

An Integrated Environmental Assessment of the St. Thomas East End Reserves (STEER)



NOAA National Centers for Coastal Ocean Science
Center for Coastal Monitoring and Assessment

Anthony S. Pait
S. Ian Hartwell
Laurie J. Bauer
Dennis A. Apeti
Andrew L. Mason



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National Oceanic and Atmospheric Administration
National Ocean Service (NOS)
National Centers for Coastal Ocean Science (NCCOS)
Center for Coastal Monitoring and Assessment (CCMA)
1305 East/West Highway (SSMC-4, N/SCI-1)
Silver Spring, MD 20910
USA

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Editors
Anthony S. Pait
S. Ian Hartwell
Laurie J. Bauer
Dennis A. Apeti
Andrew L. Mason



United States Department
of Commerce

Penny Pritzker
Secretary

National Oceanic and Atmospheric
Administration

Kathryn Sullivan
Under Secretary

National Ocean
Service

Russell Callender
Assistant Administrator

About this Document

This report represents the culmination of three years of research by NOAA's National Centers for Coastal Ocean Science (NCCOS), Center for Coastal Monitoring and Assessment (CCMA) and local partners, in the St. Thomas East End Reserves (STEER) in the U.S. Virgin Islands (USVI). The purpose of this work was to provide local resource managers with a spatially comprehensive characterization of stressors including chemical contaminants, nutrients, and sedimentation along with their effects, and a biological survey of the entire STEER.

The work was requested by local resource managers, and the data and information generated from this project establishes a baseline of conditions within the STEER, and identifies challenges to be addressed in order to protect and conserve the valuable natural resources within the STEER.

Funding for this project was provided by NOAA's Coral Reef Conservation Program (CRCP). The efforts discussed here were led by NCCOS with significant participation from partners, including CRCP, the USVI Department of Planning and Natural Resources (DPNR) Divisions of Coastal Zone Management, Fish and Wildlife, and Environmental Protection, along with the University of the Virgin Islands, and The Nature Conservancy. NCCOS has been proactive in collaborating with other NOAA line offices as well as federal, state and nongovernmental organization partners to maximize cost-sharing efforts and reach its goals. Their efforts and extramural funding has made it possible to complete assessments that would have otherwise been unobtainable through federal funding alone.

For more information on this work and other CCMA and NCCOS projects, please see:

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<http://coastalscience.noaa.gov/>

Direct questions or comments to:

Mark Monaco, PhD, Director

Center for Coastal Monitoring and Assessment

NOAA National Centers for Coastal Ocean Science

Telephone: 240.533.0327

E-mail: Mark.Monaco@noaa.gov

Executive Summary

The St. Thomas East End Reserves, or STEER, is a collection of Marine Reserves and Wildlife Sanctuaries (MRWS) located on the southeastern end of the island of St. Thomas, U.S. Virgin Islands. With an area of approximately 9.6 km², the STEER contains extensive mangroves and seagrass beds, along with coral reefs, lagoons and cays. Within the surrounding watershed are numerous marinas and hotels/resorts, a landfill serving both St. Thomas and St. John, an EPA Superfund site, residential areas with individual sewage treatment systems, and in the nearshore environment live-aboard and derelict boats, all of which can be sources of pollution to the STEER.

Discussions with environmental managers from the USVI Department of Planning and Natural Resources (DPNR) at a meeting in 2009 highlighted the STEER as a priority area. It was noted during these discussions that the input of pollutants to the STEER, many from land-based sources of pollution (LBSP) were thought to be impacting the health of the natural resources living there. DPNR managers also noted there were significant data and information gaps, particularly in terms of the chemical contaminants present, their concentrations, effects, along with the overall health of the biological communities within the STEER. To address these needs, NOAA/NCCOS' Center for Coastal Monitoring and Assessment (CCMA) worked with DPNR and other local partners to design a project that was subsequently funded by NOAA's Coral Reef Conservation Program (CRCP), to develop an integrated chemical and biological characterization of the STEER. Partners in the project included the USVI DPNR Divisions of Coastal Zone Management, Fish and Wildlife, and Environmental Protection, along with the University of the Virgin Islands, and The Nature Conservancy.

In 2011, sediment samples were collected throughout the STEER using a stratified-random sampling design and subsequently analyzed for chemical contaminants, toxicity, and benthic infaunal community condition. This approach allowed for statistical comparisons between the strata established in the STEER. Beginning in 2012, the University of the Virgin Islands began monthly monitoring of nutrients, sedimentation and total suspended solids (TSS) at selected targeted sites. Also in 2012, the first ever biological survey of the entire STEER was conducted by CCMA, NCCOS' Center for Coastal Fisheries and Habitat Research (CCFHR) and The Nature Conservancy SCUBA divers. During that field mission, samples of coral and conch were also collected at targeted sites, for analysis of chemical contaminants.

Results from the analysis of sediments collected in 2011 indicated elevated levels of certain contaminants in northern Benner Bay. A series of follow-up conversations with DPNR managers resulted in additional field work, in which both surface and sediment core samples were collected and analyzed for chemical contaminants in 2013.

The results from the project in the STEER are contained in this report, which is organized into 10 chapters, representing the interrelated studies conducted with local partners. Chapter 1 provides background information on the project and an overview of the study area. Chapter 2 describes the benthic habitat mapping effort, which resulted in a revised high resolution map of the benthic environments in the STEER. Chapter 3 presents the results from the survey of fish communities and associated benthic habitats. Chapter 4 summarizes the first-ever quantification of sediment contaminants and sediment toxicity throughout the STEER. Chapter 5 presents the results of a follow-up effort in northern Benner Bay, to better characterize the distribution of contaminants in surface sediments, and through sediment coring, assess chemical contaminants in deeper, older sediments. Chapter 6 contains an assessment of water soluble contaminants at six sites in the STEER, including an upstream site in Turpentine Gut, using a series of passive water samplers. Chapter 7 summarizes the results of work to analyze coral, conch and fish samples for chemical contaminants. Chapter 8 presents a histologic examination of tissues from the coral *Porites astreoides*, the same coral species analyzed for chemical contaminants. Chapter 9 summarizes the results of monitoring nutrients, sedimentation and total suspended sediments at six sites throughout the STEER for nearly two years. Finally, Chapter 10 provides a summary from the project along with a series of conclusions. Highlights from each chapter are summarized below.

Chapter 1. Introduction

- The STEER contains a variety of habitats and an abundance of natural resources.
- The western half of the STEER contains a greater amount of commercial, residential and industrial activities.
- The goals of the project were to quantify chemical contaminants and effects, and conduct a biological survey, establishing a baseline that can be used to assess change over time, as management actions are implemented to preserve and restore the STEER.

Chapter 2. Benthic Habitat Mapping

- High resolution habitat maps were developed using a combination of semi-automated classification methods. Habitats were interpreted from aerial photographs and LiDAR (Light Detection and Ranging) imagery.
- One hundred ten distinct combinations of habitat classes describing the geology and biology of the seafloor were identified from the aerial photographs and LiDAR imagery.
- The overall map accuracies (corrected for proportional bias) were 93.0% for major structure, 75.1% for detailed structure, 86.2% for percent hardbottom, 86.5% for major cover and 74.5% for detailed cover and 83.3% for live coral cover.
- Unconsolidated sediments were dominant inside the STEER's boundary, with sand colonized by seagrass being the most common habitat type.
- Live coral cover rarely exceeded 10% inside the STEER's boundaries. However, habitat features with >10% live coral cover were located in the eastern portion of the study area, specifically west of Great St. James Island in the STEER, and south and west of Deck Point.

Chapter 3. Fish Communities and Associated Benthic Habitats

- A total of 80 sites were surveyed during a two-week field mission in June 2012.
- Hard coral cover averaged 5.2%, with the greatest coverage observed in the southern study area, particularly on the southwest reef tract near Long Point. Several of the survey sites with relatively high coral cover were located outside the STEER.
- Mustard hill coral (*Porites astreoides*) was the most abundant coral species, followed by boulder star coral (*Orbicella annularis* complex), lesser starlet coral (*Siderastrea radians*), symmetrical brain coral (*Pseudodiploria strigosa*), massive starlet coral (*S. siderea*), and finger coral (*P. porites*).
- In general, both benthic and fish community metrics in the STEER were similar to other U.S. Caribbean monitoring locations sampled with the same methodology.
- Due to low visibility and concerns for diver health, no biological surveys could be conducted in the northern portions of Mangrove Lagoon and Benner Bay, which appear most impacted by LBSP. Previous studies have indicated that species richness and diversity are quite low in these areas, which would have impacted the overall benthic and fish community metrics for the STEER.
- Surveys conducted in the southern portion of Mangrove Lagoon and adjacent coral reefs ranked high in

regards to hard coral cover, fish species richness, total fish density and total fish biomass. Further research is needed to fully examine the current and potential nursery function of the entire lagoon.

Chapter 4. Sediment Chemical Contaminants and Toxicity

- One hundred and eighty-five chemical contaminants were analyzed in sediments, and a series of sediment toxicity bioassays were conducted along with a characterization of the benthic infaunal community, as part of the Sediment Quality Triad, to assess the presence and effects of chemical contaminant stressors within the STEER.
- Higher levels of chemical contaminants were found in Mangrove Lagoon and Benner Bay in the western portion, than in the eastern part of the STEER.
- Copper at one site in Benner Bay was above a NOAA guideline indicating that effects on benthic organisms were likely.
- The banned antifoulant paint ingredient tributyltin or TBT, which had widespread use on boat hulls in the past, was found at the third highest concentration in the history of NOAA's National Status and Trends (NS&T) Program.
- The benthic infaunal communities in Mangrove Lagoon and Benner Bay appeared to be severely diminished.
- Results of the bioassays indicated significant sediment toxicity in Mangrove Lagoon and Benner Bay using multiple tests.

Chapter 5. Butyltins and Metals in Sediment Cores

- The analysis of sediments in the STEER (Chapter 4) prompted a follow-up study in the marina area of northern Benner Bay.
- Results indicated that surface and deeper sediments are highly contaminated with butyltin paint residues, copper, and other toxic metals. The concentration of total butyltins at a depth of 6-8 cm was 8,871 ng Sn/g (tributyltin or TBT was over 5,000 ng Sn/g) in a sample at depth; the NOAA National Status and Trends (NS&T) mean for butyltins in sediments is 8.9 ng/g.
- The system is also receiving fresh inputs of TBT as evidenced by the percentage of undegraded TBT residues in some surface sediments; TBT has been banned for use on most vessels in the U.S. since 1989.
- If dredging to deepen the navigation channels or for remediation purposes occurs in the future in northern Benner Bay, a series of protocols would be needed to prevent resuspension of contaminated sediments into the water column, along with the proper disposal of dredged materials, so as not to impact marine or terrestrial environments.

Chapter 6. Water Soluble Chemical Contaminants

- Passive water samplers (POCIS) were deployed in the STEER at five locations in 2012 to detect the presence of water soluble contaminants.
- A total of 26 stormwater contaminants were detected at least once in the STEER.
- From an earlier (2010) deployment in Turpentine Gut, 31 stormwater contaminants were detected.
- Ambient water concentrations could be estimated for a number of contaminants including the detergent/surfactant metabolite 4-tert-octylphenol, phthalate ester plasticizers DEHP and DEP, bromoform, personal care products including menthol, indole, n,n-diethyl-toluamide (DEET), along with the animal/plant sterol cholesterol, and the plant sterol beta-sitosterol.
- The plasticizer DEHP appeared to be the only compound quantified that exceeded a water quality guideline for the protection of aquatic organisms.

Chapter 7. Contaminants in Coral, Conch and Fish

- Contaminant body burdens in coral (*Porites astreoides*), conch (*Lobatus gigas*) and fish (longspine squirrelfish, *Holocentrus rufus*, and the schoolmaster snapper, *Lutjanus apodus*) were quantified at five sites throughout the STEER.
- Contaminants found in coral from the STEER were similar to the concentration ranges reported in corals from other reef areas in the U.S. Caribbean.
- Conch from the STEER had lower contaminant body burdens relative to published data from south Florida and some other areas of the Caribbean.
- The conch samples from the STEER had contaminant body burdens lower than available FDA action levels for molluscan shellfish consumption.
- PCBs in one fish exceeded an EPA screening value for recreational fishers.
- A significant correlation between higher concentrations of butyltins closer to shore existed for conch, despite relatively low overall concentrations as compared to previous results from the region.

Chapter 8. Histologic Analysis of *Porites astreoides*

- Examinations of the health of the scleractinian coral, *Porites astreoides*, were performed on samples collected at five sites from the STEER.
- Zooxanthellae were present in all samples and were in excellent condition microscopically.
- Mangrove Lagoon had fewer colonies and developing gonads than other nearshore sites; the deepest offshore site in the STEER had the most larvae.
- Some of the samples had single cell or segmental necrosis or apoptosis of the epidermis, gastrodermis, or

calicodermis; colonies from Benner Bay were the most affected.

- Due to the lack of a clear pattern in association of lesions with sites, the results do not support the hypothesis that exposure to land-based sources of pollution was affecting the corals, at least based only on the histopathological examinations.

Chapter 9. Nutrients, Sedimentation and TSS

- As part of the project, the University of the Virgin Islands monitored nutrients, sedimentation, and total suspended solids (TSS) monthly at six sites in the STEER for nearly two years.
- Concentrations of nutrients in the western part of the STEER and in nearshore areas were significantly higher than at sites in the eastern portion of the STEER.
- Using a set of nutrient threshold concentrations noted to sustain macroalgal blooms in Caribbean coral reefs, approximately 60% of the samples collected in the STEER were above the threshold for orthophosphate (HPO_4^-), while 55% of samples were above the dissolved inorganic nitrogen (DIN) threshold.
- Benner Bay had the highest sedimentation rate of any site in the STEER, including Mangrove Lagoon.
- There was also an east/west and north/south gradient in sedimentation, indicative of higher sedimentation rates in the western, more urban areas surrounding the STEER, and in the more nearshore sites.
- TSS also tended to be higher in the western and nearshore areas of the STEER.

