Etiology and Distribution of Coral Diseases in the Florida Keys National Marine Sanctuary

Final Project Report

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I. Introduction

Activities conducted under this project fell into two components addressing coral diseases in the Florida Keys: (1) etiologic studies and (2) reef surveys. Each is discussed in turn below. Note that activities were coordinated by and based at Mote's "Pigeon Key Marine Research Center" which has been relocated and renamed the "Center for Tropical Research" since completion of the project. All future correspondence regarding this project should be to the current address.

This project involved personnel from a number of institutions in addition to those of the grant recipients. A brief description of project roles follows. Erich Mueller (Mote Marine Laboratory - MML) was the project director. In addition to project management and report generation, he conducted all drilling to establish stations, conducted coral and disease (June 1998 only) counts on surveys, mapped the LK03 site, drilled holes in lesions and secured pins. Esther Peters (Tetra Tech., Inc. - TTI) was Quality Assurance (QA) officer. She also conducted or supervised all sample collection activities and conducted coral disease surveys (August, 1998 only). James Porter (Univ. of Georgia – UGA) and Doug Marcinek (UGA) took photographs of photoquadrats at the Eastern Dry Rocks and of the disease lesions at the LK03. D. Marcinek also assisted E. Mueller with installing pins in disease lesions. Karen Porter (UGA) was responsible for the *Coralliophila* experiment and conducted some coral counts. Kathryn Patterson (UGA) assisted with coral collections, conducted light and electron microscopic analyses of the collected samples and performed image analysis of the white pox lesions. Lillian Becker (Florida International University – FIU) conducted coral disease counts (June, 1998 only) and assisted with surveys. Shay Viehman (MML), Susan Thornton (Nova Southeastern Univ.), Tim Howe (FIU), and Harry McCarty assisted with surveys; McCarty and Howe also assisted with quality control checks on coral and disease counts.. Elizabeth Glynn (MML) assisted with mapping and photography at the LK03 site.

II. Project Activities and Methods

A. Etiologic Studies

1. Colony Marking and Recording

Two studies were planned to focus on white-band and white pox (may be synonymous with "patchy necrosis") diseases affecting *Acropora palmata*, one April 13-20, 1998 and the other August 2-8, 1998. It was planned that *A. palmata* colonies at a specific site would be marked and imaged so that progress of the diseases could be assessed. Within each of the study periods, short-term measurements would be made and by returning several months later, disease progress over a longer time period could be measured. The project investigators and assistants gathered at Pigeon Key in April for the "Coral Disease Field Study 98a", however, high wind conditions prevented us from getting to the reef. Thus, all marking activities took place in August during the "Coral Disease Field Study 98b" only . Schedules for these activities are presented in Appendices Ia and Ib, respectively.

Stations for assessing the incidence of coral disease were established during the Eastern Keys Survey (see section below). One of these stations (LK03; see Table 1) in the back reef area of Looe Key was selected for the study site. On August 3, 1998, all *A. palmata* colonies and fragments larger than 20 cm within a 10-m (314.6 m^2) radius of the LK03 pivot pipe were mapped. One team began tagging diseased colonies for collections and monitoring; those outside the 10-m radius were mapped on August 4 (Figure 1). Tags were prenumbered plastic tags secured to dead portions of the colonies with cable ties. Yellow tags were used for the ten colonies (Y1 to Y10) to be monitored. Only one colony (Y8/W4) had white-band disease; all others had white pox. White tags were used to mark eight colonies (W1 to W8) from which collections were to be made. With one exception (Y8/W4), these colonies were not the same as those used for the monitoring study above.

On August 5, small (1/8" diameter x 1/2" deep) holes were drilled in or near several lesions on each of the tagged colonies. PVC pegs were then secured in each of the holes; the total number of lesions marked was 38 . The pegs were made from 1/2" square bar stock, 1" long. Into the base had been secured a 1/8" PVC rod that protruded ~3/4". This rod was inserted into the drilled holes with a small amount of underwater epoxy putty. The top of each peg was marked with a stamped "V", generally oriented upwards. All pegs with their associated disease lesions on three colonies (Y1 to Y3) were photographed with a Nikonos camera. The remaining colonies (Y4 to Y10) were photographed on August 6. All colonies for monitoring were also videotaped with digital camera. All tags were also re-secured and cable ties used for temporary marking removed.

Marked lesions were re-photographed by J. Porter on August 20. The images were digitized and analyzed by Kathryn Patterson at the Univ. of Georgia. The site was re-visited by E. Mueller, L. Becker and Lauri MacLaughlin (Florida Keys National Marine Sanctuary)on October 13, 1998 about 2 ½ weeks following Hurricane Georges. The site was barely recognizable. Many *A. palmata* were displaced and those left had no living tissue; mortality was 100% for this species. The stainless steel stake was gone and only two of the 18 tags were found and recovered.

2. Collection Activities

Six loose fragments of *A. palmata* exhibiting signs of white pox were collected along with six *Coralliophila abbreviata* snails for observation at the Pigeon Key Marine Research Center. They were transported "dry" (in wet plastic bags) in coolers back to the lab. Time lapse videography was set up in an aquarium to observe feeding of the snails on the coral surface. Problems with this system led to switching to still photography on August 5. The snails and corals were returned to the study site on August 6.

Lesions to be collected on tagged corals in the field were photographed with a Nikonos camera. Fragments with these lesions as well as apparently healthy regions were collected on August 6 for electron microscopy. Coral samples were removed with a masonry hammer and chisel, placed in resealable 4-liter plastic bags, and collected in a cooler containing seawater for transport by boat to Pigeon Key.

3. Electron Microscopy

Corals were broken into small pieces with a masonry hammer and chisel and preserved in a glutaraldehyde (3.1%)/paraformaldehyde (2%) fixative mixture containing sodium cacodylic acid (0.1 M) and sodium chloride (1.75%) (D.L. Santavy, personal communication). Following 24 hours of fixation at 4°C, samples were rinsed three times (15 minutes each) with sodium cacodylate buffer and stored in fresh buffer. Coral samples were transported to the University of Georgia, Athens.

In the fall of 1998, white pox-diseased and apparently healthy coral samples collected from a single coral colony (W1) were processed for scanning and transmission electron microscopy analysis. The other coral samples will be analyzed at a later date. Preserved corals were trimmed to smaller pieces (approximately 1 cm^2) using a masonry hammer and chisel and post-fixed in 1% osmium tetroxide (OsO_4) plus 0.1 M sodium cacodylate buffer for one hour at room temperature (Rützler and Santavy, 1983; Martin and Le Tissier, 1991). Corals were then rinsed in three changes of 0.1 M sodium cacodylate buffer for 15 minutes each. Coral samples for TEM and SEM were stored in a fourth change of buffer in the refrigerator for five days and several weeks, respectively.

a. Scanning Electron Microscopy (SEM)

Following removal of sodium cacodylate buffer, samples were dehydrated with a graded ethanol series (30%, 50%, 70%, 85%, 95%, 100%, 100%) for 15 minutes each (Steiner and Cortes, 1996). Samples were then critical point dried with liquid CO_2 in a Samdri Critical Point Dryer. Dried samples were mounted on aluminum stubs using carbon tape and carbon paint. Samples were coated with gold

(Au) using a SPI sputter coater and viewed with a JEOL JSM 5800 scanning electron microscope. Micrographs were prepared.

b. Transmission Electron Microscopy (TEM)

Following removal of sodium cacodylate buffer, corals were decalcified in a 10% EDTA and 0.03 N NaOH solution (Glynn et al., 1985). The decalcifying solution was changed every 24 hours until all calcium carbonate coral skeleton was removed. All coral samples were decalcified within 10 to 14 days. Decalcified corals were rinsed three times for 15 minutes each in 0.1 M sodium cacodylate buffer (Peters, personal communication). Samples were stored in a fourth change of buffer for four days.

Coral tissue was kept moist with 0.1 M sodium cacodylate buffer while the tissue was cut into 1 mm cubes with a razor blade. Samples were dehydrated with a graded ethanol series (30%, 50%, 70%, 85%, 95%, 100%, 100%) for 10 minutes each (Steiner and Cortes, 1996). Samples were then washed in two changes (10 minutes each) of propylene oxide (PO). Coral tissues were embedded in Epon 812 epoxy (E) using a five-step process. Tissues were incubated on a rotator for 60 minutes each in (1) a 2:1 mixture PO/E, (2) a 1:1 mixture PO/E, and (3) a 1:2 mixture of PO/E. Tissues were then transferred to (4) pure Epon 812 and incubated overnight on a rotator. Following overnight incubation, (5) each cube of coral tissue was placed in a beam capsule, embedded in pure Epon 812, and polymerized for 48 hours in a 60°C oven. Embedded tissue was trimmed with razor blades and sectioned with glass knives on an ultramicrotome. Approximately 60-80 nm sections were collected on formvar-coated 300 square mesh copper grids and stained with uranyl acetate (2%) and Reynolds lead citrate (Reynolds, 1963). Stained sections were viewed under a JEOL 100CX II transmission electron microscope and micrographs were prepared.

B. Coral Disease Surveys

Two coral disease surveys, referred to as the "Eastern Keys Coral Disease Survey 98a" and "98b", were conducted under this project. Cruise plans are included in Appendices IIa and IIb. The EPA-approved Quality Assurance Project Plan (QAPP) and appropriate permits were present in the original copies given to senior project personnel but are not included here for brevity. As the project was originally proposed, the surveys of reefs for coral disease would take place on the Coral Reef/Hardbottom Monitoring cruise. It turned out that this was not possible; however, the Suncoast Seabird Sanctuary (Tampa, FL) provided the M/V *Whisker* for two cruises of 7 and 5 days, respectively at no cost other than food expenses. Mote Marine Laboratory vessels were used to survey several stations as well. These trips were temporally coordinated with Dr. Deborah Santavy who surveyed reefs from Western Sambo to the Dry Tortugas on the O.S.V. *Peter W. Anderson* (Western Keys Survey).

1. Station Installation

Figure 2 shows the sites (reefs) used in the Eastern and Western Keys Surveys in 1998. This project concerns only those sites in the Eastern Keys Survey. In most cases, there were several stations at each site that were pre-selected using EMAP protocols. Using the NOAA-FMRI Benthic Habitat maps, zones were selected at each site (reef) to constrain the randomized coordinates. Where possible, three zones were targeted at each site: back reef *A. palmata* zone, fore reef and transitional reef. At several reefs, the back reef zone was not present so only the other two zones were examined. Several coordinates were provided by Mr. Jed Campbell (U.S.EPA, Gulf Breeze) for each station. During the first survey (Eastern Keys Survey 98a - May 20 to June 3), the provided coordinates were visited in sequence and the first set of coordinates located on appropriate habitat was selected for station installation and survey (although constrained using the habitat map, some coordinates were located on sand or grass and were eliminated from consideration). Differential GPS (Garmin 12XL) was used to locate the coordinates and to provide averaged values on the actual station.

Upon selection of a station, a suitable area of bare substrate was chosen to install the station pivot pipe. A pneumatic drill with a 1" masonry bit was used to drill a hole 4-6" deep. In some cases, it was possible to hammer the pipe in directly. The pivot pipe was 12" long x 1" diameter stainless steel and was secured using Gunnybo Liquid Rock epoxy. After the surveys, the pipe was capped with an orange "safety cap" (normally used to cap rebar) to which a small float was attached (Figure 3). In high visitation areas, only the cap without the float was installed. The definitive station coordinates (Table 1) were obtained by floating the DGPS over the pipe and using an averaging function to increase the position accuracy. Figure-of-merit (FOM) values (an estimate of accuracy) provided by the unit were typically 6-8 feet.

2. Station Surveys

The Eastern Keys Survey was conducted twice in 1998: May 20 - June 3 (EKS98a) and August 7-14 (EKS98b). After installing or re-locating the station pivot pipe, the cap and buoy (if present) were removed and a stainless steel pivot rod inserted into the pipe. A 10 m-long transect line was clipped to a swivel on the rod and the height adjusted over a range of 2 meters. This allowed the transect to be as close to the substratum as possible while preventing snagging on gorgonians, coral heads or sponges. A small buoy on a weight was placed on the bottom arbitrarily (generally down-current) as a start/end point. Weighted flagging tape was used to highlight the 8- and 10-m radius points. An assistant swam the line in a counter-clockwise direction while a coral colony counter and coral disease counter swam between the 8- and 10-m marks, recording on pre-printed underwater data forms. The survey area was 113.1 m^2. Eighteen species of scleractinian coral known to be susceptible to disease and sea fans (*Gorgonia* spp.) were counted (> 10 cm maximum dimension) and examined. After completing the 360° arc, the line was reeled up, the marker buoy retrieved, the pivot rod removed and the cap and buoy replaced, usually with fresh ones. In some cases, only non-differential coordinates had been obtained during the EKS98a and these were redefined using averaged DGPS coordinates as described above during the EKS98b.

Table 1: Eastern Keys Survey stations. Region codes: LK = Lower Keys, MK = Middle Keys, UK = Upper Keys. Reef zone codes: BR = back reef, FR = fore reef, TR = transitional reef. All positions determined by DGPS. Date "established" coincides with the first survey.

station			reef		position		date	
region	name	code	zone	depth (ft)	latitude (N)	longitude (W)	established	2nd survey
LK	Eastern Sambo 1	ES01	TR	24-28	24° 29.513'	81° 39.674'	5/26/98	8/14/98
	Eastern Sambo 2	ES02	FR	12-16	24° 29.495'	81° 39.808'	5/26/98	8/14/98
	Eastern Sambo 3	ES03	BR	8-10	24° 29.532'	81° 39.919'	8/14/98	8/14/98
	Looe Key 1	LK01	TR	45	24° 32.543'	81° 24.877'	5/25/98	8/7/98
	Looe Key 2	LK02	FR	10-25	24° 32.750'	81° 24.350'	5/25/98	8/7/98
	Looe Key 3	LK03	BR	3	24° 32.819'	81° 24.358'	8/3/98	8/7/98
MK	Sombrero Reef 1	SR01	TR	23-25	24° 37.550'	81° 06.508'	6/3/98	8/8/98
	Sombrero Reef 2	SR02	FR	9-23	24° 37.573'	81° 06.568'	6/3/98	8/8/98
	Alligator Reef 1	AR01	TR	25	24° 50.790'	80° 37.244'	5/21/98	8/10/98
	Alligator Reef 2	AR02	FR	15-20	24° 50.764'	80° 37.289'	5/21/98	8/10/98
UK	Molasses Reef 1	MR01	TR	55-75	25° 01.057'	80° 21.832'	5/24/98	8/13/98
	Molasses Reef 2	MR02	FR	12-22	25° 01.087'	80° 22.078'	5/24/98	8/13/98
	Carysfort Reef 1	CR01	TR	15-22	25° 13.461'	80° 12.493'	5/23/98	8/12/98
	Carysfort Reef 2	CR02	FR	15-25	25° 13.308'	80° 12.595'	5/23/98	8/12/98
	Carysfort Reef 3	CR03	BR	5-8	25° 13.496'	80° 12.562'	5/23/98	8/12/98
	Elkhorn Reef 1	ER01	BR	10	25° 21.806'	80° 09.927'	5/22/98	8/11/98
	Pacific Reef 1	PR01	TR	45	25° 21.735'	80° 08.370'	5/22/98	8/11/98
	Pacific Reef 2	PR02	FR	15-22	25° 22.225'	80° 08.387'	5/22/98	8/11/98

III. Results and Discussion

A. Etiologic Studies

1. Changes in White Pox Lesion Size

It had originally been intended that lesions would be initially imaged in the Spring and then followed over two time scales: days and weeks (until late summer). Because poor weather prevented imaging in the Spring, this was done in August and the lesions re-imaged after two weeks. It had been hoped that we could image the lesions later to obtain the longer time scale data; however, Hurricane Georges severely damaged the study area. Thus, the data presented represent changes over the 2-week period from August 5-20 1998.

