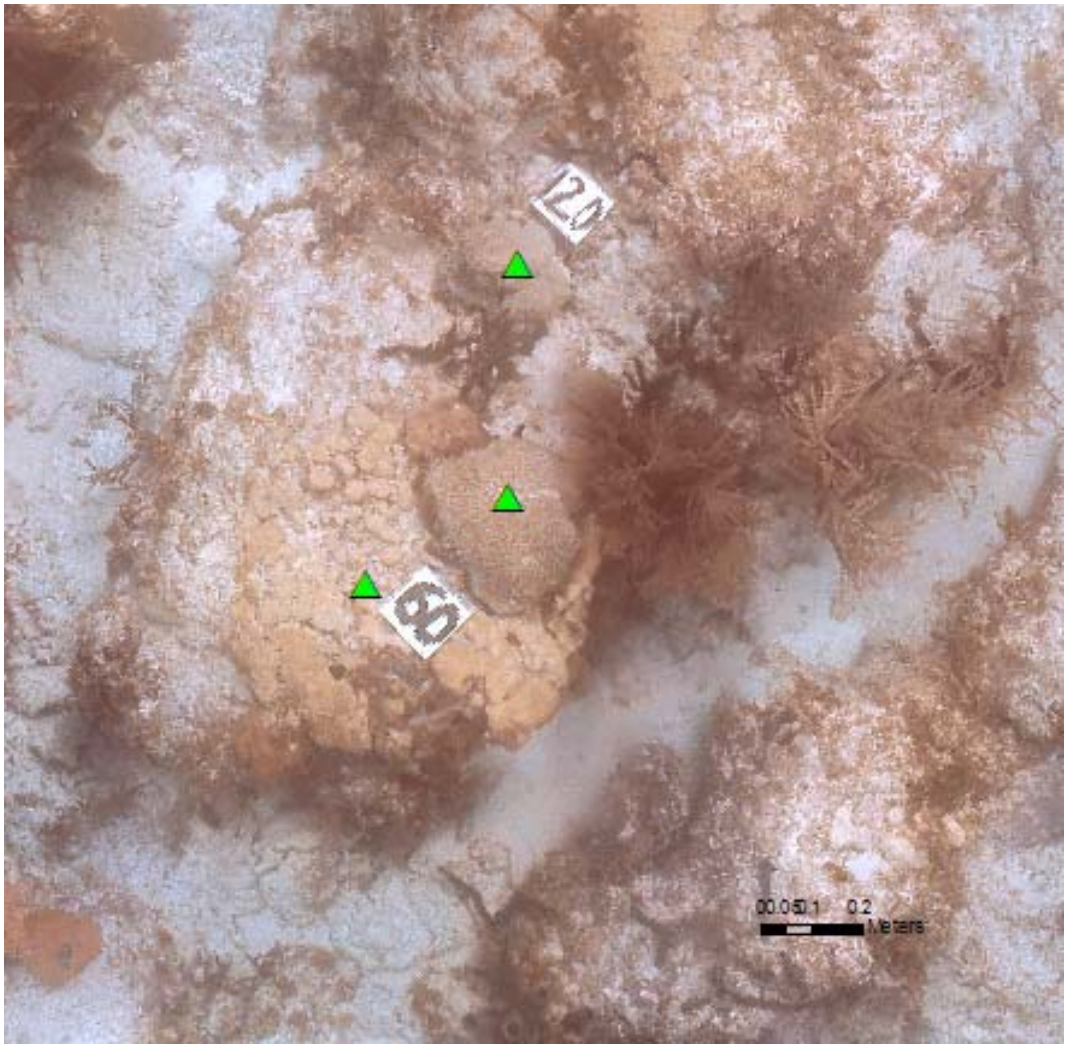
- June 16th - 17th 2008
- 2 reef sites in Florida
- Coral Markers Used for Spatial Integration
- Individual FiRe measurements
  - Hard corals, soft corals, zoanthids, sponges
- Mosaiced survey area

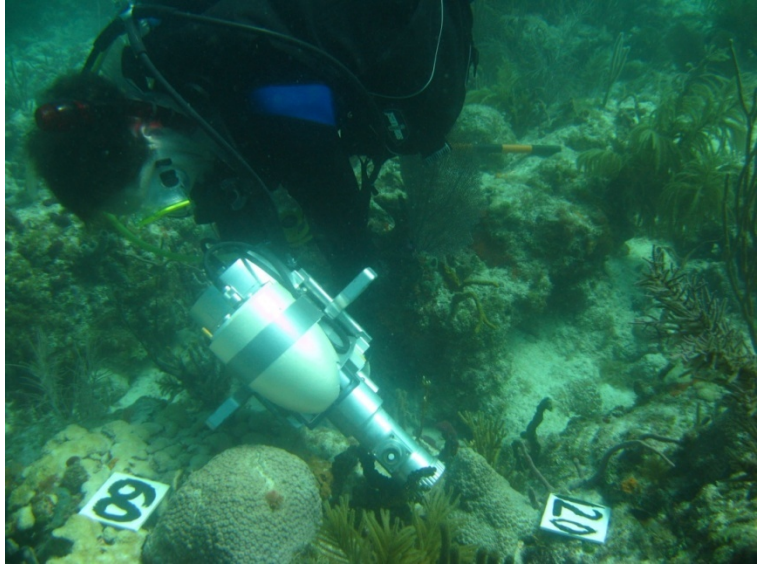
Results

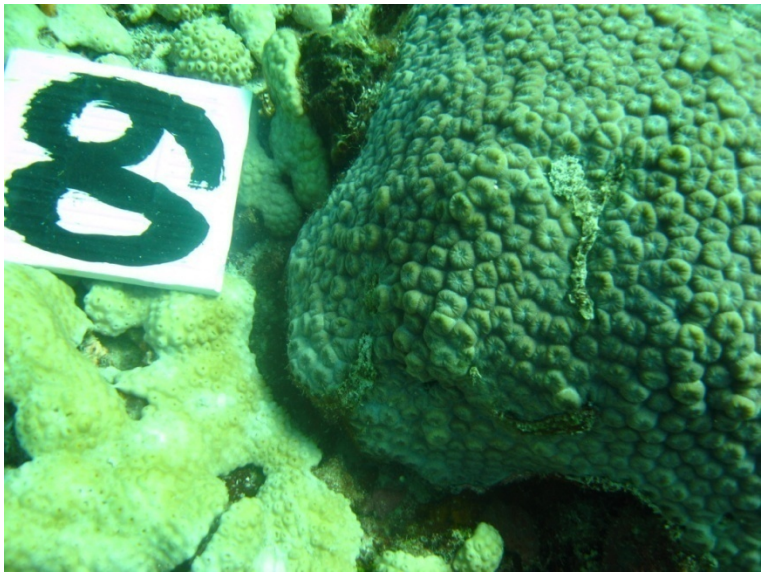


Results



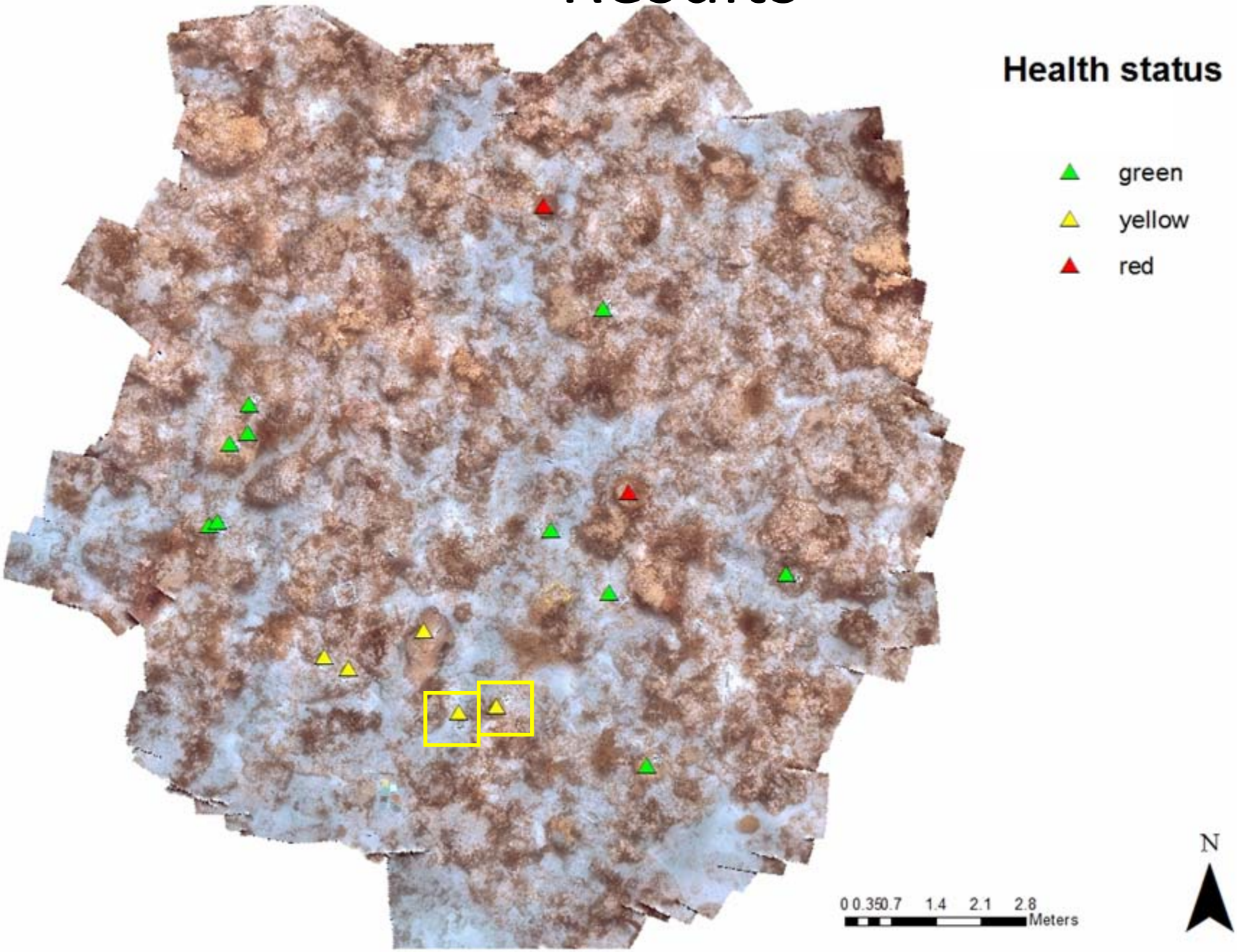


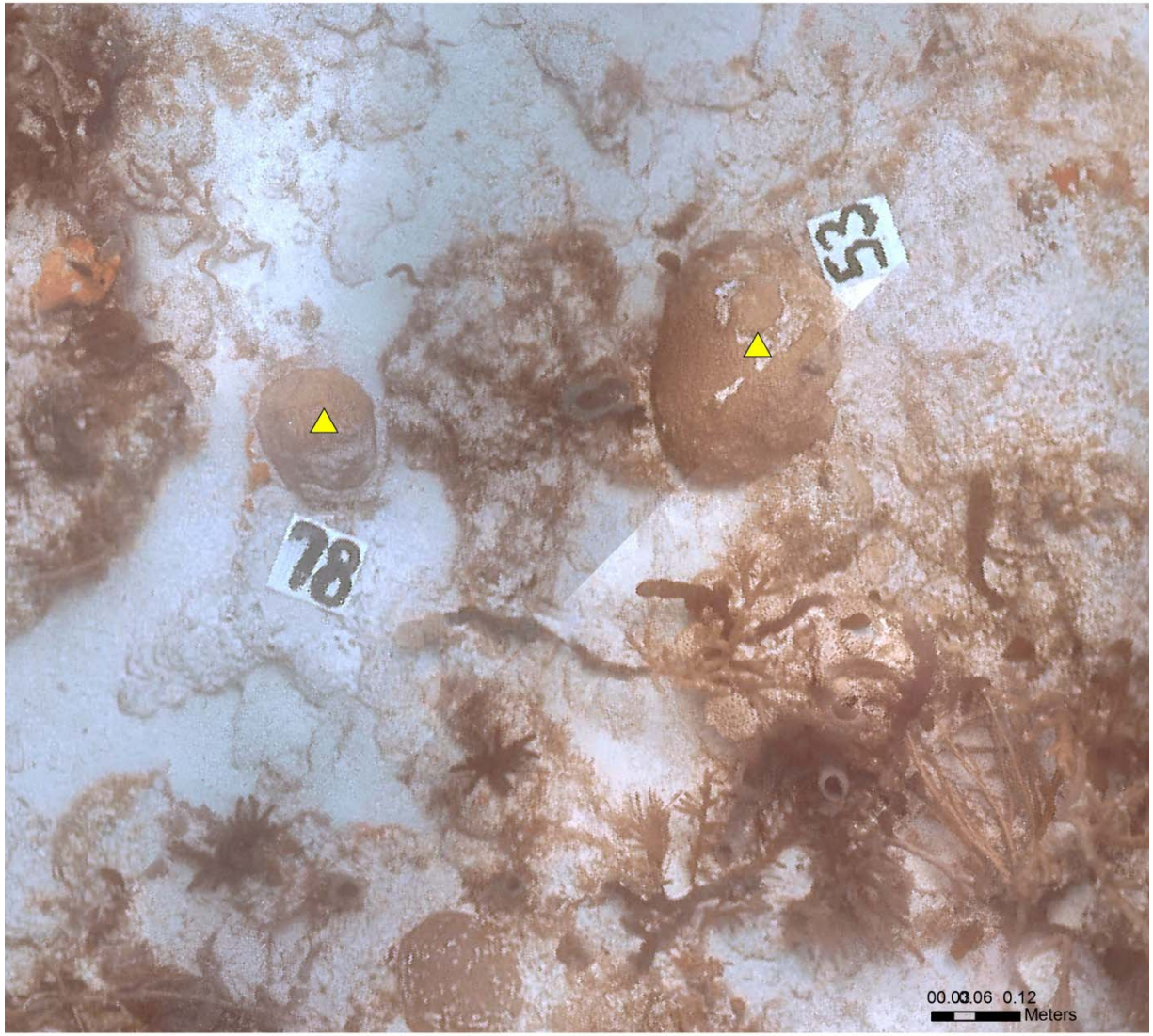


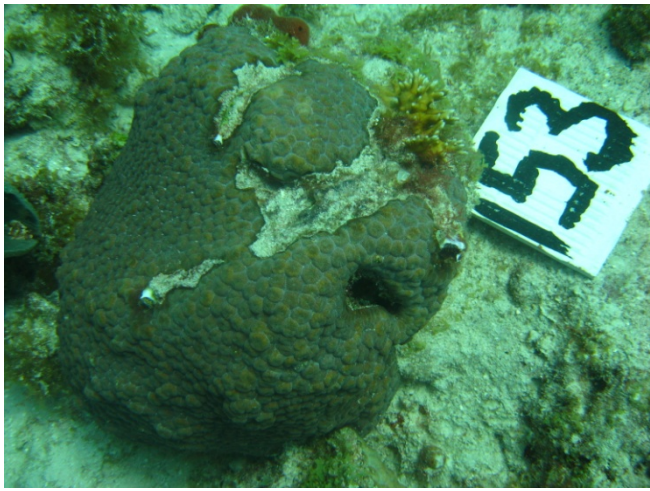


Species	Fo	Fm	Fv	Fv/Fm	Sigma	Tau1	Green/Yellow/Red flag
M. cavernosa	228	353	126	0.36	211	586	green
P. caribaeorum	304	433	129	0.30	266	501	green
M. cavernosa	338	561	223	0.40	280	547	green

Results

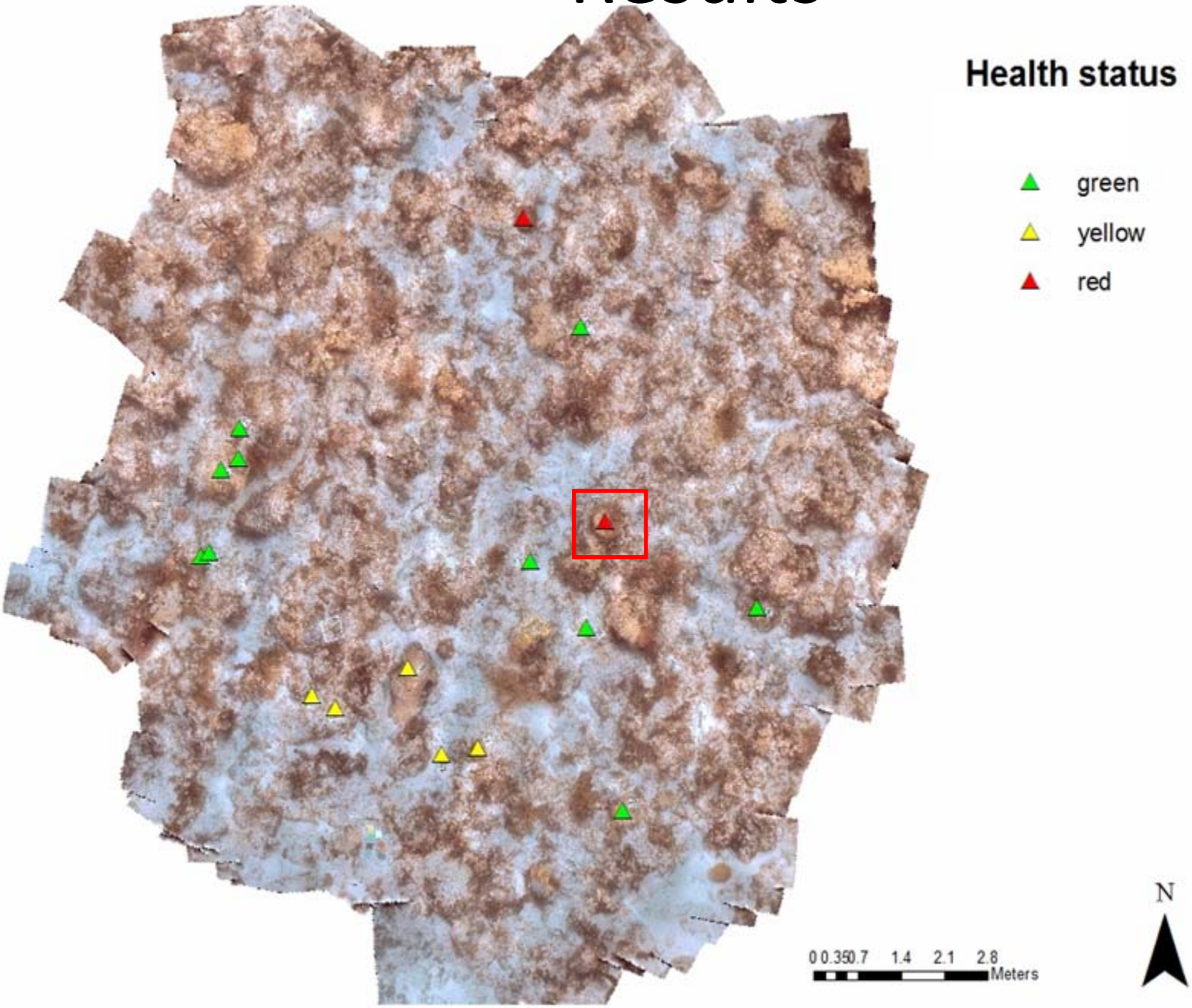


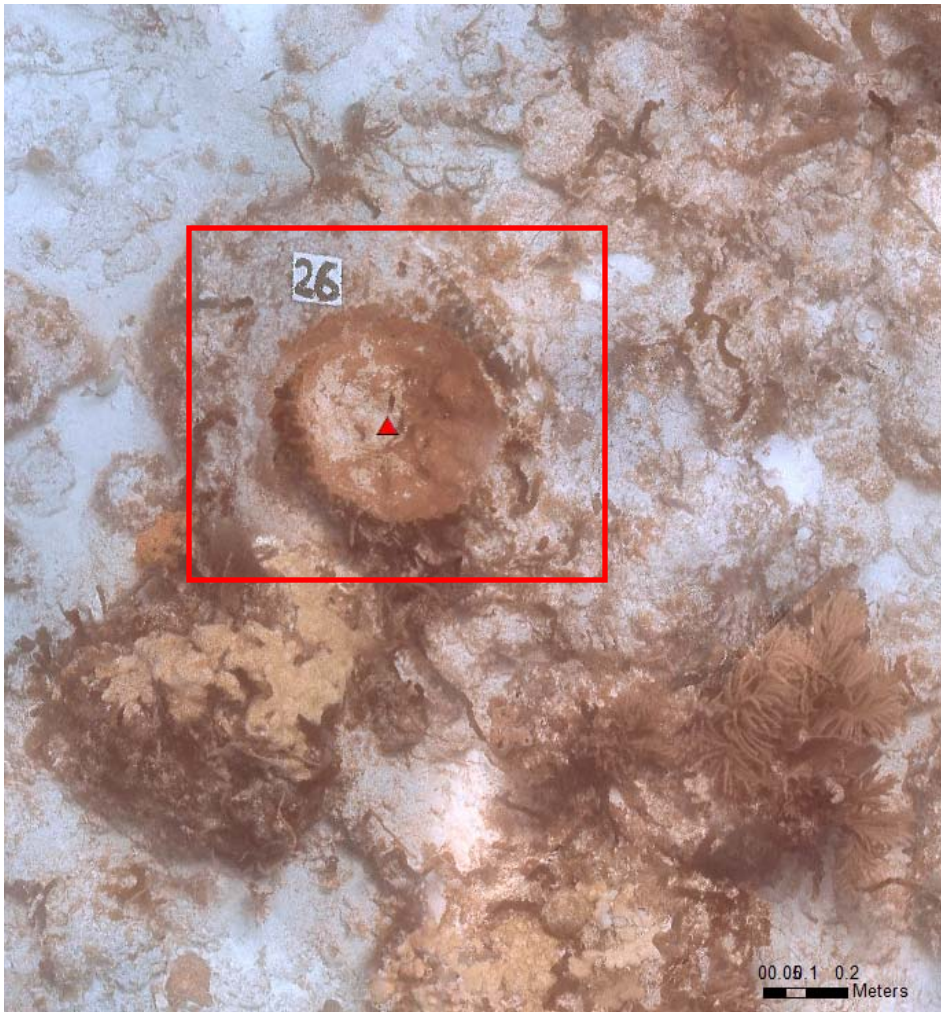


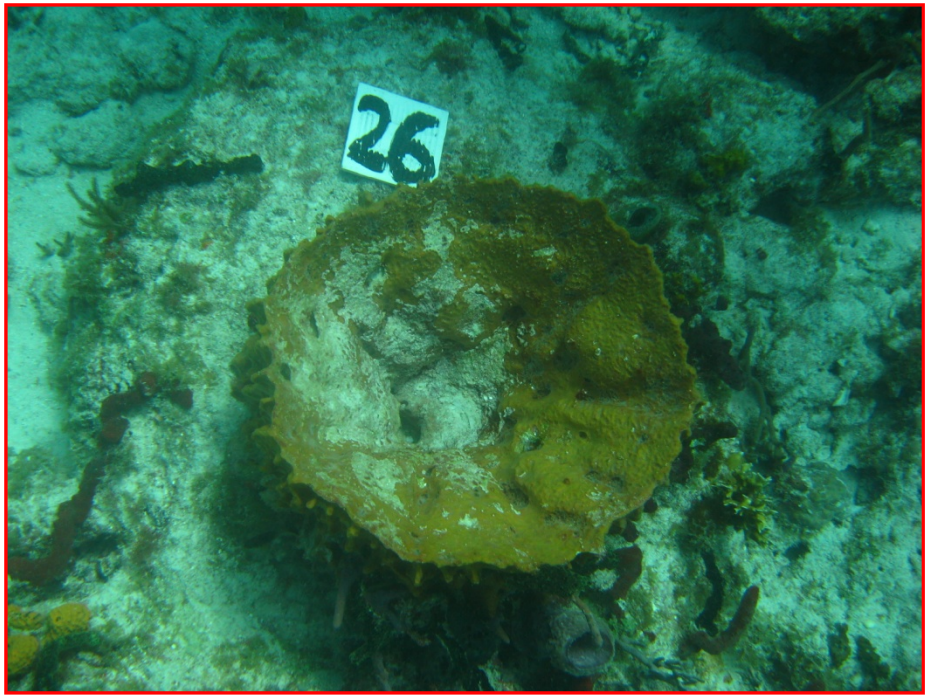


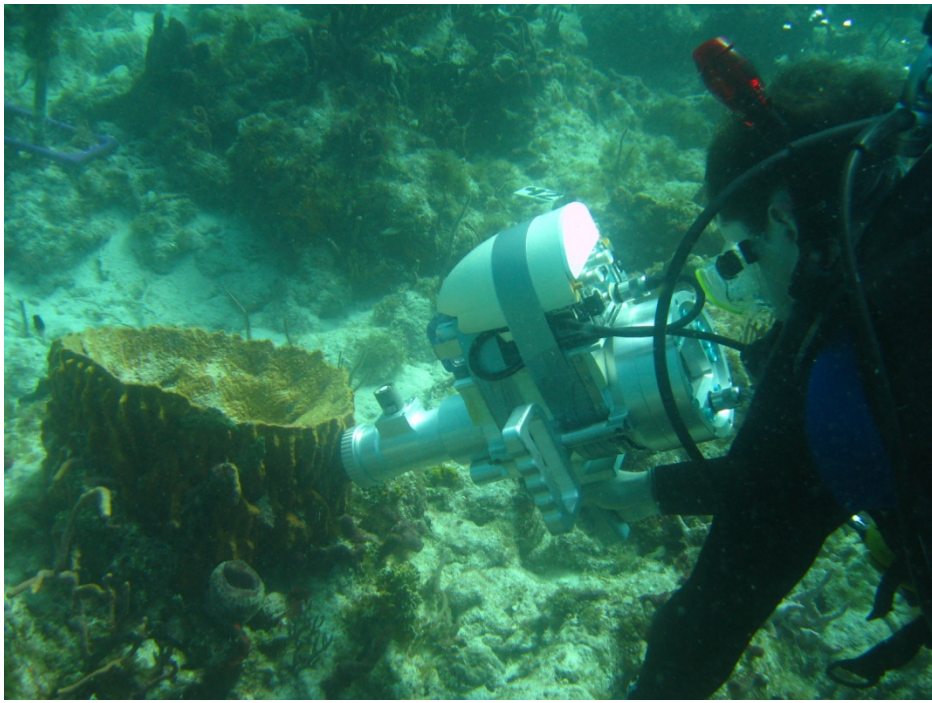
Species	Fo	Fm	Fv	Fv/Fm	Sigma	Tau1	Green/Yellow/ Red flag
D. stokesii	304	456	152	0.33	277	655	yellow
M. cavernosa	450	670	220	0.33	334	625	yellow