Chapter 10. Summary and Conclusions

- The overall condition of the habitats and biological resources that could be surveyed within the St. Thomas East End Reserves appears similar to other coral reef areas in the U.S. Caribbean investigated by NOAA's NCCOS.
- The proximity of the Reserves to the Bovoni Landfill, marinas, and other commercial and industrial activities, combined with likely inputs from residential sewer systems are additional stressors on the STEER.
- No biological surveys could be conducted in the northern portions of Mangrove Lagoon and Benner Bay, due to low visibility and concerns for diver health.
- The presence of chemical contaminants and other stressors (e.g., sedimentation and nutrients) in Mangrove Lagoon and Benner Bay are of particular concern as the mangroves serve as an important nursery area for a variety of vertebrate and invertebrate marine species.
- Loss of suitable habitat further impacts the ability of the STEER to be a source of new recruits of marine resource species for St. Thomas and beyond.

Table of Contents

About this Document	i
Executive Summary	iii
Chapter 1: Introduction	1
Literature Cited	5
Chapter 2: Benthic habitat mapping within the St. Thomas East End Reserves	7
2.1. Introduction	7
2.2. History of benthic habitat mapping in STEER	7
2.3. Habitat Mapping	8
2.4. Results and Discussion	10
Literature Cited	15
Chapter 3: Characterization of fish communities and associated benthic habitats in the St. Thomas East End Reserves	17
3.1. Introduction	17
3.2. Methods	17
3.3. Results and Discussion	21
3.4. Conclusions	69
Literature Cited	71
Chapter 4: An assessment of chemical contaminants, toxicity and benthic infauna in sediments from the St. Thomas East End Reserves (STEER)	75
4.1. Introduction	75
4.2. Methods	78
4.3. Results and Discussion	85
4.4. Conclusions	111
Literature Cited	113
Chapter 5: An assessment of tributyltin and metals in surface sediments and sediment cores from Benner Bay, St. Thomas East End Reserves	119
5.1. Introduction	119
5.2. Methods	120
5.3. Results and Discussion	122
5.4. Conclusions	129
Literature Cited	130
Chapter 6: An assessment of chemical contaminants detected in passive water samplers deployed in the St. Thomas East End Reserves (STEER)	133
6.1. Introduction	133
6.2. Methods	134
6.3. Results and Discussion	136
6.4. Conclusions	142
Literature Cited	142

Table of Contents (continued)

Chapter 7: An assessment of contaminant body burdens in coral (<i>Porites astreoides</i>), conch (<i>Lobatus gigas</i>) and fish (<i>Holocentrus rufus</i> and <i>Lutjanus apodus</i>) from the St. Thomas East End Reserves (STEER).....	145
7.1. Introduction.....	145
7.2. Methods	145
7.3. Results and Discussion	149
7.4. Conclusions.....	166
Literature Cited.....	167
Chapter 8: Histological examination of coral tissue samples from St. Thomas East End Reserves (STEER), USVI.....	173
8.1. Introduction.....	173
8.2. Methods	174
8.3. Results and Discussion	179
8.4. Conclusions.....	189
Literature Cited	190
Chapter 9: An assessment of nutrients, sedimentation, and total suspended solids (TSS) in the St. Thomas East End Reserves (STEER).....	193
9.1. Introduction.....	193
9.2. Methods	193
9.3. Results and Discussion	196
9.4. Conclusions.....	213
Literature Cited.....	214
Chapter 10: Summary and Conclusions.....	217

List of Tables

Table 2.1. The classification scheme used to classify benthic habitats in the STEER in 2013 (Costa <i>et al.</i> 2013).....	8
Table 2.2. Qualitative comparison of the 2001 and 2013 benthic habitat maps in the STEER using the modified STEER boundary to include mangrove habitat in Mangrove Lagoon and Benner Bay.....	14
Table 3.1. Abiotic and biotic variables measured in five quadrats along fish transects.....	19
Table 3.2. Number and type of fouling organisms on marine debris items found during STEER transects.....	33
Table 3.3. Top five families in abundance and total biomass, shown as percent of total, by habitat strata.....	38
Table 3.4. Top five species in abundance and total biomass, shown as percent of total, by habitat strata.....	38
Table 3.5. Pairwise Analysis of Similarities (ANOSIM) comparisons between habitat types based on fish species density and biomass data, and list of top five species contributing to the dissimilarity from the SIMPER analysis.....	39
Table 4.1. Compounds analyzed in sediments from the St. Thomas East End Reserves.....	81
Table 4.2. Comparison of higher concentrations of individual PAHs at STEER sites with NOAA NS&T data.....	87
Table 4.3. Comparison of higher concentrations (ng/g) of pesticides at STEER sites with NOAA NS&T data.....	91
Table 4.4. Toxicity bioassay summary results from the STEER.....	102
Table 4.5. Average (n = 3) P450 bioassay response following 6 and 24 hour incubation with STEER sediment extracts.....	104
Table 4.6. Spearman Rank correlation coefficients (bold) and significance level for community parameters and selected physical and chemical parameters, and toxicological results.....	108
Table 4.7. Results of SIMPER analysis for Strata 1 and 2.....	109
Table 4.8. Top 15 species in each stratum plus excluded sites.....	109
Table 4.9. Average per-station abundance of selected taxonomic groups in Mangrove Lagoon and Benner Bay without stations 1-4P and 2-19P, stations 1-4P and 2-19P together, and the remaining strata in 3 (Nazareth Bay), 4 (Cowpet/St. James Bay), and 5 (Great Bay).....	111
Table 5.1. Concentrations of butyltins (ng Sn/g) and metals (µg/g) in surface sediments.....	123
Table 5.2. Estimated ages of sections within sediment cores from the STEER.....	125
Table 6.1. Location and conditions during deployment of the POCIS canisters in February 2012 and May 2010.....	135
Table 6.2. Estimated water column concentrations of stormwater contaminant residues sampled by POCIS in the STEER and in Turpentine Gut.....	137
Table 6.3. Comparison of estimated concentrations of stormwater contaminants with available water quality criteria.....	141
Table 7.1. Chemical contaminants analyzed in STEER coral, conch and fish samples.....	148
Table 7.2. Summary of means, standard error, and maximum values for organic chemical contaminants analyzed in STEER coral, conch and fish.....	150
Table 7.3. PAHs, TBT and total butyltins in coral tissues from NOAA Caribbean studies.....	151
Table 7.4a. Summary statistics for contaminants (µg/g) in coral tissue, including comparison with other studies.....	157
Table 7.4b. Summary statistics for contaminants (µg/g) in coral tissue, including comparison with other studies.....	157
Table 7.5. Mean metal body burden (µg/g) in coral by stratum.....	158

List of Tables (continued)

Table 7.6. Summary statistics for contaminants ($\mu\text{g/g}$) in conch tissue ($n=10$), including comparison with mean concentration values derived from study of conch contamination in South Florida (Glazer <i>et al.</i> , 2008) and Johor Straits, Malaysia (Said <i>et al.</i> , 2013)	158
Table 7.7. Mean metal body burden in conch collected from the five strata in the STEER ($\mu\text{g/g}$).....	159
Table 7.8. Trace and major element concentrations ($\mu\text{g/g}$) in fish from the STEER.....	160
Table 7.9. Comparison of average elemental composition in fish tissues ($\mu\text{g/g}$) in STEER and Vieques.....	160
Table 8.1. Characteristics and scoring of changes noted in cells and tissues using light microscopic examination of <i>P. astreoides</i> samples collected during this study	176
Table 8.2. Summary statistics for histopathological observations for each parameter for each stratum.....	187
Table 8.3. Summary of results of the histopathological examinations, means of Specific Condition Scores Sum and percent of colonies with selected condition at each site	188
Table 9.1. Location of sites and mean water quality values from STEER sites	195
Table 9.2. Summary statistics for nutrients measured in the STEER	197
Table 9.3. Comparison of STEER nutrient results with those from other U.S. Caribbean locations	198
Table 9.4. Comparison of TSS in the STEER with other locations in the USVI.....	212

List of Figures

Figure 1.1. Individual Marine Reserves that comprise the STEER.....	2
Figure 1.2. Primary watersheds adjacent to the STEER.....	3
Figure 1.3. Marina area in Benner Bay.....	4
Figure 2.1. Spatial extent of benthic habitats in the STEER in the 2001 NOAA map (Kendall, 2001).....	7
Figure 2.2. Aerial photos (left), LiDAR bathymetry (middle), and LiDAR reflectivity (right) used to delineate and characterize the benthic habitats inside STEER (Costa <i>et al.</i> 2013).....	9
Figure 2.3. Summary statistics describing the total amount of mapped area by geographic zone within the entire mapped area and for the portion within the STEER (adapted from Costa <i>et al.</i> 2013).....	10
Figure 2.4. Summary statistics describing the total amount of mapped area by percent hardbottom categories within the entire mapped area and for the portion within the STEER (adapted from Costa <i>et al.</i> 2013)...	10
Figure 2.5. Summary statistics describing the total amount of mapped area by major and detailed geomorphological structure within the entire mapped area and for the portion within the STEER (adapted from Costa <i>et al.</i> 2013).....	10
Figure 2.6. Summary statistics describing the total amount of mapped area by major and detailed biological cover categories within the entire mapped area and for the portion within the STEER.....	10
(adapted from Costa <i>et al.</i> 2013).	
Figure 2.7. Summary statistics describing the total amount of mapped area by percent live coral and dominant coral cover types within the entire mapped area and for the portion within the STEER (adapted from Costa <i>et al.</i> 2013).....	11
Figure 2.8. Spatial extent of geographic zones in the STEER in the 2013 NOAA map (Costa <i>et al.</i> 2013).....	11
Figure 2.9. Spatial extent of major and detailed geomorphological structure types in the STEER in the 2013 NOAA map (Costa <i>et al.</i> 2013).....	12
Figure 2.10. Spatial extent of percent hardbottom categories in the STEER in the 2013 NOAA map (Costa <i>et al.</i> 2013).....	12
Figure 2.11. Spatial extent of major and detailed biological cover types in the STEER in the 2013 NOAA map (Costa <i>et al.</i> 2013).....	13
Figure 2.12. Spatial extent of percent live coral and dominant coral cover types in the STEER in the 2013 NOAA map (Costa <i>et al.</i> 2013).....	13
Figure 3.1. Benthic habitat strata and site locations of the June 2012 survey of benthic habitat composition, fish communities, invertebrates, and marine debris.....	17
Figure 3.2. a) Diver collecting data on benthic habitat, b) diver collecting data on fish composition, and c) schematic representation of the placement of the 1m ² quadrat along a 25 m transect tape during fish and benthic community surveys.....	18
Figure 3.3. Mean (\pm SE) percent cover of abiotic substrate by bottom type.....	22
Figure 3.4. Mean (\pm SE) percent cover of major cover groups by bottom type.....	22
Figure 3.5. Mean rugosity (hardbottom sites only).....	23
Figure 3.6. Mangrove prop roots colonized with algae and benthic fauna.....	23
Figure 3.7. Percent live cover of hard corals.....	24
Figure 3.8. Hard coral species richness.....	24
Figure 3.9. a) Mean (\pm SE) percent cover of hard corals by species at hardbottom sites (N=44).....	25
Figure 3.10. Percent cover of coral species listed as threatened under the Endangered Species Act (ESA) or proposed for future listing.....	26
Figure 3.11. Percent gorgonian cover.....	27
Figure 3.12. Percent sponge cover.....	28
Figure 3.13. Percent macroalgae cover.....	29
Figure 3.14. Percent turf algae cover.....	29
Figure 3.15. Percent filamentous algae and cyanobacteria (FA & CB) cover.....	30
Figure 3.16. Percent seagrass cover.....	30
Figure 3.17. Percent cover of seagrass by species on unconsolidated sediments.....	31

List of Figures (continued)

Figure 3.18. Density of longspined sea urchin (<i>Diadema antillarum</i>)	31
Figure 3.19. Density of mature and immature queen conch (<i>Lobatus gigas</i>)	32
Figure 3.20. Density of total observed marine debris	32
Figure 3.21. Mean (\pm SE) fish species a) Shannon diversity, b) richness, c) density, and d) biomass by habitat type	34
Figure 3.22. Fish species richness	34
Figure 3.23. Fish species diversity	35
Figure 3.24. Total fish density	35
Figure 3.25. Total fish biomass	36
Figure 3.26. Proportional a) abundance and b) biomass of trophic groups across habitat types	36
Figure 3.27. Piscivore biomass	37
Figure 3.28. Non-metric multidimensional (nMDS) scaling ordination based on between site similarity composition using species a) density and b) biomass data	39
Figure 3.29. Grouper (Family Serranidae) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency	40
Figure 3.30. Red hind (<i>Epinephelus guttatus</i>) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency	42
Figure 3.31. Graysby (<i>Cephalopholis cruentata</i>) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency	43
Figure 3.32. Snapper (Family Lutjanidae) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency	44
Figure 3.33. Gray snapper (<i>Lutjanus griseus</i>) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency	45
Figure 3.34. Schoolmaster (<i>Lutjanus apodus</i>) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency	46
Figure 3.35. Yellowtail snapper (<i>Ocyurus chrysurus</i>) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency	47
Figure 3.36. Mahogany snapper (<i>Lutjanus mahogoni</i>) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency	48
Figure 3.37. Grunt (Family Haemulidae) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency	49
Figure 3.38. French grunt (<i>Haemulon flavolineatum</i>) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency	50
Figure 3.39. Bluestriped grunt (<i>Haemulon sciurus</i>) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency	52
Figure 3.40. Tomtate (<i>Haemulon aurolineatum</i>) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency	53
Figure 3.41. Surgeonfish (Family Acanthuridae) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency	54
Figure 3.42. Ocean surgeonfish (<i>Acanthurus bahianus</i>) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency	55
Figure 3.43. Blue tang (<i>Acanthurus coeruleus</i>) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency	56
Figure 3.44. Parrotfish (Family Scaridae) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency	57
Figure 3.45. Redband parrotfish (<i>Sparisoma aurofrenatum</i>) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency	58
Figure 3.46. Stoplight parrotfish (<i>Sparisoma viride</i>) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency	59
Figure 3.47. Striped parrotfish (<i>Scarus iseri</i>) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency	61