Two matched images of white pox lesions are shown in Figure 4. Most lesions increased in size (Table 2) over the study period although two lesions decreased in size (Figure 5). This suggests that these lesions no longer had active disease and the tissue was regrowing over the dead area. Three lesions increased over 300%, considerably higher than any others. The increase in area expressed as $cm^2\ day^{-1}$ and as a percentage are shown (Table 2) with and without the three high values. The more conservative values still indicate a rapid loss of tissue of ~1.5 $cm^2\ day^{-1}$ per lesion. There did not appear to be a strong relationship between lesion size and the rate of increase in size. Numerous subjective observations suggest that these lesions do not grow indefinitely as occurs with other diseases which can spread until the entire colony is dead. The largest recorded in the study was 183 cm^2 and although this size was unusual in our study, it must be assumed that larger lesions may occur. As multiple lesions are usually present on a colony, it should be noted that coalescing lesions are not uncommon and could be interpreted as being larger lesions. So, although individual lesions do not appear to be very large, the total effect of multiple lesions can lead to substantial loss of coral tissue area.

Table 2: Changes in white pox lesion size.

parameter	mean	SD	N
initial lesion area (cm^2)	38.00	25.99	36
final lesion area (cm^2)	63.89	47.65	36
all data			
increase in area ($cm^2\ day^{-1}$)	1.80	2.15	36
increase in area (%)	75.50	95.52	36
without III-3, VI-1 and X-2			
increase in area ($cm^2\ day^{-1}$)	1.45	1.62	33
increase in area (%)	49.78	41.84	33

2. Ultrastructural Effects of White Pox on Coral Tissues

a. Scanning Electron Microscopy

Scanning electron micrographs of apparently healthy and white pox-diseased *Acropora palmata* showed the tissue of apparently healthy coral to be more intact than tissue of white pox-diseased coral. Loss of coral tissue was apparent on diseased samples and the calcium carbonate coral skeleton was clearly visible. Coral skeleton was not visible in apparently healthy samples.

SEM analysis suggests that there is a difference between the surface appearance of apparently healthy coral and that of white pox-diseased coral. Additional diseased and apparently healthy corals need to be examined with SEM techniques before any conclusions can be drawn regarding the differences observed between the apparently healthy and white pox-diseased corals. The apparent differences may be artifacts of chemical fixation and/or SEM preparation procedures.

b. Transmission Electron Microscopy

No apparent differences between apparently healthy and white pox-diseased *Acropora palmata* were observed in the TEM micrographs. However, during preparation of the coral tissue for TEM analysis, apparently healthy tissue was easily cut into 1-mm cubes, while diseased tissue fell apart and was not easily cubed. Further analysis of apparently healthy and white pox-diseased coral tissue using TEM techniques is necessary before any conclusions can be made about the lack of apparent differences between white pox-diseased and apparently healthy coral samples.

B. Coral Disease Surveys

1. Incidence of Coral Disease

The overall proportion of examined corals with disease, other than bleaching, is shown by study area in Figure 6. Included here are data from the Western Keys although statistical comparisons are not made between the Eastern and Western Keys study areas. Disease incidence was lowest in the Dry Tortugas and New Ground study areas, both likely to receive the least anthropogenic influence. The Key West and Lower Keys areas appear to have had the highest concentration of coral disease and increased significantly from May to August. In other areas, there were no temporal difference in 1998.

A comparison of study areas within the Eastern Keys Survey is presented in Table 3. Disease incidence did not differ between areas in May but differences were observed in August when the Lower and Middle Keys were significantly different at the 0.05 level. The Upper Keys were intermediate in overall disease incidence and not significantly different than either of the other two areas.

Table 3: 1-way ANOVA results using study area as the class variable, examining difference of percent coral disease across all regions (p and R values) in the Eastern Keys Surveys. Mean percent diseased corals, standard error and (N) are presented. (N) is number of sites surveyed within each region at each sampling period. Tukey's Studentized Range Test was used to separate classes (α=0.05).

Sampling time	Study Area	Mean % Coral Diseased (N)	± SE	p value	Significance (α=0.05)	R value
May 1998	Lower Keys	8.6 (5)	2.3	p<0.31	1	0.15
	Middle Keys	6.0 (4)	5.4		1	
	Upper Keys	14.4 (8)	3.7		1	
August 1998	Lower Keys	21.0 (6)	6.0	p<0.011	1	0.45
	Middle Keys	1.8 (5)	0.76		2	
	Upper Keys	9.7 (7)	1.9		1 2	

The second major data stratum of interest was reef type (Table 4). It was clear in 1998 that the back reef stations had the highest prevalence of disease. Although the fore reef and transitional reef areas were not significantly different from each other, there may be a general trend towards lower disease incidence with increasing depth. The higher disease incidence in the back reef, and to some extent the fore reef, was due to white pox/patchy necrosis on *Acropora palmata*. Back reef stations were largely dominated by this species while it is more sparse on the fore reef and absent from the transitional reef environments. There were no temporal trends found with respect to reef type between the May and September surveys.

Table 4: 1-way ANOVA results using reef type as the class variable, examining difference of percent coral disease across all regions (p and R values) in the Eastern Keys Surveys. Mean percent diseased corals, standard error and (N) are presented. (N) is number of sites surveyed within each region, at each sampling period. Tukey's Studentized Range Test was used to separate classes (α=0.05).

Sampling time	Reef Type	Mean % Coral Diseased (N)	± SE	p value	Significance (α=0.05)	R value
May 1998	Back Reef	27.2 (2)	8.0	p<0.004	1	0.55
	Fore Reef	11.7 (8)	2.5		2	
	Transitional Reef	5.0 (7)	2.0		2	
August 1998	Back Reef	24.3 (4)	8.6	p<0.03	1	0.38
	Fore Reef	7.6 (7)	2.0		2	
	Transitional Reef	7.5 (7)	2.9		2	

The distribution of diseases by site is presented in Figure 7 a,b. Aspergillosis was the most commonly reported disease while black-band the rarest. Aspergillosis appears to increase in the Lower Keys from May to August while Upper Keys reefs had the reverse trend. White pox was not adequately surveyed in May as the back reefs at Eastern Sambo and Looe Key were not examined. Both sites had a considerable incidence of this disease when surveyed in August.

2. Coral Counts

The coral counts were essential to estimate the disease incidence within various coral species populations but also provide interesting information about the frequency of coral occurrence. although coral cover is generally more meaningful with respect to reef functional ecology, colony frequency provides autecological information and an indication of the potential for increasing or decreasing trends in coral cover.

During each of the Eastern Keys surveys, *Acropora cervicornis* or *Dendrogyra cylindrus* were not found within the survey stations although they were observed outside of transect belts. This is not surprising for *D. cylindrus* which is uncommon, but is puzzling for the more common *A. cervicornis*. *Montastraea annularis, M. faveolata* and *M. franksi* were separately counted but analyzed together as "*M. annularis* complex." Frequencies (100 m^{-2}) of the 14 species, or group, are plotted separately as a function of reef site and type in Figure 8a-bb. While we do not consider these data of sufficient resolution to detect changes in population frequency between the May and August cruises, these plots can show any type preferences by a given species and general patterns of their distribution along the Keys. Clear

examples of reef type preference include *A. palmata* in the back reef (BR – blue bars), *Mycetophyllia* spp. in the fore reef (FR – green bars) and *Solenastrea bournoni* on transitional reefs (TR – gray bars). Regional patterns include generally more *Colpophyllia natans* and *Diploria strigosa* in the Lower Keys while *Gorgonia* spp. were more common in the Upper Keys. As many others have shown, the Middle Keys are generally lower in coral frequency.

The variations between these two surveys are much more likely due to observer error. In May, a total of 1183 corals were counted and examined for disease. After correcting for the additional two stations reached in August (LR03 and ES03 were not surveyed in May because of rough conditions), 1205 colonies were counted, a difference of 22 colonies representing an overall error of ~1.8 %. Most of these errors are likely undercounts. The error rate is undoubtedly higher on stations where there are many colonies, usually including a dominant species such as *Gorgonia* or acroporids where colonies are often difficult to discern and subject to fragmentation. At the beginning of the first cruise in May, repetitive counts (three) were made at Sand Key (SK01) to assess inter- and intra-observer error. E. Mueller, who counted all corals on both Eastern Keys Surveys, had a total coral count of 127.7±6.5. This is around the maximal limit (5%) set for intraobserver error in the QAPP. Each subsequent transect survey of the SK01 station had a higher count although this was not true for all who participated.

It appears that the effective counting error rate is sufficiently low to detect substantial changes, or lack thereof, to coral populations. Although surveys in 1999 were not part of the Special Studies project, some examples from Looe Key, which was hit hard by Hurricane Georges in late September 1998 show the ability to detect major changes. The LK03 station, in the back reef, was completely obliterated and had 100% loss of *A. palmata* (56 colonies or fragments >10 cm were found in early August 1998). The nearby fore reef station, LK02, had 67 total colonies in August 1998 and 70 in June 1999. A similar situation occurred at Eastern Sambo where the back reef went from 47 to 20 colonies after the storm (the *A. palmata* count went from 35 to 5) and the fore reef had virtually no changes (50 before and 46 colonies after). The lack of change on the fore reef indicates that Hurricane Georges effects were mostly on only the shallowest reef areas.

IV. Conclusions

The objectives of this study were: (1) to examine white-band disease and white pox, particularly its etiology and patterns of small scale distribution, (2) establish permanent stations for disease surveys in the Eastern Keys and (3) to obtain quantitative data on disease incidence at those stations in the late spring and late summer. Objective 1 remains elusive. Very few white-band infections were found and only one at the Looe Key study site. We did measure substantial rates of white pox lesion growth on a short time scale (days) but were unable to obtain longer temporal scale data due to loss of the site in Hurricane Georges. We were unable to draw conclusions regarding the spread of white pox within or between reef systems.

The establishment of disease stations in the Eastern Keys and their survey was successful. These stations complement many others from the Key West area to the Dry Tortugas, thus, providing fairly comprehensive coverage of the Florida Reef Tract. These stations were surveyed in 1999 and most again in 2000. Plans are underway to ensure that all are surveyed again in 2001 and beyond to provide temporal information regarding the incidence of disease in the Florida Keys. In 1998, we found two significant trends in disease distribution: (1) the back reef stations had a higher disease incidence than the fore reef or transitional reef areas and (2) in August, the Lower Keys had a higher incidence of disease than the Middle Keys; the Upper Keys had an intermediate value and was not different from either of the other study areas.

Ms. Katie Patterson's doctoral dissertation research was partially supported by this funding. She has continued her studies on white pox with other funding and has found the putative pathogen. A manuscript will be submitted shortly with her results.

V. References

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VI. Appendices

Ia Coral Disease Field Study 98a – schedule

Ib Coral Disease Field Study 98b – schedule

IIa Eastern Keys Survey 98a – cruise plan

IIb Eastern Keys Survey 98b – cruise plan

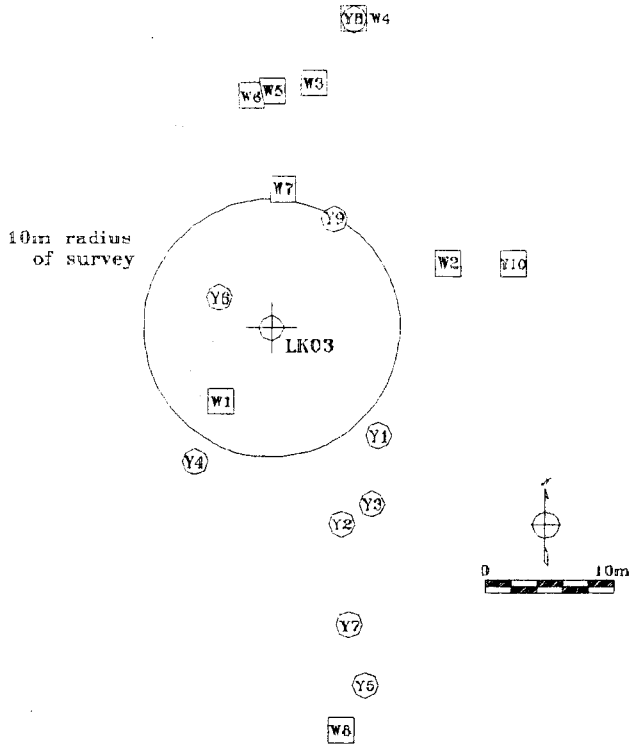


Figure 1. Map of colonies used in the study at the LK03 station. Colonies monitored for disease progression indicated by "Y" codes within circles. Colonies from which collections were made indicated by "W" codes within squares. Other colonies mapped within the 10 m radius not shown for clarity.

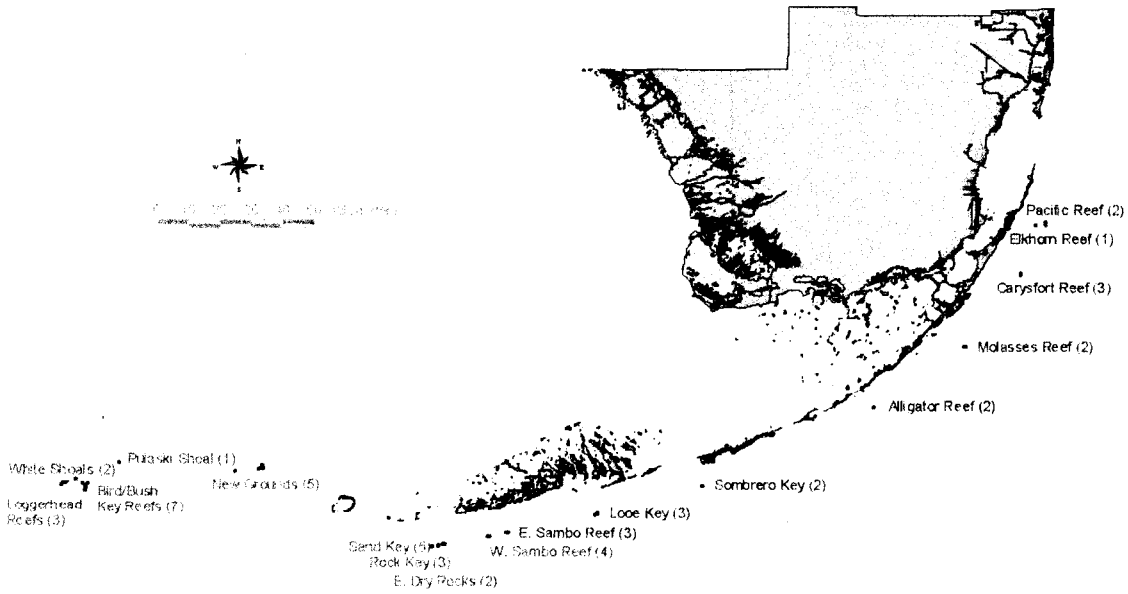


Figure 2. Coral disease survey sites in the Florida Keys. Number of stations at each site shown in parentheses. Only the Eastern Keys sites (labeled in blue) were surveyed in this project.