Results



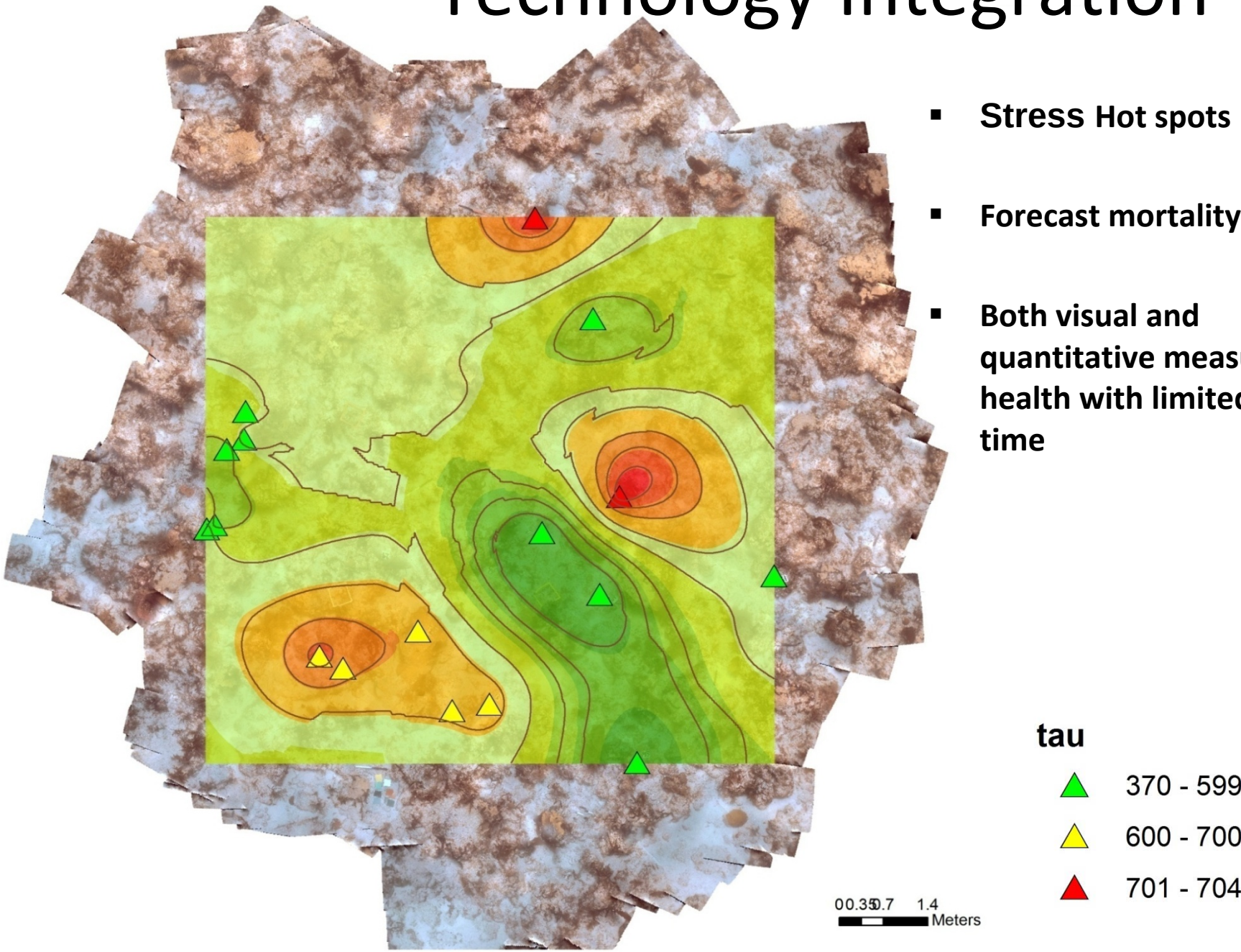






Fo	Fm	Fv	Fv/Fm	Sigma	Tau1	Green/Yellow /Red flag
39	55	15	0.28	126	704	red

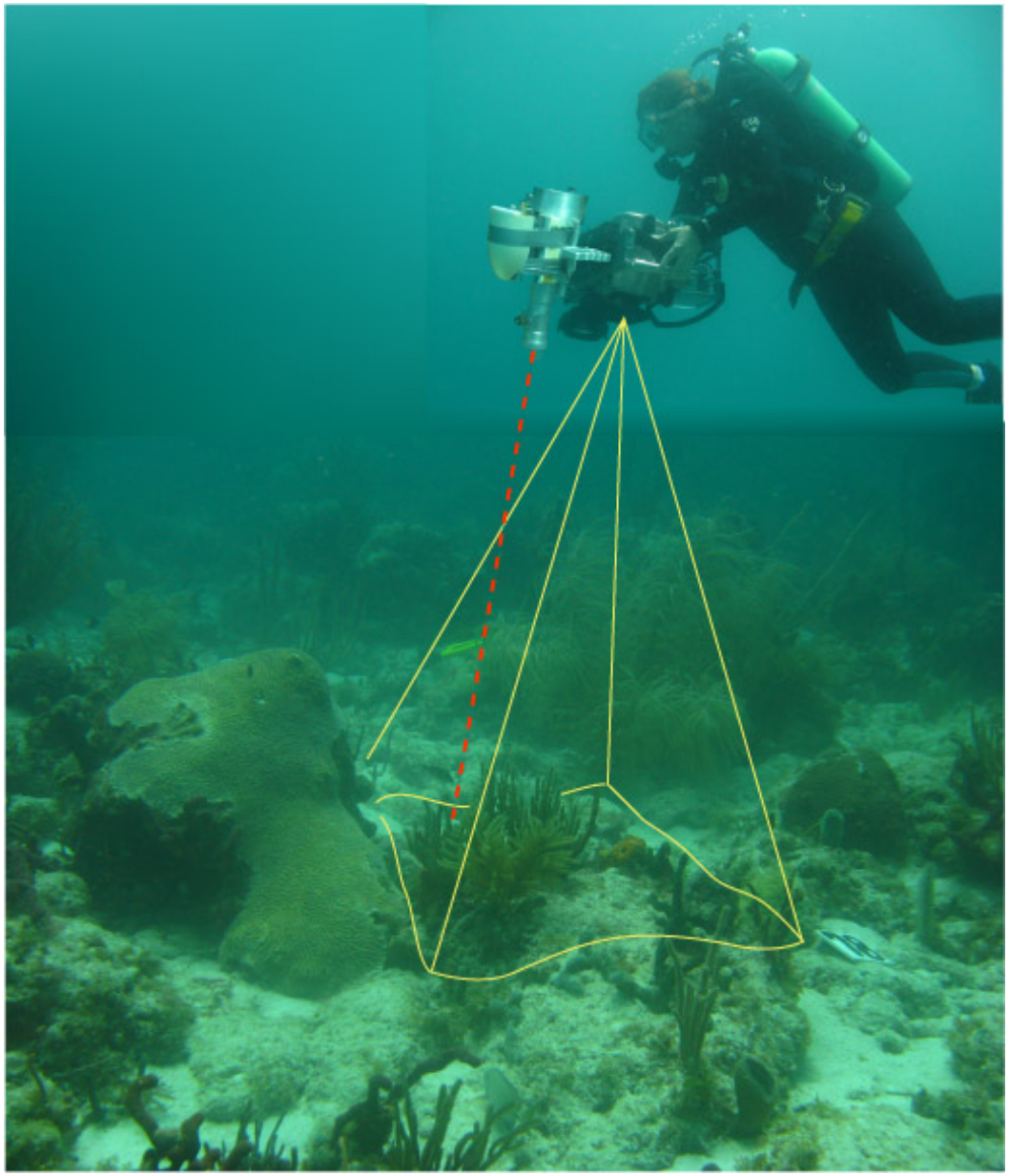
Technology Integration



- **Stress Hot spots**
- **Forecast mortality?**
- **Both visual and quantitative measures of health with limited field time**

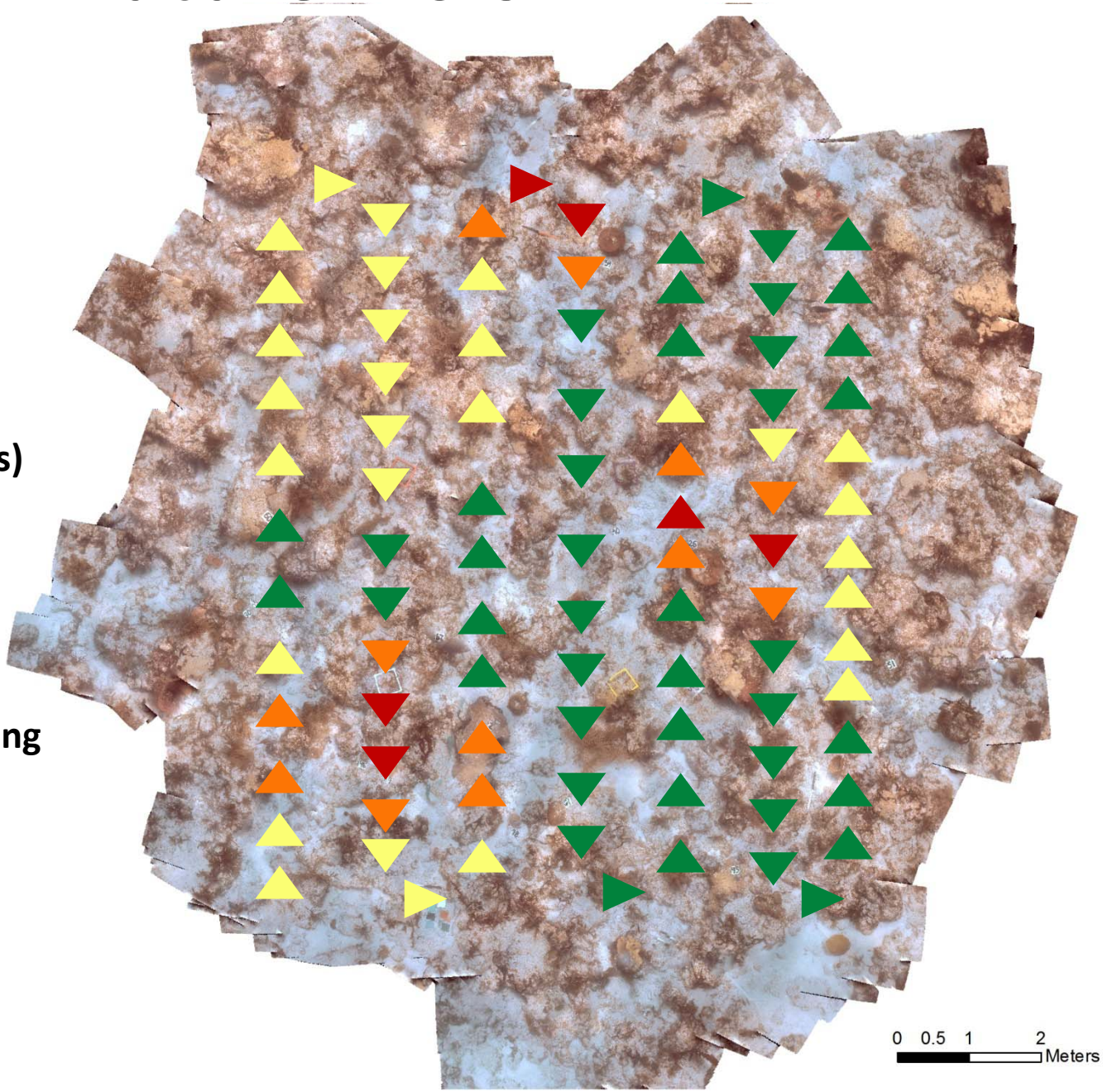
Future Vision

- Integrated platform
- New Fire instrument
- Distance measurement capability
- Simultaneous or near simultaneous sampling



Future Vision

- **Integrated sampling (spatially referenced measurements)**
- **3 scales of reef health information**
  - **Landscape (mosaics)**
  - **Colony (High res stills)**
  - **Microscale (FiRe)**
- **Future of coral monitoring (status, trends, and forecast capabilities)**



SERDP
Strategic Environmental Research and Development Program

RUTGERS
MARINE & COASTAL SCIENCES
Environmental Biophysics & Molecular Ecology Program

Development and Applications of Variable Fluorescence Technique for Monitoring and Assessing Coral Reef Communities

Analysis of Biophysical, Optical, and Genetic Diversity of DoD Coral Reef Communities using Advanced Fluorescence and Molecular Biology Techniques (SERDP SI-1334)

Drs. Maxim Gorbunov & Paul Falkowski
Institute of Marine and Coastal Sciences
Rutgers University, New Jersey

November 19, 2008

Background

- SON # CSSON-03-02 "Assessment of Benthic Communities for the Department of Defense"
- Executive Order 13089 "Protection of Coral Reefs" directs Federal agencies including DoD to study, restore, and conserve U.S. coral reefs.
- ~ 60% of the world's reefs are at risk from human activity.
- Most of U.S. reefs are threatened by habitat destruction, including 90% of reefs in Florida, 98% in PR and U.S. Virgin Islands, and 40% in Hawaii (World Resources Institute).

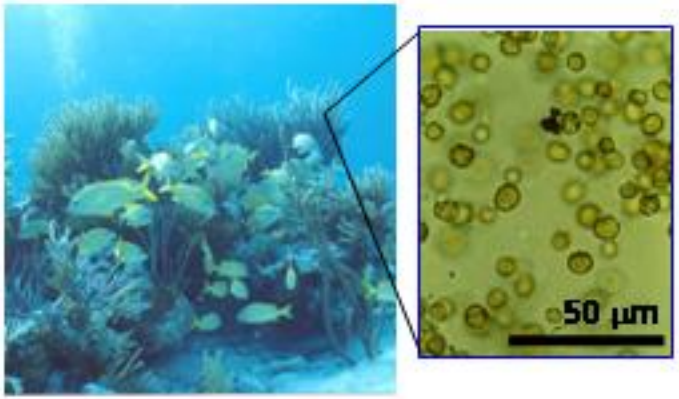
The development and implementation of advanced environmental monitoring programs requires

- an understanding of how different environmental factors affect the key elements of the ecosystems and
- the selection and validation of specific monitoring protocols that are most appropriate for the identification and quantification of environmental stressors.

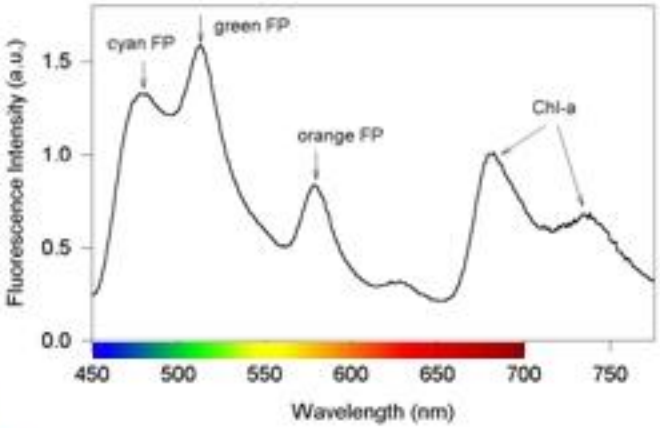
Technical Objectives

- Develop bio-optical techniques for non-destructive assessment of the health of coral reef communities with the capabilities of selective identification of natural and anthropogenic stressors.
- Develop prototypes of Fluorescence Induction and Relaxation (FIRe) Sensors for underwater monitoring stations and Remote Operated Vehicles.
- Collect a library of baseline data on physiological, biophysical, optical, and genetic diversity of coral reef communities.

Technical Approach



Fluorescence Emission Spectrum of Coral (under UV excitation)

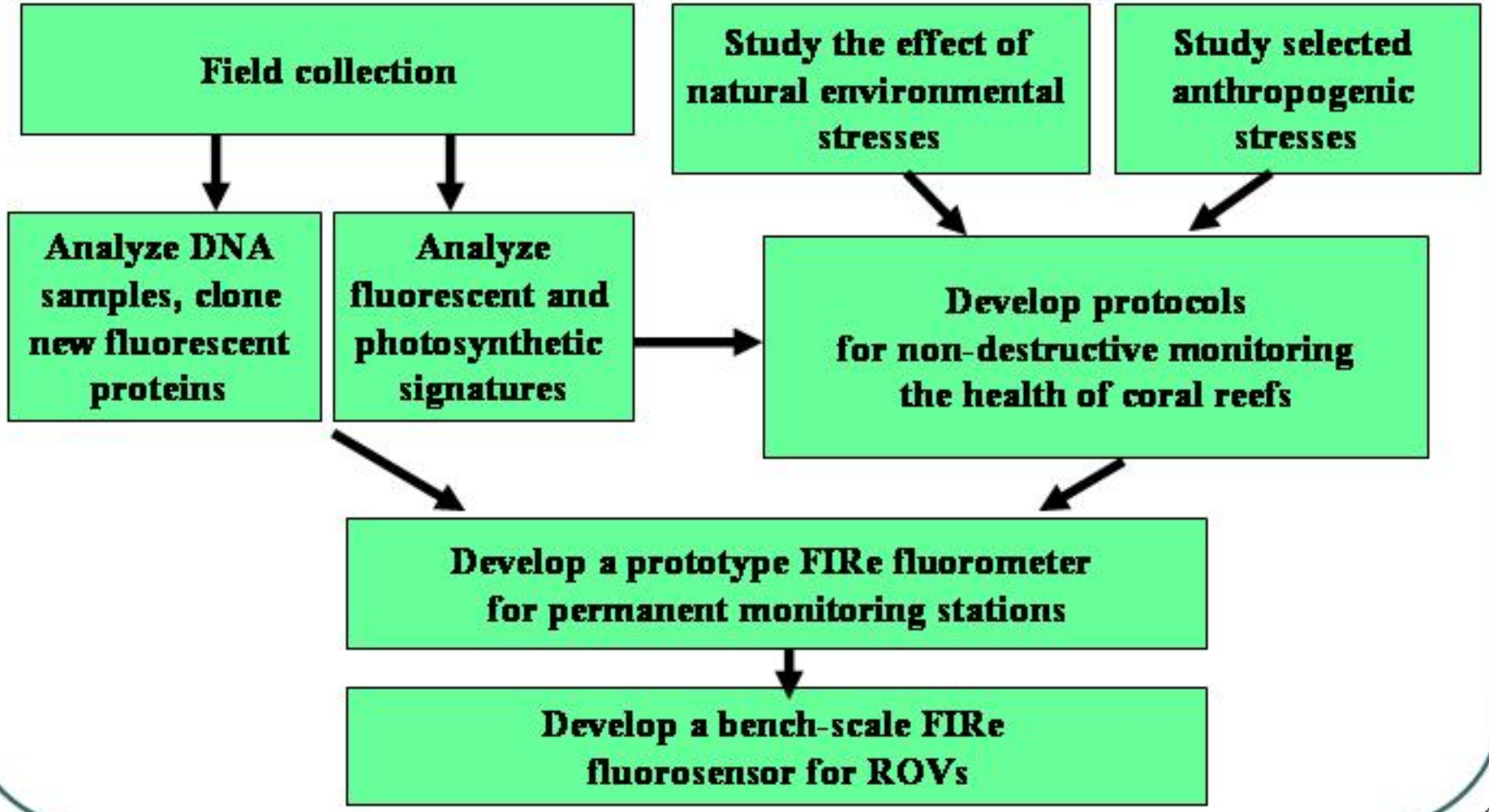


- Because photosynthesis is the ultimate source of energy for all shallow water communities, photosynthetic organisms are absolutely critical components in coral reef ecosystems.
- Corals are symbiotic associations between an invertebrate host and a photosynthetic alga (zooxanthellae).
- The chlorophyll-a fluorescence yield depends on the photosynthetic efficiency and is a sensitive indicator of the physiological state (=> "variable fluorescence" technique)

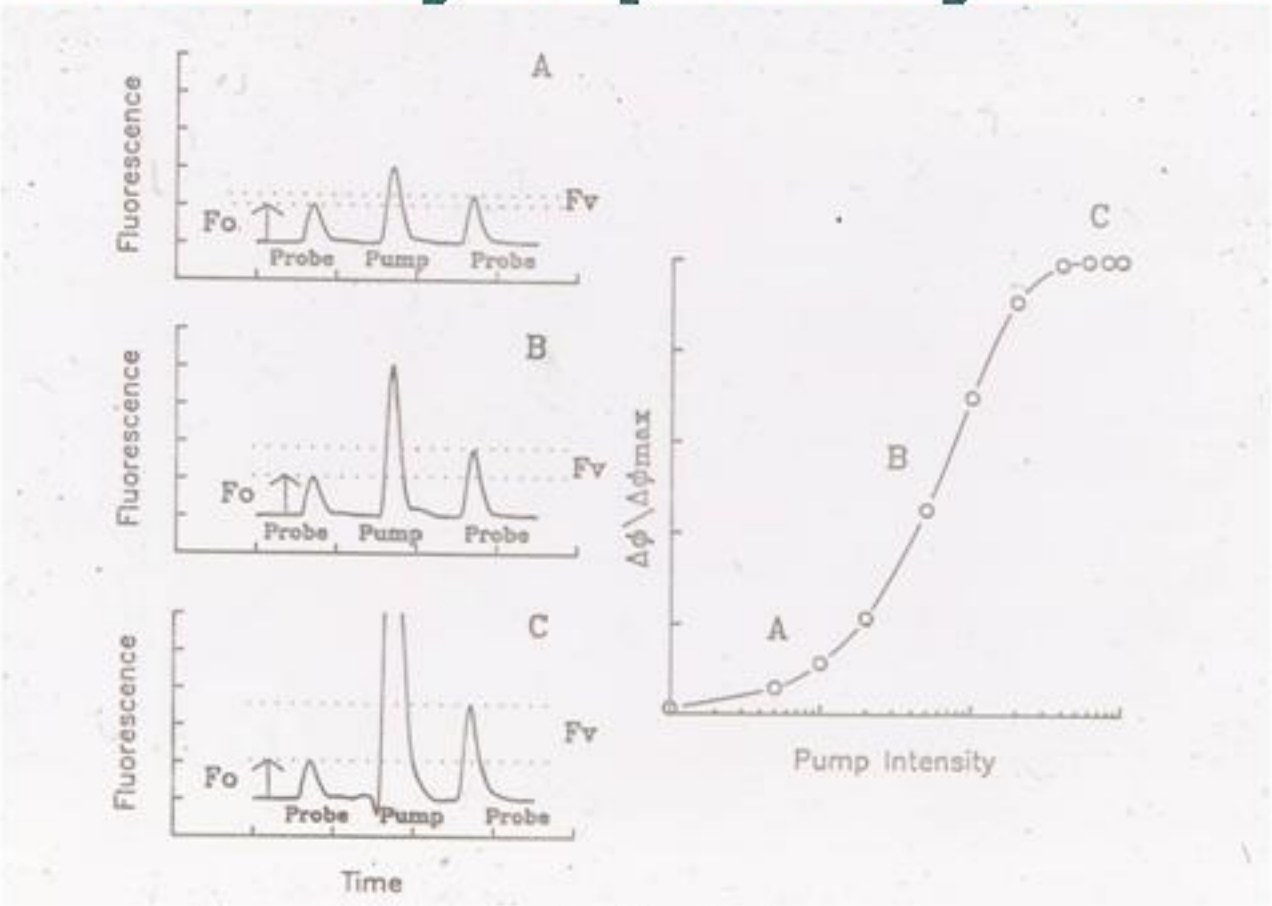
Methods

- Develop and use variable fluorescence technique (FRR and FIRe fluorometry) for non-invasive assessment of the physiological state of coral and of the impact of environmental stresses.
- Complement the FIRe fluorometry with biochemical and genetic laboratory techniques:
  - lipid analysis, optical and electron microscopy,
  - caspase activity (an indicator of Programmed Cell Death),
  - chromatorgaphy and mass spectrometry,
  - DNA sequencing and phylogenetic analysis,
  - spectrofluorometry and fluorescent microscopy.
- Understand the molecular mechanisms of the diverse color pallete of coral by sequencing, cloning, and characterization of color determinants in coral (GFP-like proteins).

Technical Approach

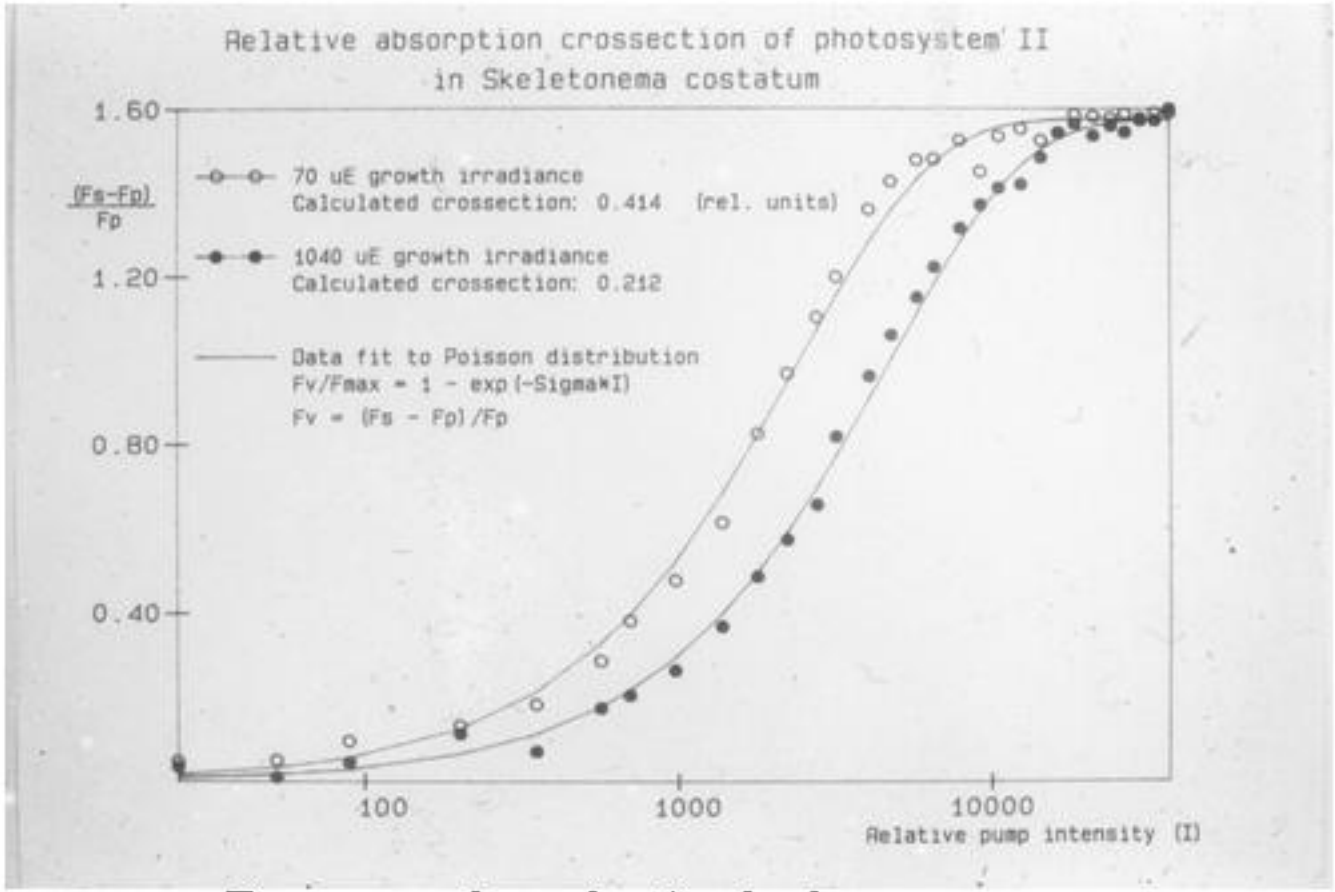


Measurements of the quantum efficiency of photosystem II



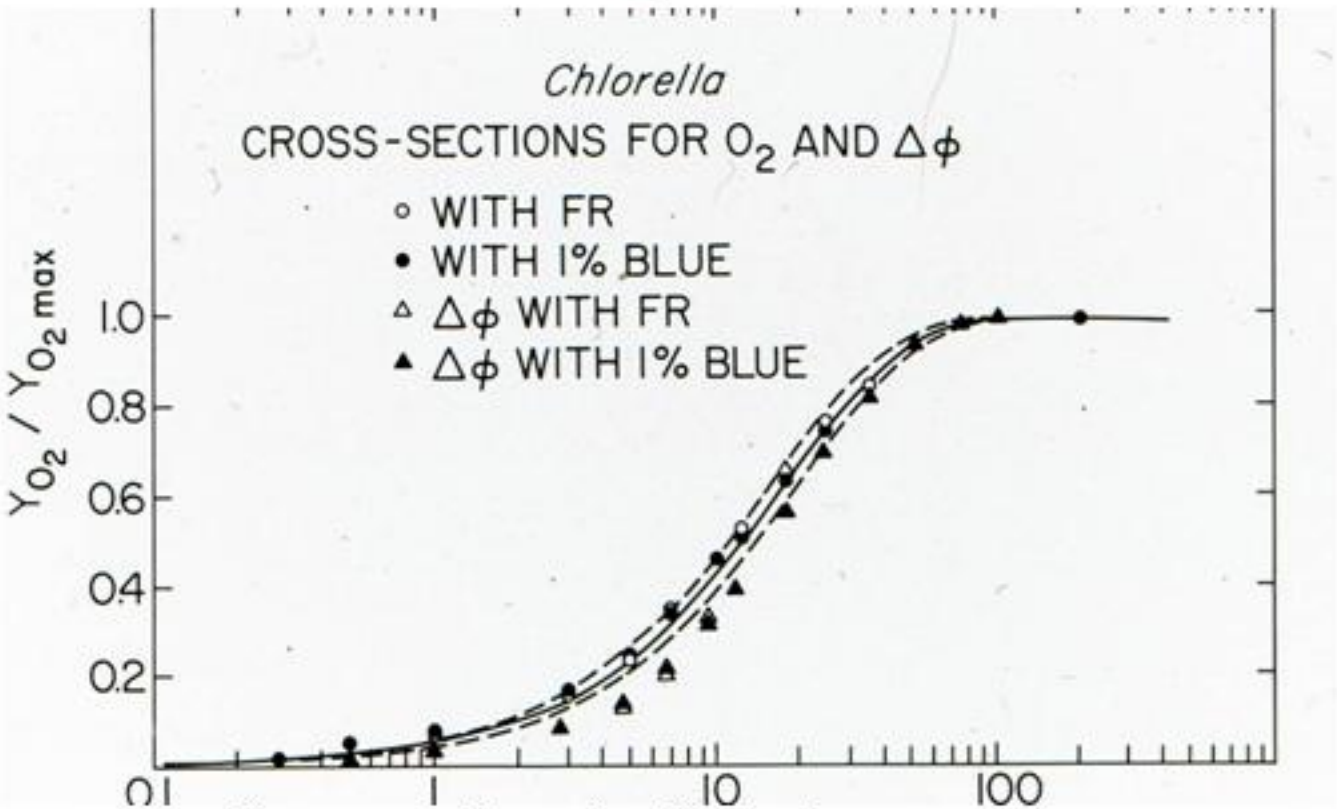
Pump-and-probe technique

Measurements of the effective absorption cross section of PSII



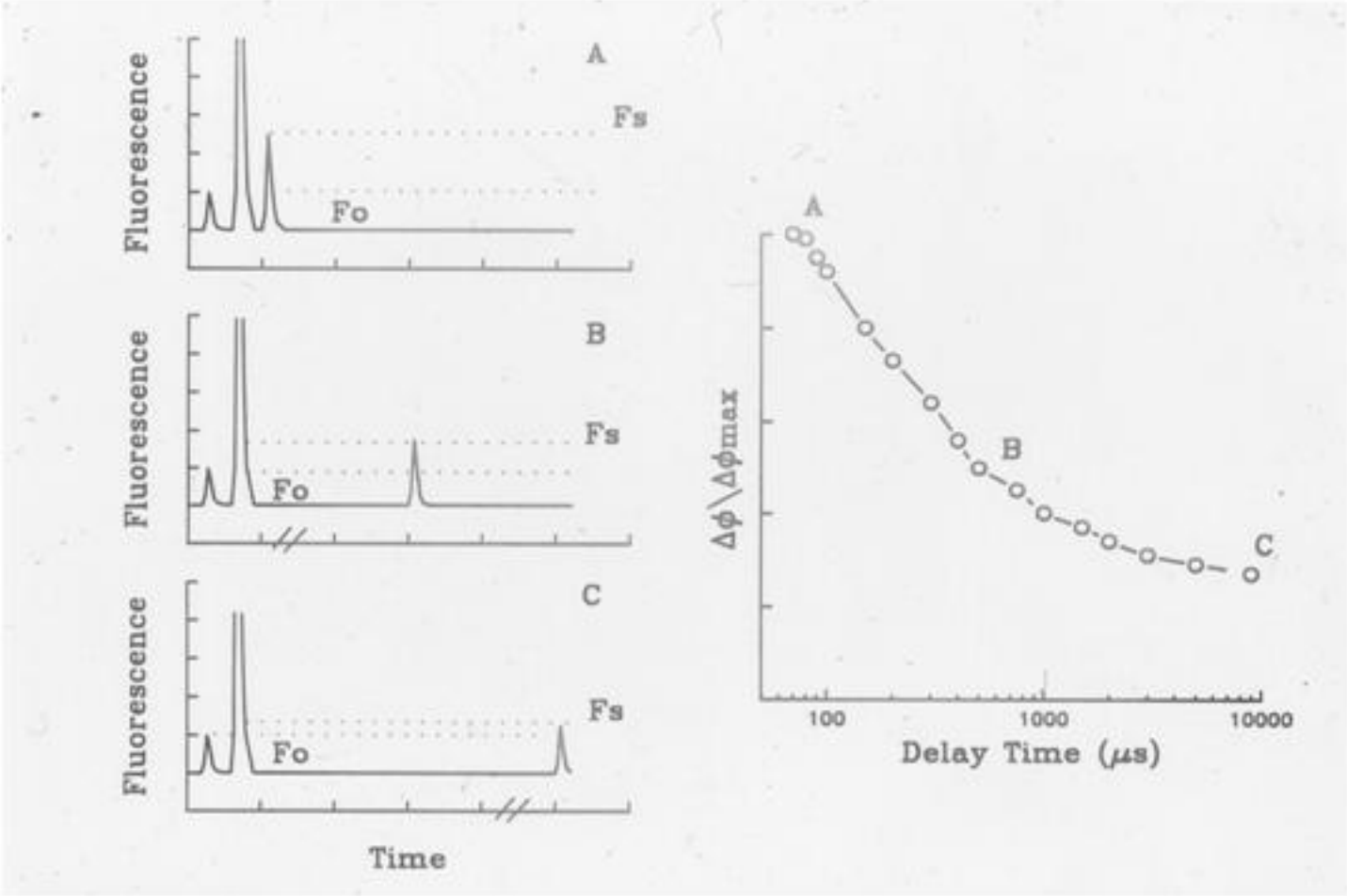
Pump-and-probe technique

The cross sections for O2 evolution and fluorescence are basically indistinguishable