List of Figures (continued)

Figure 3.48. Princess parrotfish (<i>Scarus taeniopterus</i>) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency	62
Figure 3.49. Wrasse (Family Labridae) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency	63
Figure 3.50. Goatfish (Family Mullidae) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency	64
Figure 3.51. Damselfish (Family Pomacentridae) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency	65
Figure 3.52. Queen triggerfish (<i>Balistes vetula</i>) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency	66
Figure 3.53. Comparison of benthic cover groups between STEER and other locations within NOAA's Caribbean Coral Reef Ecosystem Monitoring Program	67
Figure 3.54. Comparison of community metrics between STEER and other locations within NOAA's Caribbean Coral Reef Ecosystem Monitoring Program	68
Figure 4.1. Strata and sediment sites sampled in the St. Thomas East End Reserves (STEER) in June 2011	79
Figure 4.2. Combined cluster analysis overlays of species clusters and site clusters.....	83
Figure 4.3. Hypothetical representation of the distribution of physicochemical habitat parameters, contaminant concentrations, and other site-specific data used to characterize site and species clusters.....	84
Figure 4.4. Percent fines (percent silt plus percent clay fractions) in sediments from the STEER	85
Figure 4.5 Total polycyclic aromatic hydrocarbons (PAHs) detected in sediments from the St. Thomas East End Reserves.....	87
Figure 4.6. Total extractable hydrocarbons (TEH) detected in sediments from the St. Thomas East End Reserves.....	88
Figure 4.7. Straight chain and branched hydrocarbons (aliphatics) in sediments from the St. Thomas East End Reserves.....	89
Figure 4.8 Total PCBs detected in sediments from the St. Thomas East End Reserves.....	90
Figure 4.9. Total DDT detected in sediments from the St. Thomas East End Reserves.....	91
Figure 4.10. Tributyltin detected in sediments from the St. Thomas East End Reserves.....	93
Figure 4.11. Tributyltin detected in sediments from southwest Puerto Rico and Vieques, Puerto Rico	94
Figure 4.12. Tributyltin detected in sediments from three sites in the STEER	94
Figure 4.13. Cadmium detected in sediments from the St. Thomas East End Reserves	95
Figure 4.14. Chromium detected in sediments from the St. Thomas East End Reserves.....	96
Figure 4.15. Copper detected in sediments from the St. Thomas East End Reserves	98
Figure 4.16. Lead detected in sediments from the St. Thomas East End Reserves	99
Figure 4.17. Zinc detected in sediments from the St. Thomas East End Reserves.....	100
Figure 4.18. <i>Clostridium perfringens</i> detected in sediments from the St. Thomas East End Reserves.....	101
Figure 4.19. Amphipod mortality vs % sand and gravel in St. Thomas STEER sediments	103
Figure 4.20. Amphipod mortality versus % total organic carbon in the STEER sediments.....	103
Figure 4.21. P450 response vs PAH concentrations (a), P450 response in terms of B[a]P eq for the subset of samples that exceeded the TCDD standard response (b).....	104
Figure 4.22. P450 response (normalized to standard 10nM TCDD) following 6 and 24 hour exposures to extracts from St. Thomas STEER sediment samples	105
Figure 4.23. Distribution of bioassays showing responses that were significantly different than controls or greater than a standard threshold (P450) in St. Thomas STEER sediments	106
Figure 4.24. Plot of total abundance of each taxa used in the analyses	106
Figure 4.25. Total animal abundance at each station in the STEER.....	107
Figure 4.26. Total number of species (taxa) and calculated diversity at each station in the STEER	107
Figure 4.27. Results of the SIMPER analysis for Strata 1-2	110
Figure 4.28. Results of the SIMPER analysis for Strata 3-5	110
Figure 4.29. Relationship between number of species and species diversity and the ERMq.....	111

List of Figures (continued)

Figure 4.30. Relationship between number of species and species diversity and percent fines	112
Figure 4.31. Relationship between mean ERMq and percent fines	112
Figure 5.1. Sampling sites for surface and sediment core samples in Benner Bay within the STEER	120
Figure 5.2. Photograph of core BB2 showing shell hash in the lower part of the core, grading to fine mud up the core in more recent sediments	122
Figure 5.3. Plots of ^{210}Pb and ^{137}Cs activities with depth for core 16P	123
Figure 5.4. Plots of ^{210}Pb and ^{137}Cs activities with depth for core BB1	124
Figure 5.5. Plots of ^{210}Pb and ^{137}Cs activities with depth for core B3	124
Figure 5.6. Plots of ^{210}Pb and ^{137}Cs activities with depth for core BB2	124
Figure 5.7. Percentage of fine grained sediment and gravel sized material (shell hash) down the length of the cores	126
Figure 5.8. Percentage of fine grained sediment and gravel sized material (shell hash) as a function of time.	126
Figure 5.9. Concentration (ng/g) of total butyltins as a function of time	127
Figure 5.10. Changes in major element concentrations over time at station B3	127
Figure 5.11. Percent tributyltin as a proportion of the total butyltins as a function of time and core depth.	127
Figure 5.12. Calcium detected in sediments collected in the STEER	128
Figure 5.13. Concentration of copper in surface sediments in northern Benner Bay	129
Figure 5.14. Copper concentrations as a function of time	129
Figure 6.1. Example of a POCIS disk	133
Figure 6.2. A disassembled POCIS deployment canister with the POCIS disks	134
Figure 6.3. Locations of passive water samplers (POCIS) in the STEER	134
Figure 6.4. POCIS deployment canister (arrow) among the sediment traps at a site in the STEER	135
Figure 6.5. Number of stormwater contaminants with estimated water column concentrations in the STEER and in Turpentine Gut	140
Figure 7.1. Coral, conch and fish sites collected in 2012 in the St. Thomas East End Reserves (STEER)	146
Figure 7.2. Total PAHs detected in the coral <i>Porites astreoides</i>	150
Figure 7.3. Total PAHs detected in the conch <i>Lobatus gigas</i>	151
Figure 7.4. Total PCBs detected in fish in the STEER	152
Figure 7.5. Total butyltins detected in the coral <i>Porites astreoides</i>	154
Figure 7.6. Total butyltins detected in the conch <i>Lobatus gigas</i>	154
Figure 7.7. Tributyltin detected in the coral <i>Porites astreoides</i>	155
Figure 7.8. Tributyltin detected in the conch <i>Lobatus gigas</i>	155
Figure 7.9. Distribution of tributyltin (TBT), dibutyltin (DBT), and monobutyltin (MBT) from fish sampled in the STEER	156
Figure 7.10. Concentrations (mean \pm SE) of metals detected in the coral <i>Porites astreoides</i>	157
Figure 7.11. Concentrations (mean \pm SE) of metals detected in the conch <i>Lobatus gigas</i>	159
Figure 7.12. Concentrations (mean \pm SE) of metals detected in the coral <i>Porites astreoides</i>	161
Figure 7.13. Concentrations (mean \pm SE) of metals detected in the conch <i>Lobatus gigas</i>	163
Figure 7.14. Scatter plot showing relationship between manganese and lead in coral	164
Figure 8.1. Map of the St. Thomas East End Reserves showing site locations (in red circles) where the samples for histopathological examination were collected in June 2012	173
Figure 8.2. Examples of fixed coral samples as photographed in the GMU Histology Laboratory	174
Figure 8.3. Examples of surface body wall and gastrodermis ruptures and lysing	180
Figure 8.4. Melanin-like granular amoebocytes (brown cells) infiltrating between cells of the epidermis and gastrodermis of tentacles (arrow), rated severe (= 5)	180
Figure 8.5. Examples of apoptosis in the epidermis (at arrows)	181
Figure 8.6. Examples of calicodermis condition	182

List of Figures (continued)

Figure 8.7. Examples of bacterial aggregates in mesoglea of tentacles (a and b), 25x; calicodermis reaction to suspect fungal hyphae (c), 40x; endolithic algae or fungi where skeleton was removed along basal gastrovascular canals (d), 10x.....	183
Figure 8.8. Examples of apicomplexan parasites of <i>P. astreoides</i>	184
Figure 8.9. Examples of apparent destruction of the basal body wall of most aboral gastrovascular canals in the tissue layer of the perforate coral, <i>P. astreoides</i>	185
Figure 8.10. Examples of other unknowns present in the STEER samples.....	185
Figure 8.11. Examples of reproductive structures present in the STEER samples.....	186
Figure 8.12. Example of black particles trapped in mucus secretions (arrows), 40x	188
Figure 8.13. Comparison of means of Specific Condition Scores Sum with standard deviations among the five sites.....	189
Figure 9.1. Sampling sites for nutrients, sedimentation, and TSS along with the STEER boundary.....	194
Figure 9.2. Mean and max concentrations of ammonium (NH_4^+) detected in water samples from the STEER	199
Figure 9.3. Mean and max concentrations of nitrate (NO_3^-) and nitrite (NO_2^-) detected in water samples from the STEER.....	200
Figure 9.4. Mean and max concentrations of urea detected in water samples from the STEER.....	201
Figure 9.5. Mean and max concentrations of dissolved inorganic nitrogen (DIN) detected in water samples from the STEER.....	202
Figure 9.6. Mean and max concentrations of total nitrogen (TN) detected in water samples from the STEER	203
Figure 9.7. Mean and max concentrations of orthophosphate (HPO_4^-) detected in water samples from the STEER.....	204
Figure 9.8 Mean and max concentrations of total phosphorus (TP) detected in water samples from the STEER.....	205
Figure 9.9. Nitrate and nitrite concentrations versus rainfall in Benner Bay during the study period	206
Figure 9.10. Percent exceedances of DIN threshold by site in the STEER	206
Figure 9.11. Herbivore biomass in the STEER.....	207
Figure 9.12. Bulk deposition of sediment in traps placed in the STEER	207
Figure 9.13. Excavation activity on Little St. James Island, a possible source of terrigenous sediments found in the sediment traps at this site.....	208
Figure 9.14. Average accumulation rates at the sediment trap sites in the STEER	208
Figure 9.15. Bulk deposition in Benner Bay sediment traps plotted against rainfall	209
Figure 9.16. Percent and rates of deposition for terrigenous, carbonate and organic materials in the sediment traps in the STEER over time for Mangrove Lagoon (a, d), Benner Bay (b, e), and Little St. James (c, f)	210
Figure 9.17. Plots of mean sediment deposition (a), rainfall (b) and wave height at Station 41052 (c)	211
Figure 9.18. Total suspended solids (TSS) means (\pm SE) at sampling sites in the STEER.....	212

CHAPTER 1: INTRODUCTION

This report contains the results of a chemical and biological characterization of the St. Thomas East End Reserves, or STEER in St. Thomas, USVI. This integrated ecological assessment represents the findings from a three-year project carried out by NOAA's National Centers for Coastal Ocean Science (NCCOS) and local partners, to assess chemical stressors and impacts, and to characterize the marine biological communities throughout the STEER. This report combines the results from five interim reports, along with data from two other assessments that were part of the project, into a final integrated assessment. The reader will be referred to these other publications on occasion, for additional information.

The St. Thomas East End Reserves, or STEER, is a collection of Marine Reserves and Wildlife Sanctuaries (MRWSs) located on the southeastern end of the island of St. Thomas, U.S. Virgin Islands (Figure 1.1). Within the STEER, there are extensive mangroves and seagrass beds, along with coral reefs, lagoons and cays.

The value of the natural resources in the Reserves has long been recognized. In 1979, the area was identified by NOAA's National Marine Sanctuaries Program as a "marine area of national significance, deserving of marine sanctuary designation" (NOAA, 1981). The same year, the Mangrove Lagoon/Benner Bay area was designated by the USVI government as an Area of Particular Concern, or APC, due to the abundance of important but threatened natural resources, and the desire to preserve and, as needed, restore these areas.

The STEER is made up of four MRWSs, including Inner Mangrove Lagoon, Cas Cay/Mangrove Lagoon, St James, and Compass Point Salt Pond Marine Reserves and Wildlife Sanctuaries (Figure 1.1). The STEER comprises an area of approximately 9.6 km², with 34 km of coastline (STEER, 2011). Boundaries for the STEER include the Mangrove Lagoon/Benner Bay MRWS with Long Point

as the western border (Figure 1.1). The St. James MRWS forms the eastern boundary of the STEER, which includes the waters surrounding Great St. James and the north shore of Little St. James Island. To the north, the boundary of the STEER runs along the coastline, from Cabrita Point westward to Benner Bay. At Benner Bay, the boundary follows a line offshore from Coculus Rock along Roto Cay to the northeastern entrance of Mangrove Lagoon; the marina areas within Benner Bay are outside of the STEER.