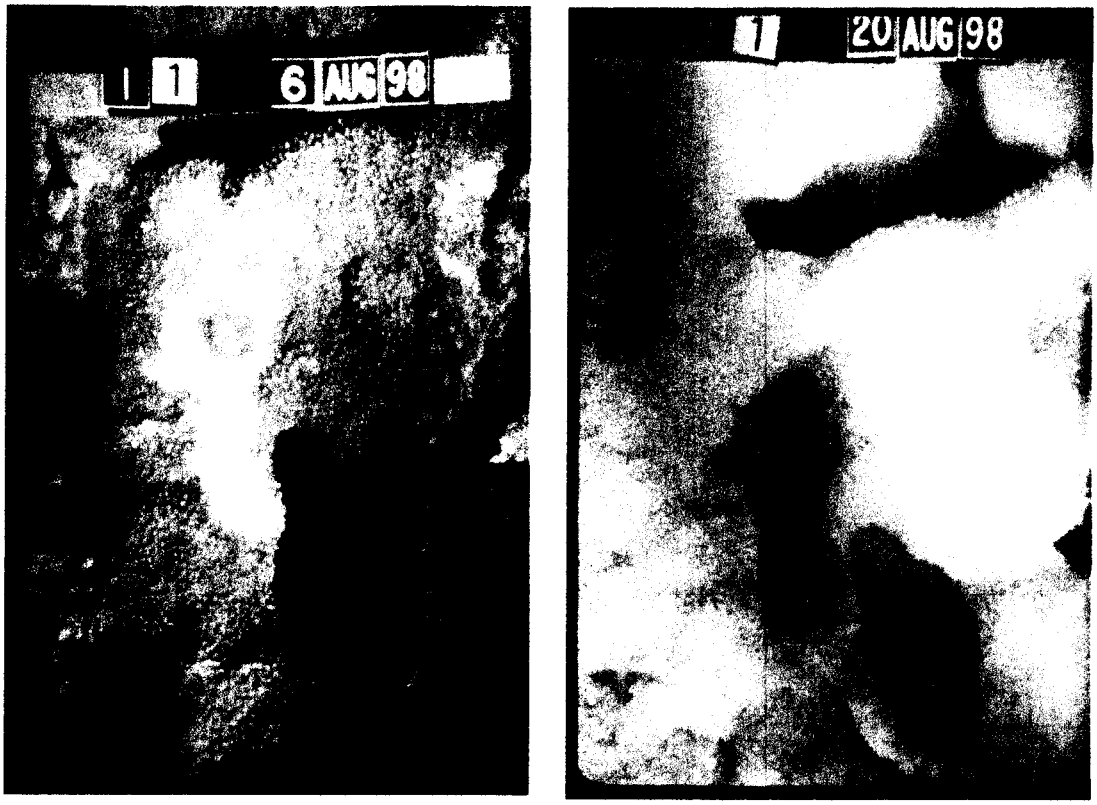


Figure 3. Matched images of white pox lesion 1 from colony 1. Note that the lesion area has been colonized by filamentous algae while the surrounding area has bleached.

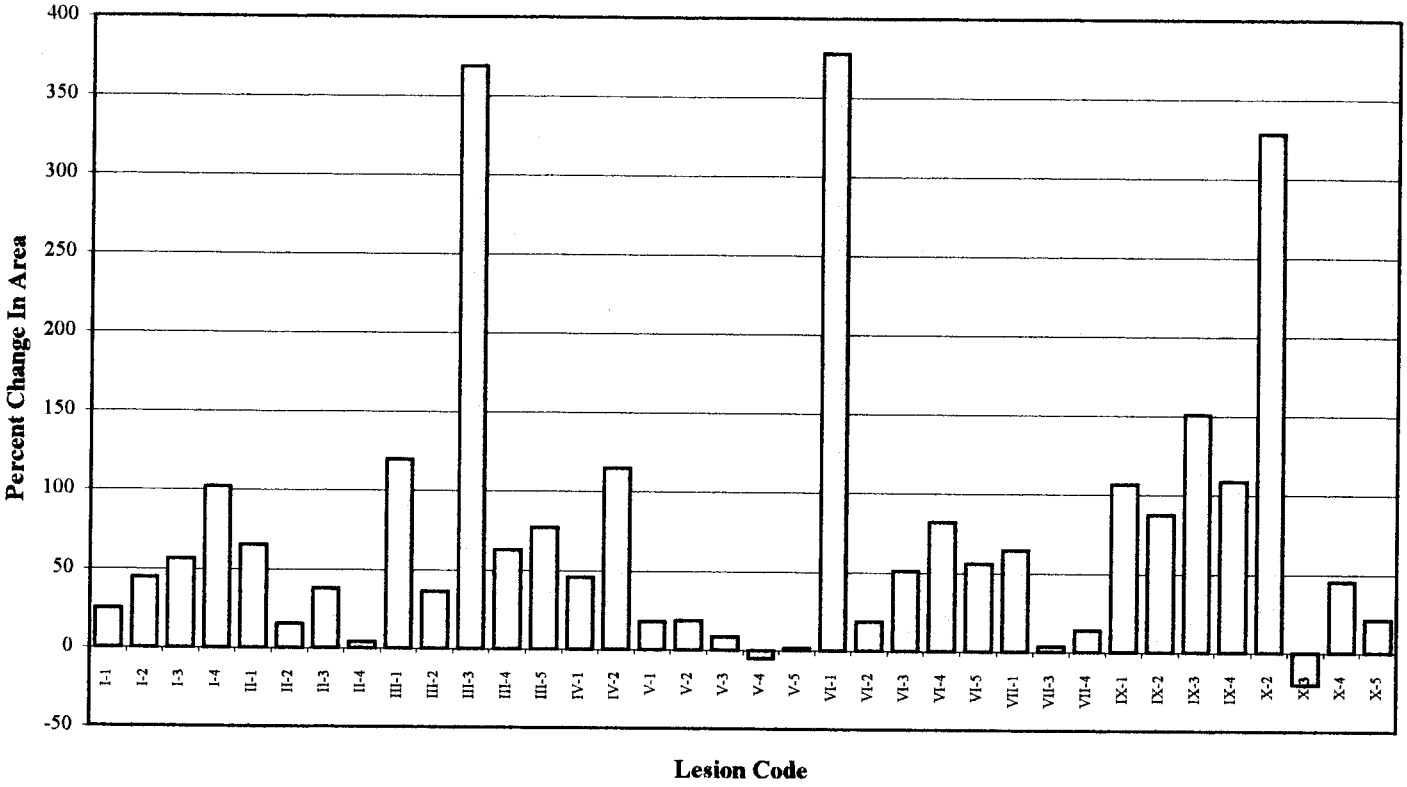


Figure 4. Percent change in size of white pox lesions. Roman numerals refer to colony and Arabic numerals to lesion number.

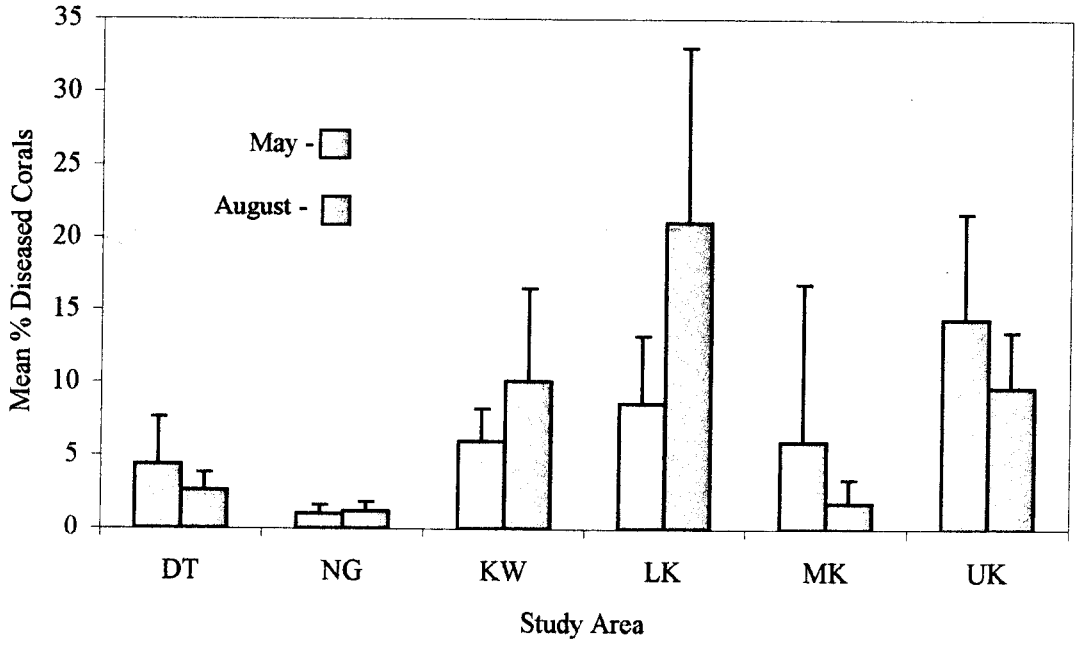


Figure 5. Overall proportion of corals in both the Eastern and Western Keys Surveys with disease during 1998 by study area. Study areas: DT = Dry Tortugas; NG = New Grounds; KW = Key West; LK = Lower Keys; MK = Middle Keys; UK = Upper Keys

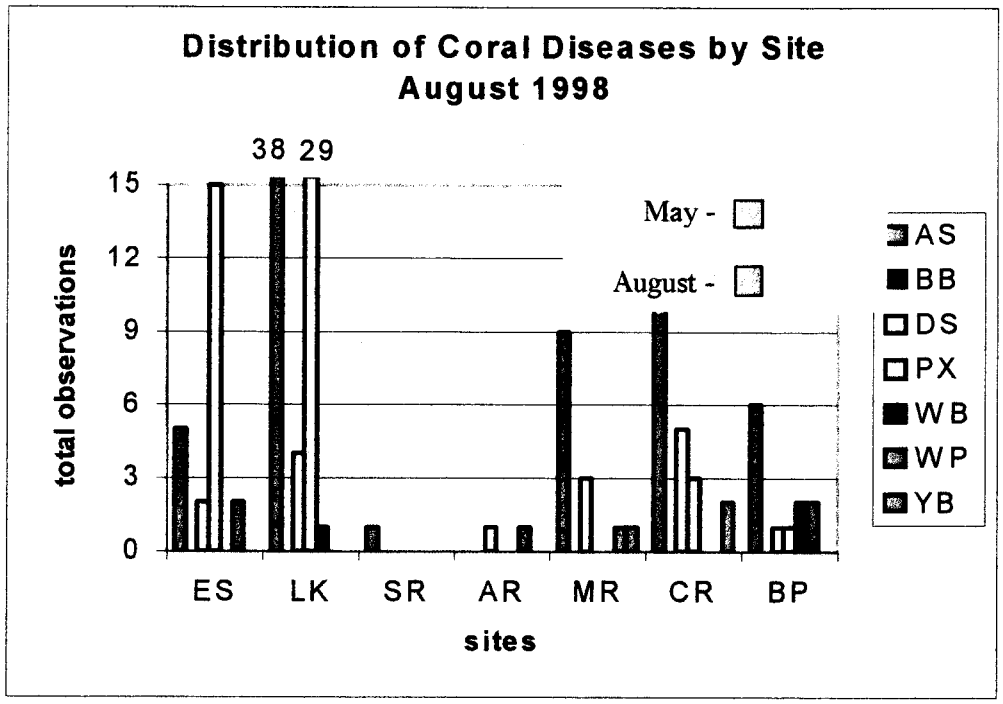


Figure 6. Overall proportion of corals in the Eastern Keys with disease during 1998 by reef type. Reef types: BR = back reef; FR = fore reef; TR = transitional reef.

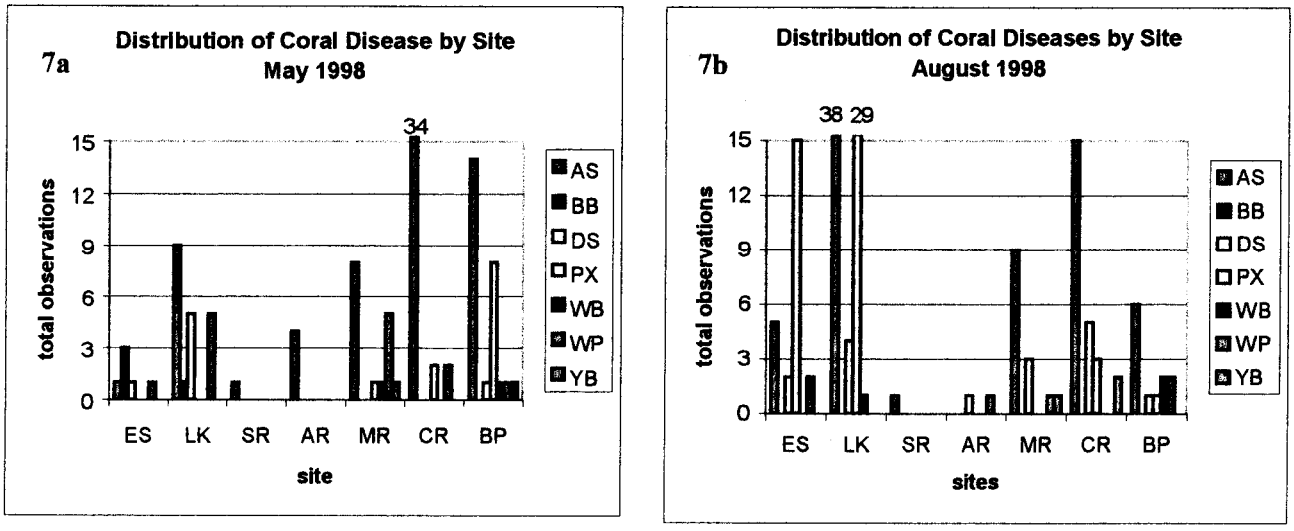
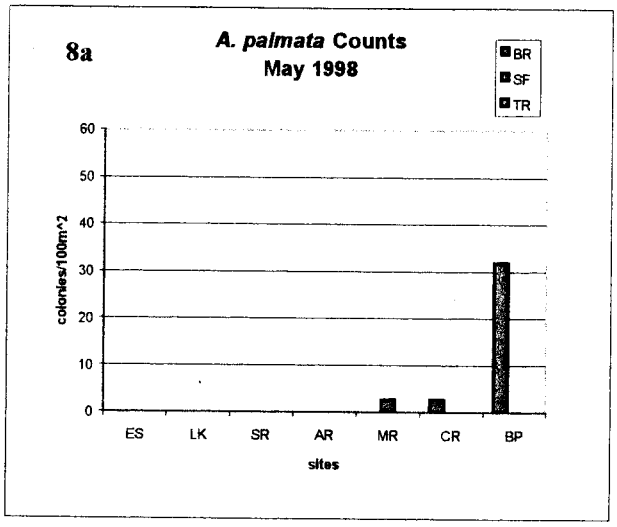
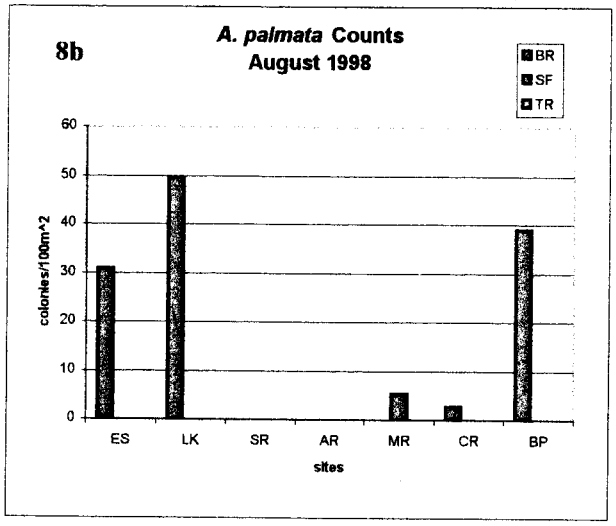
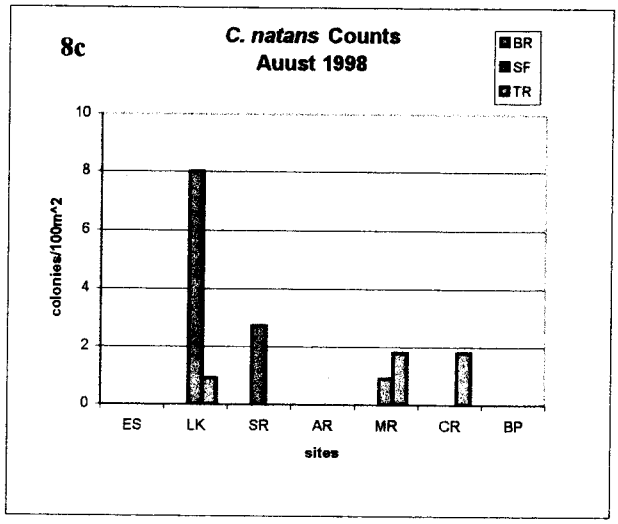
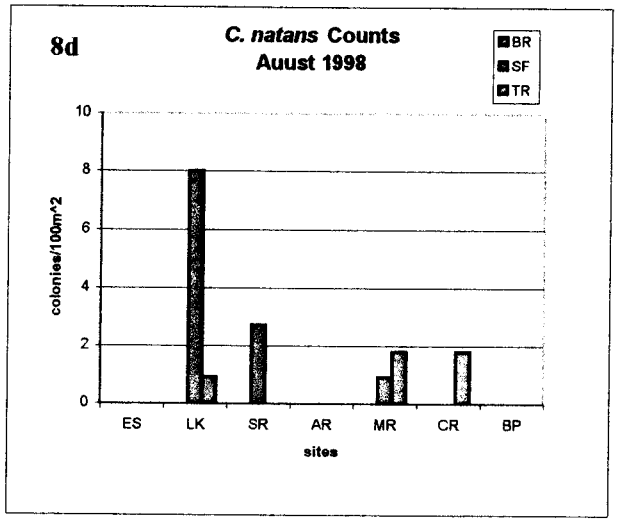