Pump-and-probe technique

Measurements of turnover times



Pump-and-probe technique

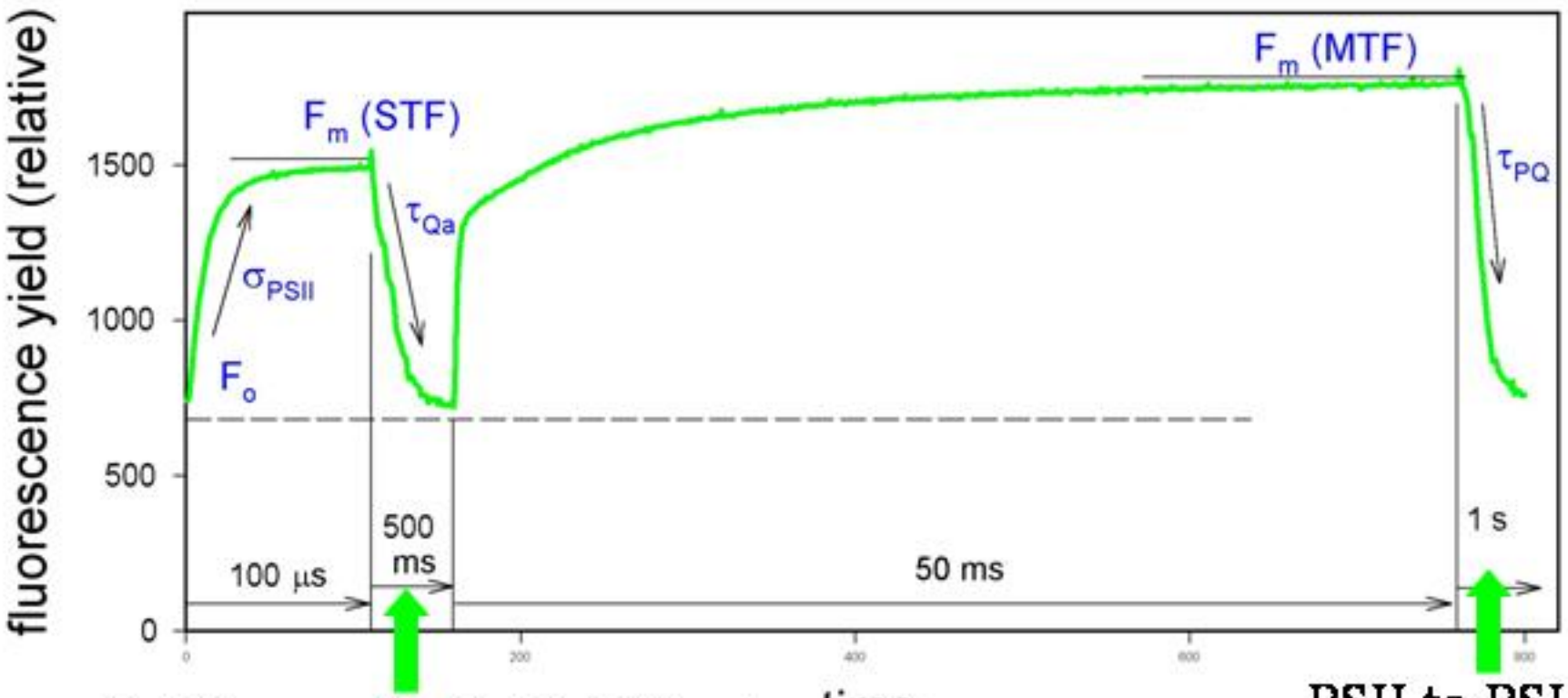
Technical Approach

- The primary stress indicator is Fv/Fm (the quantum yield of photochemistry in PSII).
- Under optimal conditions Fv/Fm is maximum (= 0.50 in coral and 0.65 is algae and plants).
- Most stresses leads to decrease in Fv/Fm.
- How can we find what stress is involved?
- Fv/Fm along in not enough.

Potential Solution:

- Additional parameters are needed for selective diagnostics of multiple stressors.

Fluorescence Induction and Relaxation (FIRe) technique



$Q_a^- Q_b \rightarrow Q_a Q_b^-$ (~200 us)

$Q_a^- Q_b^- \rightarrow Q_a Q_b^=$ (~800 us)

$Q_a^- __ \rightarrow Q_a^- Q_b \rightarrow Q_a Q_b^-$ (~2000 us)

PSII to PSI e^- transport (>5000 us)

Photosynthetic and Physiological Characteristics from Variable Fluorescence Measurements

FIRe parameter	*Description*
Fo, Fm	Minimum and maximum yields of chlorophyll-a fluorescence measured in a dark-adapted state
Fv	Variable fluorescence (= Fm – Fo)
Fv/Fm	Maximum quantum yield of photochemistry in PSII, measured in a dark-adapted state
Fo', F', Fm'	Minimum, steady-state, and maximum yields of chlorophyll-a fluorescence under ambient light,
σ_{PSII}	Functional absorption cross section of PSII in a dark-adapted state
σ_{PSII}'	Functional absorption cross section of PSII in a light-adapted state
ΔF'/Fm'	Quantum yield of photochemistry in PSII, measured under ambient light (=(Fm'-F')/Fm')
Fv'/Fm'	Quantum efficiency of photochemistry in open reaction centers of PSII, measured in a light-adapted state
p	"Connectivity factor", defining the exciton energy transfer between individual photosynthetic units
(Fm- Fm') /Fm	Quantum efficiency of non-photochemical quenching (i.e., thermal dissipation of excess energy)
τ_{Qa}	Time constant of photosynthetic electron transport on the acceptor side of PSII (Qa re-oxidation)
τ_{PQ}	Time constant of photosynthetic electron transport between PSII and PSI (re-oxidation of plastoquinone pool, PQ)

Instrument Development

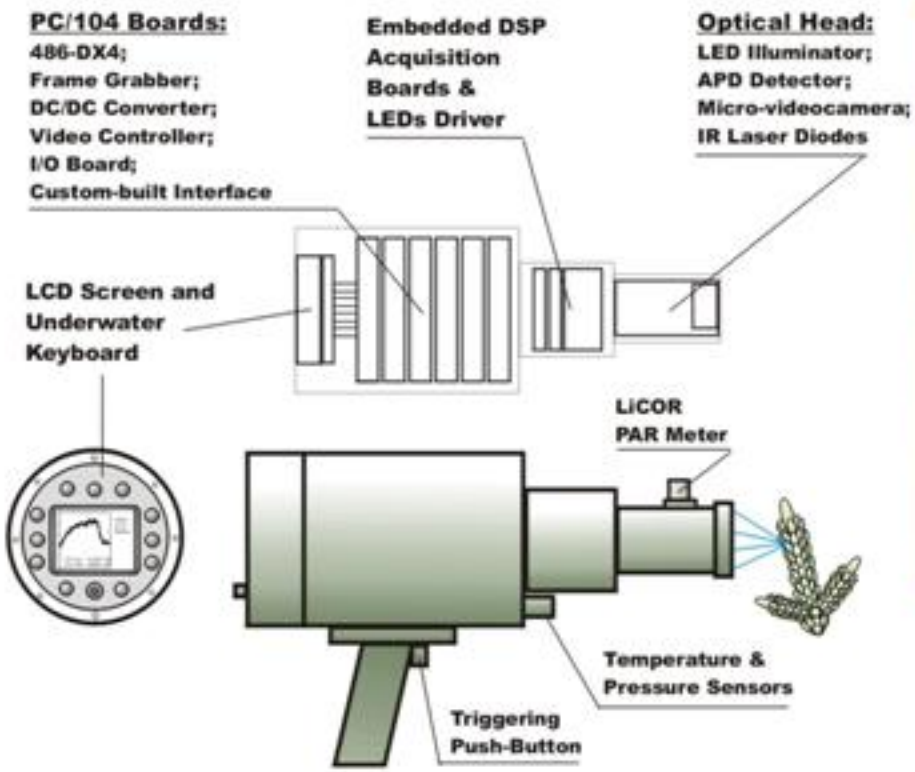
Developed FIRe System (Fluorescence Induction and Relaxation)

- FIRe technique records changes in the fluorescence yield induced by precisely controlled excitation light
- FIRe technique provides a comprehensive suite of photosynthetic characteristics
- the possibility of selective and non-invasive diagnostics of different environmental stresses
- *Basic Hypothesis*: Different stresses lead to specific modifications in the physiological state and can be diagnosed by using the FIRe technique

Developed Instrumentation

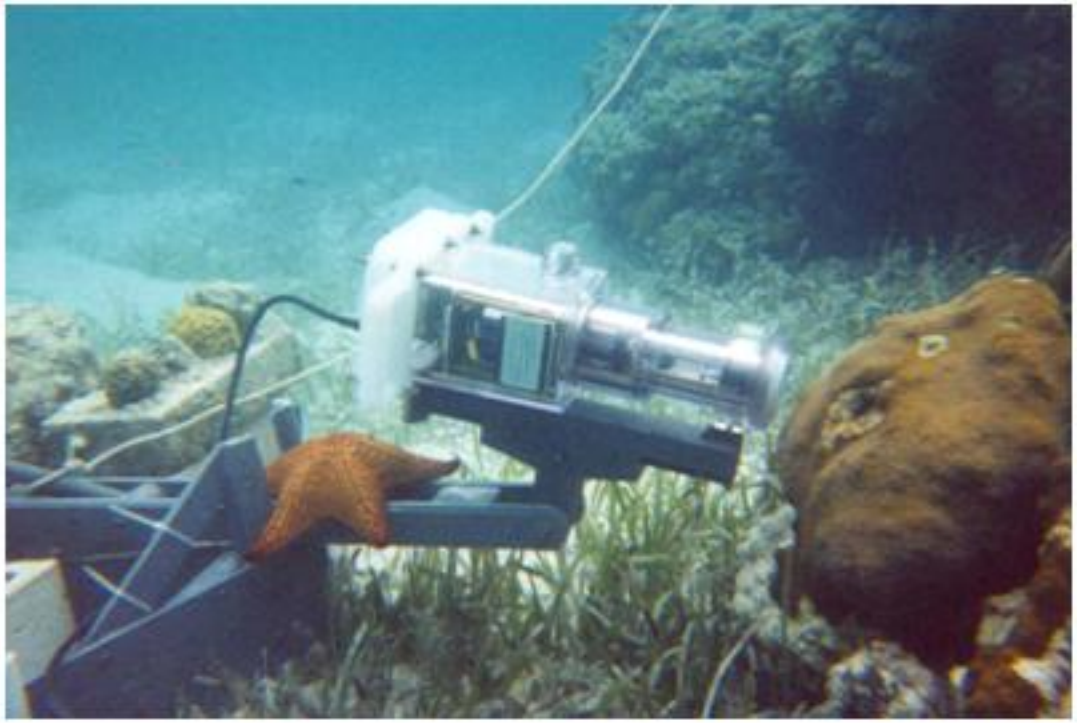
- Diver-operated fluorometer
- Moorable FRR fluorometer
- Bench-top FIRe System (with a fiber probe)

Diver-operated Fluorometer



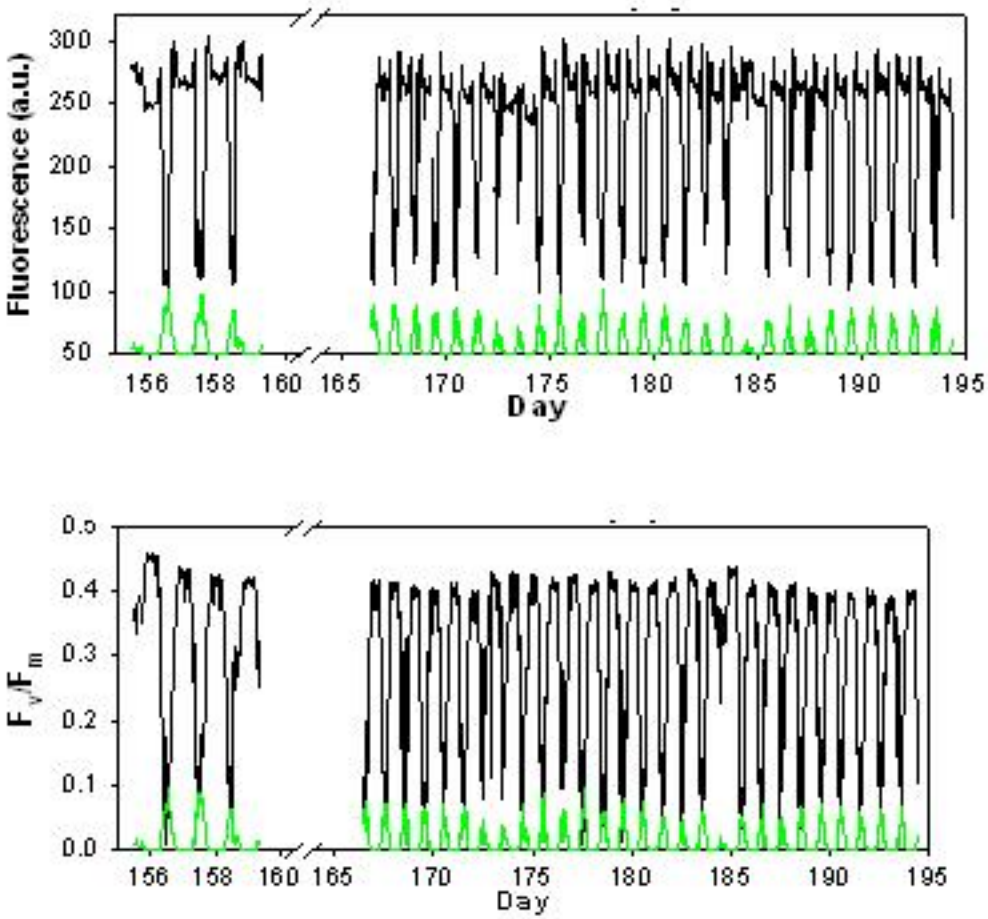
Moorable Fluorometer

- The moorable instrument has been developed to record *in situ* the temporal dynamics (diel to seasonal) in coral physiology



Long-term Monitoring

Temporal variability in fluorescence and photosynthetic efficiency ($\Delta F/F_m'$), measured in the coral *M.faveolata* in shallow waters (2 m depth).



Coral Cultivation Facilities at Rutgers

Thermal Stress and Coral Bleaching

- a major threat to coral ecosystems world-wide
- It is induced by a small (1 to 2°C) increase in water temperature
- may be superimposed to anthropogenic stresses related to military activity at DoD installations;
- the thermal stress leads to loss of symbiotic algae from coral and subsequent death
- the thermal sensitivity of corals varies strikingly between species and morphs

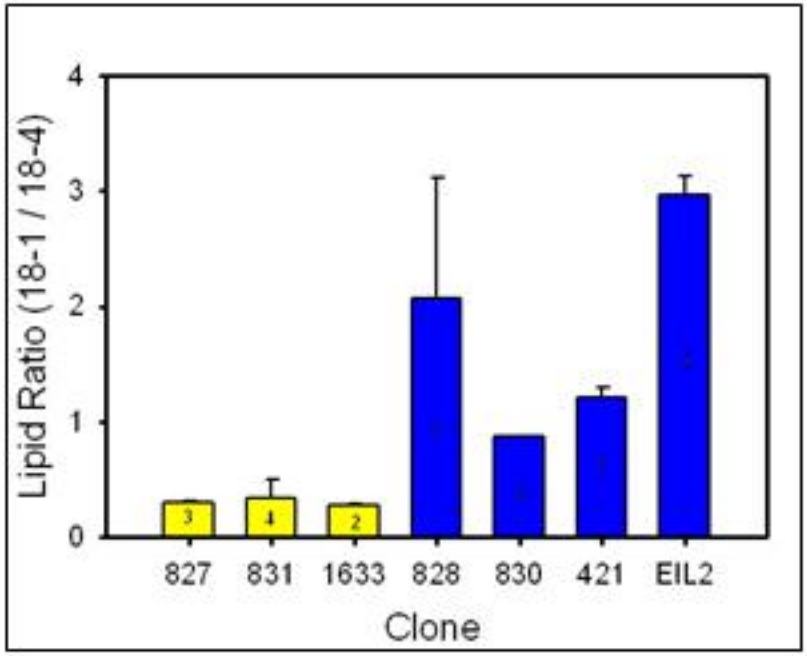
- T stress resembles photoinhibition and often considered as T-enhanced photoinhibition
- Our research revealed that T stress differs from photoinhibition and is controlled by different molecular mechanisms (Tchernov et al, 2004, PNAS).

Thermal Stress: Elucidated Mechanisms & Bio-Optical Signatures

Our research revealed that

- thermal sensitivity is controlled by the membrane lipid composition of symbiotic algae,
- thermal stress starts with disruption of thylakoid membranes and results in damage to the photosynthetic machinery (PSII);
- accumulation of Reactive Oxygen Species (ROS) produced by the stresses algae => coral death (via Programmed Cell Death);
- the stress development is accompanied by unique variable fluorescence signatures and can be readily diagnosed by the FIRe technique, even at early stages.

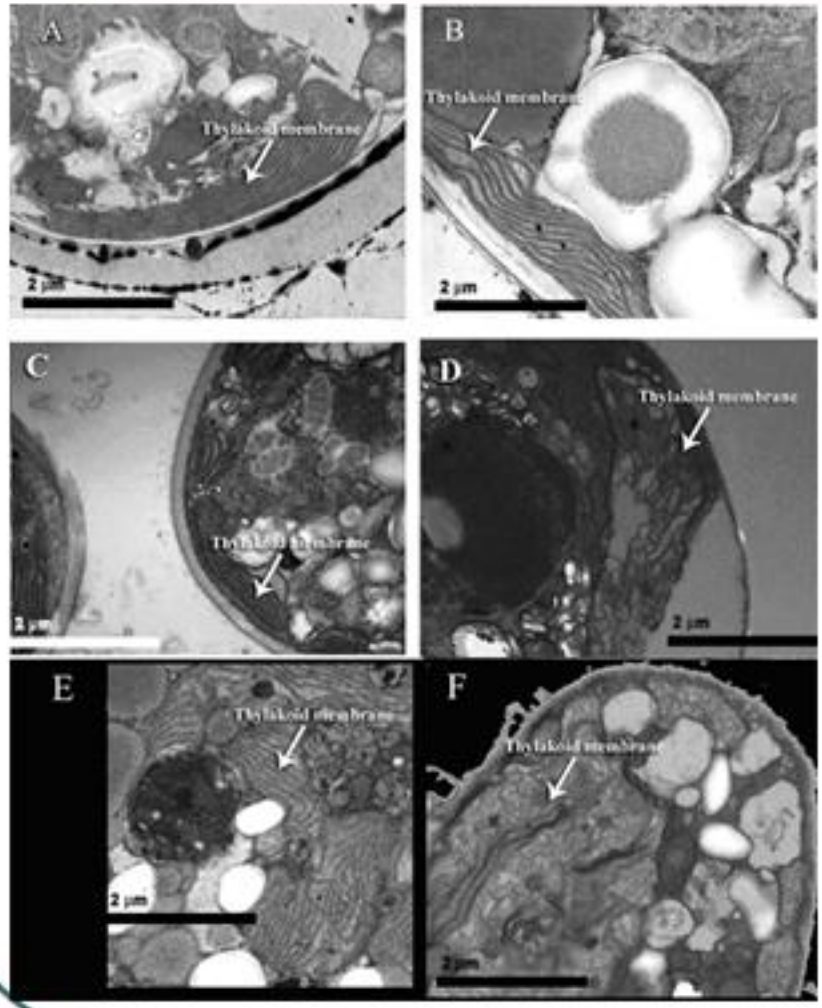
Lipid composition and Thermal Sensitivity of Coral



- The research revealed that thermally resilient algae contain different, more stable composition of lipids
- The thermal sensitivity is controlled by the saturation of the membrane lipids
- Differential sensitivity to thermal stress is related to the genes in the lipid biosynthesis pathways.

(Tchernov et al, 2004, PNAS)

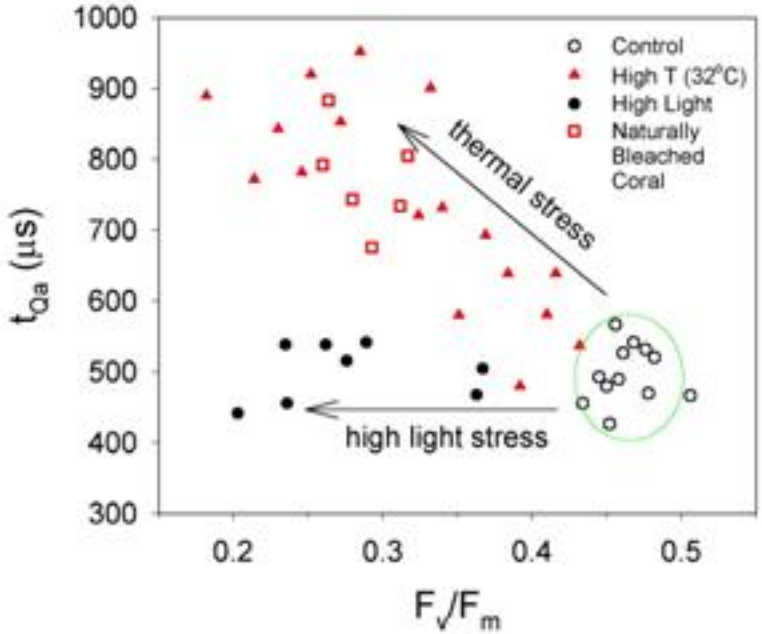
Thermal Stress via Electron Microscopy



- photosynthetic thylakoid membranes are disrupted under high T;
- photochemical energy transduction is compromised;

(Tchernov et al, 2004, PNAS)

FIRe Fluorescent Diagnostics of Thermal Stress



- a characteristic decrease in the quantum yield of photochemistry in PSII (Fv/Fv) under both T and High Light stresses
- but the thermal stress is accompanied by a striking increase in the time constant of Qa reoxidation (t_{Qa}).
- FIRe technique distinguishes between the two common stresses

Impact of Heavy Metal Contamination on Coral

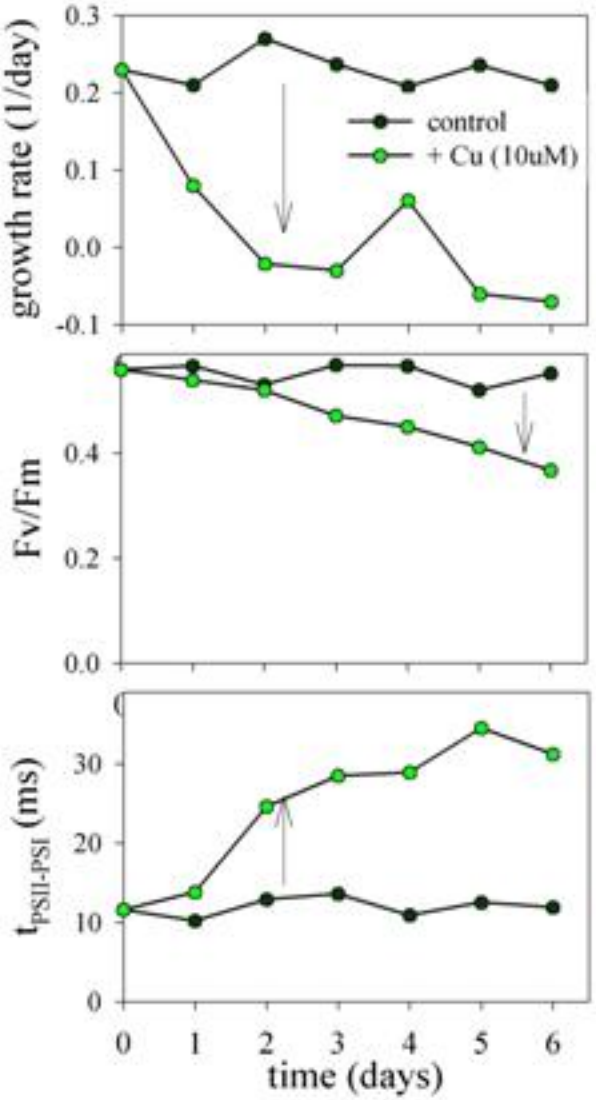
Problem Statement:

- Copper is one of the most toxic metals;
- Copper and other heavy metals are used in anti-fouling paints and are released from ships, sewage, and industrial waste.

Objectives:

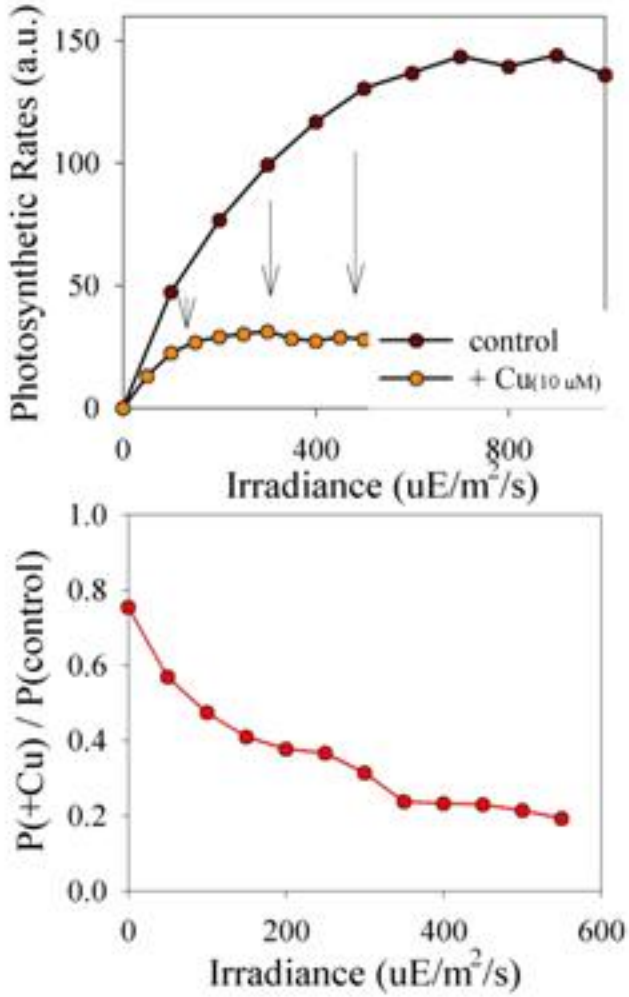
- To develop a method of recording the physiological response of corals to metal contamination;
- To elucidate physiological mechanisms of stress development;
- To find an early marker for metal contamination impact.

Impact of Heavy Metal Stress on Coral Zooxanthellae



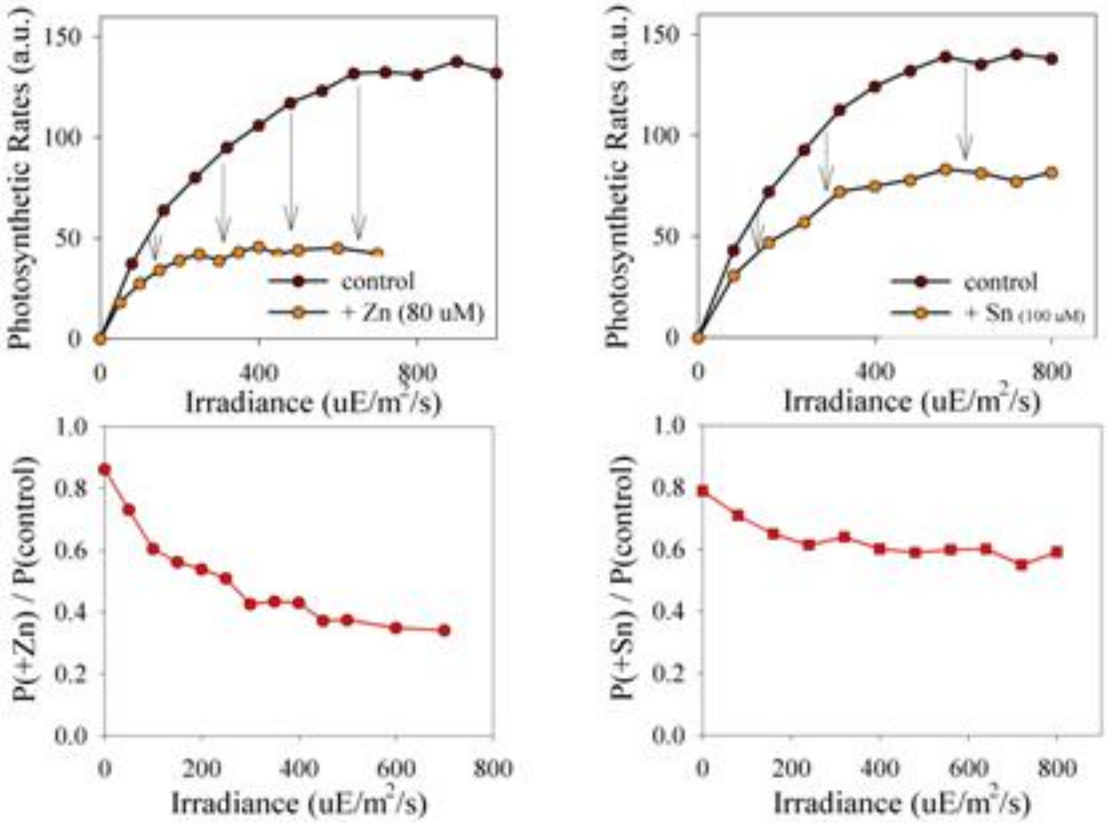
- Copper poisoning impairs the growth rates, whereas the efficiency of primary photosynthetic reactions (Fv/Fm) remains high;
- The stress development is accompanied by a characteristic reduction in the rates of electron transport between PS II and PS I (PQ pool re-oxidation).
- The data suggest that the secondary photosynthetic reactions are the primary target of the metal stress.