With mangroves, seagrass beds, lagoons, salt ponds, coral reefs and a number of cays, the STEER contains a multitude of natural habitats and resources. The STEER is

thought to be one of the most valuable nursery areas remaining in St. Thomas, with many species of fish and shellfish spending some portion of their lives in the protected areas around the mangroves and in the extensive seagrass beds (STEER, 2011). Fishing is not allowed in most parts of the STEER, and where it is allowed (e.g., for baitfish), a permit is required.



View into the St. Thomas East End Reserves (STEER).

The abundance of natural resources has contributed to the STEER being a popular destination for recreational activities ranging from swimming, camping, snorkeling and SCUBA, to boating and ecotourism.

The largest remaining mangrove system in St. Thomas occurs along the shores of Mangrove Lagoon/Benner Bay (IRF 1993; STEER, 2011). Mangrove species include the red mangrove (*Rhizophora mangle*), black mangrove (*Avicennia germinans*), and white mangrove (*Laguncularia racemosa*). Mangroves offer many ecological benefits. They serve as natural buffers against shore erosion, floods and hurricane waves. Mangroves also provide protection for juvenile fish and other organisms around the roots, and generate detrital material that enters the food chain, becoming a food resource for a number of marine organisms (DPNR-DFW, 2005). Sampling with fish traps along

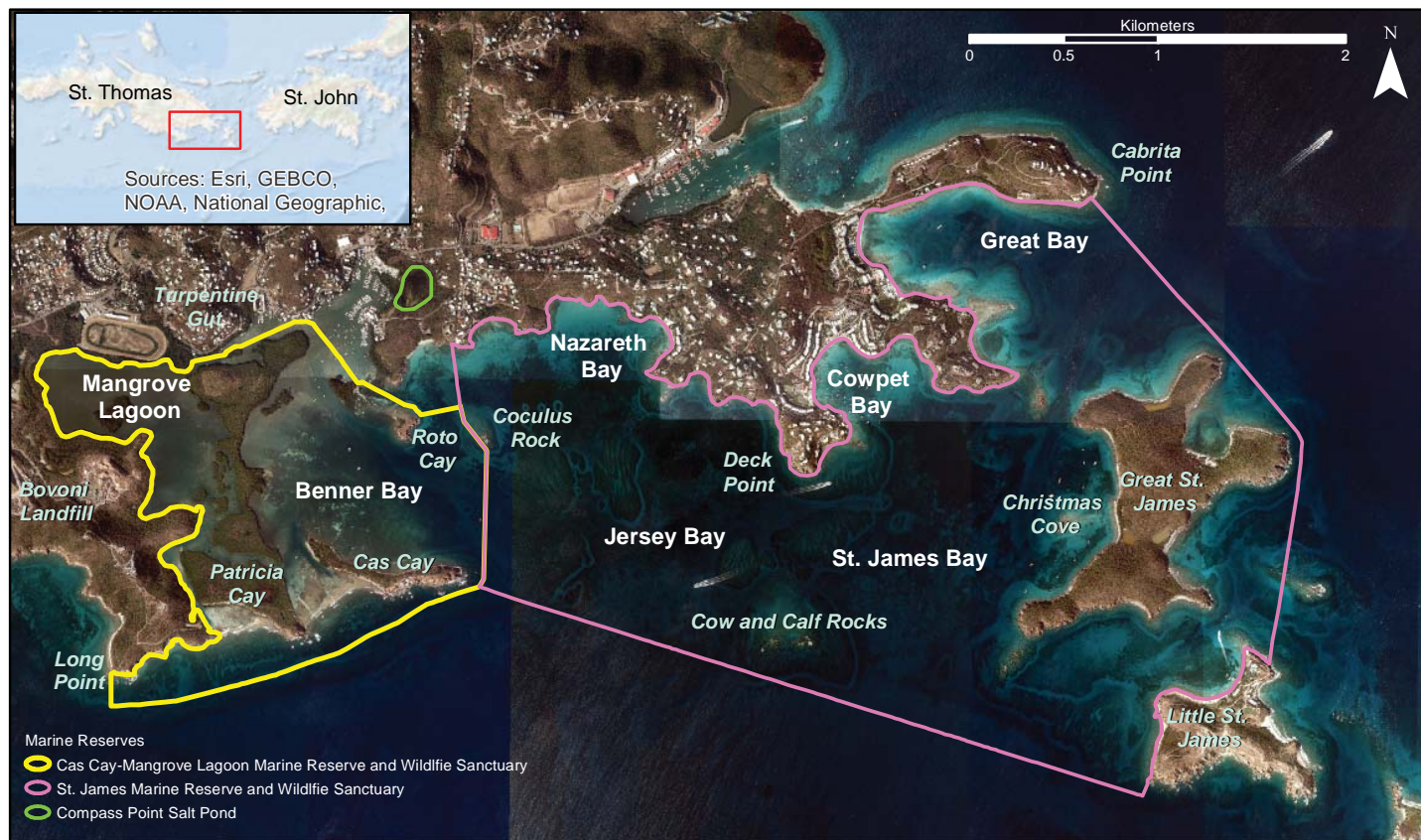


Figure 1.1. Individual Marine Reserves that comprise the STEER.

the mangrove fringe in Benner Bay/Mangrove Lagoon has yielded as many as 40 fish species (Boulon, 1992; Colletti, 2011).

There are extensive areas of seagrass in Benner and Jersey Bays. Although seagrasses were also abundant throughout Mangrove Lagoon in the 1970s, macroalgae presently dominates the benthos (Colletti, 2011). Seagrasses in Mangrove Lagoon appear to be limited to the middle portion of the lagoon, near the mangroves on the eastern edge. In the rest of the STEER, turtle grass (*Thalassia testudinum*) and manatee grass (*Syringodium filiforme*) are two of the major seagrass species found in the STEER (DPNR-DFW, 2005). Seagrasses provide habitat for many organisms, including juvenile fish. Seagrasses are also an important food source for parrotfish, surgeonfish, and pinfish, along with turtles, birds, and sea urchins. Conch feed off the epiphytes on the seagrass leaves (DPNR-DFW, 2005). Finally, seagrasses in the Caribbean as in other areas, help to improve water clarity and light penetration, by trapping fine sediments and allowing the sediment particles to settle out of the water column.

There are significant coral reef areas in Jersey Bay and also south of Cas and Patricia Cays. Many species of coral can be found in the STEER, including the star coral complex (*Orbicella annularis*, *O. faveolata* and *O. franksi*), mustard

hill coral (*Porites astreoides*), lesser and massive starlet corals (*Siderastrea radians* and *S. siderea*), finger coral (*P. porites*), symmetrical brain coral (*Diploria strigosa*), and great star coral *Montastraea cavernosa* (DPNR-DFW, 2005). Also within the STEER are the threatened elkhorn (*Acropora palmata*) and staghorn (*A. cervicornis*) corals. The STEER is a relatively shallow system, supporting both patch and fringing reefs. The coral reefs provide habitat and food for many organisms.

Not surprisingly, the habitats in the STEER are closely linked to one another. For example, a variety of fish and invertebrates move between the mangroves, seagrass beds and coral reefs, either during the course of their lives (e.g., juvenile fish living among the mangrove prop roots for protection, with adults moving out onto the reefs), or as part of a diurnal cycle (e.g., invertebrates and fish feeding in the seagrass beds at night and returning to the protection of the coral reefs during the day) (STEER, 2011).

Degraded environmental quality in any one of these habitats affects the others. Poor water and sediment quality in mangrove areas can result in lower dissolved oxygen concentrations from an overabundance of nutrients and organic matter, reducing habitat quality for fish species and their prey. Elevated levels of suspended sediments in the water column can lead to a reduction in the amount of light

reaching seagrasses and corals, which can result in reduced growth or even die-off. Sediments that settle out of the water column can also act to smother corals, or at the very least, result in the corals having to expend more energy to remove the sediment. Elevated nutrient levels can result in increased algal growth in the water column and, as with sediments, reduce the amount of light reaching corals or seagrasses. Excess nutrients also promote the growth of epiphytic algae that may smother coral and seagrass. Chemical contaminants can impact a variety of organisms and life stages.

Two watersheds, Jersey Bay and Red Hook drain to the STEER (Figure 1.2). The largest is the Jersey Bay watershed which empties into Mangrove Lagoon/Benner Bay and Jersey Bay. The Frenchman Bay watershed drains to the west of the STEER. The area of the STEER east of Deck Point (Figure 1.1) receives input from the Red Hook watershed.

Approximately one-third of the population of St. Thomas resides within the Mangrove Lagoon/Benner Bay APC (IRF, 1993). The area is considered highly impacted and urbanized, with numerous point and non-point sources of

pollution (DPNR 2003; Horsley Witten, 2013a,b). These sources include the unlined Bovoni Landfill (Figure 1.1), adjacent to Mangrove Lagoon, which serves all of St. Thomas and St. John, an EPA Superfund site (EPA, 2011), numerous marinas and boatyards (Figure 1.3), a number of resorts, various commercial/industrial activities, and a horse racetrack. In addition, it has been estimated (Horsley Witten, 2013b) that approximately 70% of the residential housing adjacent to Mangrove Lagoon and Benner Bay is on septic systems, many of which are failing (IRF, 1993). There is also a deteriorating sewer infrastructure for those connected to the public sewer system and at one time, a wastewater treatment plant emptied directly into Mangrove Lagoon (Grigg *et al.*, 1971). Elevated levels of chemical contaminants have been documented in the STEER watershed (EPA, 2011).

Turpentine Gut, which was channelized during construction of the nearby Clinton Phipps racetrack, drains approximately 60% of the watershed, and discharges of untreated stormwater and sewage overflows go directly into Mangrove Lagoon. Elevated sedimentation, nutrient and bacterial levels have been detected in the lagoon, particularly following storm events (STEER, 2011). The EPA Superfund site (Tutu Wellfield Superfund Site) was established in 1996 due to contamination of groundwater and wells in the area by chlorinated volatile organic compounds (CVOC) (EPA, 2011). Finally, sediments are delivered to the STEER through conveyances such as Turpentine Gut, or directly to the STEER as a result of runoff from the steep island slopes surrounding the northern part of Mangrove

Lagoon and Benner Bay. All of these have the potential to contribute both point and nonpoint source pollution to the STEER.

Due to the threats along with the ecological value of the wetlands and adjacent marine ecosystems, the Jersey Bay and Red Hook watersheds have been designated as “priority watersheds” in St. Thomas (Platenberg, 2006). In addition, a management plan (STEER, 2011) was developed through

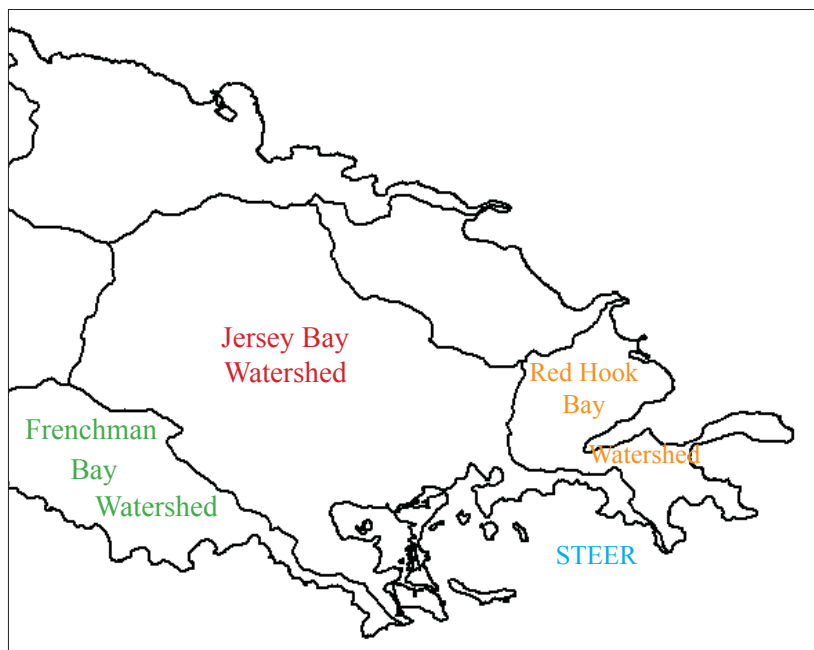


Figure 1.2. Primary watersheds adjacent to the STEER.

the collaboration of the Department of Planning and Natural Resources (DPNR), the University of the Virgin Islands (UVI), The Nature Conservancy (TNC), and community groups (e.g., Friends of Christmas Cove). The goal of the STEER Management Plan is to “restore and maintain a functional coastal ecosystem that promotes sustainable recreational opportunities and compatible commercial uses with community engagement through effective management” (STEER, 2011).

At a NOAA Coral Reef Ecosystem Integrated Observing System (CREIOS) Meeting in San Juan, Puerto Rico in 2009, NOAA scientists met with resource managers and local scientists from the U.S. Caribbean, including representatives from the USVI, to elicit priority information needs, and to highlight important issues of concern. At that

meeting, representatives from DPNR's Divisions of Coastal Zone Management and Environmental Protection identified the STEER as a priority area, and noted that there were significant data and information gaps in terms of the chemical contaminants present, effects, and the overall health of the biological communities within the STEER.

Representatives from DPNR stated that the extent of chemical contamination and biological effects in the STEER were unknown, but that data would be critical in helping to make informed decisions on coastal resource management. With these needs in mind, NOAA's NCCOS, in partnership with the STEER Core Planning Group made up of scientists and managers from DPNR's Divisions of Coastal Zone Management, Fish and Wildlife, and Environmental Protection, along with the University of the Virgin Islands, and The Nature Conservancy, proposed a multiyear project to NOAA's Coral Reef Conservation Program (CRCP), to develop an integrated assessment of the chemical and biological conditions within the STEER.