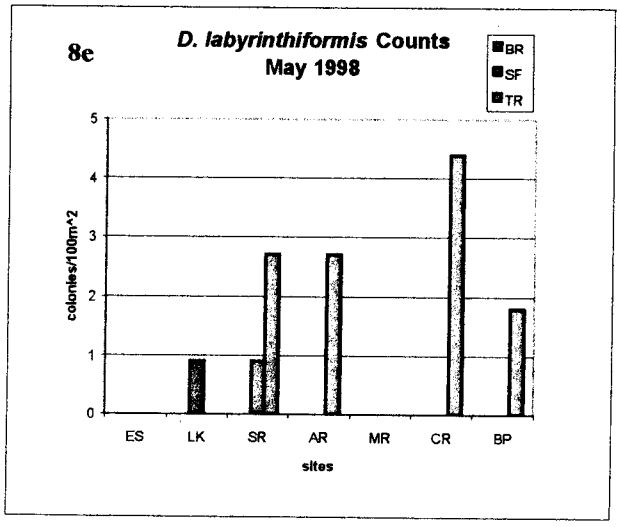
Figure 7a,b. Distribution of coral diseases by site during the Eastern Keys Surveys of 1998. Note that all hosts affected by a given disease are combined. Counts at Eastern Sambo (ES) and Looe Key (LK) are under-represented in May, particularly white pox (PX), because back reef stations were not surveyed.











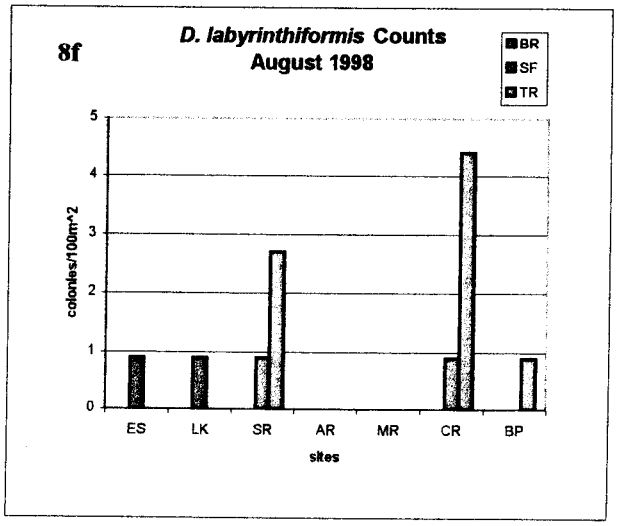
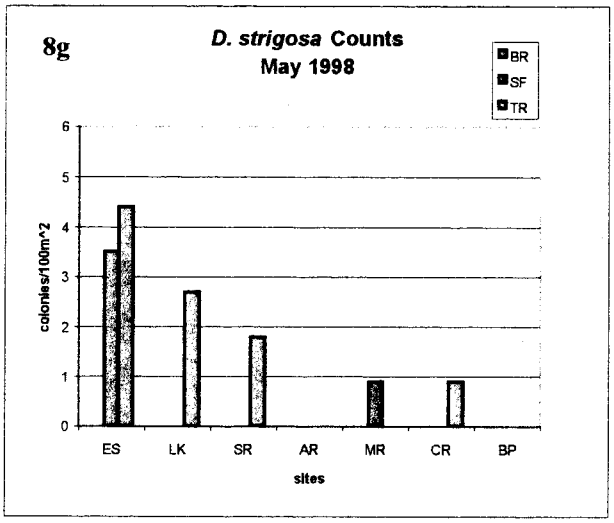
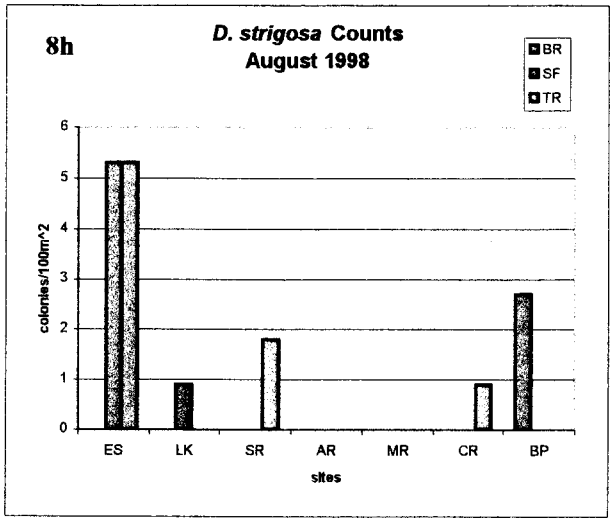
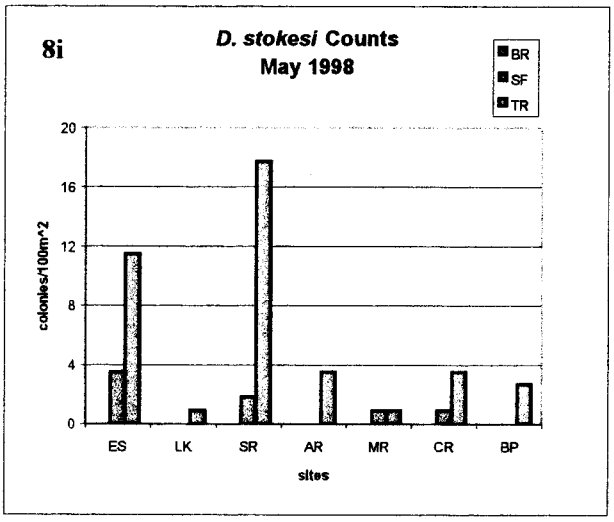
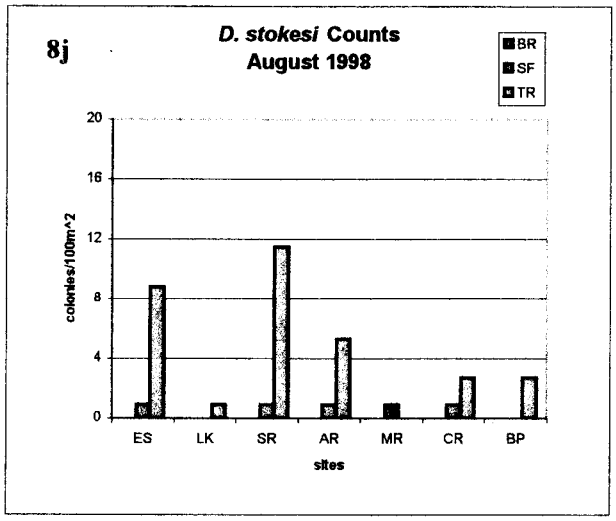


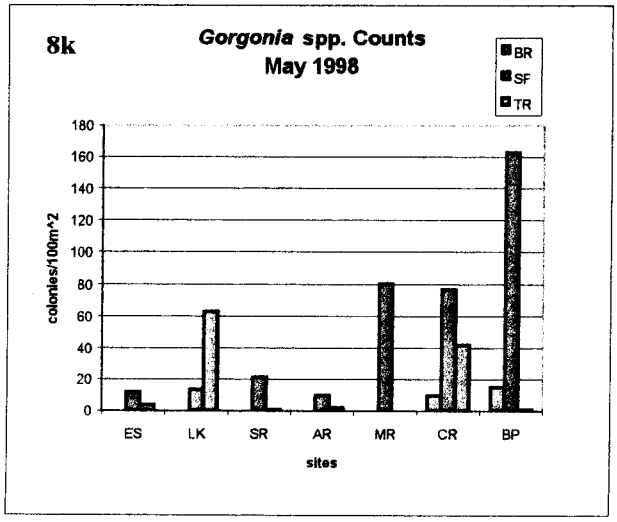
Figure 8a-f. Coral counts during the 1998 surveys. Biscayne N.P stations ER01, PR01 and PR02 designated as "BP" site. Note that back reef stations at Eastern Sambo and Looe Key were not surveyed in May. No back reef stations exist for Sombrero Reef (SR), Alligator Reef (AR) or Molasses Reef (MR).
Reef types: BR = back reef SF = fore reef TR = transitional reef











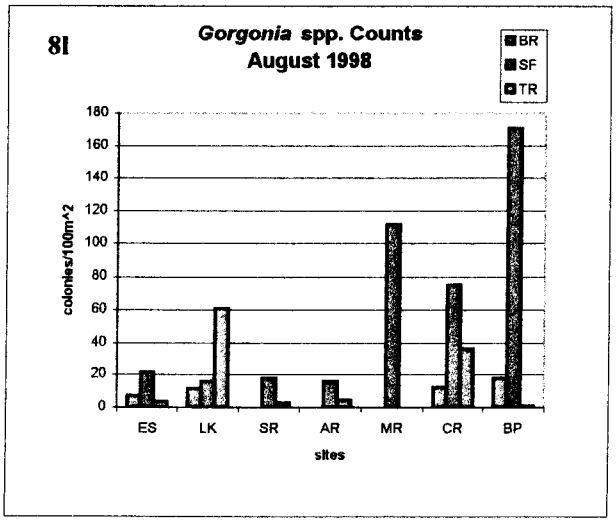
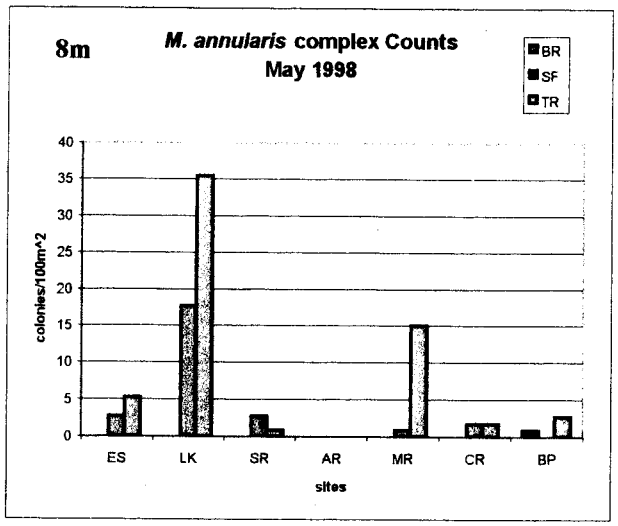
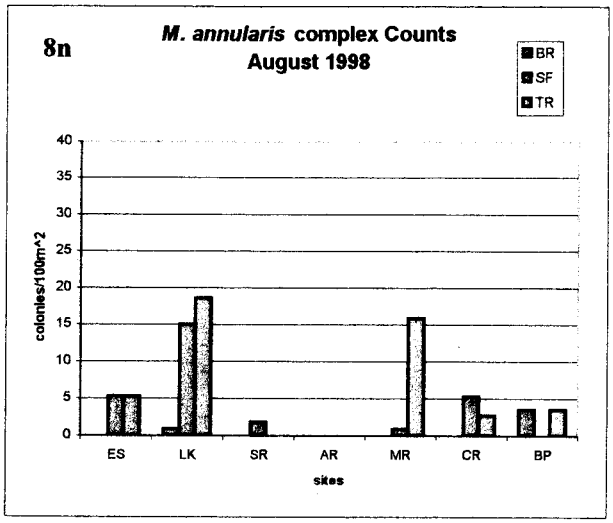
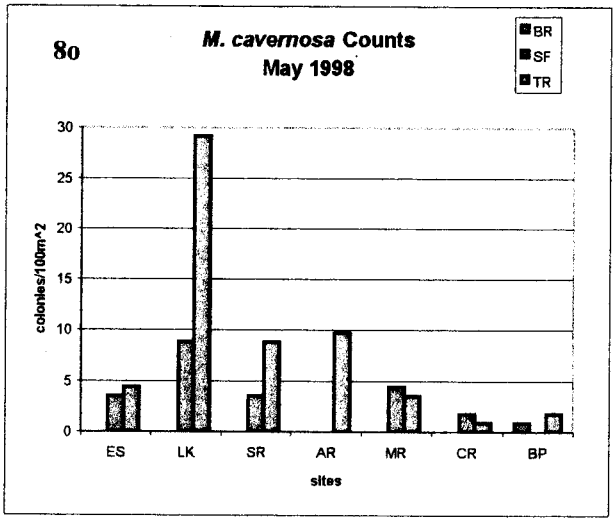


Figure 8g-l. Coral counts during the 1998 surveys. Biscayne N.P stations ER01, PR01 and PR02 designated as "BP" site. Note that back reef stations at Eastern Sambo and Looe Key were not surveyed in May. No back reef stations exist for Sombrero Reef (SR), Alligator Reef (AR) or Molasses Reef (MR).
Reef types: BR = back reef SF = fore reef TR = transitional reef