Impact of Heavy Metal Stress on Coral Zooxanthellae



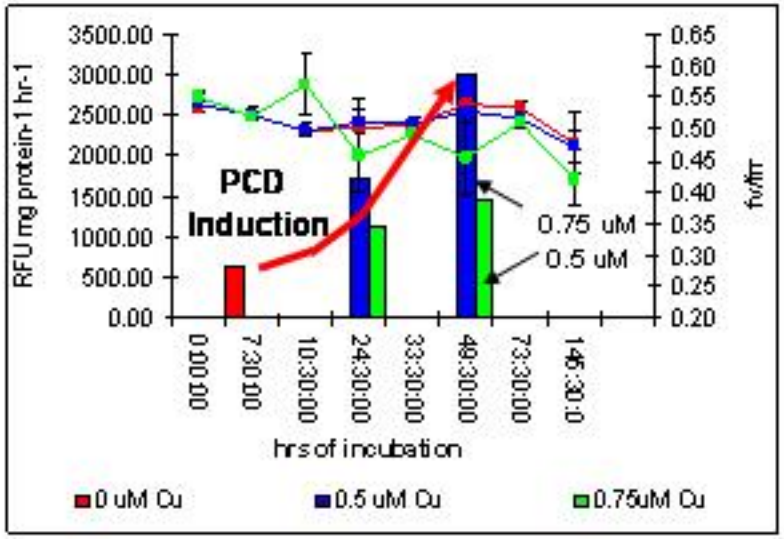
- The potential photosynthetic efficiency (at low light) remains high, while the maximum photosynthetic rates (at high light) are dramatically reduced.
- This pattern differ strikingly from the signatures of common natural stressors (elevated temperature and photoinhibition) that primarily impact the primary photosynthetic reactions.

Impact of Zn and Sn on Coral Zooxanthellae

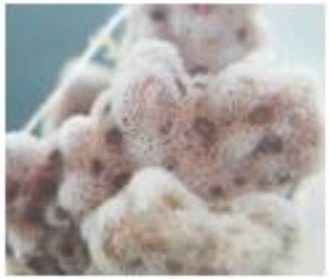


- **Zn effect is similar to that of Cu**
- **The impact of Sn is different; Sn appears to affect both primary and secondary photosynthetic reactions**

Impact of Heavy Metal Stress on Coral



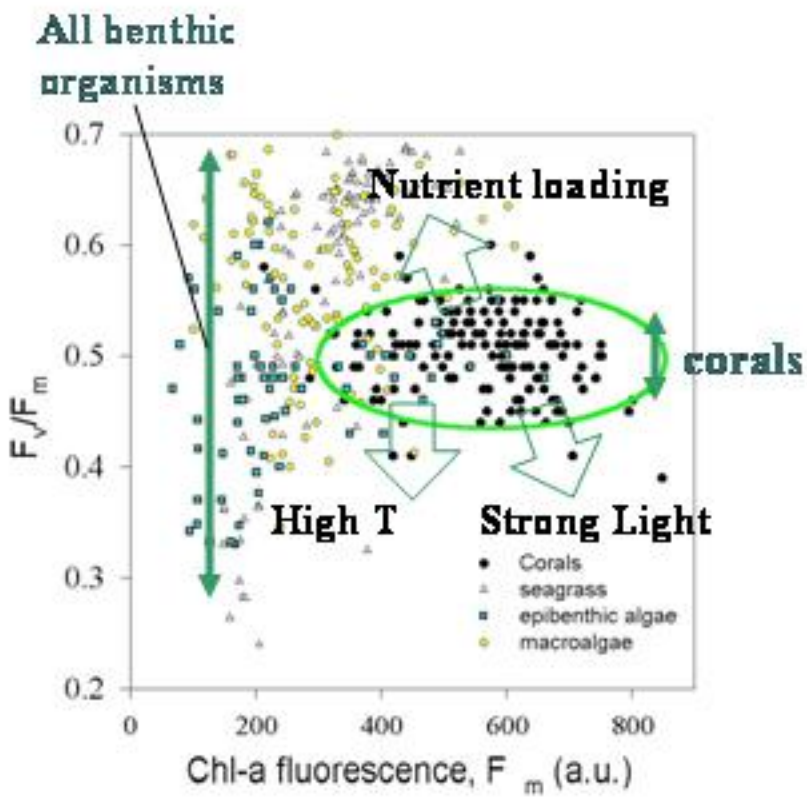
Control



0.75uM Cu, 24 hrs

- Copper induces Program Cell Depth (PCD) of the animal host, as evident from elevated caspase activity and tissue degradation;
- Thereby, the photosynthetic activity of algal symbionts (zooxanthellae) remain high.
- The signatures of copper stress differ strikingly from the signatures of common natural stresses (elevated temperature and photoinhibition).

Variability in photosynthetic efficiency in corals and the impact of stresses

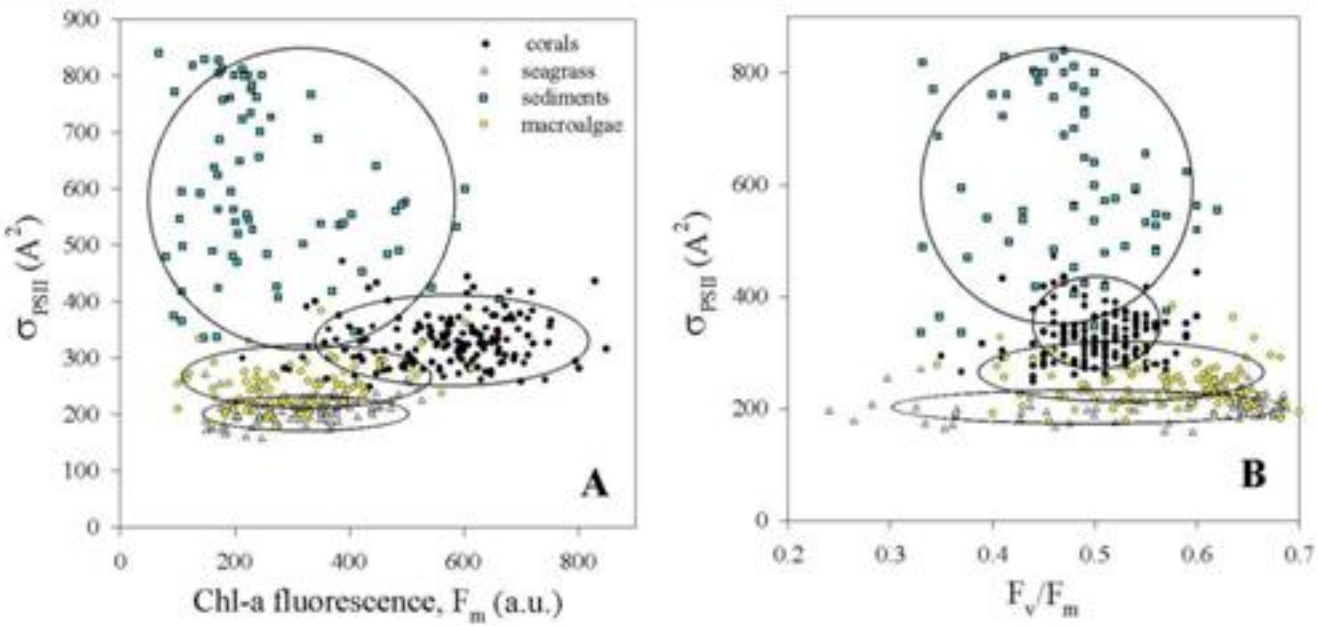


- A very narrow range of variability of Fv/Fm in healthy corals (Fv/Fm ~ 0.5).
- Stress leads to deviation of Fv/Fm from its normal value

Identification of stresses - Decision making algorithms

- **Thermal stress:**
  - Algal symbionts are the primary target
  - decrease in Fv/Fm
  - decrease in the electron transport rate in PSII (t_{Qa})
  - irreversible (> months).
- **High light stress (photoinhibition):**
  - decrease in Fv/Fm
  - no change in the electron transport rate in PSII (t_{Qa})
  - recovers within ~ hours (dynamic photoinhibition) or ~ a few days (chronic photoinhibition)
- **Nutrient (primarily nitrogen) load:**
  - Increase in Fv/Fm and pigment density
- **Heavy metal poisoning:**
  - Starts with degradation of coral tissue (Cnidarian host is the primary target);
  - Increase in caspase activity (Programmed Cell Death pathway)
  - No change in Fv/Fm at early stages; Secondary photosynthetic reactions are the first target in zooxanthellae.

Identification of Benthic Organisms using the FIRe Signatures

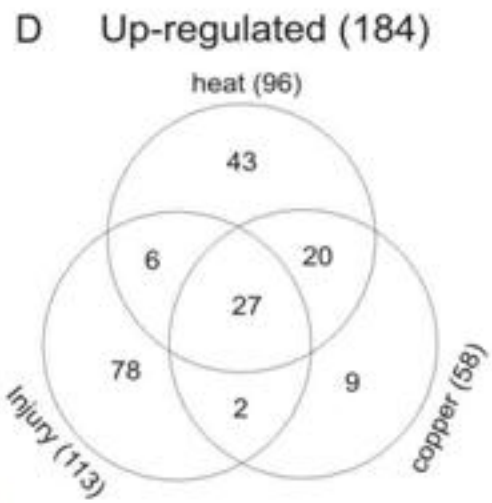


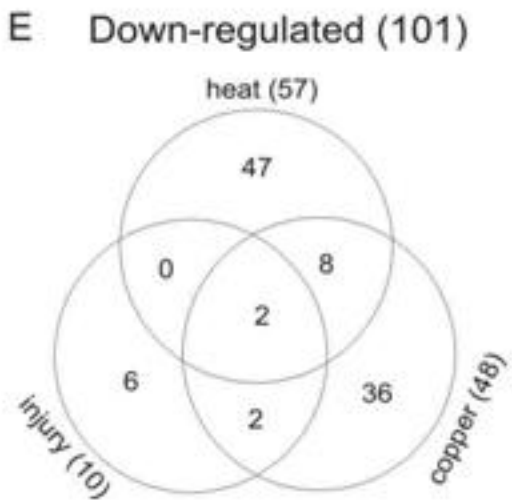
- Fluorescence yields are highly variable within each group of benthic organisms and overlaps between the groups.
- However, FIRe-derived photosynthetic parameters are constrained and specific for each group.
- Different benthic organisms exhibit unique sets of FIRe signatures.
- Implications: possibility to identify functional groups of organisms by FIRe fluorescence mapping and to assess the coral coverage from AUV platforms.

Functional Genomics of Coral Stress (Hunting for Stress-Specific Genes)

Objective: develop a laboratory-based technique for classification of stresses in coral.

- Three types of stress (elevated temperature, copper poisoning, and mechanical injury) have been evaluated.
- Array of 3460 clones from the substracted libraries has been fabricated.
- 285 differentially expressed clones were found.





Analysis revealed that

- there are responding genes common to all stresses.
- there are also genes specific to a certain stress.

Functional genomic analysis of coral stress response

Gene	N	Heat	Cu	Infl.
Stress / Immunity				
heat shock protein 16.1	5	4	6	
complement component C3	3	2	-1.6	-1.5
Oxidative phosphorylation				
Cytochrome oxidase (Symbiodinium)	1	22	20	5
NADH dehydrogenase sub 5 (coral)	2			6
ubiquinone-binding protein	1	4		
Cell shape / motility				
collagen	1	3	3	
Rho	1	8	9	2
Rho kinase	1	1.2		
actin	27		-4	-5
tubulin	4	-3		
Signal transduction				
Ser/Thr protein kinase	1			6
nuclease/phosphatase family	2	3	5	
MAD	1		2	
protein tyrosine kinase	1	1.2	1.2	
SH2 domain	1		-4	
14-3-3 protein	1		-3	
General metabolism				
transferrin-containing protein	1			5
solute carrier protein	1			7
D-aspartate oxidase	1	4	5	
glutaredoxin	1	2		
ferritin	1			2
Arginine kinase	1	-4	-3	-2
voltage-dependent anion channel	2	-3	-4	-3

Gene	N	Heat	Cu	Infl.
Protein synthesis and degradation				
Ubiquitin-like protein	1	3	5	
ubiquitin specific protease 34	1	-4	-1.5	
ribosomal protein L8e	2	-5	-8	-5
ribosomal protein S12	3	-3	-5	-2
cathepsin	2	-2.7	-2.5	
Mucus / glycoprotein metabolism				
beta-1,3-glucanase	3	2		3
ribophorin	1		-3	
Neurogenesis				
synaptotagmin	1	3		
spondin	1	4		
distalless	1	-1.3		
Skeleton deposition				
galaxin	1	4		2
carbonic anhydrase	2	-6	-14	
Reproduction				
Zona Pellucida (ZP) protein	1			3
vitellogenin	4	-2	-5	
Development / transcription regulation				
LIM-domain protein	1	2	2	
CCAAT/Enhancer binding protein	7	-3	-3	
sine oculus homeobox protein	1	-1.5		
Unidentified function				
unknown	49	32	33	3
yippee protein	1			4
hypothetical protein	4	-6	-6	-2
similar to androgen-induced	1		-4	

Conclusions and Summary

- Completed lab & field studies of the impact of common natural stresses on coral.
- Built Coral Cultivation Facilities at Rutgers to conduct controlled manipulation experiments in the lab.
- Established the molecular and cellular mechanisms and optical signatures of thermal stress in coral.
- Collected the database of baseline optical properties of corals in two geographic provinces.
- Completed sequencing, cloning, and characterization of 50 new GFPs from corals

Conclusions and Summary

- Developed the new FIRe Fluorometer System to measure photosynthetic parameters in corals and other photosynthetic organisms
- The bench-top version of FIRe System has been transferred to a small high-tech company.
- Designed the multi-receiver FIRe Fluorometer System for permanent monitoring stations and a prototype FIRe sensor for AUVs
- Identified the signatures of copper stress to coral.
- Developed and evaluated new bio-optical algorithms for detection and assessment of the natural and selected anthropogenic stresses.

Technology Transfer

- We envision that our technology will be integrated in a long-term multi-platform monitoring program.
- **Multiscale** approach:
  - *Macroscale*: Remote sensing and hyperspectral imaging
  - *Mesoscale*: ROV-type FIRe sensors; diver swum FIRe System; Video plots
  - *Microscale*: Detailed monitoring on selected sites and targets (diver swum and moored FIRe systems; bio-optics, and genetics) with high temporal resolution.

Technology Transfer

- Technology transfer of the bench-top FIRe System to a small hi-tech company, Satlantic Inc. has been completed (www.satlantic.com/fire).
- Diver swum FIRe and moored FIRe Systems will be transferred to the NAVY through ESTCP project (collaboration with SPAWAR, Space and Naval Warfare Systems Center, San Diego).
- Development of a compact low-power FIRe sensor for Glider AUV is in progress (project funded by National Oceanographic Partnership Program).
- We also envision that the new FIRe sensor will be modified for inclusion into a compact underwater sensor for vertical profiling of the water column.

Publications

Peer-Reviewed Publications:

1. Tchernov D, Gorbunov MY, de Vargas C, Yadav SN, Milligan AJ, Haggblom M, Falkowski PG. (2004) Membrane lipids of symbiotic algae are diagnostic of sensitivity to thermal bleaching in corals. – Proc. Nat. Acad. Sci., U.S.A., 101 (37): 13531-13535.
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4. Field, S. F., Bulina, M. Y., Kelmanson, I. V., Bielawski J. P. and Matz, M. V. (2006) Adaptive evolution of multi-colored fluorescent proteins in reef-building corals. - J. Mol. Evol., 62 (3): 332-U15,.
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7. Bibby T, Gorbunov MY, Wyman K, and Falkowski PG - Photosynthetic community responses to upwelling mesoscale eddies in the subtropical north Atlantic and Pacific Oceans. – Deep Sea Research II, in press.

Conference Presentations:

- SERDP/ESTCP "Partners in Environmental Technology Technical Symposium & Workshop" 2003, 2005;
- 13th International Congress of Photosynthesis, Montreal, Aug.29-Sept. 3, 2004
- 2004 ASLO Aquatic Science Conference, Honolulu, February 2004
- 2005 Ocean Science Conference, Salt Lake City, February 2005
- 2006 AGU/ALSO/TOS Aquatic Science Conference, Honolulu, February 2006
- AquaFluo'2007 - Chlorophyll Fluorescence in Aquatic Sciences Conference, Nove Hrady, Czech Republic, May 2007
- Bio-Sensing in Ocean Observation, Sarasota, Fl, June 2007

Appendix D- Technology Integration Matrix

Table 2. Summary of information provided by presenters at the SERDP Coral Monitoring Workshop held in Miami, Florida on November 18-19, 2008. The different colors appearing in the "Metrics and Indicators" column refer to the potential overlay of the SERDP-funded technologies onto existing coral reef monitoring programs based on the descriptions of monitoring needs described by the agency representatives. Coral monitoring programs that could potentially benefit from the application of video mosiacs appear in green. Programs that could benefit from the use of the Fluorescence Induction and Relaxation (FIRe) Sensors appear in yellow. Programs that could benefit from the joint use of both SERDP-funded technologies appear in purple.

AGENCY	Mandate/Mission	Location and Habitats	Depth Range	Temporal Frequency	Methods	Coral Metrics / Indicators	Gaps / Limitations	Future Directions	Notes
Minerals Management Service (Sinclair)	1) Safe and environmentally sound exploration and extraction of offshore resources (oil, gas, sulfur, sand) within federal waters, 2) Protection and Monitoring of Benthic Communities in the Gulf of Mexico, 3) Chracterization of areas prior to exploration and extraction	Gulf of Mexico. Live-bottoms, Coral Reefs, Seagrass beds, Sand, Hardbottom	Shallow to Deep (> 100 m)	Not Reported	1) Random video transects, 2) Permanent photo quadrats, 3) Coral growth studies using photographs, 4) Fish, urchin, lobster visual surveys, 5) WQ monitoring	Coral cover, coral diversity, coral growth	Not Reported	1) Completion of detailed coral monitoring protocol, 2) Improved cross calibration testing of data, 3) Upgrade to High Definition videography, 4) Use of interferometric sonar and LIDAR for mapping, 5) Use of drop camera for deep water evaluation of habitat, 6) Expanded monitoring for marine fish communities, seagrass, lobster and conch, recruitment	
National Park Service (Patterson, Ruttenberg)	Monitor natural resources within National Parks	South Florida (Biscayne National Park, Everglades National Park, Dry Tortugas), USVI (St. John, St. Croix) Coral Reefs, Seagrass	Shallow (< 20 m)	Annual	Visual surveys, video and photographic surveys at permanent (sites of interest or index sites) and random sites	Coral cover, Disease and Bleaching prevalence	1) Detailed mapping, 2) Circulation models with larval transport, 3) Coral disease causation and infection research, 4) Ocean acidification, 5) Lionfish eradication research	Not Reported	
Fish & Wildlife Service (Wolfe)	Conserve, protect and enhance fish, wildlife, and plants and their habitats for the continuing benefit of the American people. FWS roles and responsibilities include law enforcement, fisheries management, wildlife conservation, species recovery, remediation of environmental contaminants, eradication of invasive species, and habitat restoration	National Wildlife Refuge system. 13 of these refuges contain coral reefs (e.g., Palmyra Island, Navassa, 2 in the Key West area). Refuges contain 3 million acres of coral reef ecosystems. Habitats protected: mangroves, wetlands, beaches, soft bottom, coral reefs	Not Reported	Annual to Biennial	Monitoring done by USGS and EPA (no inhouse monitoring). Towed video surveys, permanent sites surveyed using video and photographs, recruitment studies, WQ sampling	Coral cover, diversity, disease and bleaching prevalence, coral recruitment	1) Enforcement of fishing regulations and other illegal activities, 2) Additional partnerships, 3) Additional transects at remote refuges, 4) Improved monitoring techniques and technologies, 5) Improved understanding of invasive species	Not Reported	
Environmental Protection Agency (Fisher)	Use Clean Water Act legislation to protect and restore watersheds. Use biocriteria to protect biological condition.	Coral Reefs in Florida, USVI.	Shallow (< 20 m)	Annual to Biennial	Visual surveys, diver measurements	Colony size, disease and bleaching prevalence, percent partial mortality, Diversity, 2D and 3D Live coral cover, Population Structure	1) Only one assemblage (stony corals), 2) Unknown connection to stressor identification, 3) Unknown connection to services and values	Development of Biocriteria for coral reefs. Biocriteria are numeric values or narrative descriptions (thresholds) established to protect the biological condition of aquatic life inhabiting waters of a given designated use. Require defensible bioassessment procedures to establish and enforce regulatory thresholds	Advantage: 1) Rapid, easily measured, easily transferred, 2) Transparent, easily interpreted, 3) Information relevant to ecosystem management, 4) Indicators responsive to human disturbance
US Navy (Swchartz, Egeland)	Sustain healthy resources for future generations while fullfilling the mission of protection and war prevention. Authorized to manage natural resources on property under its control. Active coral reef ecosystem protection through NEPA, assessment and monitoring	Worldwide. Natural resources on properties under control (30 million acres altogether). Preserves in Kingman Reef and Palmyra Atoll managed as marine sanctuaries	Shallow to Mid-Depth (30 m)	Varies depending on location	Visual surveys, diver measurements	Coral Status (cover, prevalence of diseases and bleaching, coral sizes, recent and old mortality)	1) Increased reef sampling assessment efficiency, 2) a multi-tier approach with different levels of detail (monitoring, restoration), 3) Need for a technology that is safe, efficient, and digital, 4) need to reduce the costs and dangers associated with using divers for assessments while keeping data integrity.	Develop a Coral Reef Monitoring Protocol that: 1) Supports Habitat Equivalency and NEPA analyses, 2) provides a broadly accepted methodology for mapping, assessment and in-situ coral reef health monitoring, 3) provides data/image archival capability and data compatibility with existing software	Natural resources have been assessed at all bases and the data entered into GIS (PMAP) program available for management and navigation. Potential projectes that may benefit from mosaic surveys: Kihlo Wharf extension, Kamehameha outfall, Guam expansion

AGENCY	Mandate/Mission	Location and Habitats	Depth Range	Temporal Frequency	Methods	Coral Metrics / Indicators	Gaps / Limitations	Future Directions	Notes
NOAA SEFSC (Miller)	Fishery Independent Monitoring of fishes and invertebrates, 2) Assess coral/habitat status, 3) Evaluate habitat value/associations for fishes (EFH), 4) Assess coral condition, population dynamics, 5) Protected Species Monitoring (Acroporid corals)	Florida, Navassa, Puerto Rico	Shallow (< 30 m)	Seasonal to Annual	Visual surveys, photo and video surveys, marked colonies and plots, coral spawning and recruitment surveys	Coral Status (cover, prevalence of diseases and bleaching, coral sizes, recent and old mortality), abundance of corallivores, coral recruitment	Need effective methods for recruitment quantification, coral disease diagnostics, methods to reduce diver time underwater, expand surveys to deep reef habitats (e.g., *Oculina* banks)	Incorporate novel survey techniques into monitoring tools	
NOAA CCMA Biogeography Branch (Warner)	Assess and forecast coastal and marine ecosystem conditions through research and monitoring. Guiding Principles: 1) Address National Issues with Local Approach, 2) Support Diverse, Collaborative Partnerships, 3) Provide Science and Research to Directly Support Management and Policy Decisions, 4) Integrate Research Across Scientific Disciplines, 5) Serve as a Link Between Science Conducted in Academia and Specific Needs of Coastal Decision-Makers	US jurisdictions, coral reefs and other benthic habitats	Shallow (< 30 m) to deep (100-1000 m)	Annual to Biennial	Mapping (remote sensing, GIS, diver surveys), and monitoring (rapid assessment methods, diver surveys, photo and video surveys)	Coral cover, fish abundance	need for automation, better small scale characterization and mapping	Not Reported	
NOAA CCMA Coastal and Oceanographic Assessment, Status , and Trends Branch (COAST) (Warner)	1) To assess chemical contaminant levels in water, sediments, and coral tissues, 2) Identify and quantify biomarkers and identify pathogens in coral tissues, 3) Develop and test hypotheses relating contaminant burdens to measures of coral health, 4) Link Results of these exercises to ongoing regional coral reef ecosystem monitoring – including coral health and diversity, reef fish distribution, abundance, and diversity, phycology, and land use practices, 5) Evaluate application of the analytical construct to other areas in the US Caribbean and Pacific basins	US jurisdictions, coral reefs and other benthic habitats	Shallow (< 30 m) to deep (100-1000 m)	Not Reported	Benthic mapping, acoustic tracking,	Biomarkers, coral diseases	Need rapid, effective, non-destructive methods to evaluate coral physiological condition	Not Reported	
NOAA Sanctuaries, Restoration Division, FKNMS (Goodwin)	Assess physical damage to benthic resources (seagrass, hardbottom, coral reefs). To prepare rapid, cost-effective, litigation-quality claims for injuries to coral resources resulting from vessel groundings and other mechanical injuries, and to implement the restoration and monitoring of coral reef ecosystem injuries	Florida Keys National Marine Sanctuary. 33 grounding sites (coral and seagrass) presently monitored	Shallow (<20 m)	Irregular intervals. Damage assessment as needed	Damage assessment: Permanent and temporary sites, visual and photo quadrats, video surveys, underwattep mapping (GPS, buoys, aerial images, Fishbone method)	Extent of injury, cover of organisms in damaged and reference sites, recovery patterns over time	1) Survey methods are time-consuming and require trained divers, 2) Video analyzed using Ravenview to create strip mosaics, 3) Most reef restoration efforts have been set ad hoc, 4) Most efforts have not been founded on scientific data, 5) Ecosystem function has been absent in the decision-making process	1) Determine Success and Efficacy (or Failure) of Past Efforts, 2) Understand what works – what doesn't – and why? , 3) Implement Adaptive Management Program, 4) Develop more automated system to accurately assess extent of damage caused by vessel groundings	
NOAA Sanctuaries, Restoration Division, FKNMS (Precht)	Assess the status and trends of benthic resources within the Florida Keys National Marine Sanctuary (FKNMS)	Florida Keys National Marine Sanctuary. All reef habitat types.	Shallow (<20 m)	Annual	Rapid reef assessment (SCREAM team methods, UNCW, random sites, visual and video surveys) / CREMP methods (permanent sites, visual, photo and video surveys, strip mosaics) / Bleachwatch Program (visual surveys)/ WQ monitoring.	Cover of benthic organisms, urchin abundance, colony sizes, partial mortality, disease and bleaching prevalence, coral recruitment, diversity of multiple taxa, topographic complexity	Limited research in deeper reef habitats (> 30 m)	Not Reported	
The Nature Conservancy (Bergh)	Preseve and protect reef resources. Assess impacts of coral bleaching. Monitor for reef resilience	Florida and the Caribbean. In Florida, areas within Biscayne National Park, FKNMS, and the region of the Southeast Florida Coral Reef Initiative (Dade to Martin Co.)	Shallow (<20 m)	Annual, during the warmest summer months	Rapid reef assessment methodology (modified AGRRA)	Coral Status (cover, prevalence of diseases and bleaching, coral sizes, recent and old mortality), abundance of urchins, topography	Current approach does a good job of characterizing stony coral condition and demographics during short time period over large spatial area with rapid reporting of results. However, it does a poor job of identifying causality of changes in coral condition and demographics that occur between annual sampling events	Previous efforts concentrated on bleaching but can be expanded to other sources of stress. Post-bleaching follow-up monitoring needs to be designed to increase understanding of causality of changes in coral condition and demographics	

Appendix E- White Papers

DEPARTMENT OF DEFENSE NEEDS FOR MAPPING, INVENTORY, AND ASSESSMENT OF BENTHIC MARINE COMMUNITIES

The Department of Defense (DoD) needs to inventory, identify, document and assess benthic reef communities and other benthic habitats in order to have baseline information to comply with regulations and resource management requirements in proximity to installations and operational areas. DoD utilizes tools such as Habitat Equivalency Analysis for performing analysis of potential impacts for construction activities. Additionally, DoD needs to conduct monitoring of benthic habitats in order to fulfill NEPA or permit mitigation, Trustee obligations or other conservation commitments. The benthic reef community includes corals, algae, and other sessile and mobile invertebrates and associated substrates.

Technologies fulfilling these needs will provide operators and natural resources personnel with comprehensive knowledge of benthic habitats and coral reef communities under DoD purview. This information is necessary for operational and environmental planning and provides decision-makers with crucial information needed to maintain compliance with statutes, regulations, and executive orders directly related to operations conducted in benthic areas, including:

- Clean Water Act (33 USC §1251 et seq.)
- Coastal Zone Management Act of 1972(16 USC §§1451-1465)
- Comprehensive Environmental Response, Compensation, and Liability Act (42 USC Chapter 103)
- Coral Reef Conservation Act of 2000 (16 USC §6401 et seq.)
- Magnuson-Stevens Fisheries Conservation and Management Act (16 USC §§1801-1882)
- Marine Protection, Research and Sanctuaries Act (16 USC §§1431-1445a)
- National Environmental Policy Act as amended (42 USC §§4321-4347)
- Oil Pollution Act of 1990, 33 USC §2701
- Rivers and Harbors Act (33 USC §403)
- Sikes Act Improvement Act of 1997 (16 USC §670a-o)
- Executive Order 13089, Coral Reef Protection
- Executive Order 12114, Environmental Effect Abroad
- Executive Order 12777, Oil Pollution Act Implementation
- Executive Order 13158, Marine Protected Areas

Obtaining baseline ecological data is an important element not only for Federal coastal management of protected resources but also to provide a foundation for environmental documentation necessary to conduct operations. Such documentation requires the assessment of environmental conditions prior to any incidents possibly resulting in damage to or loss of habitat. Successful and legally defensible documentation requires the assessment of environmental conditions prior to conducting operations and implementation of mitigation measures. Assessment information is also necessary in resolving Federal trustee matters related to damage assessments. Legally defensible data is necessary to communicate and negotiate all regulatory actions in the marine environment.