Several other CRCP-funded projects in the STEER have been completed. In 2012, a survey of human uses was documented in a coastal use mapping project (Dillard and D'lorio, 2012). In another CRCP-funded project, the goal of which was to develop a watershed management plan for the STEER, information from the current project was incorporated into the management plan. Reducing impacts from the landfill, along with improving wastewater and storm water management, pollution prevention, and wetland restoration, were seen as key items for STEER watershed restoration (Kitchell, 2012).

NOAA's CRCP also funded an effort to develop a new high resolution map describing the distribution, quantity and type of seafloor habitats inside the STEER. This map was developed from LiDAR (Light Detection and Ranging) bathymetry and reflectivity imagery collected by Fugro LADS Corporation. The new habitat map and related products are described in Chapter 2, and will be used to: update nautical charts in the area to; help fill critical information gaps about the seafloor in a priority area identified by NOAA's CRCP; support fisheries-related performance measures outlined in the USVI Jurisdictional Working Group Priority Settings document; and to support best management practices inside

the STEER, related to permitting activities, restoration, fisheries, climate change and scientific research.

The USVI Territorial Coral Reef Monitoring Program (TCRMP) has been surveying reefs around St. Thomas since 2001 but only has one permanent site within the STEER, at Cocus Rock in Benner Bay, and an additional site located just outside STEER near Little St. James (Smith *et al.*, 2011). Benthic metrics have been surveyed annually since 2001, and an annual fish census was added at the Cocus Rock monitoring site in 2009. The TCRMP data is collected using frame grabs from digital video to estimate benthic cover to the species level. While the data produced by both of these studies are robust, a STEER-wide assessment characterizing the fish and benthic communities across all habitat types has been lacking.

Other recent research in the STEER has focused on limited geographic areas. A recent one-year study of fish communities was conducted in Benner Bay/ Mangrove Lagoon. Fish traps were used to compare fish diversity and abundance among three geographic strata within the lagoon (Murray 2009; Colletti, 2011). This study also produced a benthic habitat map of the lagoon using broad categories (coral reef, mangrove,

cyanobacteria, seagrass, macroalgae, and coral rubble) (Colletti, 2011).

EPA's Environmental Monitoring and Assessment Program (EMAP) collected sediment samples in 2004, at a number of locations around St. Thomas. Four of the sites were in the STEER; two sites were in Benner Bay, and one site each in Jersey Bay and Great Bay. Data from the EMAP work are highlighted in Chapter 4.

The overall objectives of the project in the STEER involving NCCOS and partners were to: 1) quantify chemical contamination in sediments, coral and conch, fish and water; 2) assess the effects that these stressors may be having on organisms living within the sediments; 3) characterize the fish communities and associated benthic habitats in the STEER; and 4) establish baseline values that can be compared with conditions following changes in land use practices or implementation of management actions.

In 2011, NCCOS scientists along with project partners collected sediments for chemical contaminant analysis,



Figure 1.3. Marina area in Benner Bay.

toxicity bioassays, and benthic infaunal analysis. In early 2012, the University of the Virgin Islands began monthly sampling for nutrients, and monthly monitoring of total suspended solids (TSS, a CRCP performance measure), and sedimentation using sediment traps placed at six locations within the STEER as part of the project. Passive water samplers were also deployed in the same locations as the sediment traps. The passive water samplers were used to quantify the presence of wastewater contaminants in the water column. Activities in the second year also included a biological survey of the entire STEER, along with the collection of coral, conch and fish for chemical contaminant analysis. In the third year (2013), a followup investigation of contaminants in surface sediments and also in sediment cores was completed in northern Benner Bay. The quantification of sediment contaminants, toxicity, and the benthic infaunal community, along with the biological survey conducted as part of this project, have provided data needed to optimize management of the STEER.

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CHAPTER 2: BENTHIC HABITAT MAPPING WITHIN THE ST. THOMAS EAST END RESERVES

Bryan Costa^{1,2}, Laurie Bauer^{1,2}, and Tim Battista¹

¹NOAA/NOS/National Centers for Coastal Ocean Science, Center for Coastal Monitoring and Assessment (CCMA), 1305 East/West Highway, Silver Spring, MD 20910

²CSS-Dynamac Consolidated Safety Services, Fairfax, VA 22030, under CSS-Dynamac Contract #EA-133C-14-NC-1384

2.1 INTRODUCTION

Scientists and managers need a baseline understanding of the benthic communities and associated living marine resources to effectively manage the STEER (STEER, 2011). Benthic habitat maps are an integral component to this process, as they support effective ecosystem-based approaches to management. Habitat maps inform local managers about the existing distribution of resources and how they have changed over time. They also help managers locate and protect sensitive marine communities, and help guide monitoring efforts and prioritize subsequent management actions. The objectives of this chapter are to 1) provide a synopsis of benthic habitat mapping efforts conducted within STEER, and 2) provide summary statistics and a description of benthic habitats within the STEER.

2.2 HISTORY OF BENTHIC HABITAT MAPPING IN STEER

A benthic habitat map for the USVI and Puerto Rico was completed in 2001 by NOAA's National Centers

for Coastal Ocean Science (NCCOS), Center for Coastal Monitoring and Assessment CCMA) Biogeography Branch (Figure 2.1, Kendall *et al.*, 2001). This map is based on 1999 aerial photography, and has a minimum mapping unit of 1 acre (4,048 m²). Major mapped categories include unconsolidated sediments, submerged vegetation, and coral reef/hardbottom.

Detailed habitat types for reef/hardbottom include colonized bedrock, colonized pavement, colonized pavement with sand channels, linear reef, patch reef (aggregated), patch reef (individual), and scattered coral/rock in unconsolidated sediment (Kendall *et al.*, 2001). Detailed habitat types within unconsolidated sediments include sand and mud. Submerged aquatic vegetation was divided into macroalgae and seagrass and mapped at various levels of patchiness. Other delineations include mangrove, land, and artificial structures.

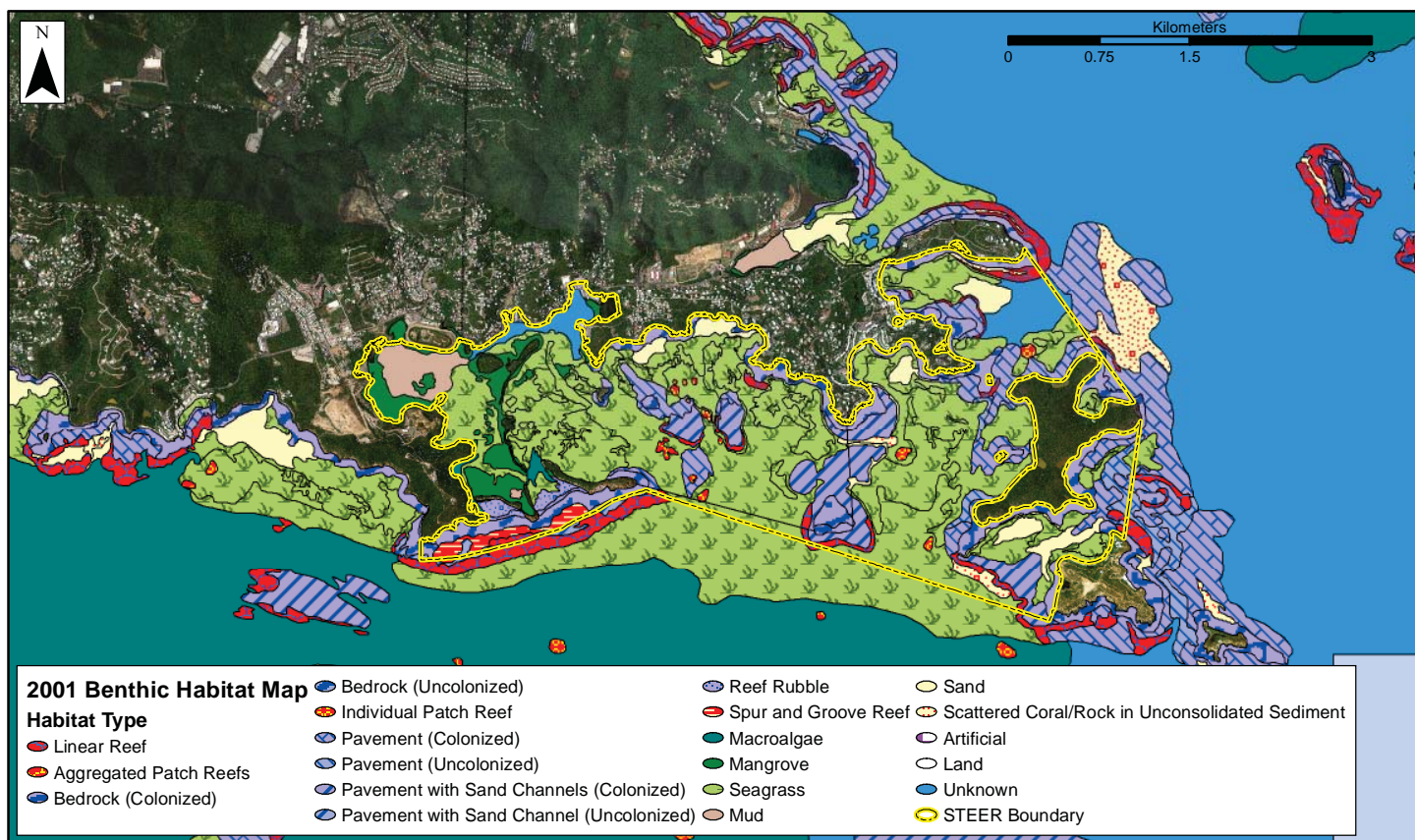


Figure 2.1. Spatial extent of benthic habitats in the STEER in the 2001 NOAA map (Kendall, 2001).

In 2011, the National Oceanic and Atmospheric Administration (NOAA) collected LiDAR (Light Detection and Ranging) imagery of the nearshore, marine areas (0-40 meters deep) around St. John and north of St. Thomas in the U.S. Virgin Islands (USVI). LiDAR sensors use lasers to collect depth and topographic information about the seafloor. This seafloor imagery was collected with a number of applications in mind (USVI and NOAA CRCP, 2010), including hydrodynamic modeling, chemical contaminant and sediment dispersal modeling, water quality monitoring, nautical chart updates and benthic habitat mapping. NOAA's Biogeography Branch used this imagery to develop benthic habitat maps for three locations identified as high priorities by local resource managers (USVI and NOAA CRCP, 2010), which included the marine areas in and around the STEER as well as Coral Bay and Fish Bay in St. John (Costa *et al.*, 2013).

The new NOAA map (Costa *et al.*, 2013) was different than the previous version (Kendall, 2001) in several ways, including the use of: 1) a revised, expanded classification scheme, 2) a finer scale of delineation, and 3) a smaller minimum mapping unit (100 m²). In addition, whereas only aerial photos were used to create the 2001 maps, the new mapping effort utilized both aerial photos and LiDAR

imagery, which when integrated provides a wealth of additional information about the seafloor.

2.3 HABITAT MAPPING

Methods

This section provides a general overview of the most recent benthic habitat mapping effort within STEER (Costa *et al.*, 2013). The habitat classification scheme used to map shallow-water benthic habitats (<30 m) is summarized in Table 2.1. This scheme was adapted from a scheme previously developed by NOAA to map benthic habitats around St. John (Zitello *et al.*, 2009; Costa *et al.*, 2009), Buck Island Reef National Monument in St. Croix (Costa *et al.*, 2012) and Southwest Puerto Rico (Bauer *et al.*, 2012). The habitat classification scheme defines benthic habitats based on six attributes: 1) broad geographic zone; 2) geomorphological structure type; 3) percent hardbottom; 4) dominant biological cover; 5) amount of live coral cover (includes both hard and soft corals); and 6) dominant coral type (hard or soft coral). Every feature in the benthic habitat map was assigned a class from each level of the scheme (Table 2.1).

The following steps were used to map shallow-water habitats (Costa *et al.*, 2013).

Table 2.1. The classification scheme used to classify benthic habitats in the STEER in 2013 (Costa *et al.*, 2013).