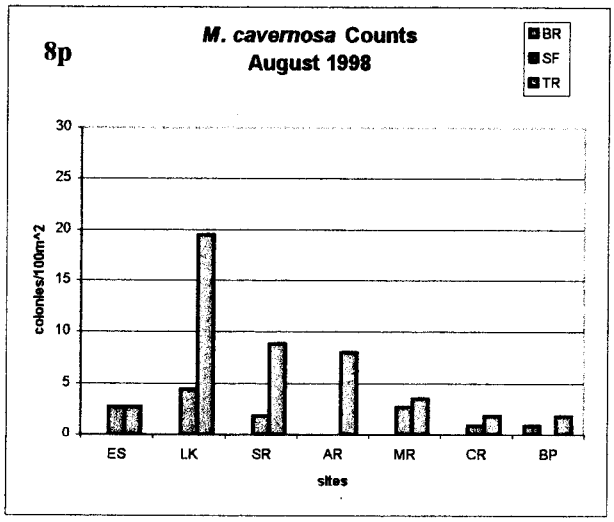
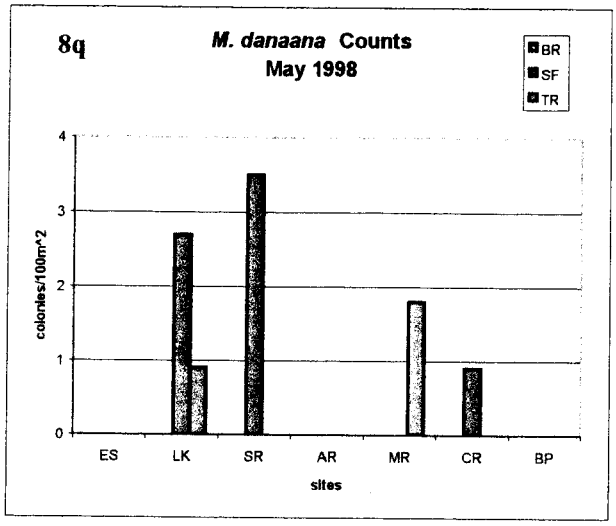
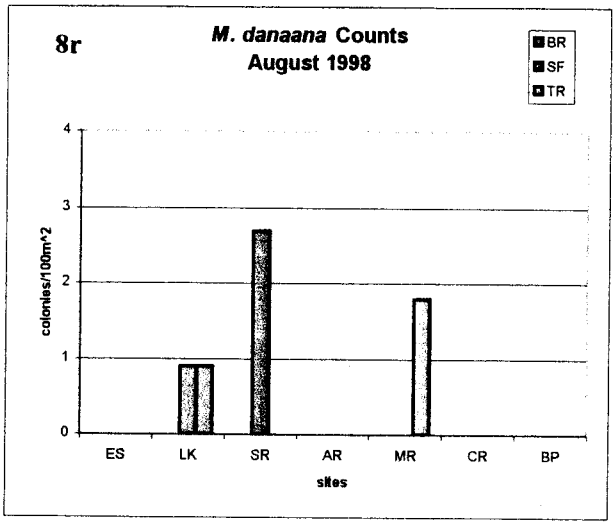
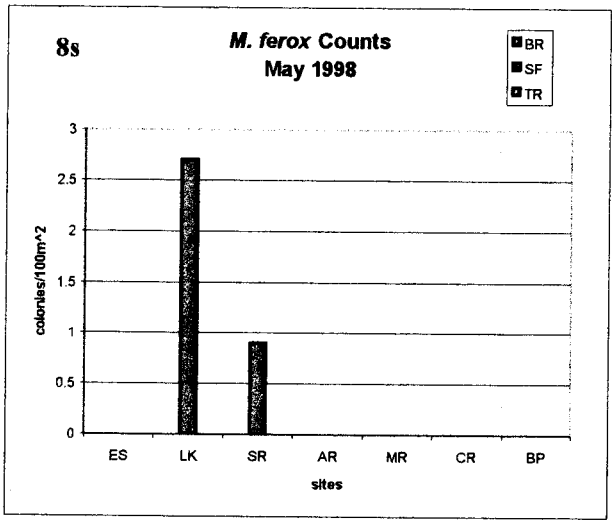
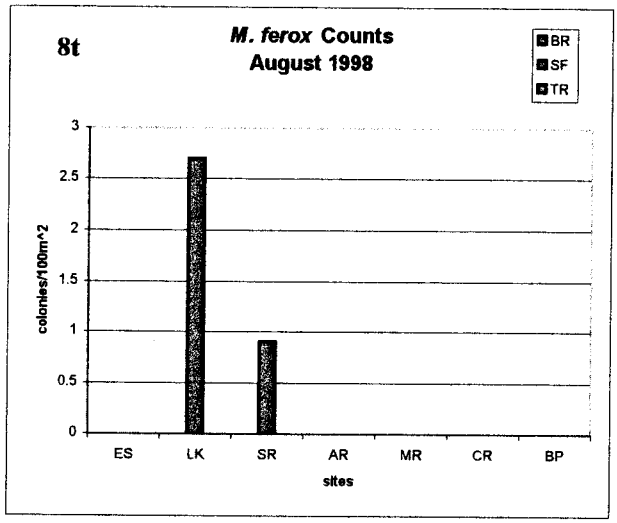


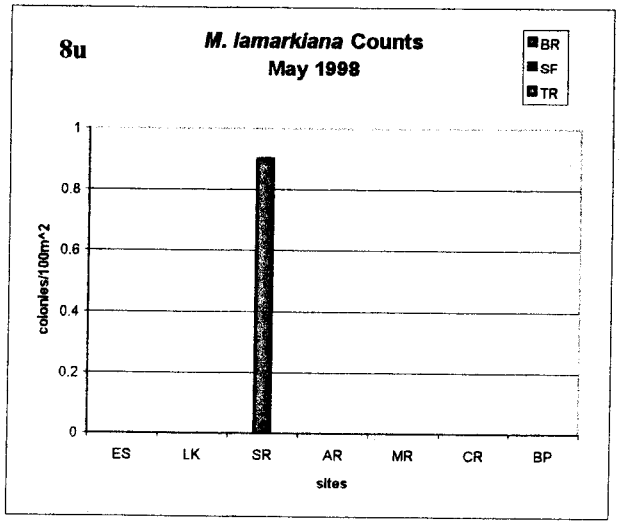
Figure 8m-p. Coral counts during the 1998 surveys. Biscayne N.P stations ER01, PR01 and PR02 designated as "BP" site. Note that back reef stations at Eastern Sambo and Looe Key were not surveyed in May. No back reef stations exist for Sombrero Reef (SR), Alligator Reef (AR) or Molasses Reef (MR).
Reef types: BR = back reef SF = fore reef TR = transitional reef











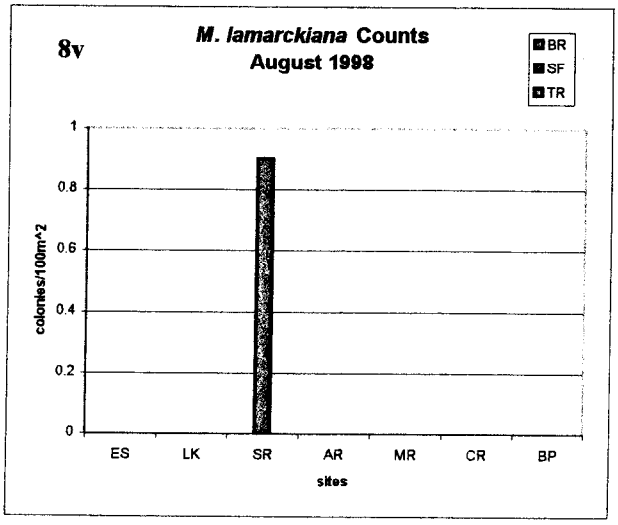
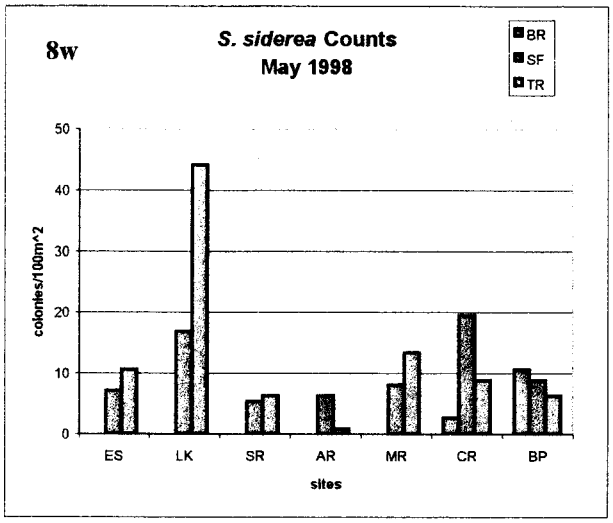
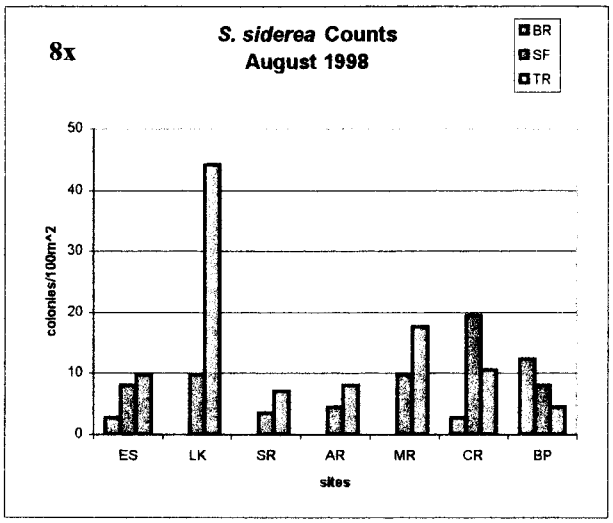
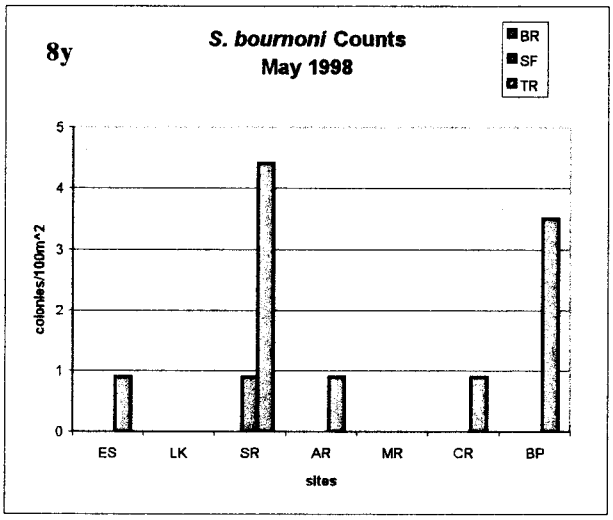
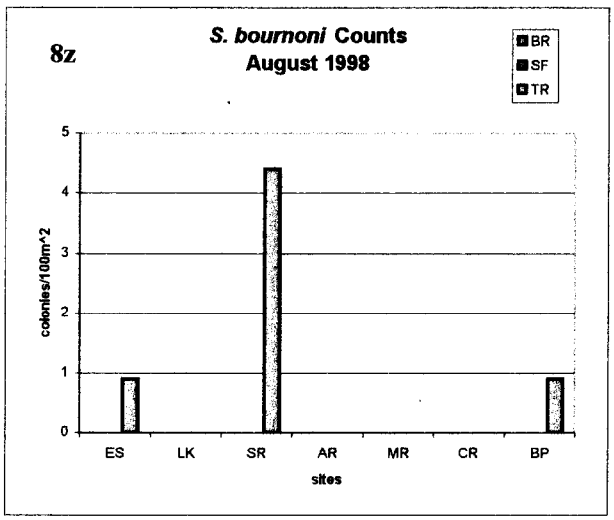


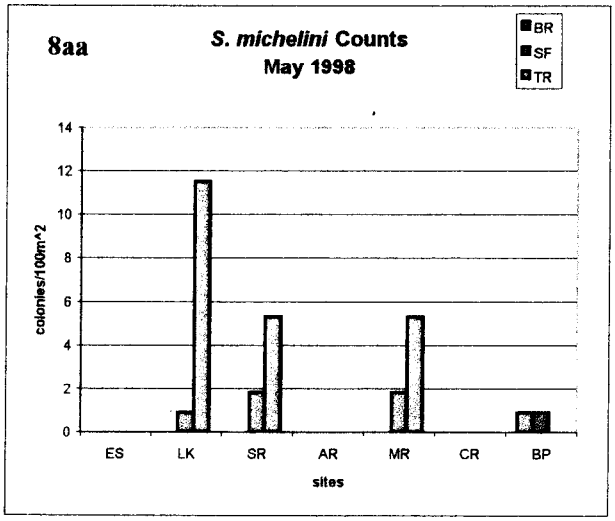
Figure 8q-v. Coral counts during the 1998 surveys. Biscayne N.P stations ER01, PR01 and PR02 designated as "BP" site. Note that back reef stations at Eastern Sambo and Looe Key were not surveyed in May. No back reef stations exist for Sombrero Reef (SR), Alligator Reef (AR) or Molasses Reef (MR).
Reef types: BR = back reef SF = fore reef TR = transitional reef











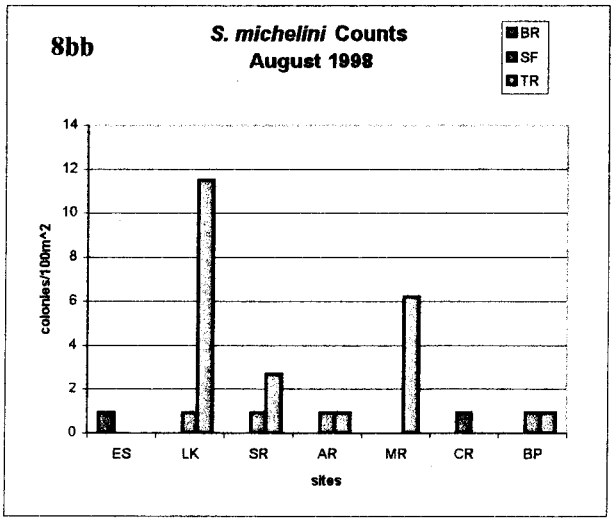


Figure 8w-bb. Coral counts during the 1998 surveys. Biscayne N.P stations ER01, PR01 and PR02 designated as "BP" site. Note that back reef stations at Eastern Sambo and Looe Key were not surveyed in May. No back reef stations exist for Sombrero Reef (SR), Alligator Reef (AR) or Molasses Reef (MR).
Reef types: BR = back reef SF = fore reef TR = transitional reef

CORAL DISEASE FIELD STUDY 98A

13-20 April, 1998
Pigeon Key, FL

Participating Institutions

Mote Marine Laboratory
Tetra Tech, Inc.
University of Georgia
U.S. EPA, Gulf Ecology Division

Prepared by:

Erich Mueller
Mote Marine Laboratory

Coral Disease Field Study 98a

Table of Contents

Participants & Roles

Addresses and numbers for some participants are listed in the QAPP appended to this schedule and not reiterated below. More detailed roles in the overall project are also listed in the QAPP. Contact points for those not in the QAPP are listed below.