- Efficient assessment of benthic habitats to support routine activity planning
- Reduced time and expense for data collection
- Reasonable operator experience and dive time requirements
- Experts spend more time in the lab analyzing data than in field collecting data
- Applicable in a wide range of locations (see military facility table below)
- Support day or night data collection as required
- Data quality to support compliance requirements
- Quantitatively and qualitatively characterize the diversity, abundance, temporal variation and spatial distribution of corals, algae and other invertebrates
- Support Habitat Equivalency Analysis tool and NEPA analyses, as well as permit and mitigation compliance
- Provide a common monitoring protocol for the benthic community was formulated with regards to location and frequency of surveys
- Robust, reliable and legally defensible
- Locate survey start and end points located using Global Position System (GPS) sensors
- Provide data/image archival capability and data compatibility with existing software including military GIS applications (EIMS and PMAP)
- Facilitate interoperability between DoD components and cooperation with other Federal and State agencies for compliance and stewardship efforts.
- Mutual benefit to use same tools
- Cost savings to share the same data for regulatory needs.
- Low cost, high benefit, ease of deployment will allow expanded benthic habitat assessment and monitoring
- Potential to leverage research needs.

Military Facilities with Adjacent Coral Reef Resources

Branch	Facility Name	Location
Air Force	Anderson Air Force Base	Guam
Air Force	Cape Canaveral Air Force Station	Florida
Air Force	Eglin Gulf Test and Training Range (EGTTR)	Florida
Air Force	Hickam Air Force Base	Hawaii
Air Force	Tyndall AFB	Florida
Air Force	Bellows Air Force Station	Hawaii
Air Force	Patrick Air Force Base	Florida
Air Force	Wake Atoll (Wake Island)	US Territory

Branch	Facility Name	Location
Air Force/ Navy	Eglin AFB	Florida
Army	Fort Buchanan	Puerto Rico
Army	Fort Shafter	Hawaii
Army	Johnston Atoll Chemical Agent Disposal System Facility (JACAD)	US Territory
Army	Kwajelein Atoll, Reagan Test Site, Marshall Islands	US Territory
Army	Pohakuloa Training Area	Hawaii
Army	Schofield Barracks	Hawaii
Army	Tripler Army Medical Center	Hawaii
Marine Corps	Marine Corps Base Hawaii	Hawaii
Marine Corps	Marine Corps Base Hawaii Ranges	Hawaii
Navy	Andros Island, AUTEC	Bahamas
Navy	Awase Transmitter Site, Okinawa	Japan
Navy	Barbers Point Family Housing and Support	Hawaii
Navy	Diego Garcia Navy Support Facility	BIOT
Navy	Diego Garcia Range Complex	BIOT
Navy	Farallon De Madinilla (FDR)	CNMI
Navy	Ford Island Naval Station Annex	Hawaii
Navy	Guam Naval Activities	Guam
Navy	Guantanamo Bay Naval Station	Cuba
Navy	Guantanamo Complex	Cuba
Navy	Gulf of Mexico Training Area	Florida
Navy	Hawaiian Range Complex	Hawaii
Navy	Japan Range Complex	Japan
Navy	Key West Range Complex	Hawaii
Navy	Key West Naval Air Station	Florida
Navy	Marianas Range Complex	CNMI
Navy	NAMFI Complex	Mediterranean
Navy	NASD, EMA & AFWTF	Puerto Rico
Navy	Naval Supply Center Red Hill	Hawaii
Navy	Okinawa Naval Activities	Japan
Navy	Okinawa Complex	Japan
Navy	Pachino Complex	Mediterranean
Navy	Pacific Missile Range Facility (PMRF) Barking Sands, Kauai	Hawaii
Navy	Panama City Coastal Systems Center	Florida
Navy	Pearl Harbor Naval Station	Hawaii
Navy	Pensacola, Naval Air Station	Florida

Branch	Facility Name	Location
Navy	Tinian Island, Military Leased Areas	CNMI
Navy	White Beach Naval Facility, Okinawa	Japan

High Resolution Landscape Mosaics for Coral Reef Mapping and Monitoring

What is a landscape mosaic? Individual underwater images taken close to the seabed (~1-2m) have high resolution and minimal water column attenuation, but cover only a small area. A landscape mosaic is a composite of many underwater images. The mosaics have the clarity and resolution of individual pictures but afford a "landscape view" of the seabed (Fig 1).

The U.S. Strategic Environmental Research and Development Program (SERDP) has supported a) the development of software tools for generating underwater landscape mosaics without relying on external navigation and b) the evaluation of these mosaics for coral reef mapping and monitoring. We are seeking to identify potential applications and partners.

Data Acquisition Requirements: Mosaics are made in one of two modes: "Standard mode" uses video data only; "Enhanced mode" uses still images acquired synchronously with the video. Both need:

- Near-nadir view video 1-2 m from seabed.
- High (~80%) overlap between swaths.

Enhanced mode additionally requires:

- Still camera synchronized with video.

Mosaic Characteristics:

- Area covered: ~ 400 m^2 (~2000 frames)
- Spatial resolution (pixel size):
  enhanced mode, sub-mm;
  standard mode, ~ 3 mm.
- Spatial accuracy: +/-5 cm (1 standard deviation)

Highly automated mosaic production requires about 4 man-hours and 24-36 hours computer time with current desktop processors.

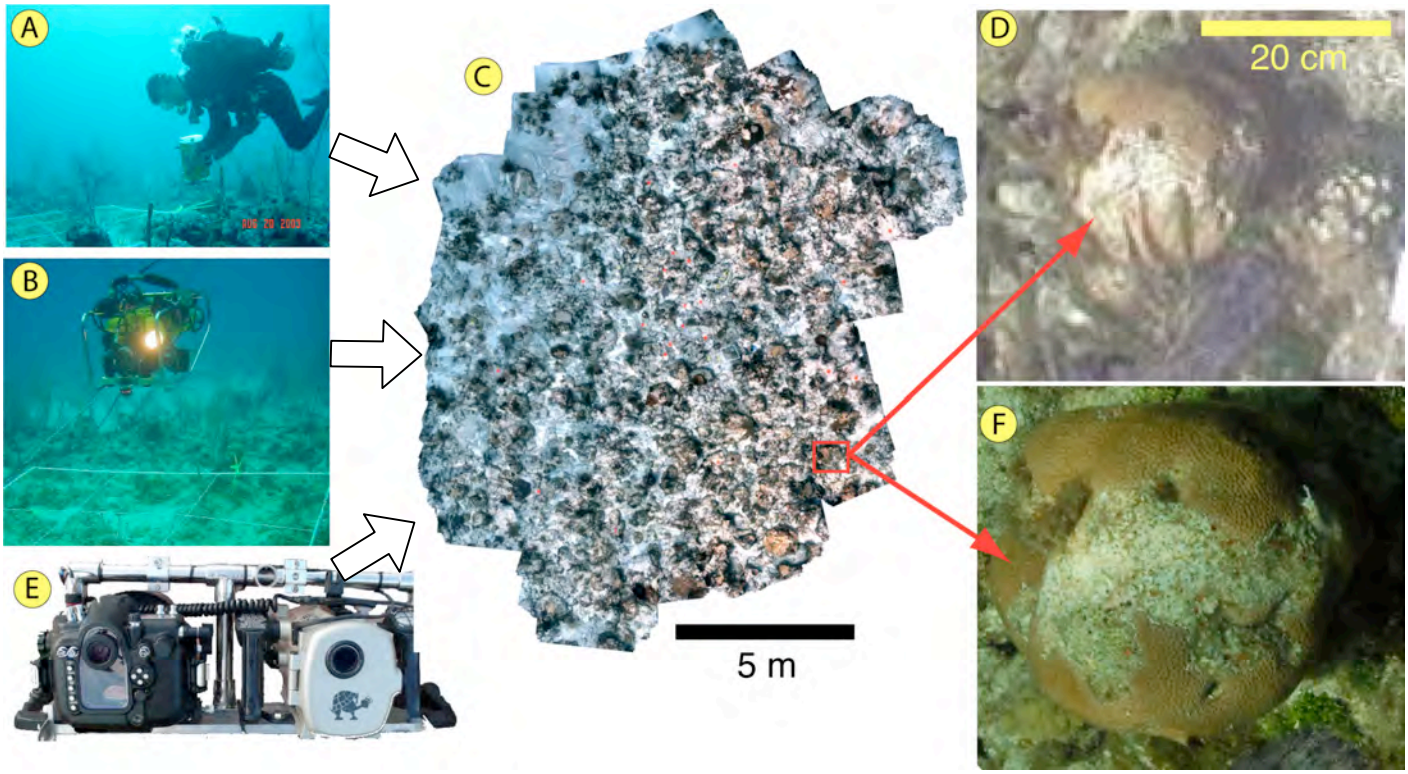

Figure 1: Mosaic overview: Video images acquired by a diver (A) or other platform such as an ROV (B) are automatically stitched together to form a landscape mosaic (C) covering a large area (about 200 m^2 in this case). "Standard mode" (i.e. video only) produces mosaics with mm-scale resolution (D). In "enhanced mode", still imagery is acquired simultaneously with the video (E) to achieve sub-mm resolution (F).

Key Benefits:

- Landscape view: Mosaics provide a landscape view of coral reefs that has previously been unobtainable. This enables new measures of reef health, such as documenting spatial relationships of disease patterns, or the effects of hurricane damage and ship groundings.
- Spatial accuracy: High spatial accuracy, combined with a landscape view, enables accurate size and distance measurements to be taken directly from the mosaic. Mosaics can be georeferenced and integrated with other data sets using Geographic Information Systems (GIS)
- Colony monitoring without tagging: Mosaics are efficient tools to track patterns of change over time. Mosaics collected in repeat surveys can be referenced to one another with only four permanent markers, allowing monitoring of individual coral colonies without the need for extensive tagging.

Compared with traditional techniques: Mosaics retain key strengths of a diver-based approach, while overcoming the limitations of diver-based or photo-quadrat / video transect methods (Table 1).

Table 1: Comparison of monitoring techniques.

Technique	Diver Survey	Photo-quadrat or Video transects	Landscape Mosaics
Strengths of the Diver-transect			
Percent cover benthic organisms			Note (1)
Diversity indices			Note (1)
Juvenile coral density			
Disease / Bleaching / Partial Mortality			Note (2)
Coral Colony Size			
Limitations of the Diver-transect			
Scientific diver required			
Long dive times			
Permanent record for reanalysis			
Repeatability (track changes over time)			
Depth limits			
Landscape view (map large features)			
Spatial accuracy			

Green indicates full capability, yellow partial capability, and red poor capability. Note (1): Enhanced mode required for species-level IDs, but identification of major functional groups (e.g., corals, sponges, algae) is done with standard mode. Note (2): Enhanced mode required.

Sample mosaics are available upon request!

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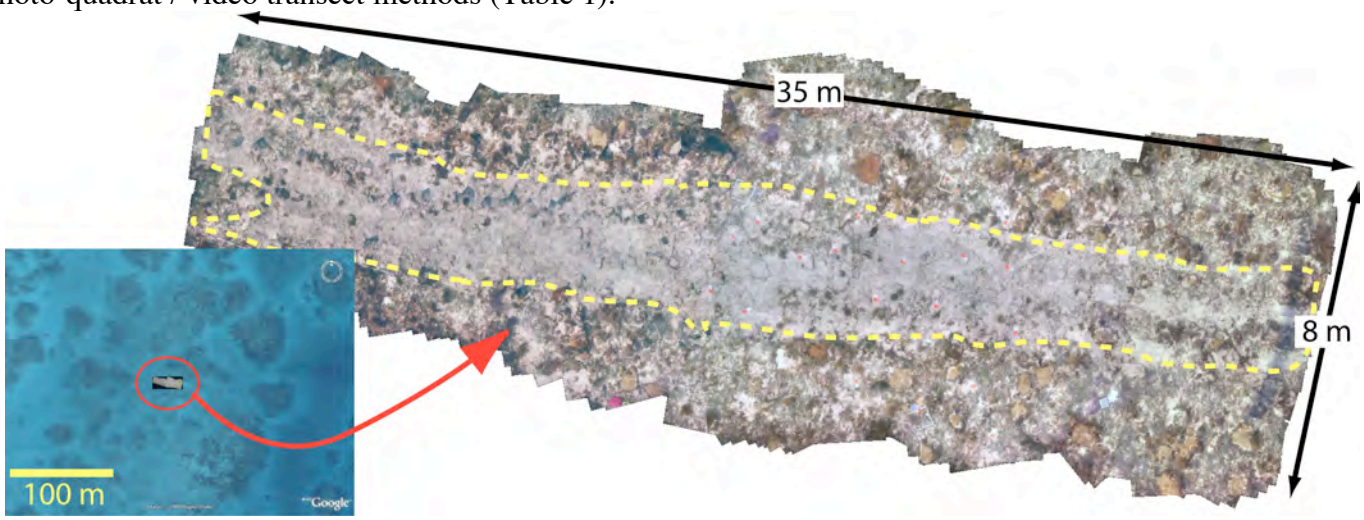

Figure 2: Mosaic of a scar created by a ship grounding on a shallow reef, Florida Keys (depth = 3 m). The dashed line marks the extent of damage. The inset shows this mosaic inserted into Google Earth, illustrating the potential to incorporate mosaics in GIS systems. Groundings are large and cumbersome to survey solely by divers.. An image conveys more information about the extent of the damage than measurements of the overall dimensions, especially when viewed by non-technical personnel (e.g. juries).

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ORIGINAL ARTICLE

Development and application of a video-mosaic survey technology to document the status of coral reef communities

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Abstract The recent decline in the condition of coral reef communities worldwide has fueled the need to develop innovative assessment tools to document coral abundance and distribution rapidly and effectively. While most monitoring programs rely primarily on data collected *in situ* by trained divers, digital photographs and video are used increasingly to extract ecological indicators, provide a permanent visual record of reef condition, and reduce the time that divers spend underwater.

In this study, we describe the development and application of a video-based reef survey methodology based on an algorithm for image registration and the estimation of image motion and camera trajectory. This technology was used to construct two-dimensional, spatially accurate, high-resolution mosaics of the reef benthos at a scale of up to 400 m^2. The mosaics were analyzed to estimate the size and percent cover of reef organisms and these ecological indicators of reef condition were compared to similar measurements collected by divers to evaluate the potential of the mosaics as monitoring tools.

The ecological indicators collected by trained divers compared favorably with those measured directly from the video mosaics. Five out of the eight categories chosen (hard corals, octocorals, *Palythoa*, algal turf, and sand) showed no significant differences in percent cover based on survey method. Moreover, no significant differences based on survey method were found in the size of coral colonies. Lastly, the capability to extract the same reef location from mosaics collected at different times proved to be an important tool for documenting change in coral abundance as the removal of even small colonies (<10 cm in diameter) was easily documented.

The two-dimensional video mosaics constructed in this study can provide repeatable, accurate measurements on the reef-plot scale that can complement measurements on the colony-scale made by divers and surveys conducted at regional scales using remote sensing tools.

Keywords Benthic surveys · Image motion · Reef condition · ROV · Video mosaics · Video surveys

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1 Introduction

The recent worldwide decline in coral reef health and extent has fueled a myriad of local and regional efforts

aimed at collecting comprehensive monitoring data that can be used to evaluate the present condition of reef communities as well as to provide a baseline against which future changes can be accurately gauged (Gardner *et al*., 2003; Kramer, 2003; Wilkinson, 2004). While sampling design and survey approaches differ among monitoring programs, the use of plot (e.g., quadrats) and line-based (e.g., line intercept) methods to estimate the percent cover of benthic organisms prevail as important components of these efforts (Hodgson, 1999; Kramer and Lang, 2003). Coral cover has historically been the predominant indicator of reef condition but recent studies have also highlighted the importance of the size-structure of coral populations as a powerful but often underused status indicator (Bak and Meesters, 1998, 1999). In response to these studies, plot and line-based methods are now commonly supplemented by colony-based methods that document the size and condition of individual coral colonies (Lang, 2003).

The rapid patterns of reef decline have also prompted the design of innovative assessment tools to document coral abundance, distribution, and condition rapidly and effectively (Solan *et al*., 2003; Fisher *et al*., 2005). With the development of better and more affordable photography and videography techniques and equipment, many programs routinely complement diver-based measurements with digital images of the bottom that are later analyzed using image analysis software (Riegl *et al*., 2001; Porter *et al*., 2002). These digital tools improve survey efficiency by: (1) reducing the time that divers need to spend underwater by shifting data capture away from the field and into the lab; and (2) providing a permanent visual record of reef condition. The use of digital video provides the added benefit of capturing a large number of digital frames in a limited amount of time.

Digital photographs and video frames provide two-dimensional images of the bottom that can be analyzed with the same methods commonly used by divers to estimate percent cover *in situ*. These methods include: (1) the point intercept method where a number of points are randomly placed over each image and the identity of the benthic organisms immediately under each point is determined; and (2) the area estimation method where the boundary of each organism is delineated. In both cases, the proportion of the total number of points or total reef area occupied by each organism is used to measure percent cover. While these methods provide an effective estimate of the areal coverage of benthic organisms, they provide only limited size-estimation capabilities because sizes can be measured only for organisms that fall completely within an image. This limitation is especially manifested in reef habitats with large corals and high topographical relief where individual colonies are rarely captured wholly within frames or video transects.

The goals of the present study are to: (1) describe the development and application of a novel, video-based reef survey methodology that provides a powerful and efficient alternative to existing photography and video-based approaches; and (2) evaluate whether the video mosaic method could provide the type of ecological information related to coral reef condition commonly obtained by trained divers *in situ*. This technique, based on a recently developed algorithm for image registration, is used to construct spatially accurate mosaics of the reef benthos that can be analyzed to estimate not only the percent cover of organisms but also their size and spatial distribution and arrangement patterns. This flexible mosaicing algorithm allows the technique to be used in a variety of applications from low cost surveys with handheld underwater video cameras to mapping deep reefs with remotely operated vehicles (ROV). A reef site in the Florida Keys, U.S., was surveyed using these two platforms and the community attributes obtained by analyzing the video mosaics are compared to similar indicators collected by trained divers to provide a direct comparison between methods.

2 Materials and methods

2.1 Video mosaic creation

2.1.1 Video acquisition

The field activities for this study were conducted at Brooke's Reef (25°40.508′N, 80°5.908′W, depth = 7–10 m), a patch reef located in the northernmost section of the Florida Reef Tract, just offshore of Key Biscayne, Florida. A square plot (3 m × 3 m) was established at this site using aluminum pipes cemented to the bottom to provide a permanent reference location for video surveys. Three video mosaics of the same reef area were created using different survey platforms (Table 1). For the first mosaic (June, 2004), video footage was acquired by a diver using a Sony TRV900 DV camcorder placed in an underwater

Table 1 Description of the three different mosaics constructed in this study based on digital video collected at a reef in the northern Florida reef tract (depth = 7–10 m)

Survey	Date	Survey platform (Camera resolution)	Altitude	Area covered	Ground resolution
1	June 04	Diver (720 × 530 pixels)	2 m	53 m^2	3.0 mm/pixel
2	April 05	ROV (1024 × 768 pixels)	2.5 m	400 m^2	2.5–3.0 mm/pixel
3	April 05	ROV (1024 × 768 pixels)	1.5 m	45 m^2	1.4 mm/pixel

camera housing. This first survey is included to illustrate that the mosaicing algorithm can produce geometrically accurate mosaics from a standard, low-cost, handheld camera. For the second and third mosaics (April, 2005), video was collected using a Flea digital camera mounted on a Phantom XTL remotely operated vehicle (ROV) (Xu, 2000) representing high and low altitude data sets from which ecological indices were assessed. The cameras were internally calibrated to reduce image distortion from the lens and housing (Bouguet, 2002). The frame resolution is 720 × 530 pixels for the handheld camcorder and 1024 × 768 pixels for the Flea camera. On all occasions, the camera followed a lawnmower's pattern of side-by-side strips, complemented by the same pattern rotated 90° to ensure full coverage of the area and high superposition among the strips.

2.1.2 Mosaic algorithm

The mosaic-creation algorithm used in this study stems from previous work on underwater video mosaicing by Gracias and Santos-Victor (2000, 2001). The method comprises four major stages. The first stage consists of the sequential estimation of the image motion, using a subset of the captured images. The set of resulting consecutive homographies (i.e., coordinate mapping between two image projections of the same 3D plane) is cascaded to infer the approximate trajectory of the camera. The trajectory information is then used to predict the areas of image overlap from non-consecutive images (i.e., neighboring video strips). To reduce the algorithmic complexity and memory requirements, a set of key frames are selected based on an image superposition criterion (typically 65–80%). Only such key frames are used in the following optimization steps.

In the second stage, a global alignment is performed where the overall camera trajectory is refined by executing the following two steps iteratively: (1) point correspondences are established between non-adjacent pairs of images that present enough overlap; and (2) the trajectory is updated by searching for the set of homographies that minimizes the overall sum of distances in the point matches.

In the third stage, high registration accuracy is obtained by re-estimating the camera trajectory using a general parameterization for the homographies. This parameterization has six degrees of freedom (DOF) for the pose and is capable of modeling the effects of general camera rotation and translation. The essential building block of this step consists of the registration of pairs of images done as follows: (1) a set of point features corresponding to textured areas are extracted from one of the images using the Harris corner detector method (Harris and Stephens, 1988); and (2) for each feature (defined as a small square image patch centered at the detected corner location), a prospective match is found in the other image using normalized cross-correlation. We assume that prior information exists on the expected image motion (typically in the form of a homography). This information is used to: (1) establish the location of the correlation window center; and (2) define the required warping of the image feature so that the search over the other image becomes essentially a translation (2D) search. This allows for the use of area-correlation for heavily rotated or slanted images. Finally, a robust estimation technique is used to remove outliers using a Least Median of Squares criterion based on a planar motion model.

The final stage of the mosaicing process consists of blending the images (i.e., choosing representative pixels from the spatially registered images to render the mosaic image). The mosaic is created by choosing the contributing pixels that are closest to the center of their frames. The image rendering method used in this study compares favorably to other traditional rendering methods, such as the average or the median, by: (1) preserving the texture of the benthic objects; (2) reducing artifacts due to registration misalignments of 3D structure; and (3) allowing for an efficient implementation in terms of memory requirements and execution speed. However, in the presence of strong illumination

changes or strong 3D content, the present version of our method can create visible seams along the boundaries of the images. The visibility of these seams may be reduced by employing more computationally intensive rendering methods, such as optimal seam finding (Uyttendaele *et al*., 2001; Agarwala *et al*., 2004) and gradient domain blending (Levin *et al*., 2004). A fast, memory-efficient method for optimal seam finding is currently being developed to address the processing of large underwater image sets with variable light conditions as included in this study.

2.1.3 Spatial accuracy

To quantify the geometric accuracy of the mosaics, a geometric distortion analysis was performed using ground-truth data consisting of a set of points of known positions that are easily located on the mosaic images. The accuracy analysis consists of two steps. In the first step, 2D positions were measured by divers taking distance measurements to the closest cm between markers placed on the bottom relative to four reference stakes using flexible underwater tapes. For this study, the area of interest is assumed to be approximately flat, so the geometric analysis is carried out in 2D. Given the fact that it is difficult to measure XY locations underwater accurately, the creation of the ground-truth data had to be done indirectly using a network of distance measurements between points (Holt, 2000). A set of ground-truth points was created within the test area by placing 24 markers (painted CDs) on the bottom with masonry nails and attaching four control stakes permanently with underwater cement (Fig. 1). The distances between each marker and the four control stakes, as well as the distances among the control stakes, were measured by divers using flexible tapes.

Given a set of distance measurements, we want to estimate the 2D locations of all points with respect to a common metric reference frame. Let $\tilde{d}_{ij}$ be the measured distance between points i and j. The observed noisy measurement relates to the ideal noise-free distance $\tilde{d}_{ij}$ as: $\tilde{d}_{ij} = d_{ij} + \varepsilon$, where ε is an additive noise term. Each point is represented by its 2D coordinates: $P_i = (x_i, y_i)$. The observations relate to the sought parameters as:

$$\tilde{d}_{ij} = \sqrt{(x_i - x_j)^2 + (y_i - y_j)^2} + \varepsilon$$

Using a Least-Squares criteria, the problem can be formulated as finding the set of $(\hat{x}_i, \hat{y}_i)$ such that:

$$(\hat{x}_i, \hat{y}_i) = \arg\min_{(x_i, y_i)} \sum_{i,j} (\tilde{d}_{ij} - \sqrt{(x_i - x_j)^2 + (y_i - y_j)^2})^2$$

To establish a reference frame for the coordinates, additional constraints need to be imposed. These can be defined as: $x_1 = y_1 = y_2 = 0$, which sets the origin at point 1 and the world X axis along the line between points 1 and 2. The coordinates of the ground-truth points were estimated using a standard non-linear least squares algorithm (Press, 1988).

In the second step of the spatial accuracy analysis, comparisons were made between distance measurements taken directly from the mosaics and the ground-truth distance measurements taken by divers in an operation known as mosaic "referencing". The computation of this step can be done by using a set of points of known world coordinates that can be located on the mosaic. The most general model for mapping the world plane into an image plane requires the knowledge of at least four points whose world coordinates are known. However, this mapping can be computed using a larger set of point correspondences, resulting in a higher-precision referencing. In this study, all 24 markers were used for referencing the mosaics.

For each ground-truth point of metric coordinates (x_i, y_i) and mosaic image coordinates (u_i, v_i) we consider the difference residue defined as:

$$\begin{bmatrix} r_{x_i} \\ r_{y_i} \end{bmatrix} = \begin{bmatrix} \frac{h_1 u_i + h_2 v_i + h_3}{h_7 u_i + h_8 v_i + 1} \\ \frac{h_4 u_i + h_5 v_i + h_6}{h_7 u_i + h_8 v_i + 1} \end{bmatrix} - \begin{bmatrix} x_i \\ y_i \end{bmatrix}$$

where $\vec{h} = [h_1 \ldots h_8]^T$ are the parameters of the world-to-mosaic projective mapping. This mapping is computed using standard least squares as:

$$\hat{\vec{h}} = \operatorname{argmin}_{\vec{h}} \sum_i \left(r_{x_i}^2 + r_{y_i}^2\right)$$

Two criteria were used to assess the geometric distortion: (1) the standard deviation of all residues $(r_{x_1}, r_{y_1}, \ldots, r_{x_N}, r_{y_N})$; and (2) the maximum distance error: $d_{\max} = \max_i \sqrt{(r_{x_i}^2 + r_{y_i}^2)}$. These indicators are useful for two main reasons: (1) they provide nominal

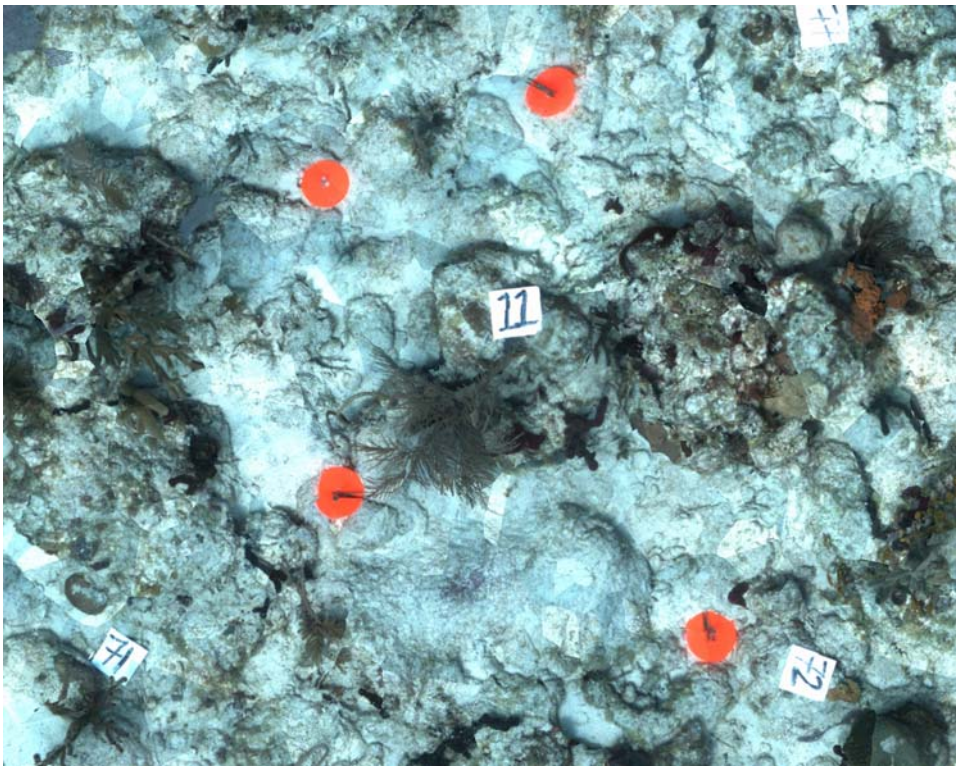

Fig. 1 Sample image from the second mosaic showing the placement of the ground-truth markers (painted CDs) used for measuring spatial accuracy. The numbered tiles show the location of coral colonies for which size measurements were obtained by divers

error bounds to metric distance measurements made over the mosaic; and (2) they can be used as quality indexes to compare mosaics created under different environmental conditions, such as varying relief, depth, illumination, and turbidity.

2.1.4 Sub-sampling mosaic images: Tile extraction and change detection

Referencing a mosaic allows for any area of the image to be delimited in metric coordinates.

Using the parameter vector $\vec{h}$, the metric coordinates of image point (u_i, v_i) are given by:

$$\begin{bmatrix} x_i \\ y_i \end{bmatrix} = \begin{bmatrix} \frac{h_1 u_i + h_2 v_i + h_3}{h_7 u_i + h_8 v_i + 1} \\ \frac{h_4 u_i + h_5 v_i + h_6}{h_7 u_i + h_8 v_i + 1} \end{bmatrix}$$

Using the location of control stakes as a reference, a sample grid can be established so that sub-sections or "tiles" of known size can be surveyed (Fig. 2). Also, if mosaics share a reference frame defined by the same four control stakes, the same locations can be retrieved from all images if desired. The capability to extract the same reef locations from mosaics collected at different times was tested here as a mechanism to document patterns of change in the abundance and spatial distribution of reef organisms. In this study, tiles covering areas of 0.25 m^2 were extracted from the mosaics to evaluate the percent cover of benthic organisms using the point intercept-method. The tiles extracted from the first mosaic were compared to the same tiles extracted from the third mosaic to evaluate changes in coral abundance from 2004–2005.