GEOGRAPHIC ZONE		GEOMORPHOLOGICAL STRUCTURE		BIOLOGICAL COVER	
Back Reef	Coral Reef and Hardbottom (Hard)	}		Major Cover	
Bank/Shelf	Aggregate Reef			Algae	
Bank/Shelf	Aggregated Patch Reefs			Live Coral	
Escarpment				Mangrove	
Channel	Individual Patch Reef			No Cover	
Dredged	Pavement			Seagrass	
Fore Reef	Pavement with Sand Channels			Unclassified	
Lagoon	Reef Rubble		Percent Hard	Unknown	
Land	Rhodoliths		0% ≤ 10%	Percent Major Cover	
Reef Crest	Rhodoliths with Scattered Coral & Rock		10% ≤ 30%	10% ≤ 50%	
Reef Flat	Rock/Boulder		30% ≤ 50%	50% ≤ 90%	
Salt Pond	Spur & Groove		50% ≤ 70%	90% ≤ 100%	
Shoreline Intertidal	Unknown		70% ≤ 90%	N/A	
	Unconsolidated Sediment (Soft)		90% - 100%	Unknown	
	Mud		N/A	Percent Coral Cover	
	Sand		Unknown	0% ≤ 10%	
	Sand with Scattered Coral & Rock			10% ≤ 50%	
	Unknown			50% ≤ 90%	
	Other Delineations			90% - 100%	
	Artificial			N/A	
	Land			Unknown	
	Unknown				

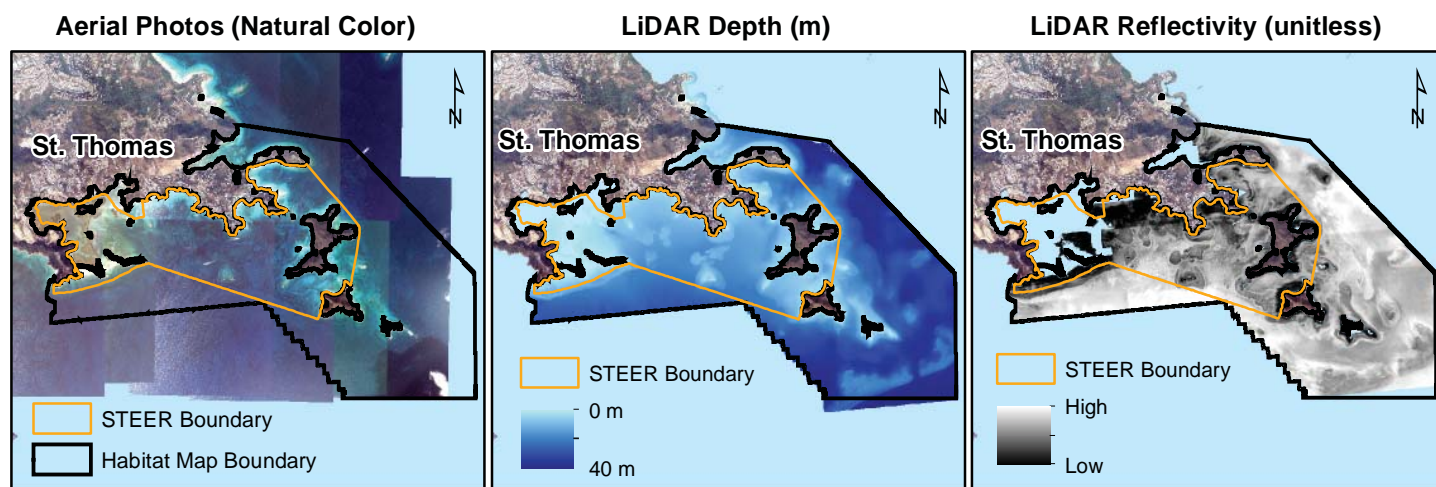


Figure 2.2. Aerial photos (left), LiDAR bathymetry (middle), and LiDAR reflectivity (right) used to delineate and characterize the benthic habitats inside STEER (Costa *et al.*, 2013).

1. Imagery Acquisition

Aerial photographs and LiDAR imagery were collected in 2012 and 2011 (respectively) covering the full geographic extent of project area (Figure 2.2). These images were used to generate the benthic habitat map for STEER.

2. Habitat Boundary Delineation

A semi-automated approach (Costa *et al.*, 2009; Costa *et al.*, 2012; Costa and Battista, 2013) was used to delineate habitat features visible in the aerial photographs and LiDAR imagery. In areas without LiDAR, habitat boundaries that were visible in the normalized aerial photos and whose area was $\geq 100 \text{ m}^2$ were manually delineated (at a scale of 1:1,000). In areas with LiDAR, features were identified and extracted habitat features on the seafloor using the ENVI Feature Extraction (Fx) toolbox. This remote sensing software uses edge detection algorithms to detect and delineate seafloor features visible in a single image or in a suite of spatially coincident images. This software defines a feature as a region of interest with unique spatial, spectral (brightness and color), and/or textural characteristics that make it visually distinct from its surroundings.

3. Ground Validation (GV) and Habitat Classification

Ground validation is the process of collecting underwater photos and/or videos at discrete locations. GV sites are selected to explore habitats which were unknown, or to verify that habitat types look the same (in the source imagery) across the entire mapped area. Underwater video cameras were used to explore select habitat features delineated in step 2, and this video was used to classify each polygon and inform the draft habitat map creation. Areas without LiDAR were attributed manually. For the areas with LiDAR, classified habitat maps were developed using features

extracted and characterized by ENVI Fx and Random Forests in R, respectively. The two maps (i.e., one generated manually and the other using Fx and Random Forests) were then merged together to produce a final, seamless map.

4. Expert Review

Local marine biologists, scientists, resource managers and community groups reviewed these draft maps online to qualitatively assess the thematic accuracy of each map based on their local ecological knowledge.

5. Accuracy Assessment (AA)

Underwater videos were collected (using a random stratified sampling plan) to independently and quantitatively evaluate the accuracy of the habitat maps. Thematic accuracy was characterized for major and detailed geomorphological structure, major and detailed biological cover, percent hard bottom, percent coral cover and dominant coral type classifications using error matrices. Error matrices quantify the number of locations correctly classified for each habitat class. These numbers were also corrected for proportional bias by accounting for the geographic area occupied by different habitat classes.

6. Final Product Creation

Errors identified during the expert review and accuracy assessment were corrected to produce final habitat maps for the STEER. Digital map products (i.e., benthic habitat map, imagery, GV and AA data) are available on the web (<http://coastalscience.noaa.gov/projects/detail?key=171>) and through an interactive, web-based GIS application (<http://maps.coastalscience.noaa.gov/biomapper/biomapper.html?id=STEER>). More details on the classification scheme and each of these steps can be found in Costa *et al.*, (2013).

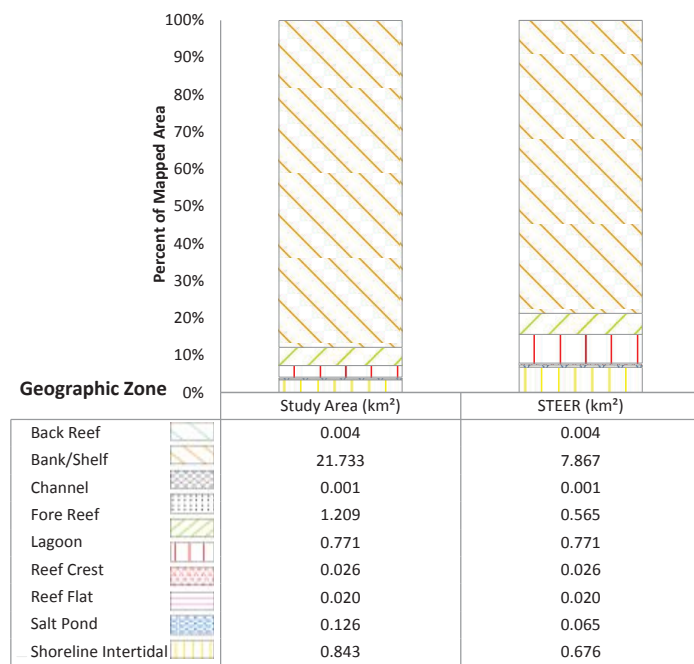


Figure 2.3. Summary statistics describing the total amount of mapped area by geographic zone within the entire mapped area and for the portion within the STEER (adapted from Costa *et al.*, 2013).

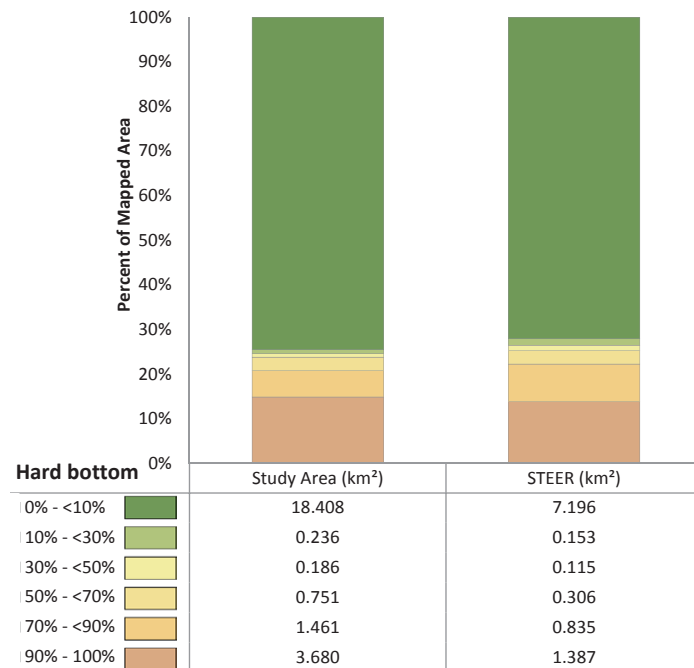


Figure 2.4. Summary statistics describing the total amount of mapped area by percent hardbottom categories within the entire mapped area and for the portion within the STEER (adapted from Costa *et al.*, 2013).

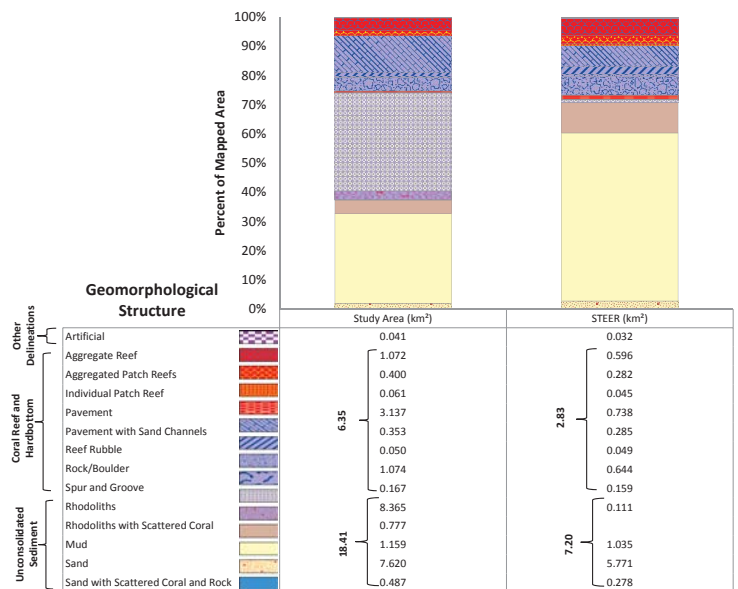


Figure 2.5. Summary statistics describing the total amount of mapped area by major and detailed geomorphological structure within the entire mapped area and for the portion within the STEER (adapted from Costa *et al.*, 2013).

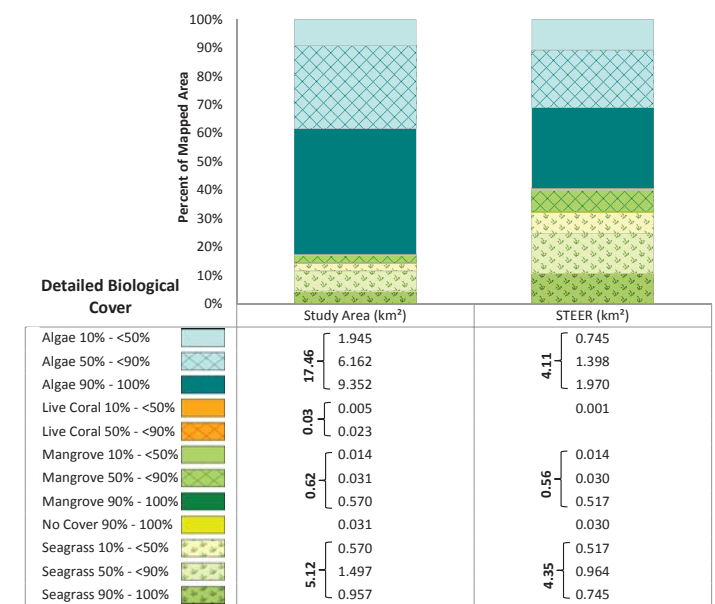


Figure 2.6. Summary statistics describing the total amount of mapped area by major and detailed biological cover categories within the entire mapped area and for the portion within the STEER (adapted from Costa *et al.*, 2013).

2.4. RESULTS AND DISCUSSION

The overall map accuracies (corrected for proportional bias) for this habitat map were 93.0% for major structure, 75.1% for detailed structure, 86.2% for percent hardbottom, 86.5% for major cover and 74.5% for detailed cover (Costa *et al.*, 2013). The live coral and dominant coral type classes had 83.3% and 88.2% accuracies, respectively. These numbers are similar to the other benthic habitat maps created by NCCOS's Biogeography Branch (Zitello *et al.*, 2009; Bauer

et al., 2012; Costa *et al.*, 2012). As a result, these digital map products can be used with confidence by scientists and resource managers for a multitude of different applications. The STEER benthic habitat map extends to the south and west of the STEER boundary. Summary statistics are presented here for both the entire mapped area and for mapped areas only within the STEER (Figures 2.3-2.7).