Erich Mueller, Mote Marine Laboratory: PI/PD Alternate QA Officer

Esther Peters, TetraTech, Inc.: Co-PI, QA Officer

James Porter, University of Georgia: Co-PI

Karen Porter, University of Georgia: Co-PI

Deborah Santavy, U.S. EPA: Cooperating Scientist

Katherine Patterson, University of Georgia: graduate student (coral & disease counting; sample preparation and lab analysis)

Jed Campbell: research assistant (coral & disease counting, water chemistry)

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1 Sabine Island Drive
Gulf Breeze, FL 32561

Lauri MacLaughlin: research assistant (coral & disease counting, videography)

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email: lmaclaughlin@ocean.nos.noaa.gov

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Key West, FL 33040

Doug Marcinek: research assistant (coral counting)

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University of Georgia
Athens, GA 30602-2602

Lillian Becker: graduate student (coral & disease counting)

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F: (305)
email: lbecke01@fiu.edu

Department of Biological Sciences
Florida International University
Miami, FL

Shay Viehman: research assistant (disease counting) Note: Starts graduate program at FIU in Fall, 1998.

P: (305) 872-2205 or 872-2331
F: (305) 872-
email:

Newfound Harbor Marine Institute
Route 3, Box 170
Big Pine Key, FL 33043

Susan Thornton: graduate student (coral counting)

P: (954) 920-1909
F: (954)
email: sthornto@ocean.nova.edu

Nova Southeastern University
Oceanographic Center
8000 N. Ocean Dr.
Dania, FL 33004

Travel Plans and Accommodations

Room codes are: Mote = Bridge Foreman's House, next to dock and BTH = Bridge Tender's House, directly facing the Mote building.

Name	Initials	Dates	Arrive Via	Room
Erich Mueller	EM	4/13 – 4/20	car	off site
Esther Peters	EP	4/14 (noon) – 4/21 (AM)	arrives Marathon @ 11:45	Mote
James Porter	JP	4/13 (AM) – 4/20 (eve)	car	Mote
Karen Porter	KP	4/13 (AM) – 4/20 (eve)	car	Mote
Deborah Santavy	DS	4/14 (eve) - 4/17 (late PM)	arrives Miami @ 19.20; renting car	BTH
Jed Campbell	JC	4/13 (AM) – 4/17 (early PM)	car	BTH
Lauri MacLaughlin	LM	4/14 (AM) - 4/15 (eve)	car	off site
Katherine Paterson	KT	4/16 (eve) – 4/20 (eve)	arrives Ft. Lauderdale @ 17.15	BTH
Doug Marcinek	DM	4/13 (AM) – 4/20 (eve)	car	BTH
Lillian Becker	LB	4/15 (eve) – 4/18 (eve)	car	BTH
Shay Viehman	SV	4/15 (AM) - 4/15 (eve)	car	off site
Susan Thornton	ST	4/14 - 4/16 (late PM)	car	BTH

GENERAL INFORMATION

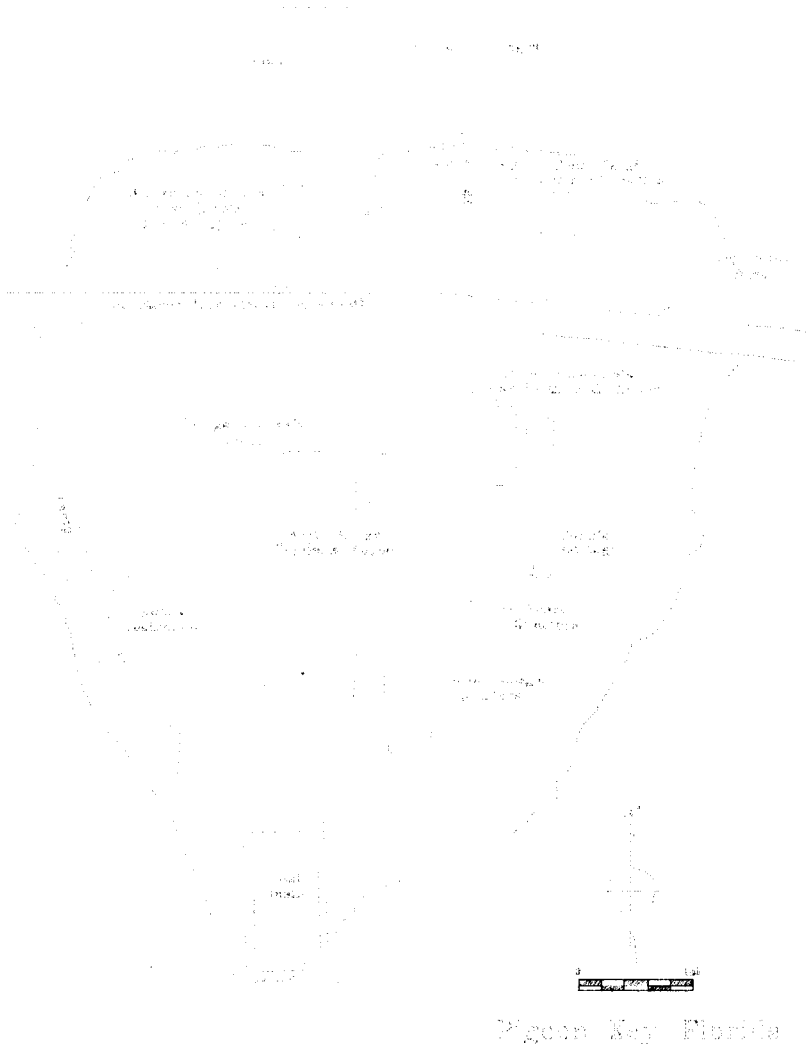
Vehicle access to Pigeon Key via the Old Seven-Mile Bridge is restricted to those with specific business on the island (including yourselves). Visitors arrive by walking, bicycles, rollerblades or the Pigeon Key train. Please car pool insofar as possible when driving off the island to minimize vehicular traffic on the bridge. Always give pedestrians the right-of-way and follow the posted speed limits (25 mph max; 15 mph around pedestrians). Many bridge users are locals who will let us know promptly if anybody drives improperly.

The PKMRC office is in the Bridge Foreman's House (see map next page) along with a small dry lab and some accommodations. Julianna (Juli) Weir is the PKMRC intern and has her room in that building. Please direct any questions to her if I am not available. Please use the outside entrances to the back room and lab rather than going through Juli's room. For those staying in the Bridge Foreman's House (Mote building), note that the toilet is NOT functional but there is a shower. Use restrooms in the bath/shower house (Paint Foreman's House) or the Section Gang Quarters. Those staying in the Bridge Tender's House should also use those facilities. It is suggested that personal valuables not be left lying about when out of your rooms. The public often wanders through in spite of any posted signs.

Those funded by the EPA Region 4 Special Studies project (EP, JP, KP & KT) have breakfast and lunch provided; there is a $12.00 /day allowance for dinner (save and submit all receipts). For all others, breakfast ($3.00) and/or lunch ($5.00) is available. Breakfast will include coffee, tea, orange juice, cereals, bagels, muffins, etc. and any other reasonable requests. Breakfast food will be placed in the Bridge Tender's House kitchen. Lunches include a variety of sandwiches and salads of your choice along with snacks and drinks packed for the boat. There is a menu card that each person should fill out in the evening for the next day. Juli will be handling the food; submit the menu cards to her and any other requests.

A pay telephone is available in the Section Gang Quarters. The phone in the Mote office is also available but please keep use to a minimum and use a calling card if calling long distance unless the call is related to this project. On-line services include Web access, Telnet and FTP. The modem line is shared with the FAX so please minimize use during business hours.

MAP OF PIGEON KEY



DIVING POLICIES

Unless previously certified as a Mote diver, please fill out the Diver Resume, Medical History and provide medical clearance for diving within the past year (need not be Mote form). Also, we will need to photocopy your certification card and conduct a brief, in-water check-out. A general waiver form also needs to be signed by those who have not done so already. Divers should report ANY problems to E. Mueller as soon as possible, both potential medical or gear difficulties.

Weights are available and tanks will be provided as needed. Tanks will be in E. Mueller's truck and should be placed there again at the end of the day. A rinse tank is located behind the Bridge Foreman's House near the dock and a dive gear rack nearby.

FOUL WEATHER ACTIVITIES

Given the El Niño effects on weather this past winter, it is unlikely that we will be able to complete the schedule that follows. Discussion of manuscripts or proposals is certainly a primary alternative for windy or otherwise poor water days. On Thursday, 4/16, and Friday, 4/15, there will be meetings in Key West regarding the development of an ecological reserve in the Dry Tortugas. I believe all would be welcome to attend if we can't get into the field on those days. An agenda for "Tortugas 2000" is attached.

all would be welcome to attend if we can't get into the field on those days. An agenda for "Tortugas 2000" is attached.

Study teams based on the Mote vessel, *Montastræa* (E. Mueller, captain). Survey teams will largely be based on the on the EPA Mako 22 (J. Campbell, captain). Photomonitoring team based on Reef Relief vessel (C. Quirilo, captain). Normal departure time from dock is 09:00 except Monday, 4/13 when a noon departure is planned. Return to dock 17:00-18:00. Please have ALL gear loaded at least 15 minutes prior to departure.

Monday, 4/13

Survey Team: none

Study Team (EM, JC, JP, KP, DM): Load Hydrolab software; calibrate DataSonde; establish study site and begin mapping; deploy DataSonde; get Mako.

Tuesday, 4/14

Study Team (JP, EM, DM): Begin marking and imaging of diseased corals; collect some corals and *Coralliophila*.

Survey Team (JC, LM, KP): Establish survey sites; begin surveys.

survey #	coral count	disease count	transect
1	KP	LM	JC
2	JC	LM	KP

ST – Pick up Esther, 11:45 AM, Marathon Airport

Wednesday, 4/15

Study Team (JP, EM, EP, KP, DS): Complete marking and imaging of diseased corals; mark and image healthy corals; complete mapping; collect coral specimens.

Survey Team (JC, LM, ST): Site surveys.

survey #	coral count	disease count	transect
1	DS	JC	LM
2	DS	LM	ST
3	DS	JC	ST

Lab Team (DM) – Monitor *Coralliophila* experiment.

Evening – Slide Exams I (ST, LM, EM, JP)

Thursday, 4/16

Survey Team *Montastræa* (EM, ST, SV, LB): Site surveys.

survey #	coral count	disease count	transect
1	ST	SV	EM
2	ST	SV	LB
3	ST	SV	LM

Survey Team *Mako* (JC, DS, EP): Site surveys.

survey #	coral count	disease count	transect
1	JC	DS	EP
2	EP	DS	JC
3	JC	DS	EP

Lab Team (DM) – Monitor *Coralliophila* experiment; pick up KT in Ft. Lauderdale.

Photomonitoring Team (CQ, JP, KP): Photograph Eastern Dry Rocks photostation.

Evening – Slide Exam II (DS, JC, LB, SV)

Friday, 4/17

Study Team (JC, JP, DS, KT): Complete collections (AM trip); sample processing.

Survey Team (EM, LB, EP, KP): Site surveys.

survey #	coral count	disease count	transect
1	LB	EP	KP
2	KP	LB	EM
3	LB	EP	EM

Lab Team (DM) – Monitor *Coralliophila* experiment.

Evening – Slide Exam III (EP, KT, KP, DM)

Saturday, 4/18

Survey Team (EM, LB, EP, KP): Site surveys.

survey #	coral count	disease count	transect
1	EP	LB	EM
2	KP	EP	EM
3	EP	LB	KP

Lab Team (DM, KT, JP) – Monitor *Coralliophila* experiment and finish processing samples.

Sunday, 4/19

Survey Team (EM, JP, KT, DM): Site surveys.

survey #	coral count	disease count	transect
1	EM	KT	JP
2	EM	KT	DM
3	EM	KT	JP

Lab Team (EP, KP) – Monitor *Coralliophila* experiment and collate data.

Monday, 4/20

Study Team (EM, EP, JP, KP, KT, DM): Re-image corals; return *Coralliophila*; retrieve DataSonde.

CORAL DISEASE FIELD STUDY 98B

2-8 August, 1998
Pigeon Key, FL

Participating Institutions

Mote Marine Laboratory
Tetra Tech, Inc.
University of Georgia
Florida International University

Prepared by:

Erich Mueller
Mote Marine Laboratory

Coral Disease Field Study 98b

Table of Contents

Participants & Roles

Addresses and numbers for some participants are listed in the QAPP appended to this schedule and not reiterated below. More detailed roles in the overall project are also listed in the QAPP. Contact points for those not in the QAPP are listed below.

Erich Mueller, Mote Marine Laboratory: PI/PD Alternate QA Officer
P: (305) 289-4282
F: (305) 289-9664
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Pigeon Key Marine Research Center
P.O. Box 500895
Marathon, FL 33050

Esther Peters, TetraTech, Inc.: Co-PI, QA Officer
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F: (703) 385-6007
email: PeterEs@tetratech-ffx.com
10306 Eaton Pl.; Suite 340
Fairfax, VA 22030

Karen Porter, University of Georgia: Co-PI
P: (706) 542-3410
F: (706) 542-3344
email:kporter@sparrow.ecology.uga.edu
711 Biological Sciences Building
University of Georgia
Athens, GA 30602-2602

Katherine Patterson, University of Georgia: graduate student
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711 Biological Sciences Building
University of Georgia
Athens, GA 30602-2602

Lillian Becker, Florida International University: graduate student
P: (305) 662-8682
F: (305) 348-1986
email: lbecke01@fiu.edu
Department of Biological Sciences
Florida International University
Miami, FL

Doug Marcinek: research assistant
P: (706) 542-3410
F: (706) 542-3344
email: marcinek@arches.uga.edu
711 Biological Sciences Building
University of Georgia
Athens, GA 30602-2602

Travel Plans and Accommodations

Room codes are: Mote = Bridge Foreman's House, next to dock and BTH = Bridge Tender's House, directly facing the Mote building.

Name	Initials	Dates	Arrive Via	Room
Erich Mueller	EM	8/3 - 8/8	car	off site
Esther Peters	EP	8/3 - 8/8	arrives Marathon on 8/2/98 @11:45	BTH
Karen Porter	KP	8/3 - 8/8	car	off site
Katie Paterson	KT	8/3 - 8/8	car; arrives Big Pine Key on 7/31/98	off site
Lillian Becker	LB	8/3 - 8/6	car	Mote
Doug Marcinek	DM	8/3 - 8/6	car	off site

GENERAL INFORMATION

Vehicle access to Pigeon Key via the Old Seven-Mile Bridge is restricted to those with specific business on the island (including yourselves). Visitors arrive by walking, bicycles, rollerblades or the Pigeon Key train. Please car pool insofar as possible when driving off the island to minimize vehicular traffic on the bridge. Always give pedestrians the right-of-way and follow the posted speed limits (25 mph max; 15 mph around pedestrians). Many bridge users are locals who will let us know promptly if anybody drives improperly.

The PKMRC office is in the Bridge Foreman's House (see map next page) along with a small dry lab and some accommodations. Elizabeth (Liz) Glynn is the PKMRC intern and has her room in that building. Please direct any questions to her, or Lill Becker, if I am not available. Please use the outside entrances to the dry lab rather than going through Liz's or Lill's room. Please note that the toilet is NOT functional in the Mote building but there is a shower. Use restrooms in the bath/shower house (Paint Foreman's House) or the Section Gang Quarters. Those staying in the Bridge Tender's House should also use those facilities. It is suggested that personal valuables not be left lying about when out of your rooms. The public often wanders through in spite of any posted signs.