2.2 Benthic characterization

2.2.1 Diver surveys

The benthic coverage of the different components of the coral reef community was quantified using the point-intercept method. This method was chosen because: (1) it is the method used by EPA's Coral Reef Monitoring Program (CRMP) which surveys >40 permanent reef sites throughout the Florida Keys National Marine

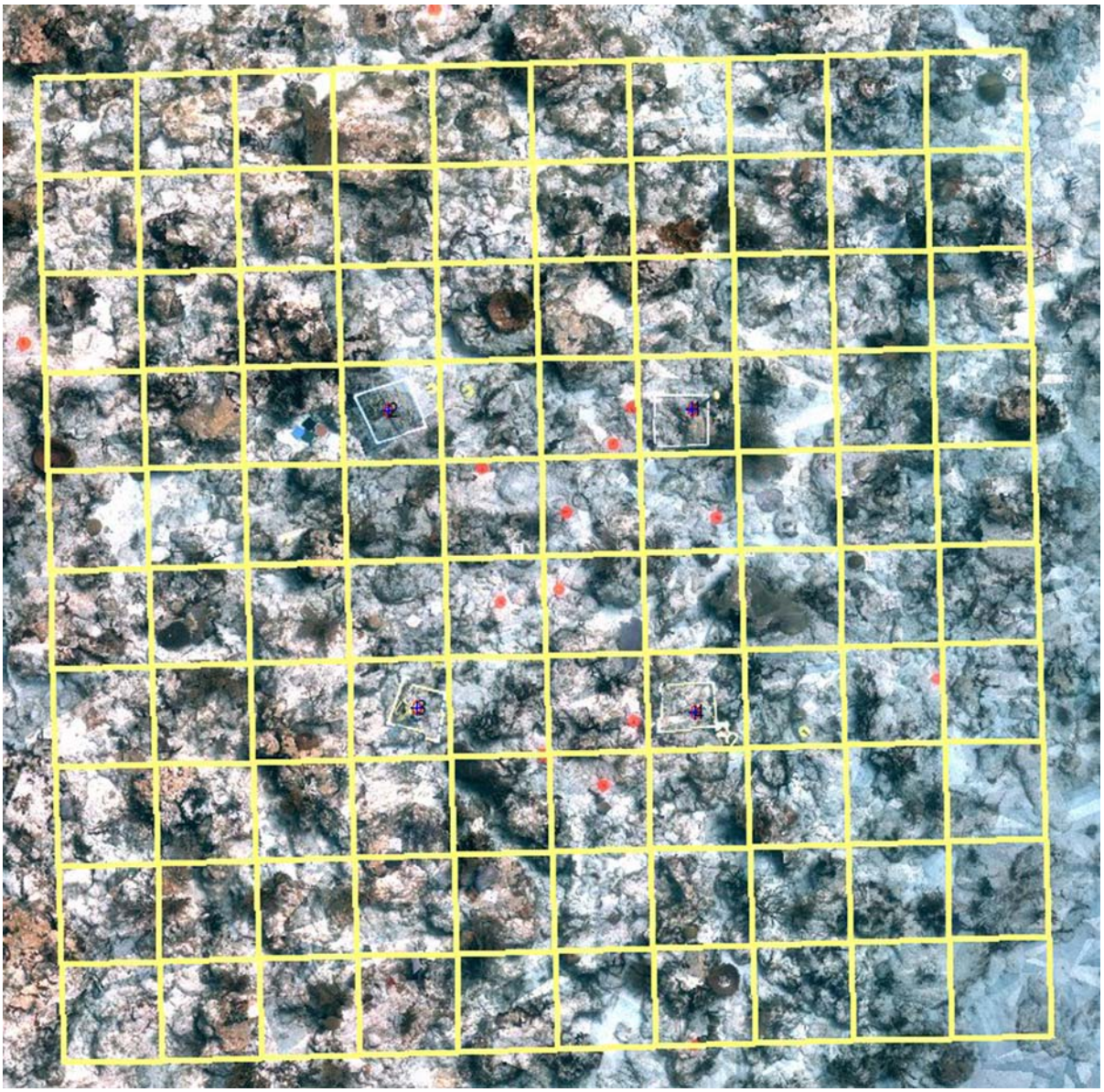

Fig. 2 Example of a sampling grid constructed to extract subsections or tiles from video mosaics. The grid is referenced using four numbered control stakes. If the same four reference points are used from multiple mosaics, the same locations can be extracted to assess change patterns in the abundance of benthic organisms over time. In this mosaic, the white PVC quadrats are placed over each of the control stakes

Sanctuary (Porter *et al*., 2002); and (2) it can be applied during *in situ* visual surveys as well as to analyze photographs and video mosaics.

The point-intercept method consists of deploying PVC quadrats (0.25 m^2) subdivided with elastic rope. In each quadrat, survey points are identified by marking a subset of the rope intersections with colored plastic ties. In the field, the quadrats are placed on the bottom haphazardly and the identity of each benthic organism lying directly under the labeled points is recorded. In this project, eight main benthic categories were identified: stony corals, octocorals, sponges, the zoanthid *Palythoa*, macroalgae (>1 cm in canopy height), crustose coralline algae, algal turfs (<1 cm in canopy height), and sand. A preliminary analysis of the minimum number of quadrats as well as the number of points per

quadrat needed to characterize the benthic community was conducted following methods outlined by Brown *et al*. (2004). Based on this analysis, 25 quadrats (covering approximately 25% of the reef area surveyed) and 25 points per quadrat were analyzed.

The number of points occupied by each category was used to determine their percent cover within quadrats and these values were averaged among quadrats to determine a mean value for each category. In addition to these measurements, the size (maximum diameter and height) of coral colonies within the survey area was quantified by divers using a flexible tape.

2.2.2 Video mosaics

To quantify the cover of benthic categories from video mosaics, each mosaic was sub-divided into 0.25 m^2 sub-sections or "tiles" (i.e., the same dimensions as the quadrats used by divers in the field) and a subset of mosaic tiles was extracted at random from the complete set to simulate the random placement of individual quadrats by divers in the field. The images were analyzed using the CPCe program developed by the National Coral Reef Institute (http://www.nova.edu/ocean/cpce/index.html). This application superimposes a user-determined number of points over a digital image. Once the points are placed, the user can identify the benthic category under each point just as it is done in the field. The program creates, as an output, a file that summarizes the information for each image and calculates the percent cover of each category by quadrat and by site.

The size (i.e., maximum diameter) of the coral colonies measured by divers (identified by a numbered tile visible in each mosaic) was estimated using the image analysis software Image J developed by the US National Institutes of Health with the scale provided by the pixel-size of each mosaic.

2.2.3 Comparison of diver surveys to video mosaics

To evaluate the performance of our video mosaics as assessment tools, indicators of reef condition measured by divers were compared directly to the same indicators obtained from mosaics created with video sequences collected at the same time. The indicators measured by a single diver (D. Lirman) were used as the standard against which all other measurements were compared.

The percent cover of the eight main benthic categories was compared among survey methods (i.e., diver surveys, high-altitude mosaic, low-altitude mosaic) using a Kruskal-Wallis test. As an additional measurement of coral cover, the boundaries of all stony corals found within the area imaged by the low-altitude mosaic were digitized and analyzed using the "particle analysis" feature in the ImageJ software that calculates the total area of polygon features within an area of interest. Finally, the abundance of juvenile corals (<4 cm in diameter) measured by divers within benthic quadrats was compared to the abundance of juvenile corals measured from the mosaic tiles.

To determine the accuracy of diver surveys and video mosaics to estimate coral colony size, the differences between the values obtained by Lirman and those obtained by a second diver (B. Gintert), or directly from the video mosaics were measured. Accuracy of the size measurements was ascertained by calculating two measurements of error as described by Harvey *et al*. (2000):

$$\text{Absolute Error} = \text{AE} = (|\text{Diver 1} - \text{Diver 2}|) \quad \text{and} \quad (|\text{Diver 1} - \text{Mosaic}|)$$

$$\text{Relative Absolute Error} = \text{RAE} = [(|\text{Diver 1} - \text{Diver 2}|)/\text{Diver 1}] \quad \text{and} \quad [(|\text{Diver 1} - \text{Mosaic}|)/\text{Diver 1}]$$

To compare the size data collected by divers and mosaics, an ANOVA with two factors, survey method and coral size category, was performed using the AE values.

3 Results

3.1 Video mosaics

The first video mosaic (Fig. 3) was created from 365 key-frames selected using a criterion of 75% overlap between consecutive images. For the second mosaic (Fig. 4), 496 key frames were selected out of the complete set of 5061 images, using 72% overlap. The registration parameters for the non key-frames were obtained by linear adjustment of the sequential matching, constrained by the registration parameters of the two

Fig. 3 Video mosaic constructed with video collected from a hand-held digital camcorder in June 2004 at Brooke's Reef in the Florida Reef Tract (depth 7–10 m). The video was collected at a distance of 2 m from the bottom. The painted CDs show the location of ground-truthing points

closest key-frames. For the third mosaic (Fig. 5), 872 key frames were selected from a set of 3439 images with a 75% overlap criterion. The colors on all mosaics were adjusted by manually selecting both a white and a black reference and linearly interpolating the red, green, and blue intensities. The algorithms were coded in Matlab 6.2, and the overall processing took between 6–12 h per mosaic using a 3.0 GHz PC.

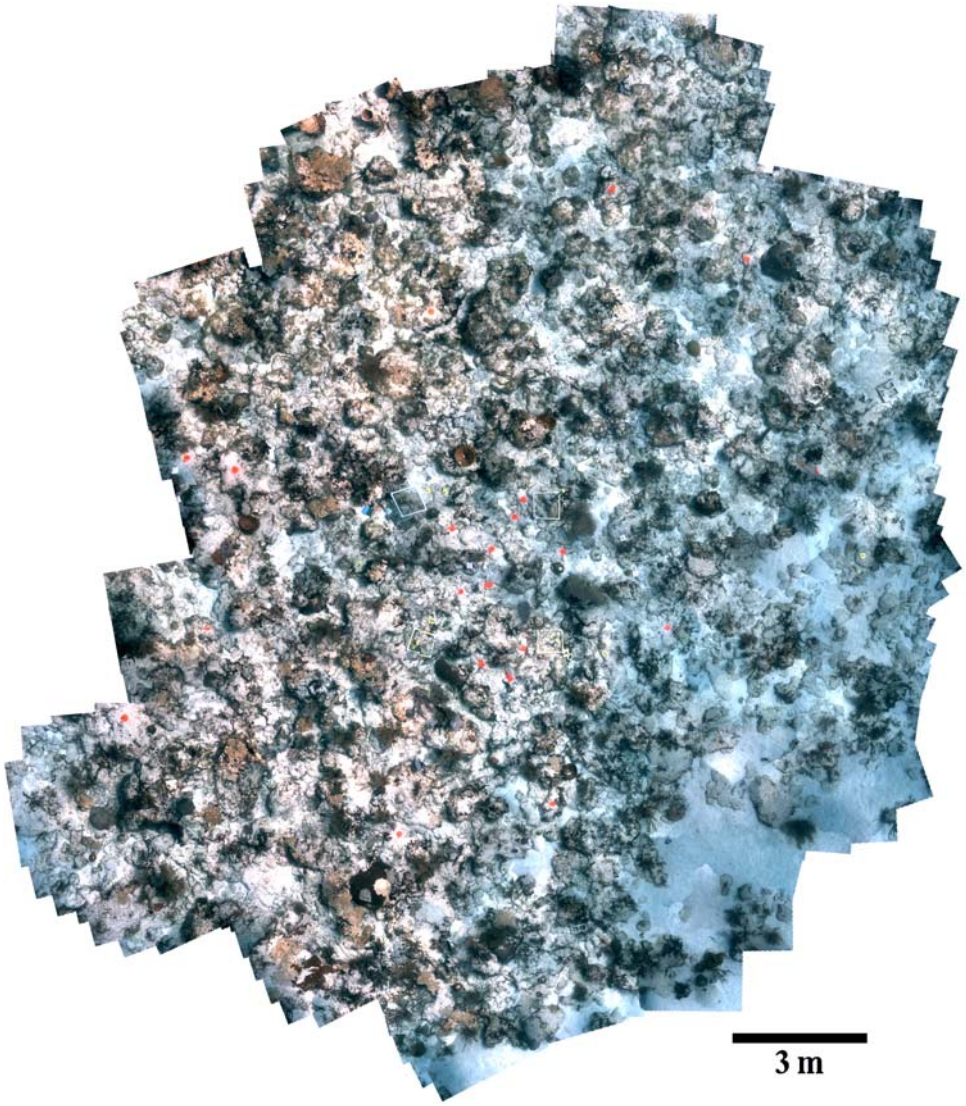

Fig. 4 Video mosaic constructed with video collected from a high resolution camera from an ROV platform in April 2005 at Brooke's Reef. The video was collected at a distance of 2.5 m from the bottom

3.2 Spatial accuracy of video mosaics

The algorithm used in this study produced three mosaics with high spatial accuracy. The distortion indicators showed an improvement in spatial accuracy (i.e., decreases in the standard deviations of the residues and maximum distance errors) going from video collected by a diver holding a digital camcorder (first mosaic) to video collected by a high-resolution camera mounted on the ROV (second mosaic). However, distortion indicators did not improve with increased image resolution as the distance to the bottom was decreased in the third mosaic. Standard deviations of the residues were 5.1, 3.9, and 5.5 cm, while maximum distance errors were 12.9, 10.7, and 13.5 cm for the first, second, and third mosaics respectively.

3.3 Comparison of diver surveys to video mosaics

Five out of the eight categories chosen (hard corals, octocorals, *Palythoa*, turf, and sand) showed no significant differences in percent cover based on survey method (Table 2, $p > 0.05$). The remaining three categories, corresponding to functional forms of reef

Table 2 Mean cover (±S.E.M.) of the different benthic categories surveyed by divers and measured from video mosaics from a reef site in the northern Florida Reef Tract (depth = 7–10 m). Divers surveyed twenty-five 0.25 m^2 quadrats. For comparison, a subset of 25 quadrats (0.25 m^2) were sampled at random form the video mosaics collected at 2 different resolutions. High-resolution mosaics were collected at a distance of 1.5 m to the bottom (2.5–3.0 mm/pixel). Low-resolution mosaics were collected at a at a distance of 2.5 m to the bottom (1.4 mm/pixel). CCA = Crustose Coralline Algae. *p* values from a Kruskal-Wallis test

Benthic categories	Diver	Mosaic – high resolution	Mosaic – low resolution	*p*
Stony Corals	1.4 (0.5)	2.0 (0.7)	1.8 (1.0)	0.6
Octocorals	7.5 (2.6)	6.2 (1.6)	4.7 (1.6)	0.6
Macroalgae	38.1 (3.4)	31.7 (3.0)	21.2 (3.1)	<0.01
CCA	1.1 (0.4)	0.3 (0.2)	0	0.02
Sponges	3.4 (1.2)	12.9 (1.9)	13.6 (1.9)	<0.01
Palyhtoa	4.2 (2.6)	1.2 (0.5)	2.7 (1.7)	0.3
Sand	5.8 (2.0)	9.2 (2.0)	7.5 (1.7)	0.6
Turf	38.9 (2.9)	36.5 (3.0)	41.6 (3.9)	0.3

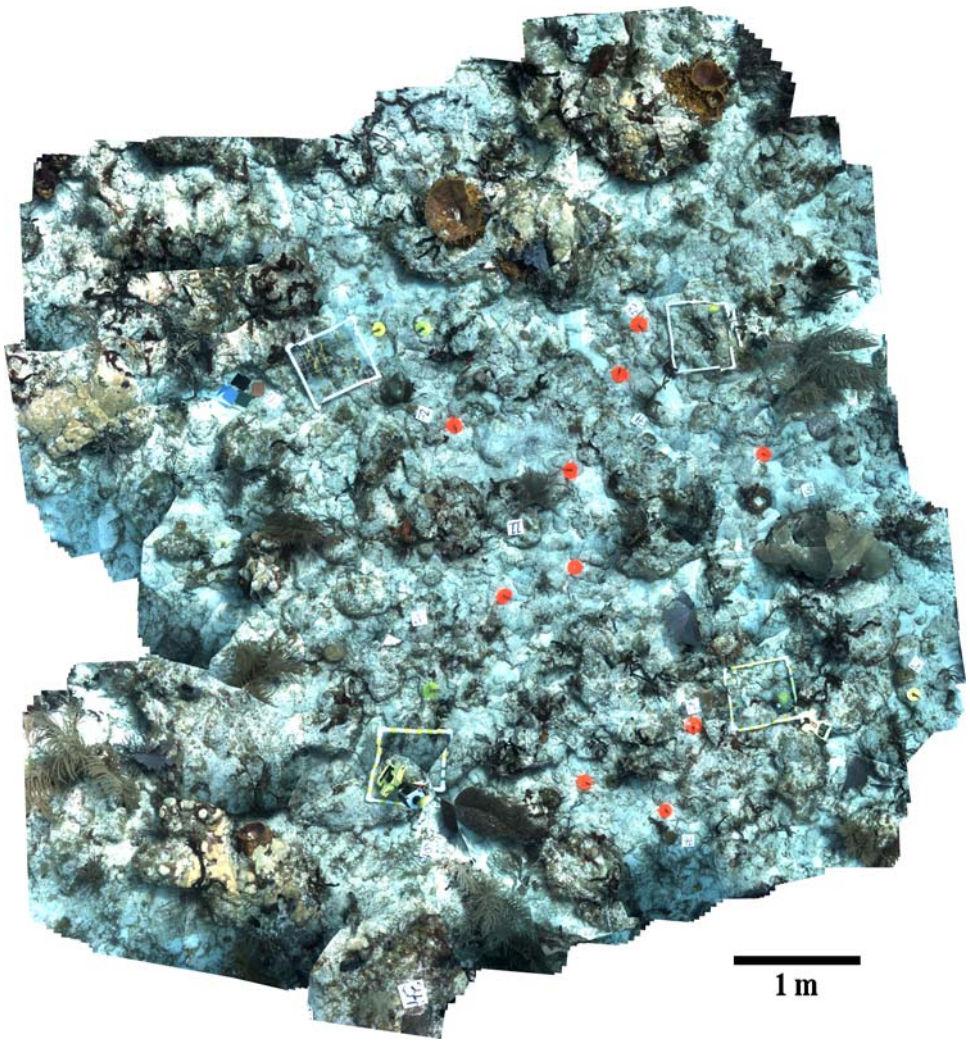

Fig. 5 Video mosaic constructed with video collected from a high resolution camera from an ROV platform in April 2005 at Brooke's Reef. The video was collected at a distance of 1.5 m from the bottom

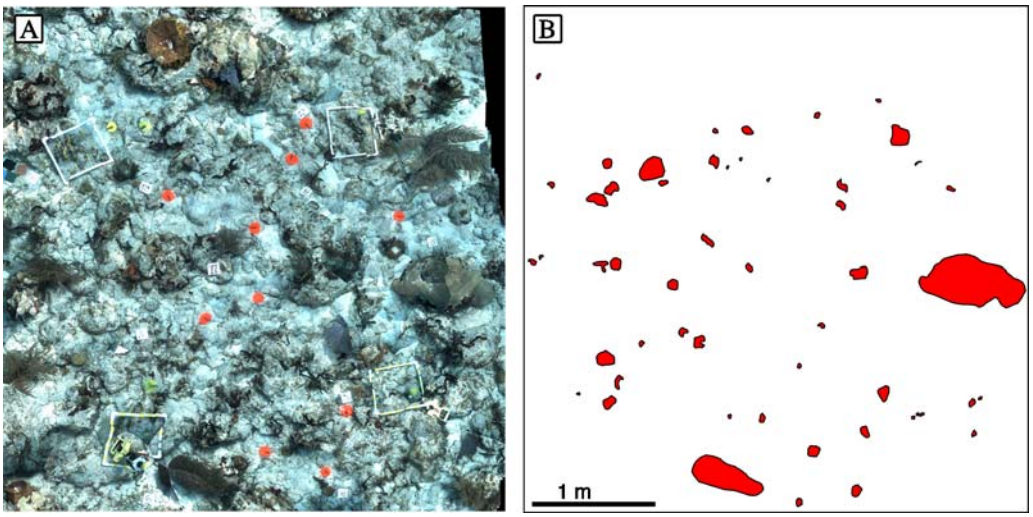

Fig. 6 Abundance and spatial distribution of stony corals obtained from a high-resolution (1.4 mm/pixel) video mosaic (A). The boundaries of each coral colony (B) were digitized and the benthic coverage of stony corals was measured using the ImageJ software. The coral cover obtained by this method (2.8%) was within the 95% confidence intervals of the values obtained by divers and from video mosaics using the point-count method

macroalgae (erect macroalgae and crustose coralline algae) and sponges did show significant differences among survey methodologies ($p < 0.05$). However, when macroalgae categories are grouped together into a single macroalgae group, no significant differences were found among survey methodologies ($p > 0.05$).

The coral cover value obtained by digitizing the boundaries of all of the coral colonies within the area imaged by the high-resolution mosaic (2.8%) was within the 95% confidence intervals of the values obtained by divers and from video mosaics using the point-count method (Table 2; Fig. 6).

Lastly, while the mean abundance of juvenile corals (<4 cm in diameter) documented by divers during visual surveys were 1.1 and 1.4 juveniles m^{-2}, no juvenile corals were detected from the mosaics.

When the accuracy of the two methods was compared using the AE, significant differences were found among the size categories, with AE increasing with colony size and height (ANOVA, $p < 0.01$) (Table 3). However, no significant differences were documented based on survey method (ANOVA, $p > 0.05$).

3.4 Change detection

The removal of coral colonies or other benthic organisms and changes in the composition of the substrate can be easily discerned by looking at the same section of the reef (Fig. 7). Using this method, the mortality or removal of four coral colonies (out of 50 colonies) was documented between 2004–2005 (mosaics 1 and 3) from an area of approximately 16 m^2 (Fig. 6).

4 Discussion

The use of digital imagery in benthic monitoring has increased dramatically in the last decade and video surveys are now routinely conducted as complements to diver-based measurements (Carleton and Done, 1995; Ninio *et al.*, 2003; Page *et al.*, 2003). Moreover, several large-scale monitoring programs are now based almost exclusively on the analysis of video imagery. One such example is the Coral Reef Monitoring Program of the Florida Reef Tract where permanent belt transects are surveyed annually and video frames are sub-sampled to obtained estimates of coral cover and condition (Porter *et al.*, 2002). The methodology presented here provides an important improvement over this technique by constructing referenced, spatially accurate landscape images of the benthos at a scale of up to 400 m^2 from which spatial distribution patterns and size measurements can be extracted.

Table 3 Comparison of coral size measurements between: (1) two divers measuring the same colonies; and (2) between diver measurements and measurements of the same colonies obtained directly from the video mosaics. AE_1 = absolute error = (|Diver 1 − Diver 2|), RAE_1 = relative absolute error = [(|Diver 1 − Diver 2|)/Diver 1]. AE_2 = absolute error = (|Diver 1 − Mosaic|), RAE_2 = relative absoluteerror = [(|Diver 1 − Mosaic|)/Diver 1]. Measurements taken by Diver 1 (Lirman) were considered here as the standard against which all other measurements were compared. Values reported are means (±S.D.)

	Diver-Diver comparison$_1$			Diver-Mosaic comparison$_2$		
Coral sizes (cm)	AE_1	RAE_1	N	AE_2	RAE_2	N
<10	0.7 (0.3)	8.9	9	1.6 (0.4)	21.0	22
10–20	1.9 (0.7)	10.6	15	2.5 (0.4)	16.5	45
>20–30	4.8 (1.2)	17.7	7	3.4 (0.8)	14.2	19
>30–80	5.4 (2.7)	11.1	7	5.6 (1.4)	13.1	20

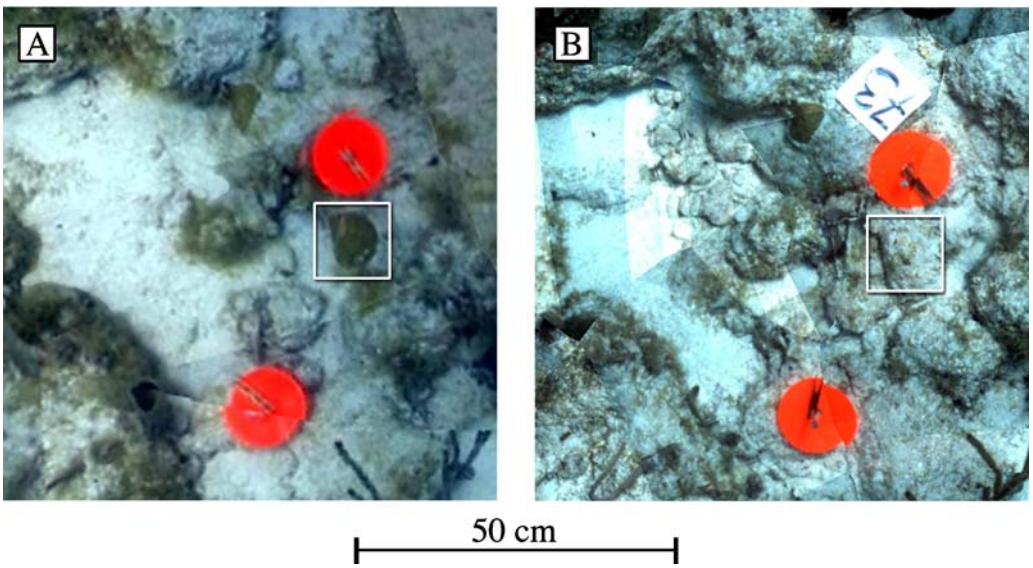

Fig. 7 Referenced mosaic sub-sections or tiles used to assess patterns of change in the abundance and distribution of benthic organisms between 2004 (A) and 2005 (B). The box highlights the removal or mortality of a small (<10 cm in diameter) coral colony between surveys

The ecological indicators collected by trained divers *in situ* compared favorably with those measured directly from the video mosaics. Percent cover of the dominant benthic organisms on reefs of the Florida Reef Tract was characterized well from the video mosaics compared to diver-based measurements. Estimates of bottom cover of hard corals, octocorals, sponges, the encrusting zoanthid *Palythoa*, and sand were statistically similar to values collected *in situ* by trained divers, while significant differences were found between the percent cover of the three dominant macroalgal groups estimated by the different methods. This pattern is a direct consequence of the increased difficulty in assigning points to these categories with decreasing image resolution. Not surprisingly, the categories that are consistent among methods are those that are the easiest to identify in the field and from photographs due to their shape, color, and clear boundaries. In contrast, those categories that have ill-defined boundaries and subdued coloration showed the highest variability among methods. Lastly, a major limitation of video-mosaic surveys is the ability to detect and identify juvenile corals (<4 cm in diameter). These small corals are often found on cryptic habitats and can only be seen in visual surveys where the observer can shift the angle of view. Future improvements in camera resolution will enhance the detection capabilities of this technique and facilitate the classification of additional benthic categories and smaller organisms.

The capability of identifying individual coral colonies and measuring their size directly from each mosaic is one of the most important benefits of this novel technique. While the accuracy of the mosaic measurements relative to the diver-based measurements was influenced by colony size, these patterns result from the difficulty that divers commonly encounter while trying to measure coral colonies in the field. Colony boundaries are easily distinguished in small (<20 cm) colonies that commonly exhibit circular shapes, but larger colonies with irregular shapes pose a challenge for divers trying to delimit live tissue boundaries. Future improvements in the 3D representation of benthic mosaics are expected to substantially improve the accuracy of this technique with respect to the measurement of larger colonies with more complex topographies (Negahdaripour and Madjidi, 2003; Nicosevici *et al*., 2005).

Previous research on the design of field programs aimed at documenting patterns of change in benthic resources over time has highlighted the increased statistical power gained by surveying precise specific locations repeatedly compared to the survey of random locations (Van de Meer, 1997; Ryan and Heyward, 2003). The demarcation of permanent plots on hard benthic substrate is commonly achieved by attaching pipes or nails on the bottom, and the number of markers needed to mark multiple colonies, quadrats, or transects at a given site can be quite large. Video mosaics provide an alternative to these labor-intensive methods. By placing a limited number of permanent markers to provide a reference frame within each video mosaic (only four permanent markers were used in this study to accurately survey an area of 400 m^2), the technique described in this study can reduce significantly the bottom-time needed to collect ecological information in the field. Moreover, by providing the ability to survey specific sub-plots repeatedly within a larger area of the benthos, video mosaics provide increased statistical power to detect small changes in abundance, cover, and size of benthic organisms. However, a trade-off exists between within-site precision and the ability to survey large areas, making the video mosaic technique an ideal method to survey areas <500 m^2 but impractical for documenting changes in the extent and condition of benthic resources at larger spatial scales. It is expected that further improvements in the mosaicing algorithms combined with the use of improved positioning modalities (e.g., acoustic transponder networks) will make this technique practical at larger scales in the near future.

Another major benefit of the algorithm described here is the ability to provide landscape-level views and analytical capabilities of benthic data collected by remotely operated platforms (i.e., AUVs, ROVs). This technique can provide unique opportunities to study the spatial arrangement, condition, and sizes of benthic organisms at locations not easily accessible to scientific divers, thus providing a crucial set of tools for the study of deep benthic communities where diver bottom-times are restricted.

The analysis of mosaics constructed over two spatial dimensions has highlighted several advantages over strip mosaics constructed along a single spatial dimension. For example, the sizes of coral colonies were accurately measured from two-dimensional mosaics, even though they are typically hard to acquire from one-dimensional mosaics where only the smallest coral colonies are completely imaged along a single transect. Moreover, two-dimensional imagery from repeated surveys was accurately referenced to assist with change-detection, unlike linear transects that are exceedingly difficult to duplicate precisely over time. Two-dimensional video mosaics can provide useful tools to assess the impacts of physical sources of disturbance to shallow reefs such as boat groundings, which can cause significant localized damage to reef resources (Lirman and Miller, 2003). The spatial extent of features such as vessel grounding scars that are often too small to map using airborne or satellite-based remote sensing tools and too large to be mapped efficiently by divers, could be measured accurately from a two-dimensional video mosaic.