The majority of mapped area (87.9%) was on the Bank/Shelf (Figures 2.3 and 2.8), which extends offshore from the shoreline or seaward edge of a coral reef to the edge of the continental shelf. This pattern was also true within the STEER, but Bank/Shelf accounted for a slightly smaller percentage (78.7%). Lagoon and Shoreline Intertidal accounted for slightly higher percentages within the STEER compared to the overall study area. Geomorphological structure types (Figures 2.5 and 2.9) and the amount of hardbottom (Figures 2.4 and 2.10) were patchy across the total mapped area. Unconsolidated Sediment dominated the seafloor overall (74.3%) and within STEER's boundary (71.8%). Rhodoliths (i.e., calcareous nodules approximately 6 cm in diameter), and Sand were the two most common types of unconsolidated sediment, comprising 33.8% and 30.8% of the total mapped area, respectively. However, while Rhodoliths dominated the area outside of

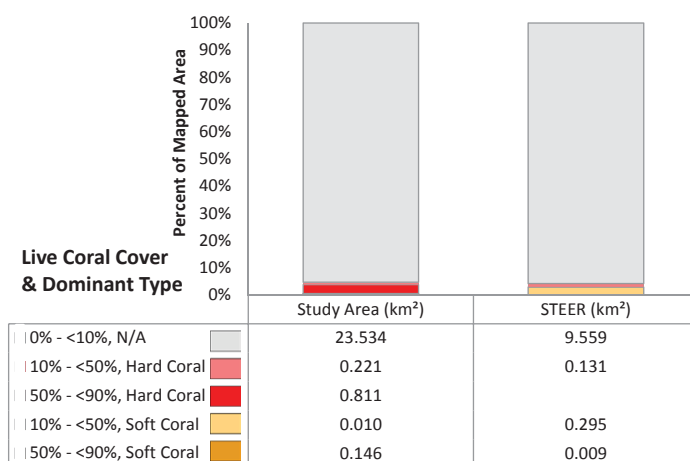


Figure 2.7. Summary statistics describing the total amount of mapped area by percent live coral and dominant coral cover types within the entire mapped area and for the portion within the STEER (adapted from Costa *et al.*, 2013).

the STEER's boundary, Sand made up the majority of the area inside the MPA. Mud habitats made up a small amount (4.7%) of the total mapped area, and were located mainly in Mangrove Lagoon. Coral Reef and Hardbottom habitats constituted 25.5% of the total mapped area with Pavement (i.e., flat, low-relief or sloping solid carbonate rock with little or no fine-scale rugosity), being the most dominant

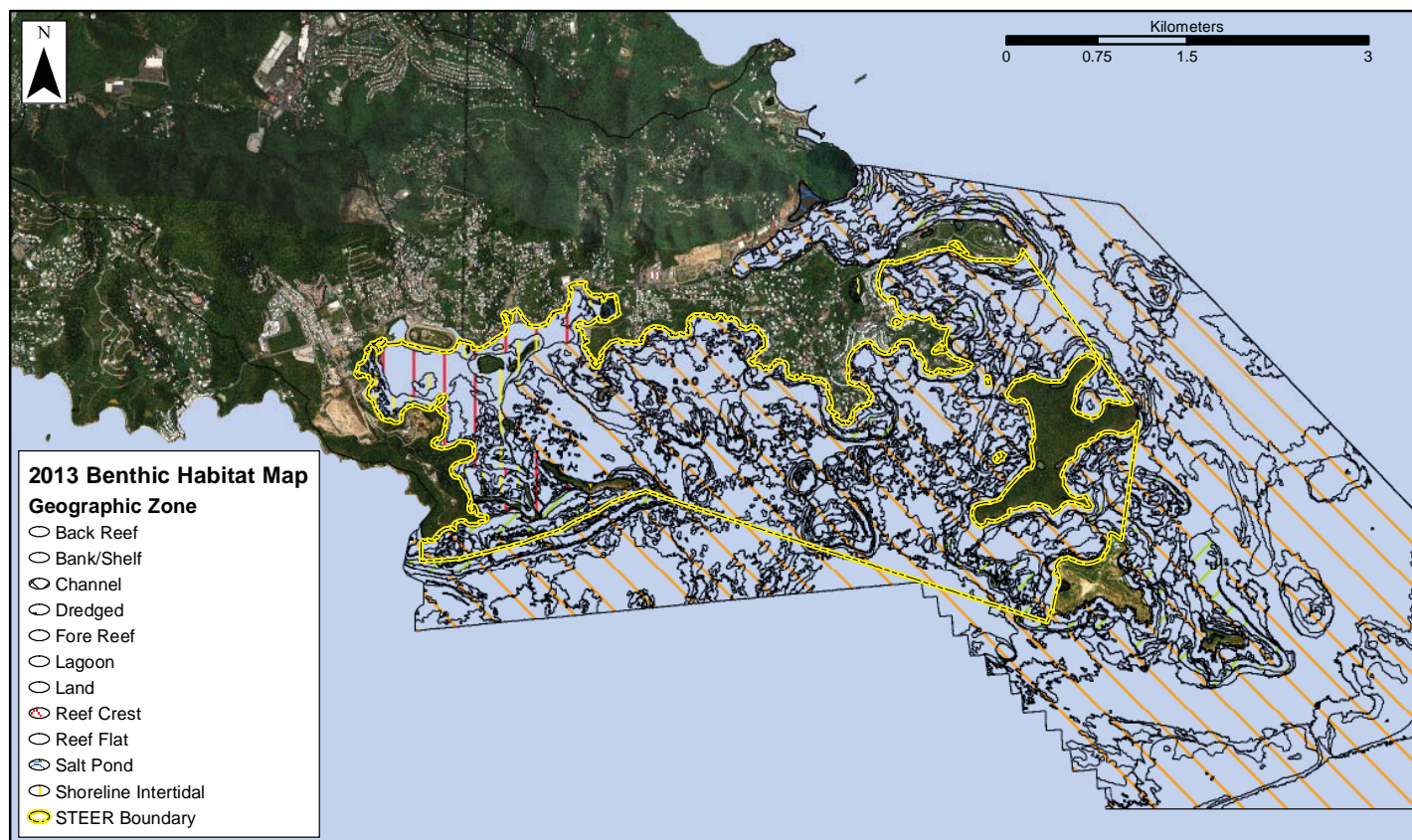


Figure 2.8. Spatial extent of geographic zones in the STEER in the 2013 NOAA map (Costa *et al.*, 2013).

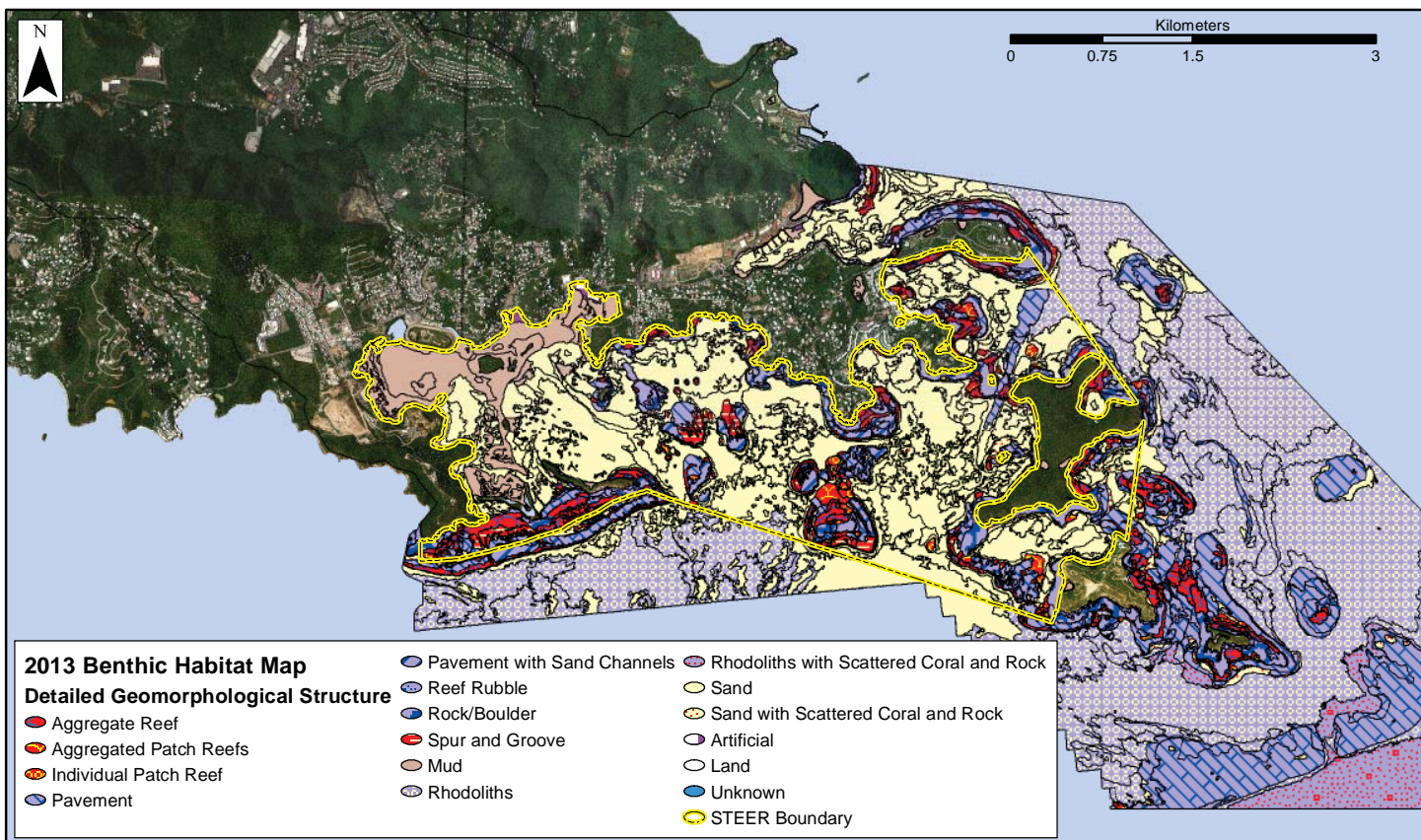


Figure 2.9. Spatial extent of major and detailed geomorphological structure types in the STEER in the 2013 NOAA map (Costa *et al.*, 2013).

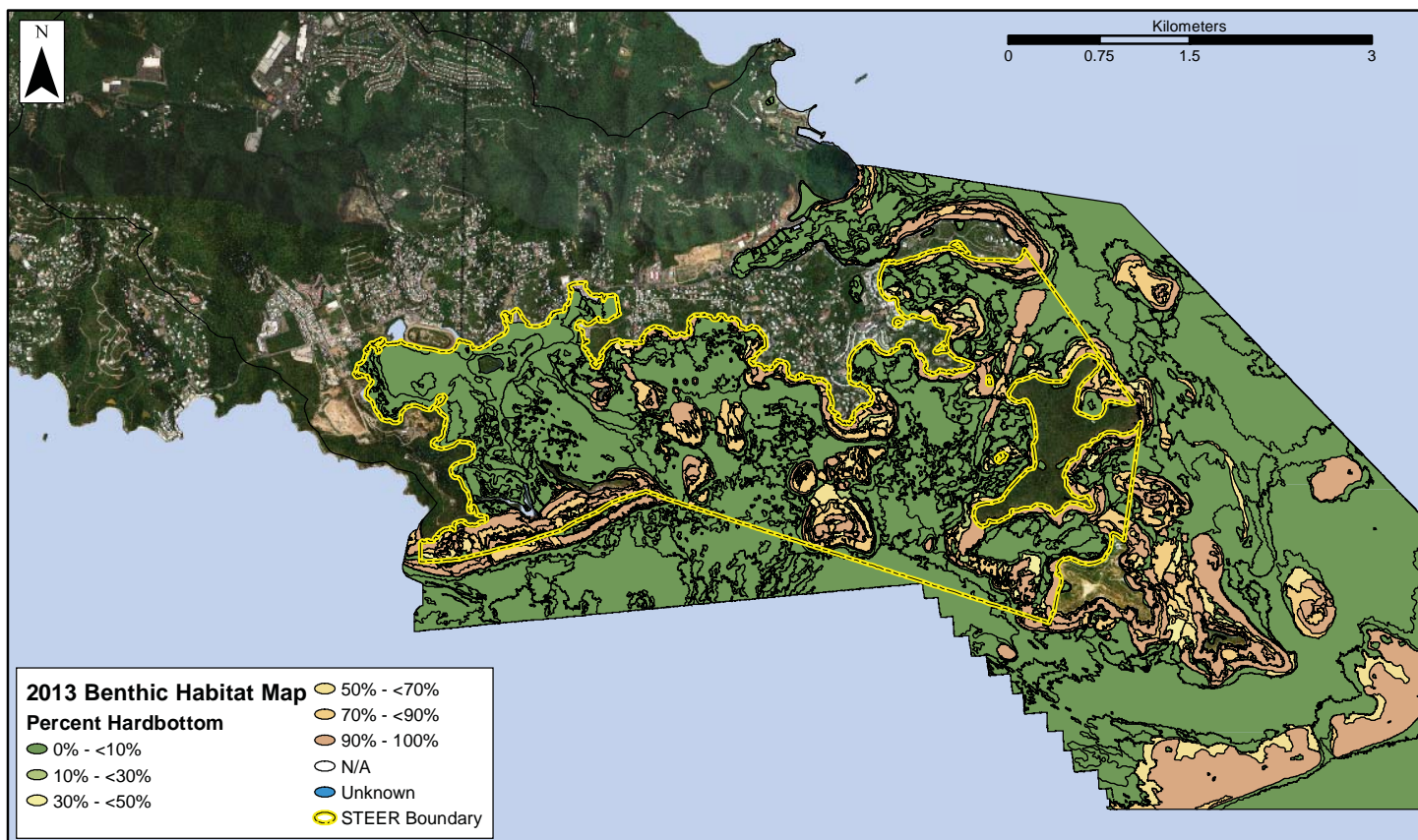


Figure 2.10. Spatial extent of percent hardbottom categories in the STEER in the 2013 NOAA map (Costa *et al.*, 2013).