Lunch will be provided for those funded by the EPA Region 4 Special Studies project (EP, JP, KP & KT). There is an additional $14.00 /day allowance for breakfast and dinner (save and submit all receipts). Lunches include a variety of sandwiches and salads of your choice along with snacks and drinks packed for the boat. There is a menu card that each person should fill out in the evening for the next day. Liz will be handling the food; submit the menu cards to her and any other requests.

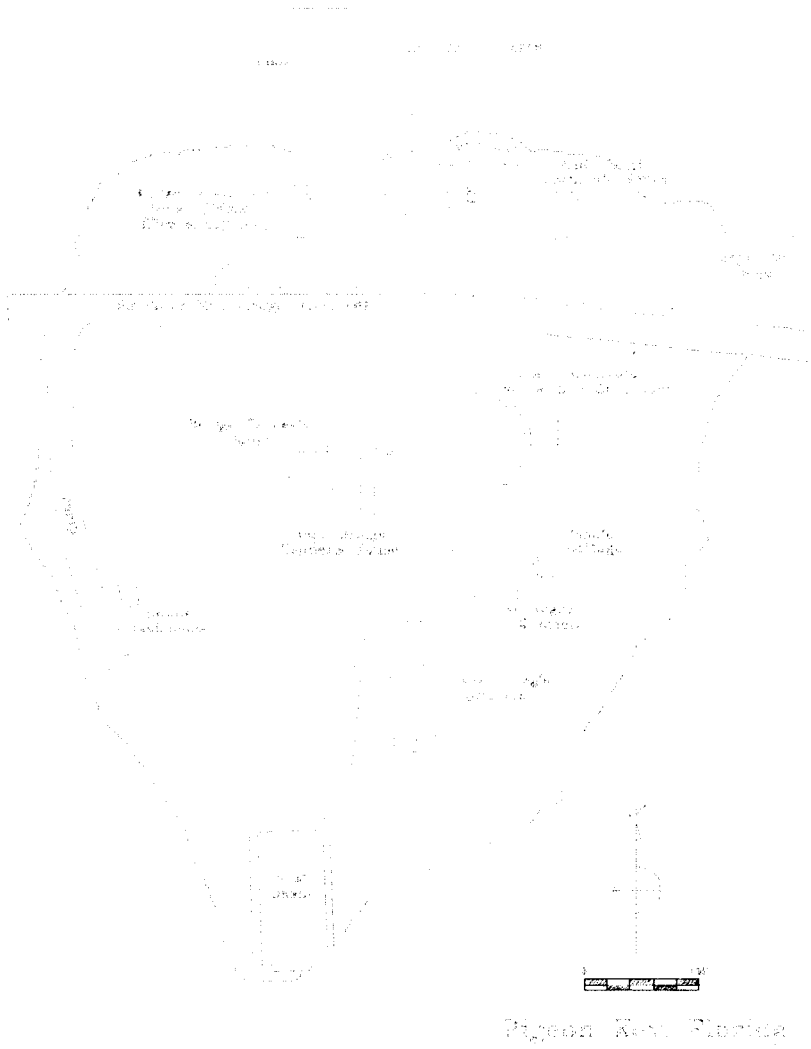
A pay telephone is available in the Section Gang Quarters. The phone in the Mote office is also available but please keep use to a minimum and use a calling card if calling long distance unless the call is specifically related to this project. On-line services include Web access, Telnet and FTP. The modem line is shared with the FAX so please minimize use during business hours (8:30-17:00).

DIVING POLICIES

Unless previously certified as a Mote diver, please fill out the Diver Resume, Medical History and provide medical clearance for diving within the past year (need not be Mote form). Also, we will need to photocopy your certification card and conduct a brief, in-water check-out. A general waiver form also needs to be signed by those who have not done so already. More detailed safety policies follow.

Weights are available and tanks will be provided as needed. Tanks will be in E. Mueller's truck and should be placed there again at the end of the day. A rinse tank is located behind the Bridge Foreman's House near the dock and a dive gear rack nearby.

MAP OF PIGEON KEY



SAFETY POLICIES

General

Vessel and dive operations are potentially dangerous activities. All members of the scientific party are expected to exercise care and diligence to safety protocols listed here or any required by the ship's crew. Injuries or unsafe conditions should be reported to the Chief Scientist as soon as possible or, in an emergency, to any appropriate persons.

Participants should report any health-related problems to the Chief Scientist. Symptoms of dive-related problems may be subtle and as discomfort after a dive should be reported, even if apparently minor. All members of the Scientific Party should be aware of vessel safety protocols and the location of first aid supplies.

Dive Procedures

Dive Team – The dive team will consist of a minimum of four persons including at least two divers, a stand-by diver and a vessel operator. Three-person buddy teams are permissible; solo diving is not.

Equipment – The dive vessel must be equipped with an operational VHF radio, tag line, basic first aid kit and O_2 supply system. All divers will be equipped with an alternate air source (octopus or pony tank), tank pressure gauge, buoyancy compensator, depth gauge and watch. A knife or diver tool is recommended.

Dive profiles – All dives will be non-decompression and utilize air tables conforming to sport diver limits (PADI Recreational Dive Planner). Bottom time will be recorded as surface-to-surface. Reverse profiles (deep dive after shallow dive) are not permissible. All dives over 40' will include a 3 minute safety stop at 10'. Ascent rates not to exceed 30'/minute.

Diver recall – In the event of an emergency in which divers must be recalled, the engine of the dive boat will be revved (in neutral) in groups of three. Maintain safe ascent rate indicated above.

Misc. – Althought the stand-by diver will record tank pressures, dive start/end times and maximum depth, divers must carefully record and monitor their time of descent, depth and bottom time. All dives must be logged each day in personal log books. Divers will be excused from dive activities if they are not feeling comfortable about diving for any reason. Use of decongestants is strongly discouraged and divers with sinus congestion should not dive.

Emergency Contacts

USCG (305) 350-5611, Ch 16, SSB 2182

Chambers: Fisherman's Hospital 24hrs - (305) 743-9891
3301 Overseas Hwy (MM 48.7)
Marathon, FL 33050
Hyperbaric physicians: Dr. Paul Buza, Dr. Dean Bard
Safety officer: Mr. Kim McDonald

Navy Base Key West (305) 293-4347
Master Sgt Dait Operations Sgt.
Status: Need to contact at commencement of operations.

FOUL WEATHER ACTIVITIES

Hopefully, this Field Study will not be a repeat of the first in April with respect to weather. At this time of year it is very possible that we could complete the schedule that follows. However, tropical weather systems could thwart our schedule. Discussion of manuscripts or proposals is certainly a primary alternative for windy or otherwise poor water days.

Schedule – 7/23/98

All Field Study activities are planned using the Mote vessel, R/V *Montastræa* (E. Mueller, captain; L. Becker, mate). Normal departure time from dock will be 09:00 with a return to the dock at 17:00-18:00. Please have ALL gear loaded at least 15 minutes prior to departure.

Monday, 8/3 (EM, EP, KP, KT, LB, DM)

Field: Field planning; calibrate DataSonde; vessel safety procedures at Pigeon Key; establish study site at LK03; deploy DataSonde; begin mapping site; collect *A. palmata* and *Coralliophila* for experiment.

Lab: Begin *Coralliophila* experiment at Pigeon Key.

Tuesday, 8/4 (EM, EP, KP, KT, LB, DM)

Field: Complete mapping; begin marking and imaging (photo/video) of diseased corals; collect coral specimens for microbiology/histology.

Lab: Begin processing specimens; monitor *Coralliophila* experiment.

Wednesday, 8/5 (EM, EP, KP, KT, LB, DM)

Field: Complete marking and imaging of diseased corals; begin marking and imaging healthy corals; collect coral specimens as necessary.

Lab: Continue processing specimens; monitor *Coralliophila* experiment.

Thursday, 8/6 (EM, EP, KP, KT, LB, DM)

Field: Complete marking and imaging healthy corals; collect coral specimens as necessary.

Lab: Continue processing specimens; monitor *Coralliophila* experiment.

Friday, 8/7 (EM, EP, KP, KT)

Field: Looe Key disease surveys. Note: also included as part of the Eastern Keys Survey 98b.

Lab: Continue processing specimens; monitor *Coralliophila* experiment.

dive #	station	coral count	disease count	transect line	stand-by
1	LK01	EM	EP	KT	KP
2	LK02	KP	EP	EM	KT
3	LK02	KP	EP	KT	EM
3	LK02	KP	EP	KT	EM
snorkel	LK03	EM	EP	KP	KT

Schedule (cont.) – 7/23/98

Saturday, 4/8 (EM, EP, KP, KT)

Field: Disease surveys. Because of dive profiles, surveys will be on stations in first OR second column shown below. Note: also included as part of the Eastern Keys Survey 98b.

Lab: Complete all sample processing; complete *Coralliophila* experiment.

dive #	station		coral count	disease count	transect line	stand-by
1	DS01	SR01	EM	EP	KT	KP
2	SR01	SR02	EM	EP	KP	KT
3	SR02	MP01	EM	EP	KT	KP

Sunday, 4/9 (EM, EP, KP, KT)

Field: Contingency day for disease surveys.

TRAINING & CERTIFICATION PROGRAM

Table 1 - Planned Coral and Disease Counts – Note: JP, KT and DM previously certified for coral counts through CRMP procedures. EM (coral & disease) & LB (coral) certified during the Eastern Keys Survey 98a.

Initials count	Friday 8/7/98	total
EP disease	3	3
KP coral	3	3

EASTERN KEYS CORAL DISEASE SURVEY 98A

19-26 May, 1998
M/V Whisker

Participating Institutions

Mote Marine Laboratory
Florida International University
Nova Southeastern University

Prepared by:

Erich Mueller
Mote Marine Laboratory

Eastern Keys Coral Disease Survey, 1998a

19-26 May, 1998

M/V Whisker

Table of Contents

Ship's Crew

Stan Jenkins – captain
Janet Matheny - first mate

Scientific Party

All will help with galley duties in rotation as requested by the first mate/cook. The captain may also request help with SCUBA tank fills. All will assist in electronic data entry.

Erich Mueller (Mote Marine Laboratory), chief scientist and primary coral counter - Organizes cruise, oversees all operations and completes reports. Will install all stations and conduct coral/disease counts.

Lillian Becker (Florida International Univ.), primary disease counter - Counts diseases on corals. Prepares slates with data forms prior to survey, handles rinsing and drying afterwards. Places and retrieves the start/end buoy during survey.

Tim Howe (Library of Congress), QC officer and assistant - Oversees quality control procedures in the field including checking data forms each day for completeness and addition errors. Resolves any discrepancies with the survey team. Files completed forms. Performs assistant roles described below.

Susan Thornton (Nova Southeastern Univ.), secondary coral counter, GIS management and assistant – Will count corals if primary counter can not. Oversees position determination and recording of station forms. Performs assistant roles described below.

T. Shay Viehman (Newfound Harbor Marine Institute), secondary disease counter, sample collection management and assistant – Will count diseases if primary counter can not. Collects samples of *Aplysina fistularis* and logs. Performs assistant roles described below.

Assistant Roles - There are several services that assistants will perform as described below, generally in rotation.

1) transect line handling - Assistant carries pivot rod and transect line to the station. The pivot rod is inserted into the station pipe, the transect attached to the swivel, height adjusted and the transect extended along the line marked by the buoy deployed by the disease counter.
2) stand-by diver - Has tank set-up and equipment ready to don in an emergency
3) top-side support - Helps divers prepare and handles gear to and from divers. Operates boat in an emergency.

Daily Schedule

07:30 - 08:00 > breakfast
08:00 - 08:30 > AM briefing
09:00 - 11:30 > survey 1
12:00 - 12:30 > lunch
13:00 - 17:00 > surveys 2 & 3
17:00 - 18:00 > gear stowage, maintenance & raw data quality check
19:00 - 20:00 > dinner
20:00 - 21:00 > electronic data entry; program GPS for next day; sample processing

Cruise Schedule

The first day will be spent in the Key West area conducting training and QC surreys on a single station established during 1997. The *O.S.V. Peter W. Anderson* (EPA) will also utilize this site on 23 May for the same purposes. For the remainder of the cruise, there are two options (listed below) depending on weather and other factors determined by the captain. The Sombrero Key stations will not be included in the cruise but will be surveyed shortly afterwards.

Day	Date	Site	Tasks
0	19 May	Conch Harbor, Key West	mobilize
1	20 May	Sand Key 1 – SK01	training & QA/QC
		Option A	
2	21 May	Palmata & Pacific Reefs – Biscayne N.P.	transitional reef, forereef, back reef
3	22 May	Carysfort Reef SPA	transitional reef, forereef, back reef
4	23 May	Molasses Reef SPA	transitional reef, forereef, back reef
5	24 May	Alligator Reef	transitional reef, forereef
6	25 May	Looe Key Reef SPA	transitional reef, forereef, back reef
7	27 May	Eastern Sambos Research Zone	transitional reef, forereef, back reef
7	27 May	Conch Harbor, Key West	demobilize
		Option B	
2	21 May	Eastern Sambos Research Zone	transitional reef, forereef, back reef
3	22 May	Looe Key Reef SPA	transitional reef, forereef, back reef
4	23 May	Alligator Reef	transitional reef, forereef
5	24 May	Molasses Reef SPA	transitional reef, forereef, back reef
6	25 May	Carysfort Reef SPA	transitional reef, forereef, back reef
7	27 May	Palmata Reef & Pacific Reef – Biscayne N.P.	transitional reef, forereef, back reef
7	27 May	Conch Harbor, Key West	demobilize

Training and QA/QC Schedule

Wednesday, 20 May at SK01

Procedures (on deck)

1) transect protocol
2) data forms
3) GPS operations

Snorkel

1) locate station
2) attach buoy
3) determine averaged DGPS position

Dive Schedule – T. Howe will serve as stand-by diver and have check-out on second dive.

Dive#	Survey #	Mueller	Becker	Thornton	Viehman
1 (AM)	1	coral & disease	disease	observer	run transect
	2	observer	run transect	coral	disease
2 (PM)	3	coral & disease	disease	run transect	observer
	4	check-out	run transect	coral	disease
3 (PM)	5	coral & disease	disease	observer	run transect
	6	observer	run transect	coral	disease

Review – Discuss field procedures and changes as necessary.

Data Management Procedures

1) U/W form rinse and dry
2) QC check
3) data entry
4) form filing

Safety Procedures

General

Vessel and dive operations are potentially dangerous activities. All members of the scientific party are expected to exercise care and diligence to safety protocols listed here or any required by the ship's crew. Injuries or unsafe conditions should be reported to the Chief Scientist as soon as possible or, in an emergency, to any appropriate persons.

Participants should report any health-related problems to the Chief Scientist. Symptoms of dive-related problems may be subtle and should not be discounted.

All members of the Scientific Party should be aware of vessel safety protocols and the location of first aid supplies.

Dive Procedures

Dive Team – The dive team will consist of a minimum of four persons including at least two divers, a stand-by diver and a vessel operator. Three-person buddy teams are permissible; solo diving is not.

Equipment – All divers will be equipped with an alternate air source (octopus or pony tank), tank pressure gauge, buoyancy compensator, depth gauge and watch. A knife or diver tool is recommended. The dive vessel must be equipped with an operational VHF radio, tag line and O_2 supply.