The ability to extract accurate distance measurements from the mosaics was evidenced by the low values calculated for the distortion indicators. Moreover, the spatial accuracy of the video mosaics presented here was similar or lower than the measurement uncertainty of diver measurements, which typically exhibits a standard deviation of 5 cm (Holt, 2000). While an improvement in camera resolution resulted in a reduction in spatial distortion, the higher distortion of the low-altitude mosaic highlighted a present limitation of the mosaic algorithm. The sources that contribute to spatial distortions in mosaics include: (1) departures from the model assumption of a flat environment; (2) amount of superposition among strips during the acquisition; (3) limited visibility underwater; (4) limited resolution of

the imaging sensors; and (5) limited accuracy of the image matching algorithm. The higher distortion recorded for the third mosaic, collected closest to the bottom, can be likely attributed to the fact that the scene's surface planarity assumptions were clearly violated at the low altitude at which the video sequence was collected and indicates that further testing is needed to determine the minimum distance to the bottom for which the 2D mosaicing algorithm can produce useful results.

In conclusion, two-dimensional video mosaics could be widely adopted as a component of reef monitoring and damage assessment programs. The flexible mosaicing algorithm developed for this study allows this technique to be used in a variety of applications from low cost surveys with handheld video cameras to mapping of deep reefs with ROV-based platforms. Two-dimensional video mosaics can fill an information gap for scientists and resource managers by providing repeatable, accurate measurements on the reef-plot scale that can complement measurements on the colony-scale made by divers as well as surveys conducted over regional scales from remote sensing platforms.

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SHORT COMMUNICATION

Documenting hurricane impacts on coral reefs using two-dimensional video-mosaic technology

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Keywords
Acropora palmata; Florida; hurricane damage; reef framework damage; video-mosaics.

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Abstract

Four hurricanes impacted the reefs of Florida in 2005. In this study, we evaluate the combined impacts of hurricanes Dennis, Katrina, Rita, and Wilma on a population of *Acropora palmata* using a newly developed video-mosaic methodology that provides a high-resolution, spatially accurate landscape view of the reef benthos. Storm damage to *A. palmata* was surprisingly limited; only 2 out of 19 colonies were removed from the study plot at Molasses Reef. The net tissue losses for those colonies that remained were only 10% and mean diameter of colonies decreased slightly from 88.4 to 79.6 cm. In contrast, the damage to the reef framework was more severe, and a large section (6 m in diameter) was dislodged, overturned, and transported to the bottom of the reef spur. The data presented here show that two-dimensional video-mosaic technology is well-suited to assess the impacts of physical disturbance on coral reefs and can be used to complement existing survey methodologies.

Problem

During the summer of 2005, an unprecedented sequence of four hurricanes impacted the reefs of the Florida Keys. Damage patterns to coral reefs are commonly influenced by the strength, path, and duration of each storm event (Harmelin-Vivien 1994; Lirman & Fong 1997; Lirman 2000). In the case of sequential storms, damage patterns can be also determined by storm frequency and prior disturbance history (Witman 1992). When the time required for live coral fragments to re-attach to the bottom and for loose rubble to stabilize exceeds the interval between storms, physical impacts can be compounded as loose pieces of coral rubble are mobilized by subsequent storms (Lirman & Fong 1997). The impacts of storms on coral colonies are often influenced by colony morphology, and the branching morphology of corals like *Acropora* spp. makes them especially susceptible to physical disturbance (Woodley *et al.* 1981). In fact, hurricane damage and coral diseases have been identified as the main source of mortality to acroporids in the Caribbean region, where this taxon has undergone such a drastic decline in abundance that the U.S. NOAA Fisheries Service has proposed listing *Acropora palmata* and *A. cervicornis* as 'threatened' species under the U.S. Endangered Species Act (Bruckner 2002; Oliver 2005; Precht *et al.* 2005).

The cumulative effects of the 2005 storms on one of the last remaining populations of *A. palmata* in the northern Florida Reef Tract were assessed with a newly developed survey methodology that is used to construct spatially accurate, high-resolution landscape mosaics of the reef benthos. Video-mosaics provide a complement to

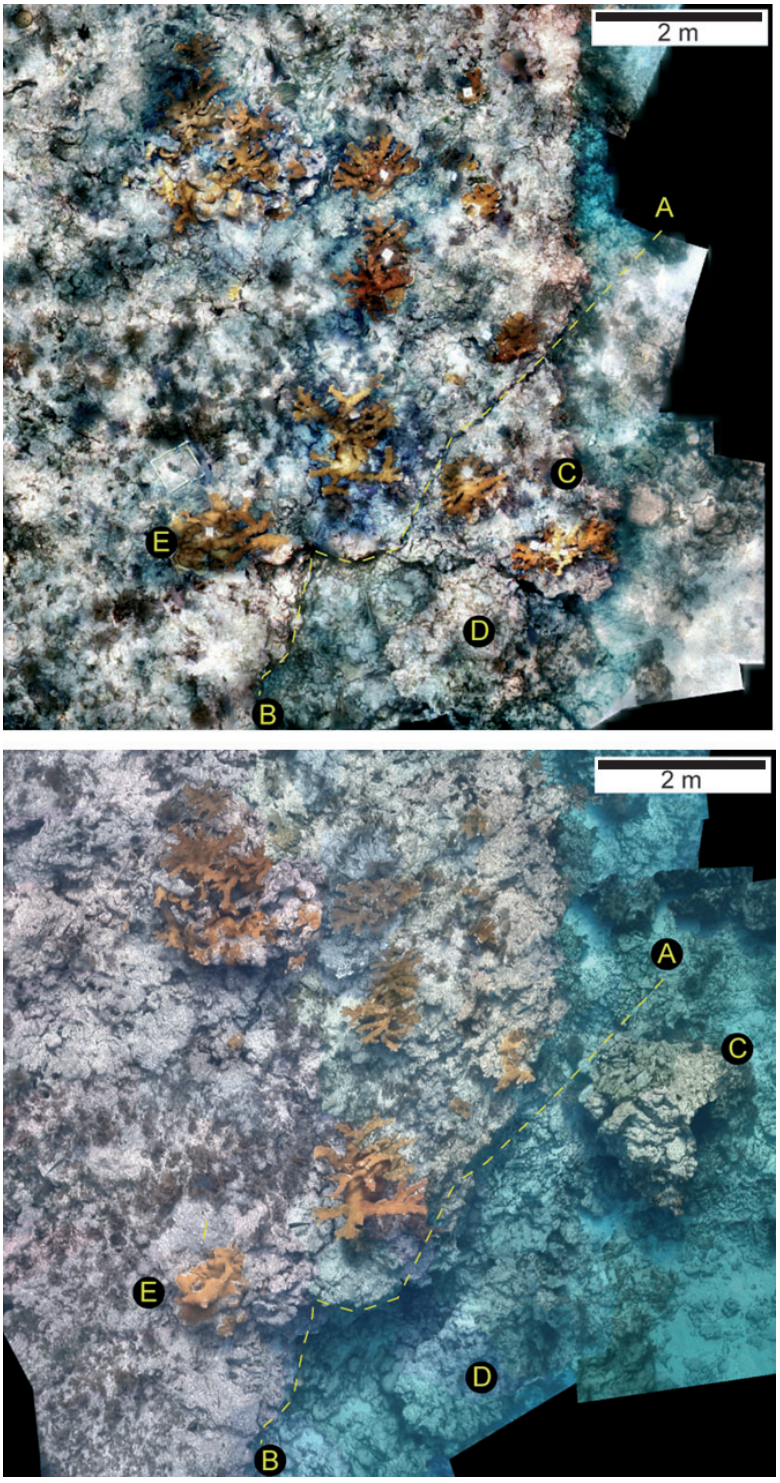

Fig. 1. Two-dimensional video-mosaics from a study plot at Molasses Reef in the Florida Reef Tract (depth 3.5–4.5 m). (Top) Mosaic from May 2005 was constructed prior to the start of the 2005 hurricane season. (Bottom) Mosaic from February 2006 following the passage of four hurricanes. The yellow line A–B shows where the reef framework was dislodged during hurricane Rita causing sections of the reef marked C and D to collapse. The section labeled C also appears in Fig. 2A. The *Acropora palmata* colonies located on section C are shown in Fig. 2B. Close-ups of the *A. palmata* colony labeled E appear in Fig. 2C and D.

standard diver-based survey methods, which require a high level of training and extended time underwater. Moreover, two-dimensional mosaics cover larger areas than one-dimensional 'strip' mosaics (Jaap *et al.* 2003) thereby allowing new types of analyses such as measuring the sizes of coral colonies and visualizing large features on the reef (Lirman *et al.* 2006).

Material and Methods

In this study, we used video-mosaic technology to document hurricane impacts on a population of the branching coral *A. palmata* at Molasses Reef (25° 0.609 N, 80° 22.397 W, depth = 3.5–4.5 m). Mosaics of the study plot (approximately 10 m × 10 m) were constructed from underwater video collected at 2 m from the bottom using a Sony TRV900 DV camcorder. The mosaicing algorithm is described in detail by Gracias *et al.* (2003), Negahdaripour & Madjidi (2003), and Lirman *et al.* (2006). Briefly, the method has four steps: (1) acquire the video in a series of parallel, overlapping swaths covering the study area; (2) estimate the image-to-image motion between pairs of sequential images to calculate an estimate of the camera trajectory; (3) refine the estimated camera trajectory by estimating motion between non-sequential but overlapping images; and (4) produce a single image by blending contributions from the individual frames. The mosaics constructed for this study have a ground resolution of 1–2 mm per pixel and coral colonies or fragments >5 cm in diameter are easily identified within each image.

Video data were collected before the passage of the hurricanes at Molasses Reef in May 2005 and again in February 2006 after hurricanes Dennis (dates of influence over the Florida Keys = July 9–10, 2005, peak wind gusts at Molasses Reef (C-MAN station) = 90 km h^{-1}), Katrina (August 25–26, 2005, 116 km h^{-1}), Rita (September 19–20, 2005, 100 km h^{-1}), and Wilma (October 24–25, 2005, 147 km h^{-1}). The video required to build the mosaics of the study plot was collected in <30 min, and production of the mosaics required approximately 10 h using a standard personal computer.

Landscape video-mosaics such as the ones produced in this study have high spatial accuracy (standard deviations of the residues = 4–5.5 cm, maximum distance error <14 cm) and thereby provide the capability to measure distances and sizes directly from the images once a scale has been established (Lirman *et al.* 2006). The scale in these mosaics is provided by PVC segments and ceramic tiles scattered throughout the images. The size of the *A. palmata* colonies found within each mosaic was measured as: (1) the maximum colony diameter (to the closest cm); and (2) the projected surface area of live tissue. The image-analysis software ImageJ was used to calculate these metrics.

Results and Discussion

The direct physical damage caused by hurricanes and tropical storms can vary significantly across scales, ranging from minimal to severe (Harmelin-Vivien 1994). Whereas changes in coral cover, abundance, and condition can be easily discerned from traditional before-and-after surveys, changes to the structure of reefs are harder to quantify. The video mosaics created in this study provide a unique view of the reef benthos that facilitates the

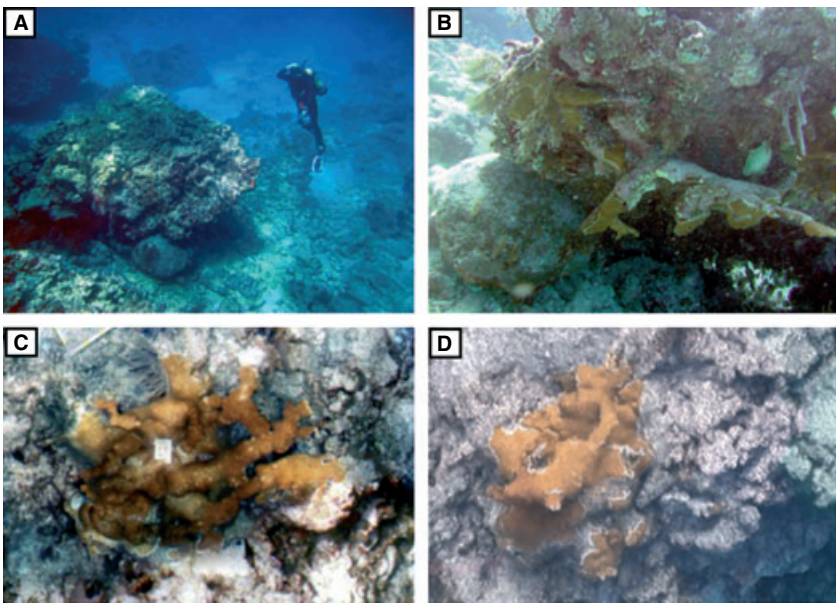

Fig. 2. A: Photograph of the reef section (labeled C in Fig. 1) that was dislodged during Hurricane Rita. B: Photograph of two *A. palmata* colonies attached to the dislodged reef section shown in A. These colonies ended up facing the sediments and died shortly after the storm. C: May 2005 and D: February 2006 photographs of an *A. palmata* colony (labeled E in Fig. 1) that experienced fragmentation and tissue losses due to the 2005 hurricanes.

documentation of colony-level impacts as well as large-scale structural changes to the reef framework.

If only coral cover and colony-based information such as abundance and size-structure had been collected prior to the onset of the 2005 hurricane season, the damage report for the *A. palmata* population at Molasses Reef after the passage of four major storms would have revealed, unexpectedly, only limited damage considering the intensity and frequency of the 2005 hurricanes. A total of 19 *A. palmata* colonies were identified from the video mosaic from May 2005, prior to the onset of the 2005 hurricane season, and 17 of these colonies remained, in the same location, in the study plot in February 2006 (Fig. 1). The two colonies that were removed from the plot were located on one of the sections of the reef framework that was dislodged during Hurricane Rita (Fig. 2A). These two colonies remained attached to the dislodged reef section but ended up in contact with bottom sediments and died shortly after this storm (Fig. 2B). The tissue on these large colonies (110 and 155 cm in maximum diameter) represented 14% of the total live *Acropora* tissue on the plot prior to the storms. For those colonies that remained, the net tissue losses between surveys were only 10%. Fifty-two percent of colonies lost live tissue, the maximum tissue loss for an individual colony was 46%. The mean diameter of colonies decreased slightly from 88.4 cm (SD ±70.1) to 79.6 (±63.3) cm. Tissue losses were mainly attributed to the removal of branches (Fig. 2C and D).

An increase in the abundance of colonies through fragment formation and reattachment after storms has been documented previously for *A. palmata* in Florida (Fong & Lirman 1995) but was not observed within the study plot at Molasses Reef. Fragment reattachment requires a minimum amount of time (Lirman 2000) and the succession of storms during the summer of 2005 may have impeded this process.

Considering the limited impacts documented for coral colonies at Molasses Reef, one of the most remarkable impacts of the 2005 hurricanes was the damage caused to the reef framework. Within the study plot, a large section of the reef (surface area = 12.7 m^2, diameter = 6 m) was dislodged and deposited on the sand at the bottom of the reef spur (Figs 1 and 2A). The shift in orientation of these sections resulted in the smothering and burial of coral colonies and the exposure of reef framework that may be further weakened by the future activities of bioeroders (Glynn 1988). The precise documentation of such large-scale modifications to the structure of the reef was only possible because of the landscape view provided by the video-mosaics.

The methods used to assess damage and recovery patterns of reef communities commonly entail the construction of underwater maps of the benthos based on diver-collected distance measurements and drawings, and the deployment of survey markers and permanent tags for coral colonies within plots. Assessing the impacts of severe physical disturbance on coral reefs can be especially challenging when large-scale modifications to the reef structure and the removal of both coral colonies and survey markers take place, as is commonly seen not only after storms but also after ship groundings (Hudson & Diaz 1988; Jaap 2000). The data presented in this study show that landscape video-mosaics provide the tools needed to accurately assess reef damage and recovery patterns and provide a significant addition to the existing survey techniques.

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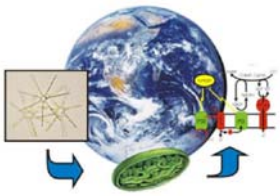
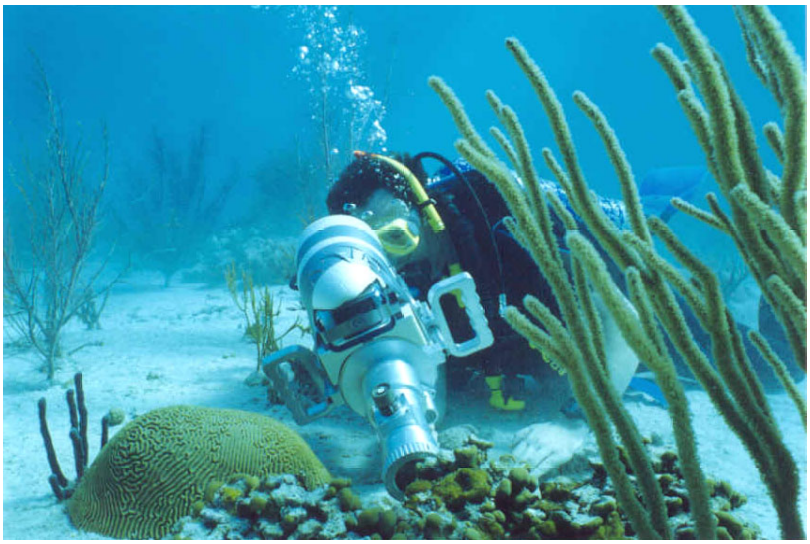
Analysis of Biophysical, Optical and Genetic Diversity of Coral Reef Communities using Advanced Fluorescence and Molecular Biology Techniques

Coral reefs are specifically susceptible to anthropogenic insult and rapidly degrade worldwide. The development of advanced technologies for environmental monitoring and assessment of benthic ecosystems requires an understanding of how different environmental factors affect the key elements of the ecosystems and the selection of specific monitoring protocols that are most appropriate for the identification and quantification of particular stresses. The objectives of this SERDP project are (1) to develop advanced techniques and protocols for rapid and non-destructive assessment of the viability and health of coral reef communities with the capabilities of identification of natural and anthropogenic stressors, (2) develop prototype bio-optical instruments for permanent underwater monitoring stations and Remotely Operated Vehicles, (3) collect a library of baseline data on physiological and genetic diversity of coral reef communities in the Caribbean and the Indo-Pacific regions.

Because photosynthesis is the ultimate source of energy for all shallow water communities, photosynthetic organisms are absolutely critical components in the viability of coral reef ecosystems. Corals are symbiotic associations between an invertebrate host and a photosynthetic alga, called zooxanthellae. Assessment of the physiological state of the photosynthetic organisms relies on the measurement and analysis of chlorophyll variable fluorescence, a property unique to the photosynthetic processes. The fluorescence emission is coupled to the photosynthetic processes and is particularly sensitive to environmental factors and stressors, including nutrient availability, irradiance, temperature, and anthropogenic insults. This provides a biophysical background for non-invasive fluorescence monitoring of the organisms.

A novel technology, called Fluorescence Induction and Relaxation (FIRe) technique, has been invented for measuring a comprehensive set of photosynthetic characteristics in corals and other benthic organisms (Gorbunov and Falkowski, 2005). The bio-optical measurements are sensitive, fast, non-destructive, and are conducted in real time underwater. Bench-top, diver-operated, and moorable instruments have been designed and developed. The bench-top FIRe System has been transferred to a small hi-tech company, Satlantic Inc. (www.satlantic.com/fire). The biophysical and biochemical research elucidated the impact of common natural stressors (such as elevated temperature and excess light) and selected anthropogenic stresses (heavy metal contamination) on coral physiology. The cellular and molecular mechanisms, together with the optical signatures of the stresses have been established (Tchernov et al, 2004). The lab and field research revealed that the FIRe parameters are very sensitive to changes in the coral physiology and alert detrimental changes at early stages of the stress development - before any visible changes in coral coloration appear. On this background, bio-optical algorithms for detection and assessment of the stresses have been developed and evaluated. This R&D project provides quantitative baseline data, as well as advanced methods and technology for the monitoring and assessment of coral reef ecosystems.





Left – Bench-top Fluorescence Induction and Relaxation (FIRe) System.
Right – Non-destructive assessment of the health of benthic organisms by using an underwater fluorometer.

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FLUORESCENCE INDUCTION AND RELAXATION (FIRE) TECHNIQUE AND INSTRUMENTATION FOR MONITORING PHOTOSYNTHETIC PROCESSES AND PRIMARY PRODUCTION IN AQUATIC ECOSYSTEMS

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Keywords: variable fluorescence, primary production, chlorophyll, phytoplankton, electron transport

INTRODUCTION

Over the last decade, the Fast Repetition Rate Fluorometry (FRRF, see Kolber et al 1998, Gorbunov et al 1999, 2000) provided tremendous insight into the factors controlling phytoplankton distributions and primary production in the ocean (e.g., Behrenfeld et al 1996, Boyd et al 2000, Falkowski & Kolber 1995, Kolber et al 1994, 2001). The use of the FRRF became an integral part of many biological oceanographic programs, but its broader use is limited by complexity and high cost of the available instrumentation. We have designed and built a new instrument, called Fluorescence Induction and Relaxation (FIRe) System, to measure a comprehensive suite of photosynthetic characteristics in phytoplankton, benthic organisms (macrophytes, corals, seagrass), and higher plants. The FIRe technique is based on similar biophysical principles as the FRRF and provides the same physiological characteristics. But the optical design has been improved, the electronic circuitries simplified, and the operational protocols extended. This permitted for the sensitivity to be enhanced and the production cost to be greatly reduced. A bench-top version of the FIRe System is used for measurements on phytoplankton or leaves. The compact design, low power consumption, and network capability of a submersible version of the FIRe System make it a robust sensor for long-term monitoring programs in coastal zones and the open ocean. Here we report the design of the FIRe System and present its first applications to study photosynthetic processes in phytoplankton and corals.

MATERIALS AND METHODS

The FIRe technique relies on active stimulation and highly resolved detection of the induction and subsequent relaxation of chlorophyll fluorescence yields on micro- and millisecond time scales (Fig. 1). To accommodate efficient excitation of diverse functional groups within phytoplankton communities including a variety of cyanobacteria, we have developed a multicolor excitation source. This source uses high luminosity blue (450 nm and 480 nm, each with 30 nm bandwidth) and green (500 nm and 530 nm, each with 30 nm bandwidth) light-emitting diodes (LEDs) to excite chlorophyll and bacteriochlorophyll fluorescence *in vivo*. A computer-controlled LED driver circuitry generates pulses with the duration varied from 0.5 μs to 50 ms. Each LED generate up to 1 W/cm^2 of peak optical power density in the sample chamber or at the leaf surface to ensure fast saturation of PSII within the single photosynthetic turnover (less than 50 μs).

The fluorescence signal is isolated by red (680 nm or 730 nm, each with 20 nm bandwidth, for Chl-a fluorescence) or infra-red (880 nm with 50 nm bandwidth, for BChl-a fluorescence) interference filters and detected by a sensitive avalanche photodiode module. A small portion of the excitation light is recorded by a PIN photodiode as a reference signal. Both the fluorescence and reference signals are amplified and digitized by 12-bit analog-to-digital converters at 1 MHz sampling rate by a custom-designed data acquisition board. To accommodate a wide range of Chl-a concentrations (0.01 to 100 mg/m3) in natural phytoplankton and laboratory cultures, the gain of the detector unit is automatically adjusted over the range of three orders of magnitudes. An embedded low-power Pentium-based board controls the excitation protocols and data acquisition and performs the real-time data analysis using a custom analysis toolbox.

An example of the FIRe protocol incorporating both Single (STF) and Multiple Turnover Flashes (MTF) is shown in Fig. 1. Analysis of fluorescence induction on microsecond time scales (Fig. 1, Phase 1) provides the minimum (F_o) and maximum (F_m) fluorescence yields, the quantum efficiency of photochemistry in PSII (F_v/F_m), the functional absorption cross-section of PSII (σ_{PSII}), and the energy transfer between PSII units ('connectivity factor', p). The recorded relaxation kinetics of fluorescence yields reflects the rates of electron transport on the acceptor side of PSII and between PSII and PSI. The photosynthetic electron transport rates as a function of irradiance, together with coefficients of photochemical and non-photochemical quenching are measured using an incorporated source of background light. The design of the electronic circuitries and operational software are extremely flexible and permit for additional excitation protocols to be implemented, including classical Kautsky induction, the FRR, pump-and-probe, pulse amplitude modulation, and potentially other protocols. The bench-top FIRe System permits the user to perform measurements on phytoplankton (on discrete samples or in flow-throw) and benthic organisms and higher plants (by using a fiber-based extension probe).

RESULTS AND DISCUSSION

The FIRe System was employed during two oceanographic cruises in the Sargasso Sea (June–August 2004) to study the impact of mesoscale eddies on primary production and the export of carbon into the ocean interior (see Bibby et al 2004 for detail). The results revealed that the cyclonic eddy-induced isopicnal displacement (i.e., upwelling of cold nutrient-rich waters) increases both Chl-a and photosynthetic efficiency in the euphotic zone (Fig. 2). The eddy-induced upwelling produced minute, but readily detectable changes in F_v/F_m (Fig. 2B). Although the eddy upwelling increases the concentration of major nutrients only at depth (~100 m and deeper), the increase in F_v/F_m was significant even at the surface (Fig. 2B). This pattern was consistently observed at most of the stations (N = 40) and suggests the sustained flux of nutrients into the surface layers, but the underlying physical mechanisms and the biogeochemical implications remain to be elucidated.

The development of submersible FIRe fluorosensors is conducted within the framework of the Strategic Environmental Research and

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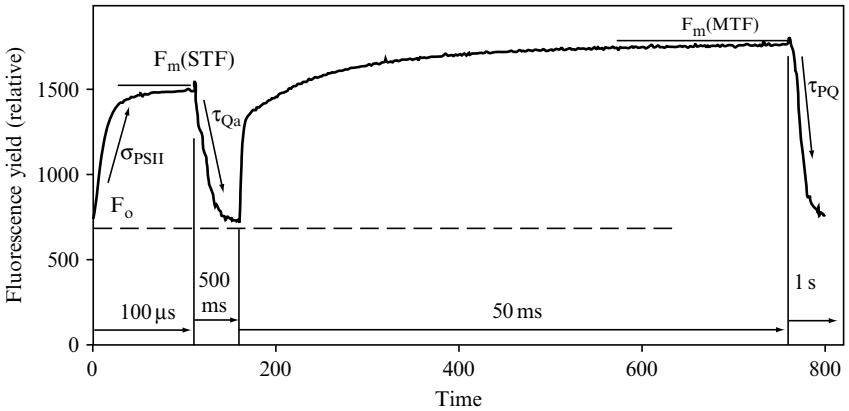

Figure 1: An example of the FIRe measurement protocol consisting of four phases: (1) a strong short pulse of 100 µs duration (called Single Turnover Flash, STF) is applied to cumulatively saturate PSII and measure the fluorescence induction from F_o to F_m(STF); (2) weak modulated light is applied to record the relaxation kinetics of fluorescence yield on the time scale of 500 ms; (3) a strong long pulse of 50 ms duration (called Multiple Turnover Flash, MTF) is applied to saturate PSII and the PQ pool; (4) weak modulated light is applied to record the kinetics of the PQ pool re-oxidation the time scale of 1 s. Analysis of the Phase 1 provides F_o, F_m, F_v/F_m(STF), σ_{PSII}, p; Phase 2 – time constants for the electron transport on the acceptor side of PSII (i.e., re-oxidation of the Qa acceptor); Phase 3 – F_m(MTF) and F_v/F_m(MTF); Phase 4 – the time constant for the electron transport between PSII and PSI (re-oxidation of the PQ pool).

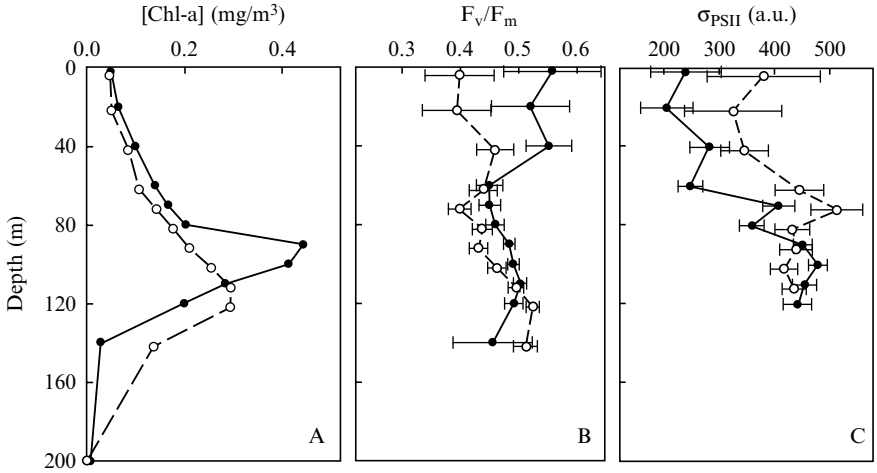

Figure 2: The effect of eddy-induced nutrient pumping on phytoplankton photosynthesis in the Sargasso Sea, assessed with FIRe fluorometry. Vertical profiles of (A) Chl-a concentration, (B) the quantum yield of photochemistry in PSII, and (C) the functional absorption cross-section of PSII measured at two stations with deep (outside the eddy, open dots) and shallow (inside the eddy, closed dots) nitrocline.

Development Program (SERDP) initiative on "Assessment of Benthic Communities at Department of Defense Installations". The objectives of our SERDP project include the development of bio-optical techniques for rapid and non-destructive assessment of the viability and health of coral reef communities and the development of submersible fluorosensors for permanent underwater observatories and Remote Operated Vehicles (http://www.serdp.org/research/cs/cs-1334.pdf).

Coral reef ecosystems are particularly susceptible to environmental changes caused by anthropogenic influences and rapidly degrade worldwide. Over the last decade, massive bleaching events of zooxanthellate corals have been occurred, bringing devastating impacts to the ecosystems. This phenomenon is triggered by small (~1 °C) increases in water temperature and starts with the impairing the photosynthetic processes in endosymbiotic zooxanthellae, but the underlying biophysical mechanisms remain poorly understood. The FIRe fluorometers, in combination with standard biochemical techniques, have been employed to elucidate the mechanisms of thermal stress and coral bleaching (see Tchernov et al 2004 for detail). The research revealed that the thermal sensitivity correlates with the lipid composition of the thylakoid membranes in symbiotic algae and is determined by the saturation of membrane lipids (Tchernov et al 2004). The thermal stress starts with disruption of the membranes, followed by impairing of the photosynthetic machinery, including PSII units. This damage is irreversible and ultimately results in cell death. The FIRe analysis revealed that the stress development is accompanied by unique variable fluorescence signatures and different from photoinhibition. Although both stresses lead to a characteristic decrease in the quantum yield of photochemistry in PSII (F_v/F_m), only thermal stress was accompanied with a striking increase in the time constant of Q_a re-oxidation, suggesting stress-specific modifications in the electron transport chain on the acceptor side of PSII. The data suggest that the FIRe technique can be used to selective identification of stresses. These approaches

can be readily used for bio-monitoring of all groups of aquatic photosynthetic organisms and we envision that the developed technology will be employed in a variety of environmental monitoring programs.