Dive profiles – All dives will be non-decompression and utilize air tables conforming to sport diver limits (PADI Recreational Dive Planner). Bottom time will be recorded as surface-to-surface. Reverse profiles (deep dive after shallow dive) are not permissible. All dives over 40' will include a 3 minute safety stop at 10'. Ascent rates not to exceed 30'/minute.

Diver recall – In the event of an emergency in which divers must be recalled, the engine of the dive boat will be revved (in neutral) in groups of three. Maintain safe ascent rate indicated above.

Misc. – Divers must carefully record and monitor their time of descent, depth and bottom time. All dives must be logged each day. Divers will be excused from dive activities if they are not feeling comfortable about diving for any reason. Use of decongestants is strongly discouraged and divers with sinus congestion should not dive.

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Hyperbaric physicians: Dr. Paul Buza, Dr. Dean Bard
Safety officer: Mr. Kim McDonald

Navy Base Key West (305) 293-4347
Master Sgt Dait Operations Sgt.
Status: Need to contact at commencement of operations.

Station Name System for Permanent Coral Disease Stations

The following system was recommended after the June, 1997 *O.S.V. Peter W. Anderson* cruise and adopted by the Coral Disease Consortium. It has been since modified to include "study region". The operational areas of "Western" and "Eastern" Keys are useful because they are presently surveyed separately on different platforms, however, these terms are NOT part of the station name designation. Additional regions, areas, sites and stations will be added as warranted.

1) study region - General operational area. The Florida Keys are "FLK". The Bahamas are "BAH".

2) study area - There are several study areas containing clusters of sites. Each study area has geographic similarity that is related to broad scale hydrologic and other similarities. Where a study area appears to be very homogeneous, it may be synonymous with a "site" [see 3)]. In the Florida Keys the following study areas have been defined: Dry Tortugas, (DT), New Grounds (NG), Key West (KW)., Lower Keys (LW), Middle Keys (MD), Upper Keys (UP) and Biscayne National Park (BP). In the Bahamas, only the Exumas area (EX) has been defined in the area around Lee Stocking Island.

3) site - A site includes a particular reef or group of coral habitats that are clustered within a study area. Although within an area of similar conditions, there would be enough differences in hydrologic or geologic conditions or exposure to anthropogenic influences to warrant their designation as a distinct site worthy of examination. On 1997 cruises, sites were referred to as "stations" and included Loggerhead Key, Western Sambo Reef, Sand Key, etc. The system used here bases site names on the reef name. Where there was no formally named reef, a generic term was used, e.g. Loggerhead Reefs.

4) zone - Most reef systems have zones defined by varying wave energy and irradiance with most other parameters being fairly equal. Distinct coral communities can be found in these zones although one can grade into the other with transitional communities. During the 1997 cruises, we identified three general zones of interest on typical bank/barrier reefs such as those found along most of the Keys. Zones of particular interest for the CDC Coral Disease Surveys include the backreef, specifically areas of *Acropora palmata* dominance, the forereef (shallow and deep forereef zones not discriminated) and transitional reef areas. Selection of the zones is important for comparison purposes, however, use of the zone code is not absolutely necessary to provide a unique station designation (see below). Zone codes presented in table below.

5) station - Conceptually, stations are where measurements or data are actually collected. With the radial transect, the station coordinates would be the point at which the pivot rod is inserted. Many stations were marked at this point by a stainless tube and float during the September, 1997 cruise. One or more stations may be established within a given zone of a given reef. The full station designation is of use during statistical coding. In practice, only the station code (site and number) need be used as it is unique within a region although adding the zone would make its character more intuitive to a reader.

Full station designation:	region-area-site#-zone
Full station designation example:	FLK-KW-SK05-BR
Station code example:	SK05 or SK05-BR

Table of Zone Codes

Zone	Code	Depth (ft)	Description
hard bottom	HB	8-40	exposed bedrock with scattered scleractinians, gorgonians and sponges; not sufficiently concentrated to be considered as a reef
inshore patch reef	IP	2-10	shallow reef without extensive internal zonation; within 1 nm of shore
mid-channel patch reef	MP	20-35	fairly deep-water patch reef; typical of Hawk Channel
offshore patch reef	OP	15-30	moderately deep patch reef on outer edge of Hawk Channel
pinnacle /platform reef	PR	35-70	found in areas of the Dry Tortugas; steep sides
backreef	BR	1-8	bank reef zone shoreward of reef crest; may include rubble or coral areas, particularly *Acropora palmata*
reef crest	RC	0-5	bank reef zone dominated by *Millepora complanata*, *Porites astreoides* and *Palythoa* spp.; NOT suitable for surveys
shallow forereef	SF	1-15	bank reef zone immediately seaward of reef crest; usually has well-developed spur-and-groove structure
deep forereef	DF	15-30	bank reef zone seaward of shallow zone; sometimes called boulder zone due to large massive coral species; zone merges with shallow forereef
transitional reef	TR	25-45	low-relief area that may or may not have spur-and-groove structure; may have scattered large corals or diffuse *Acropora cervicornis* thickets
intermediate reef	IR	35-60	give-up reef zone usually associated with bank reefs; usually has low-relief spur-and-groove structure
deep reef	DR	60-110	deep reef area often associated with bank reefs; often ledge-like with "walls" of 5-20' height

Radial Belt Transect Standard Operating Procedure

Introduction

The radial belt transect method involves surveying 2-3 m wide belts in concentric circles around a pivot point. During 1997, this method was extensively tested during cruises of the *O.S.V. Peter W. Anderson* from Key West to the Dry Tortugas and at Lee Stocking Island in the Bahamas. Originally, the complete area out to a radius of 10 m (314.16 m^2) was surveyed. There were initially five 2m intervals (0→2; 2→4; 4→6; 6→8; 8→10) and this was reduced for efficiency to four intervals (0→3; 3→6; 6→8; 8→10). Analysis of the data by Dr. D. Santavy showed that it was not necessary to survey such a large area to be representative of the site. The standard procedure to be used in 1998 includes only the 8→10 belt (113.1 m^2). The same area could be examined from 0-6 m, however, that approach can lead to overrepresentation because of the "patchiness" of some certain coral species and, particularly, their diseases. This approach spreads the survey over a larger area. Also, eliminating contiguous belts eliminates the possibility of double counting.

Station Establishment

Random latitudes and longitudes are generated by computer using a program developed by the EPA. Using benthic habitat maps recently produced by the Florida Marine Research Institute (DEP)_ and NOAA, zones are defined within which five random coordinates are produced. These are visited in numeric order and the first suitable station is selected to represent that zone. The other coordinates are discarded. If the coordinates are on unsuitable substrate (i.e. sand, or grass), then the next is visited and so on until a suitable survey station is located. Stations are established as follows:

1) boat is anchored on the GPS coordinates or as close as possible in sand or grass
2) the dive team swims 20 kicks on a random compass bearing
3) a 1" hole is drilled with a pneumatic drill about 4-6" deep on the closest bare substrate
4) a stainless steel pipe (1" x 12") is driven into the hole
5) the survey is conducted as described below; the station location is surveyed with a floating DGPS unit using the averaging function to obtain figures of merit (FOM) around 6-7'
6) the pipe is secured with Liquid rock epoxy
7) the pipe is capped (safety caps used for rebar) and buoyed

Standard Survey Procedure (3-person team)

1) remove cap & float (if present) and place in gear bag (coral counter)
2) insert pivot rod (transect line handler)
3) attach transect line to swivel and adjust height to clear near-by corals. large sponges or reef structure keeping as low as possible (transect line handler)
4) extend line to 10 m; slide weighted flagging tape to 8m and 10 m (transect line handler)
5) place buoy at the start/end point; generally should be a down-current point (disease counter)
6) line handler starts on coral counter's signal, swimming in a counter-clockwise direction; counters record (all)
7) after completing belt, buoy is retrieved and general notes recorded (disease counter)
8) retrieve transect line, unclip and place in gear bag (transect line handler)
9) remove pivot rod (transect line handler)
10) replace buoy and cap with new (coral counter)

Collection of Sponges for Chemical Contaminant Analysis

Aplysina fistularis will be collected in 50-100g (wet weight) or 50-100 ml (volumetric displacement) quantities at locations throughout the FKNMS (conversion from wet volume to dry weight is approximately 175mg/ml for each sponge, Chanas and Pawlik, 1996). Sponge specimens will be cut with a knife or scraped from an encrusting substrate with a knife and placed in the zip lock bags in the field. A fifty-ml displacement volume will be determined and the material will be transferred to a pre-cleaned, wide-mouth polycarbonate jars (250ml) with polypropylene tops. Samples will then be frozen.

The quantity of each sponge specimen and location collected will be recorded on the Sample Tracking Form. Procedures for sample labeling are shown below:

Record Number: region site # year month day-specimen# specimen type

Definitions:

region: FLK = Florida Keys
site: 2 letters to designate reef
#: station number (91, 92, 93, etc.)
year month day: 2 digits each
specimen #: 1-3 digits to designate unique specimen
specimen type: C=coral, S=sponge, W=water parameter.

Example of record number: **FLKLK91980518-2S** The specimen was collected in the Florida Keys at site LK (Looe Key), on 18 May, 1998 at station 91 (first collection station at this site). It is designated as sponge specimen 2 collected on that day. A site form is completed for that station as described on p. 6 and 7.

EASTERN KEYS CORAL DISEASE SURVEY 98B

7-14 August, 1998
R/V Montastraea
M/V Whisker

Participating Institutions

Mote Marine Laboratory
Tetra Tech, Inc.
University of Georgia
Florida International University

Prepared by:

Erich Mueller
Mote Marine Laboratory

Eastern Keys Coral Disease Survey, 1998b

7-14 August, 1998

R/V Montastraea and *M/V Whisker*

Table of Contents

Eastern Keys Coral Disease Survey, 1998b

Vessel Crews

R/V Montastraea

Erich Mueller – captain

Lillian Becker – mate

M/V Whisker

Stan Jenkins – captain

Janet Matheny - mate

Scientific Party

All will help with galley duties in rotation as requested by the morale officer.

Erich Mueller (Mote Marine Laboratory), chief scientist and primary coral counter - Organizes cruise, oversees all operations and completes reports. Will install all stations and conduct coral counts.

Esther Peters (Tetra Tech, Inc.), project QA Officer and primary disease counter - Places and retrieves the start/end buoy during survey. Counts diseases on corals. Oversees collection of and any specimens such as *Aplysina fistularis*. Oversees all QA procedures.

Lillian Becker (Florida International Univ.), backup coral/disease counter and assistant (*M/V Whisker* only) - Prepares slates with data forms prior to survey, handles rinsing and drying afterwards. Will count corals or diseases if one of the primary counters cannot. Performs assistant roles described below.

Tim Howe (Library of Congress), GIS management, morale officer and assistant (*M/V Whisker* only) - Oversees position determination and recording of station forms. Coordinates food service planning and keeps everyone in good cheer. Performs assistant roles described below.

Harry (Chip) McCarty, cruise QA officer and assistant (*M/V Whisker* only) - Conducts quality checks of data forms each day for completeness and addition errors. Resolves any discrepancies with the survey team. Files completed forms. Performs assistant roles described below.

Katherine Patterson (Univ. of Georgia), backup disease counter, GIS management and assistant (*R/V Montastraea* only) – Counts diseases if primary counter cannot. Oversees position determination and recording/updating of station forms. Performs assistant roles described below.

Karen Porter (Univ. of Georgia), backup coral counter, cruise QA officer and assistant (*R/V Montastraea* only) – Counts corals if primary counter cannot. Conducts quality checks of data forms each day for completeness and addition errors. Resolves any discrepancies with the survey team. Files completed forms. Performs assistant roles described below.

Assistant Roles - Assistants will perform duties as described below, generally in rotation.

1) transect line handling - Assistant carries pivot rod and transect line to the station. The pivot rod is inserted into the station pipe, the transect attached to the swivel, height adjusted and the transect extended along the line marked by the buoy deployed by the disease counter.

2) stand-by diver - Has tank set-up and equipment ready to don in an emergency.

3) top-side support - Helps divers prepare and handles gear to and from divers. Operates boat in an emergency.

Eastern Keys Coral Disease Survey, 1998b

Daily Schedule

07:30 - 08:00 > breakfast

08:00 - 08:30 > AM briefing

09:00 - 11:30 > survey 1

12:00 - 12:30 > lunch

13:00 - 17:00 > surveys 2 & 3

17:00 - 18:00 > gear stowage, maintenance & raw data quality check

19:00 - 20:00 > dinner

20:00 - 21:00 > electronic data entry; prepare GPS and forms for next day; sample processing

Cruise Schedule

The first two days will be based at Pigeon Key, using the *R/V Montastraea* to survey Looe and Sombrero Keys. On the first day, planned for Looe Key, three replicate surveys will be conducted by E. Peters and K. Porter. A detailed schedule for this day follows the cruise schedule. The second day includes two surveys at previously established stations at Sombrero Key. Time permitting, an additional survey will be conducted at either Delta Shoals (DS01; 35') or the Marathon Patch Reefs (MP01; 15'). August 9 will be a contingency day for Looe or Sombrero Keys, if necessary, but is planned as a lay day with boarding of the *M/V Whisker* after dinner (18:30 at Turtle Kraals). There are two basic options for the *M/V Whisker* cruise which will be at the Captain's discretion in consultation with the Chief Scientist.

Day	Date	Site	Tasks
1	7 Aug.	Looe Key	QA/QC; surveys
2	8 Aug.	Sombrero Key and DS01 OR MP01	surveys
3	9 Aug.	Conch Harbor, Key West	mobilize for *M/V Whisker* cruise
		M/V Whisker cruise - Option A	
4	10 Aug.	Elkhorn & Pacific Reefs – Biscayne N.P.	surveys
5	11 Aug.	Carysfort Reef SPA	surveys
6	12 Aug.	Molasses Reef SPA	surveys
7	13 Aug.	Alligator Reef	surveys
8	14 Aug.	Eastern Sambos Research Zone	surveys; install ES03 station
8	14 Aug.	Conch Harbor, Key West	demobilize
		M/V Whisker cruise - Option B	
4	10 Aug.	Eastern Sambos Research Zone	surveys; install ES03
5	11 Aug.	Alligator Reef	surveys
6	12 Aug.	Molasses Reef SPA	surveys
7	13 Aug.	Carysfort Reef SPA	surveys
8	14 Aug.	Palmata Reef & Pacific Reef – Biscayne N.P.	surveys
8	14 Aug.	Conch Harbor, Key West	demobilize

Eastern Keys Coral Disease Survey, 1998b

Training, QA/QC and Survey Schedule for Friday, 7 August at Looe Key

Procedures (on deck)

1) transect protocol
2) data forms
3) GPS operations

Snorkel at LK02 Prior to Survey (E. Mueller and K. Patterson)

1) locate station
2) attach buoy
3) determine averaged DGPS position

Dive Schedule

dive #	station	coral count	disease count	transect line	stand-by
1	LK01	EM	EP	KPatt	KPort
2	LK02	KPort	EP	EM	KPatt
3	LK02	KPort	EP	Kpatt	EM
3	LK02	KPort	EP	KPatt	EM
snorkel	LK03	EM	EP	KPort	Kpatt

Recover DataSonde

Review – Discuss field procedures and changes as necessary.

Data Management Procedures

1) U/W form rinse and dry
2) QC check
3) data entry
4) form filing