ACKNOWLEDGMENTS

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Membrane lipids of symbiotic algae are diagnostic of sensitivity to thermal bleaching in corals

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Over the past three decades, massive bleaching events of zooxanthellate corals have been documented across the range of global distribution. Although the phenomenon is correlated with relatively small increases in sea-surface temperature and enhanced light intensity, the underlying physiological mechanism remains unknown. In this article we demonstrate that thylakoid membrane lipid composition is a key determinate of thermal-stress sensitivity in symbiotic algae of cnidarians. Analyses of thylakoid membranes reveal that the critical threshold temperature separating thermally tolerant from sensitive species of zooxanthellae is determined by the saturation of the lipids. The lipid composition is potentially diagnostic of the differential nature of thermally induced bleaching found in scleractinian corals. Measurements of variable chlorophyll fluorescence kinetic transients indicate that thermally damaged membranes are energetically uncoupled but remain capable of splitting water. Consequently, a fraction of the photosynthetically produced oxygen is reduced by photosystem I through the Mehler reaction to form reactive oxygen species, which rapidly accumulate at high irradiance levels and trigger death and expulsion of the endosymbiotic algae. Differential sensitivity to thermal stress among the various species of *Symbiodinium* seems to be distributed across all clades. A clocked molecular phylogenetic analysis suggests that the evolutionary history of symbiotic algae in cnidarians selected for a reduced tolerance to elevated temperatures in the latter portion of the Cenozoic.

Coral bleaching on a global scale is a growing concern because of both the reduction in essential ecological services provided by zooxanthellate corals within reef communities (1, 2) and the potentially devastating economic impacts accompanying the phenomenon (3). Small, positive deviations in temperature of <2°C can trigger massive losses of symbiotic algae, *Symbiodinium* spp., from their cnidarian host cells (4). However, not all corals within a reef are equally susceptible to elevated temperature stress (5, 6). Although elevated temperatures often lead to a reduction in the quantum yield of photochemistry, a concomitant increase in the rate of protein turnover in oxygen-generating reaction center, photosystem (PS)II (7–9), and an increase in the production of reactive oxygen species (ROS) (10–12), no mechanism has been elucidated. Here we show that thermal sensitivity in isolated clones of zooxanthellae and in symbiotic animal hosts is correlated with the degree of saturation of the lipids in the thylakoid membranes in the algal plastids. Our results provide a mechanistic basis for understanding and diagnosing coral bleaching patterns in nature.

Materials and Methods

Cultures and Corals. Cultures of *Symbiodinium* spp., obtained from culture collections or isolated from hosts, were grown in F/2 medium under a 10/14-h light/dark cycle and illuminated with 100 μmol quanta $m^{-2}\cdot s^{-1}$. Corals were grown at 26°C in 800 liters of aquaria with running artificial seawater (Instant Ocean sea salt, Aquarium Systems, Mentor, OH) as described (13). For thermal-stress experiments, duplicate colonies were transferred to 300 liters of aquaria that were heated to 32°C and maintained at that temperature for 2 months or until the colonies died. Light, at 200 μmol quanta $m^{-2}\cdot s^{-1}$ on a 12/12-h light/dark cycle was provided by 400-W metal halide bulbs (Iwasaki Electric, Tokyo). Nutrients (NO_3^-, NO_2^-, NH_4^+, and PO_4^{3-}) were kept at submicromolar concentrations by foam fractioning and biological filtration (e.g., live sand).

Variable Fluorescence. Variable chlorophyll fluorescence kinetic transients were measured with a custom-built fast repetition-rate fluorometer using protocols described by Kolber *et al*. (14).

Lipid Analysis. Lipids were saponified, methylated, and extracted into hexane/methyl tertiary butyl ether as described (15). Fatty acid methyl esters were analyzed by GC/MS with an Agilent series 6890 GC system and 5973 mass selective detector, equipped with an HP5MS capillary column (i.d., 30 m × 0.25 mm; film thickness, 0.25 μm) with helium as the carrier gas.

Membrane Inlet MS. Light-dependent production and consumption of oxygen was measured by using a membrane inlet system attached to a Prisma QMS-200 (Pfeiffer, Nashua, NH) quadruple mass spectrometer with closed ion source recording at mass/charge (m/z) ratios of 32 ($^{16}O^{16}O$), 36 ($^{18}O^{18}O$), and 40 (Ar). The membrane inlet system was modified from a water-jacketed DW/2 oxygen electrode chamber (Hansatech Instruments, Pentney King's Lynn, U.K.) in which the electrode base plate was replaced by a stainless-steel base plate with a gas port drilled through the center. The standard Teflon membrane (thickness, 12.5 μm) supplied with the DW/2 oxygen electrode system was used. Illumination was provided by a high-pressure halogen arc source at 300 μmol quanta $m^{-2}\cdot s^{-1}$. Temperature was maintained at 26°C. Oxygen signals were calibrated with O_2-saturated water and zero (plus sodium dithionite) O_2 water and normalized to Ar. Oxygen production and consumption rates were calculated by linear regression analysis.

ROS. Cultures were harvested by centrifugation and resuspended in culture medium that had been stripped of O_2 by bubbling with N_2 gas. Subsamples were incubated for 3 h at 150 μmol quanta $m^{-2}\cdot s^{-1}$ in 96-well plates in the presence of 15 μM dihydrorhodamine 123, a dye that fluoresces green in the presence of ROS (10). Fluorescence (i.e., ROS production) was measured kinetically with a plate reader (Molecular Devices) at excitation λ = 488 nm and emission λ = 525 nm.

This paper was submitted directly (Track II) to the PNAS office.

Abbreviations: PS, photosystem; ROS, reactive oxygen species; LSU, large subunit; rDNA, rRNA-encoding DNA.

Data deposition: The sequences reported in this paper have been deposited in the GenBank database (accession nos. AY684261–AY684270).

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Transmission Electron Microscopy. Cells were harvested by centrifugation (15 min at 7,000 × *g*) and fixed in cacodylate buffer containing 4% glutaraldehyde and 8.6% sucrose. Pellets were washed in a series of cacodylate buffers with descending sucrose concentration and postfixed in OsO_4 for 2 h. After dehydration in an ascending ethanol series (70–100%), samples were embedded in agar and Epon, sectioned (50-nm thickness) with a Reichert ultramicrotome, stained with uranyl acetate and lead citrate, and examined with a JEOL 100 CX transmission electron microscope.

Large Subunit rRNA-Encoding DNA (rDNA) Sequencing and Phylogenetic Analyses. Genomic DNA was extracted from zooxanthellae by using the DNeasy plant minikit (Qiagen, Valencia, CA). Standard PCR amplification of nuclear ribosomal DNA was performed by using two sets of primers: (*i*) S-DINO (cgctcctaccgattgagtga) and L-DIN-1 (aacgatttgcacgtcagtaccgc), which are *Symbiodinium*-specific and cover the ITS-1/5.8S/ITS-2/partial large subunit (LSU) rDNA, and (*ii*) D1R (acccgctgaatttaagcatat) and D2C (ccttggtccgtgttt), which are dinoflagellate-specific and target a 5′ fragment of the LSU rDNA. PCR products were purified by using shrimp alkaline phosphatase and exonuclease I and directly sequenced by using an Applied Biosystems 3100-Avant automatic sequencer.

The D1 and D2 sequences of the LSU rDNA were aligned manually to the 294 homologous gene fragments from *Symbiodinium* spp. available in GenBank. All redundant, identical se-

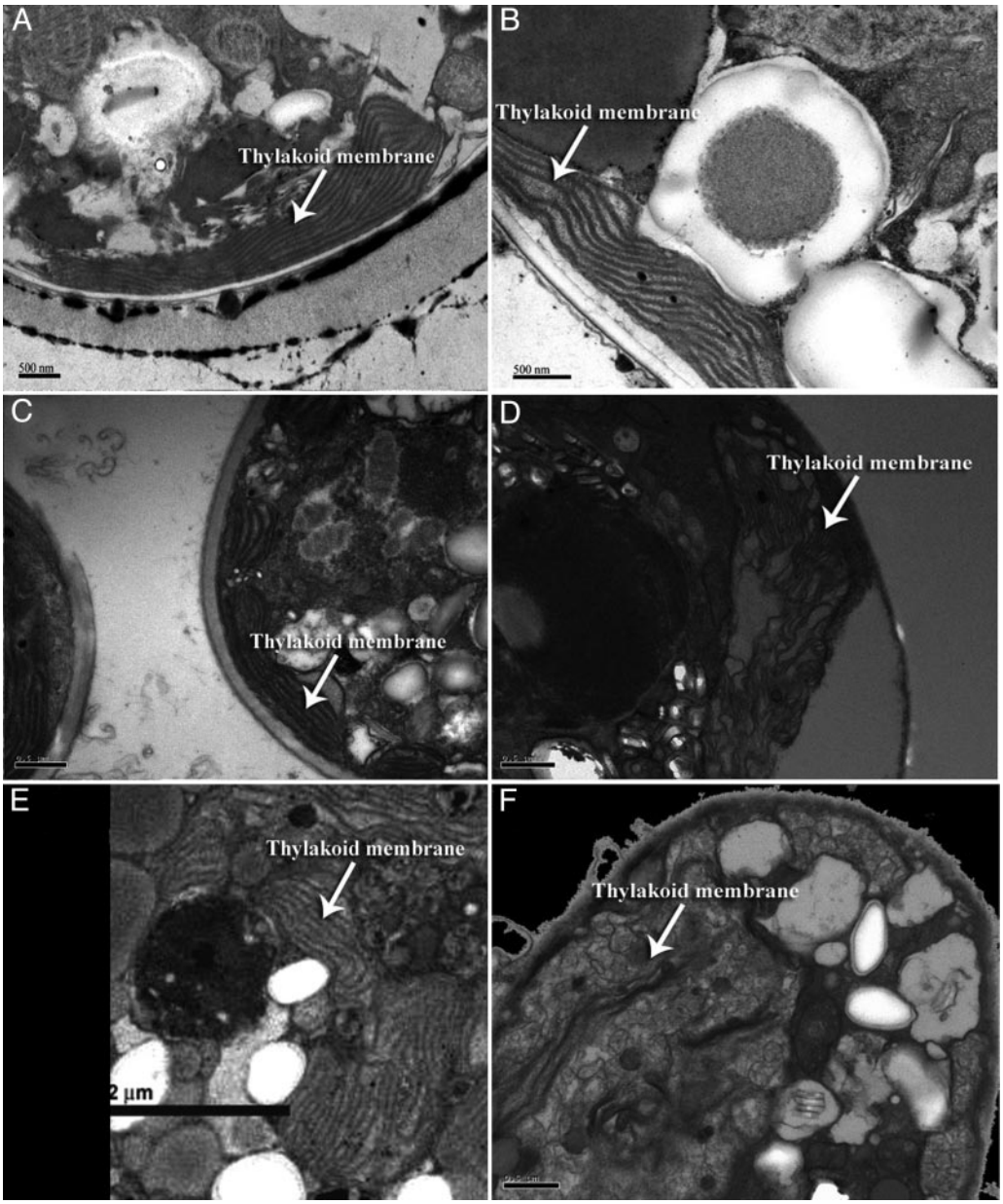

Fig. 1. Effects of elevated temperatures on the structure of thylakoid membranes in zooxanthellae. Transmission electron micrographs of thin sections of *Symbiodinium* spp. isolated from *Tridacna* spp. [Provasoli–Guillard National Center for Culture of Marine Phytoplankton (CCMP) (West Boothbay Harbor, ME) no. 828] (*A* and *B*), the sea anemone *Aiptasia* sp. (CCMP no. 831) (*C* and *D*), the coral *M. samarensis* (*E*), and the coral *S. pistillata* (*F*). Samples were incubated at 26°C (*A* and *C*) and 32°C (*B* and *D–F*). All cultures were grown in F/2 medium (36) under a 12/12-h light/dark cycle. The corals were grown in a closed system supported by a biological filtration system under a 10/14-h light/dark cycle. Note the degradation of the thylakoid membranes within the plastids of the heat sensitive strains.

quences were removed from the alignment, which resulted in a final DNA matrix containing 84 sequences and 556 nucleotide sites (297 parsimony informative characters). Hierarchical likelihood ratio tests were applied to our data set to select the most appropriate DNA substitution model: a general time-reversible model considering the proportion of invariant sites as well as rate heterogeneity among sites (γ-shaped distribution, $\gamma = 1.2581$) (16). Phylogenetic trees were inferred by using Bayesian (1 million MCMC generations, substitution model parameters = GTR+G+I), maximum-likelihood (substitution model parameters = TIM+G+I), and neighbor-joining (substitution model parameters = Tamura and Nei+G) statistics with MRBAYES, PAUP*, and LINTREE, respectively (17, 18). To give a time dimension to our tree, the 13 consensus, highly resolved clades (thick branches in the tree of Fig. 4) were tested for molecular clock deviation by using relative rate tests (20), with clade A used as an outgroup. None of the LSU rDNA *Symbiodinium* clades evolve significantly faster than others (threshold risk for 12 clades and 66 tests, $P < 0.08\%$). Consequently, we used LINTREE to infer a clock-enforced, linearized tree (see Fig. 4), which was calibrated in time by a "dinoflagellate" rate of LSU rDNA substitution based on a previously published DNA–fossil comparative data set (19).

Results and Discussion

Representative transmission electron micrographs, selected from thousands of zooxanthellae cells, revealed that when thermally tolerant clones of *Symbiodinium* spp. grown at 26°C were transferred to 32°C (a thermal stress that induces bleaching), the stacking properties and ultrastructural integrity of thylakoid membranes remained unaffected (Fig. 1 *A–C* and *E*; Table 1, which is published as supporting information on the PNAS web site). In contrast, thylakoid membranes of thermally sensitive clones subjected to the higher temperature were significantly disrupted, and the organized stacking pattern, which is essential for efficient photochemical energy transduction, was compromised (Fig. 1 *D* and *F*). This process is not reversible and was further observed in zooxanthellae *in hospite* in heat-sensitive corals cultivated in the laboratory before bleaching.

The effect of thermal stress on the photochemical energy-conversion efficiency was confirmed by fast repetition-rate fluorometer measurements (14) on a variety of isolated, cultured clones of zooxanthellae (Fig. 2). Thermally induced changes in membrane integrity were initially accompanied by both an increase in the rate of electron transport on the acceptor side of PSII and a simultaneous decrease in the maximum quantum yield of photochemistry within the reaction center (Table 2, which is published as supporting information on the PNAS web site). In energetically coupled thylakoids, the fastest component of fluorescence decay corresponds to a single electron transfer from the primary electron acceptor, Q_A, to the secondary quinone, Q_B or Q_B^- (21), and occurs with a time constant ranging from 300 to 500 μs (22). In temperature-sensitive clones of zooxanthellae, the measured time constant fell from an average of 304 ± 54 to 200 ± 46 μs, whereas in thermally tolerant clones the time constant remained statistically unchanged, averaging 318 ± 24 μs at 26°C and 341 ± 9 μs at 32°C. The marked change in electron-transfer times in thermally sensitive clones was accompanied by a 40% decrease in (but not loss of) photochemical energy-conversion efficiency in PSII reaction centers. These two phenomena are diagnostic of an energetically uncoupled system in which the transmembrane proton gradient, established by the photochemical reactions in the functional reaction centers, is dissipated without generating ATP (23). This fluorescence kinetic pattern, uniquely found in thermally sensitive zooxanthellae, qualitatively differs from photoinhibition (24–26), with which the time constant for electron transfer increases as the reaction centers become increasingly impaired (27). Moreover, in thermally sensitive clones of zooxanthellae, the pattern of change in photochemical energy conversion occurs over a very narrow thermal window of <2°C. These results not only demonstrate that high-resolution, kinetic measurements of variable chlorophyll fluorescence can be used to rapidly assess the sensitivity of zooxanthellae to thermal stress, but moreover suggest that thylakoid membrane integrity is potentially a critical determinant of thermal tolerance.

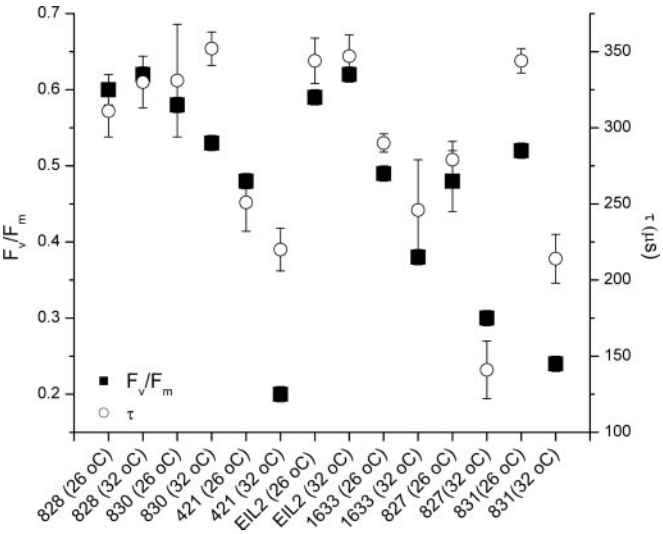

Fig. 2. Maximum quantum yields of fluorescence (F_v/F_m, dimensionless) and electron-transfer rates (τ, μs) from the primary electron acceptor in PSII, Q_A, to the secondary quinone, Q_B, for all clones of zooxanthellae. Fluorescence parameters were derived from measurements with a custom-built fast repetition-rate fluorometer (14, 24). All cultures were grown in F/2 medium; cultures were incubated for up to 224 h (to verify resilience and nonreversibility of thermally damaged cultures) under a 10/14-h light/dark cycle at 26 and 32°C for each species tested. Maximum quantum yields of photochemistry (F_v/F_m) of the thermally tolerant clones averaged 0.57 ± 0.05 at 26°C and 0.55 ± 0.01 at 32°C; the corresponding electron-transfer rates (τ) were 318 ± 24 and 341 ± 9 μs. In heat-sensitive clones, the maximum quantum yields averaged 0.50 ± 0.07 at 26°C and 0.31 ± 0.03 at 32°C; the corresponding electron-transfer rates were 304 ± 54 and 200 ± 46 μs.

We further examined the patterns of thermal sensitivity and bleaching in colonies of the zooxanthellate corals *Stylophora pistil-*

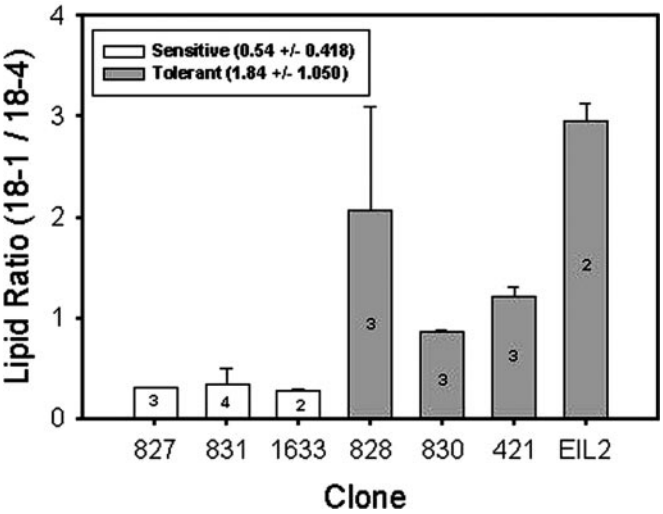

Fig. 3. Ratios of Δ9-*cis*-octadecatetraenoic (18:1) acid to Δ6,9,12,15-*cis*-octadecatetraenoic acid (18:4) for seven clones of *Symbiodinium* spp. ANOVA of the log-transformed data indicates a statistically significant difference between heat-sensitive and heat-tolerant clones.

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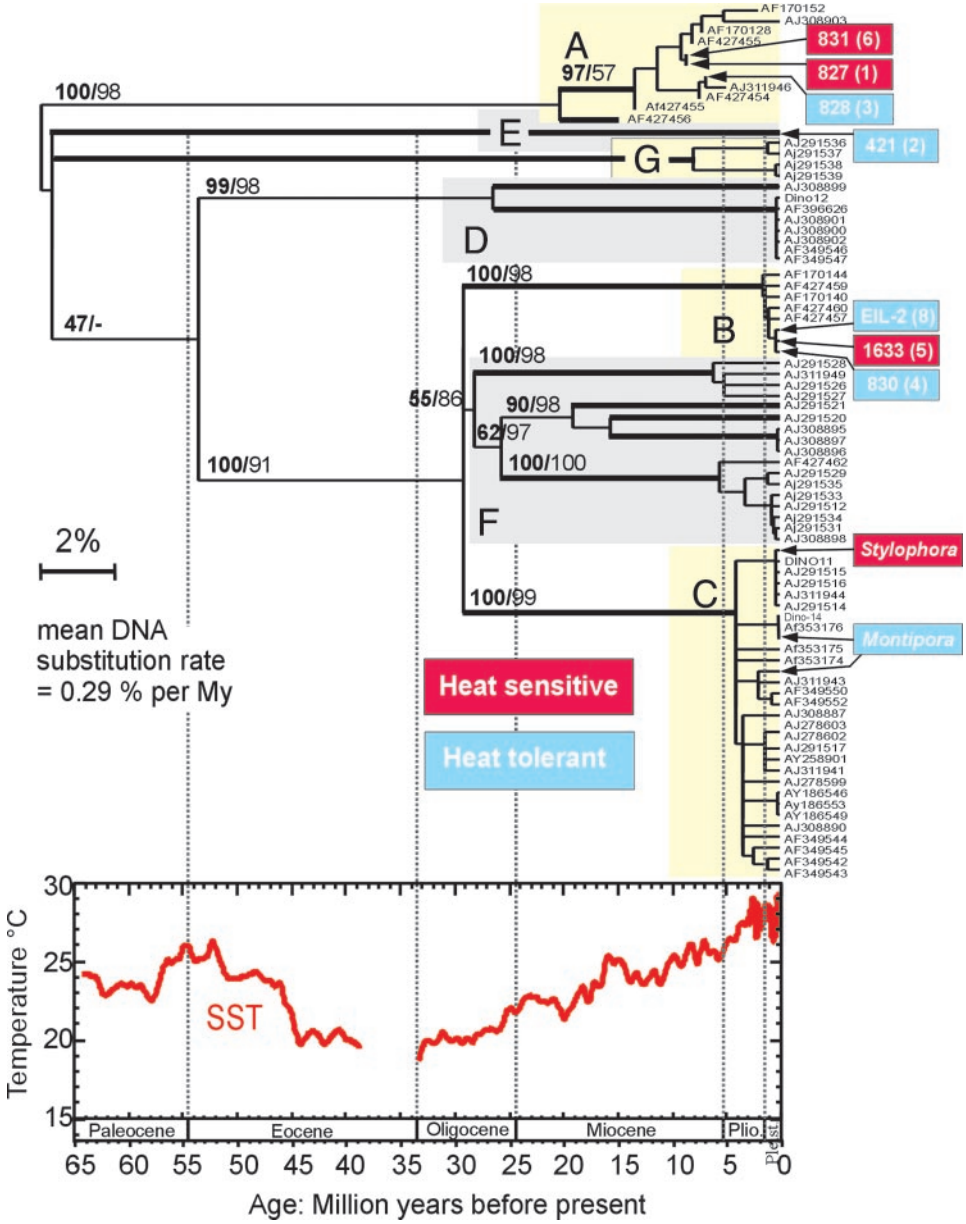

Fig. 4. LSU rDNA-based evolution of the *Symbiodinium* species complex (SSC) and phylogenetic position of the zooxanthellae isolates analyzed in Figs. 1–3. Heat-sensitive and resilient phylotypes are shown in red and blue, respectively. Clades A–G are the seven recognized *Symbiodinium* phylogenetic groups (35), with A and B (shaded yellow) being typically considered as bleaching-resistant, shallow-water types, and C (shaded pink) as bleaching-sensitive, deeper-living types. Our analysis suggests that at least 13 clades can be recognized based on genetic distances (thick branches in the tree) and that thermal sensitivity is not clade-specific. The ultrametric, linearized tree shown here allowed us to apply a crude clock and calibrate the evolution of the SSC in time. The sea-surface temperature curve, based on tropical planktonic foraminifera $\delta^{18}O$, serves as an approximate time scale for SSC evolution. Note that two to three DNA substitutions in the LSU rDNA correspond to 1 million years of evolution; thus, speciation events in the last 500,000 years may not be detectable by using this genetic marker. Neighbor-joining (1,000 replicates) and Bayesian (1 million generations) statistical values are indicated on the main internal branches.

lata and *Montipora samarensis* and the symbiotic anemone *Aiptasia* sp. cultivated *ex situ*. *S. pistillata* and *Aiptasia* sp. both lost >50% of their symbiotic algae within 72 h after exposure to waters of 32°C. In contrast, *M. samarensis* retained zooxanthellae at the elevated temperature for >2 months. In the thermally sensitive species, not only was there a change in membrane integrity (e.g., Fig. 1*F*) and loss of photochemical competence, but production of ROS in isolated zooxanthellae also increased by >2-fold at high irradiance levels. The production of ROS corresponded to a light-dependent increase in O_2 consumption as measured by membrane inlet MS using 10% $^{18}O^{18}O$ as a tracer (data not shown) (28). These results strongly suggest that the production of ROS is caused by the Mehler reaction, i.e., the photochemical reduction of O_2 in photosystem I (29). Moreover, the dye-tracer measurements clearly indicate that ROS produced in the algae leaks out of the cells. If this phenomenon happens *in hospite*, ROS would be transferred directly to the animal host, inducing a physiological stress (12).

GC/MS analysis of seven zooxanthellae isolates revealed a striking contrast in the relative composition of lipids associated with thylakoid membranes between thermally sensitive and resilient clones (Table 3, which is published as supporting information on the PNAS web site). Specifically, thermally tolerant, cultured *Symbiodinium* clones and zooxanthellae freshly isolated from corals that did not bleach after experimen-

tal thermal stress (Table 1) have a markedly lower content of the major polyunsaturated fatty acid, Δ6,9,12,15-*cis*-octadecatetraenoic acid (18:4), in relation to Δ9-*cis*-octadecatetraenoic (18:1) acid, independent of the experimental temperature (Fig. 3). The differences in this lipid profile are statistically significant at the 0.001 level (ANOVA). The higher relative concentration of the saturated polyunsaturated fatty acid enhances thermal stability in eukaryotic thylakoid membranes (30) and simultaneously reduces the susceptibility of the membrane lipids to attack by ROS (31–33). These experimental results strongly suggest that the wide variety of *Symbiodinium* spp. we analyzed have a limited ability to acclimate physiologically to changes in temperature by significantly modifying their thylakoid lipid composition and hence, unlike most eukaryotic algae, are confined to relatively narrow thermal regimes. The absence of qualitative differences in thylakoid lipid composition between the heat-sensitive and tolerant species suggests that differential susceptibility to elevated temperature results from changes in lipid biosynthetic pathways not associated with lipid desaturases *per se* but rather with regulatory elements of the enzyme(s) that controls the relative amount of desaturation in specific pools of fatty acids.

Phylogenetic analyses of the zooxanthellae isolates used in this study clearly show that thermal tolerance is not associated with a single, monophyletic clade. Heat-sensitive *Symbiodinium* spp. are found in totally different subdivisions of the LSU rDNA-based tree (Fig. 4 *A–C* and *E*), in which thermally tolerant phylotypes systematically branch as closely related sister species. This evolutionary pattern suggests that the reduced physiological ability to acclimate to elevated temperatures by enhancing thylakoid lipid-saturation levels was either acquired in the common ancestor of all modern *Symbiodinium* clades and was subsequently lost independently in individual taxa within each clade or was selected multiple times in independent lineages belonging to different clades.

The application of a molecular clock to the *Symbiodinium* spp. phylogenetic tree suggests that the ancestor of the species complex appeared at the Cretaceous–Tertiary boundary, which corresponds to a major transition time from the extinct Mesozoic, rudist-based, reefs to the modern scleractinian-dominated reefs. Juxtaposition of the clocked *Symbiodinium* spp. phylogenetic tree with a sea-surface temperature curve derived from oxygen isotope analysis of tropical planktonic foraminifera for the last 65 million years (34) suggests that for the first several million years in the Cenozoic Era, zooxanthellate-based symbioses evolved in warm tropical waters. We hypothesize that extensive cooling periods, starting in the Eocene, selected for cold-tolerant, heat-sensitive, *Symbiodinium* species, which may have been subject to negative selection (bleaching) later in the Pleistocene and even more strongly in the contemporary Anthropocene period.

Our combined physiological, biochemical, and molecular data confirm that the widely accepted but rather arbitrarily defined *Symbiodinium* taxonomic "clades" (35), often referred to as genetic or functional units, are in fact multimillion-year-old groups containing a broad diversity of modern species that are differentiated physiologically. Phylotypes belonging to different "clades" can present similar patterns of sensitivity to elevated temperatures but differ from their closely related sister phylotypes. This analysis clearly indicates that *a priori* rDNA genotyping is not diagnostic of thermal sensitivity in zooxanthellate symbiotic associations.

Our results suggest that the physiological basis of bleaching is initiated when thylakoid membrane integrity is compromised at elevated temperatures, leading to an uncoupling of photosynthetic energy transduction. The accompanying proton leak and loss of ATP restricts photosynthetic carbon assimilation; however, O_2 generated by PSII can react with the photochemically generated electrons in PSI to form ROS, which in turn oxidizes membrane lipids. The oxidized lipids initiate a positive feedback of ROS production that is accelerated by high light. Ultimately the ROS kills the intracellular algal symbionts and damages the host cells. The symbiotic algae literally are bleached and/or expelled from their hosts. These results provide an experimental demonstration of a biochemical adaptation associated with thermal tolerance in zooxanthellae and suggest that lipid analysis could potentially provide a rapid, sensitive tool for diagnosing the susceptibility of corals to thermally induced bleaching.

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