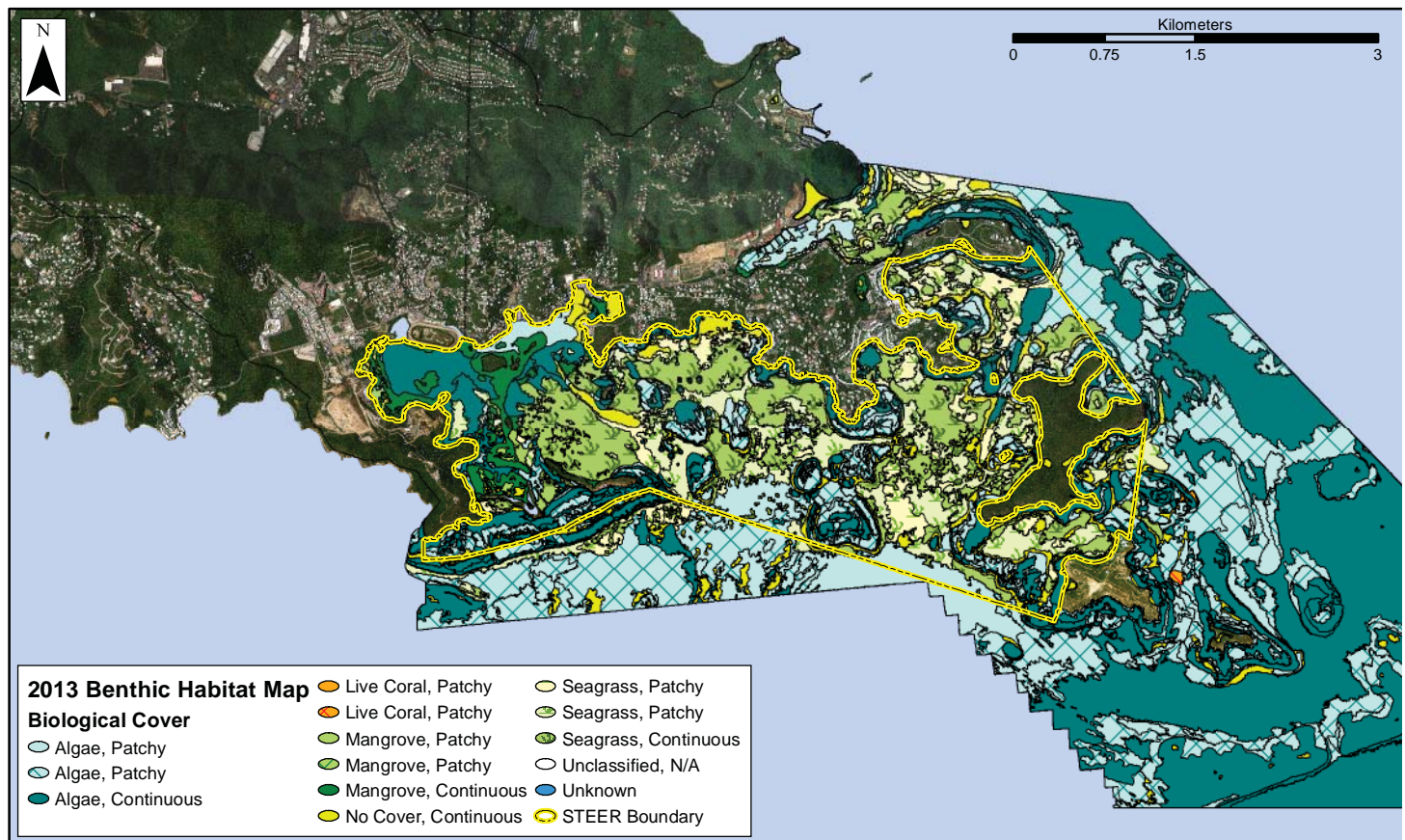


Figure 2.11. Spatial extent of major and detailed biological cover types in the STEER in the 2013 NOAA map (Costa *et al.*, 2013).

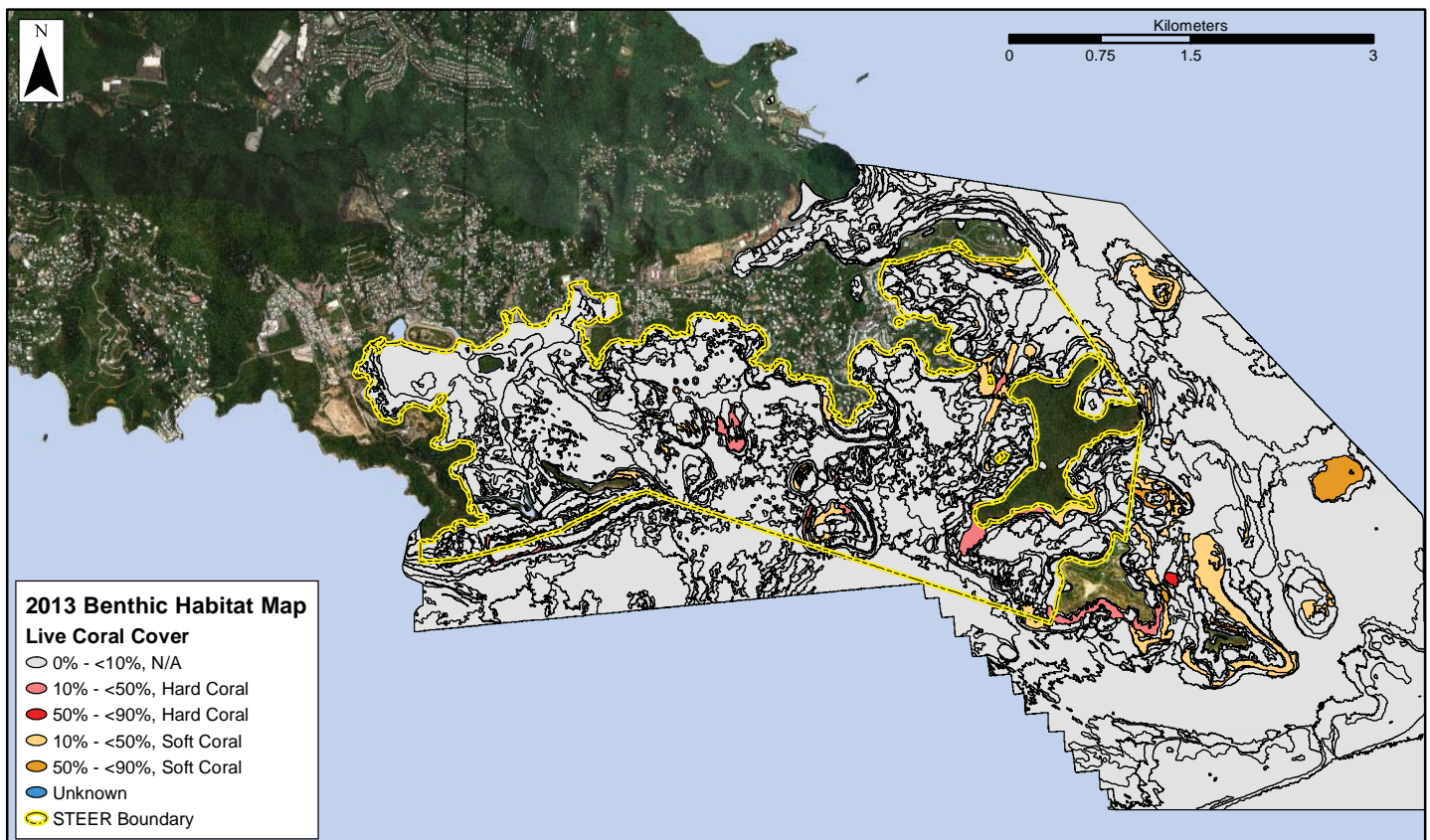


Figure 2.12. Spatial extent of percent live coral and dominant coral cover types in the STEER in the 2013 NOAA map (Costa *et al.*, 2013).

Table 2.2. Qualitative comparison of the 2001 and 2013 benthic habitat maps in the STEER, using the modified STEER boundary to include mangrove habitat in Mangrove Lagoon and Benner Bay.

	Class Names		Area (km ²)	
	2001 (4,046 m ² MMU)	2013 (100 m ² MMU)	2001	2013
Major Structure	Coral Reef and Hardbottom	Coral Reef and Hardbottom	3.20	2.80
	Unconsolidated Sediment	Unconsolidated Sediment	6.12	7.20
Detailed Structure	Linear Reef	Aggregate Reef	0.22	0.60
	Patch Reef (Aggregated)	Aggregated Patch Reefs	0.01	0.28
	-	Artificial	--	0.03
	Patch Reef (Individual)	Individual Patch Reef	0.05	0.05
	Mud	Mud	0.22	1.04
	Colonized/Uncolonized Pavement	Pavement	0.85	0.74
	Colonized/Uncolonized Pavement with Sand Channels	Pavement with Sand Channels	0.70	0.29
	Reef Rubble	Reef Rubble	0.09	0.05
	-	Rhodoliths	--	0.11
	Colonized/Uncolonized Bedrock	Rock/Boulder	1.07	0.64
	Sand	Sand	0.38	5.77
	Sand with Scattered Coral and Rock	Sand with Scattered Coral and Rock	0.09	0.28
	Spur and Groove Reef	Spur and Groove	0.12	0.16
	Algal Dominated (on softbottom)	Algae	0.05	4.11
Major Cover	-	Live Coral	--	0.00
	Mangrove	Mangrove	0.48	0.56
	-	No Cover	--	0.96
	Seagrass	Seagrass	5.10	4.35

(12.7%) structure type overall. Roughly 44% of this hard-bottom habitat was inside the STEER's boundaries, located mainly southeast of Long Point, south of Deck Point and surrounding the nearshore waters of Cabritta Point. These areas were dominated by Pavement, Rock/Boulder and Aggregate Reef (i.e., continuous, high-relief coral formation that occurs in various shapes and lacks sand channels) structure types, covering 7.4%, 6.4% and 5.9% of the area inside the STEER's boundary, respectively. However, some of the largest and most continuous hardbottom features were located just outside of the MPA boundaries around Little St. James and Dog Islands. These areas were mainly comprised of Pavement features, although Aggregate Reefs were also present along the shoreline of both islands.

Biological cover differed greatly inside versus outside of the STEER's boundary (Figures 2.6 and 2.11). Rhodolith habitats were dominated by Algae (39.2%), the majority of which was continuous (21.0%) covering 90-100% of a polygon. This cover type included various types of turf, fleshy, coralline or filamentous species, which were found primarily in Mangrove Lagoon and outside of the STEER's boundary. Hardbottom habitats inside and outside the STEER's boundary were also colonized primarily by algae. Sand habitats were dominated by continuous (90% - 100%) seagrass beds, comprising about 5.4% of the total mapped area and 21.4% of the mapped area inside the STEER's

boundary. Continuous (90% - 100%) and patchy (50% - <90%) mangrove habitats constituted only 1.4% of the total mapped area, and were located mainly inside the STEER's boundary in Mangrove Lagoon. For live coral, the majority (95.2%) of mapped area was colonized by 0% - <10% hard and soft corals (Figures 2.7 and 2.12). Areas with >10% live coral cover were rare (4.8%).

The new 2013 NOAA benthic habitat map differed from the 2001 version in several ways. First, only aerial photos and satellite imagery were used to map habitats on the seafloor in 2001, whereas both aerial photos and LiDAR imagery were collected and used to characterize benthic habitats in the 2013 map. Second, the 2013 map characterized more area than the previous map. For instance, parts of Benner Bay and an area between Cabrita Point and Great St. James Island had previously been classified as "Unknown" (Figure 2.1). Third, the new map characterized benthic habitats at a higher spatial and thematic resolution than the previous map. It did so because it used: 1) a new classification scheme with more classes, 2) a finer scale to delineate the boundaries of habitats, and 3) a smaller minimum mapping unit. These changes were made because a smaller geographic area was mapped using better quality source imagery. These different classification schemes, delineation scales and minimum mapping units prohibit a quantitative comparison between the 2001 and 2013 benthic habitat

maps because coral reef habitat polygons and attributes (e.g., polygon area) are sensitive to the resolution at which they are mapped (Kendall and Miller, 2008).

However, a broader qualitative comparison was conducted to highlight differences between benthic habitats maps within STEER (Table 2.2). For geomorphological structure, slightly less Coral Reef and Hardbottom was delineated in the 2001 map. This decrease in coral reef and hardbottom habitat was most likely due to the coarser scale at which features were delineated and the large areas on the seafloor that were left uncharacterized. For major biological cover, a substantial amount of more algae was delineated in the 2013 map as compared to the 2001 maps. This increase is most likely due to more seafloor area being characterized in 2013, as well as to the inclusion of macro, crustose, turf and filamentous algae in the Algae class in the 2013 map. The 2001 map only included macroalgae on softbottom and ignored the other algal classes. Interestingly, more area dominated by seagrass was delineated in the 2001 map, than in the 2013 map. While this is likely partly due to the coarser MMU, areas of Mangrove Lagoon and Benner Bay that were previously classified as seagrass in 2001 (Figure 2.1) were mapped as algae in the most recent effort (Figure 2.6) and may denote changes in seagrass distributions.

The final deliverables for the 2013 NOAA mapping effort, which include the benthic habitat map, ground validation and accuracy assessment data, and associated aerial imagery, are available to the public: 1) on the web (<https://coastalscience.noaa.gov/projects/detail?key=171>), 2) through an interactive, web-based map application (<http://maps.coastalscience.noaa.gov/biomapper/biomapper.html?id=STEER>), and 3) by request through the University of the Virgin Islands, Center for Marine and Environmental Studies. Potential uses for the STEER benthic habitat map include updating the management plan of the STEER, evaluating different zoning options for multiple use areas, evaluating the efficacy of future management actions, and mapping ecosystem services and estimating economic value of goods and services across the seascape. Looking forward, these map applications may help scientists and managers to better understand the benthic communities, and their relationship with particular species and groups of species within the STEER. A better understanding of these ecological relationships is key to forecasting how the distribution of these benthic communities and their associated animals may change in the future.

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CHAPTER 3: CHARACTERIZATION OF FISH COMMUNITIES AND ASSOCIATED BENTHIC HABITATS IN THE ST. THOMAS EAST END RESERVES

Laurie Bauer^{1,2}, Jenny Vander Pluym³, Chris Jeffrey^{1,2}, Chris Caldw¹, Amy V. Uhrin³

¹NOAA/NOS/National Centers for Coastal Ocean Science, Center for Coastal Monitoring and Assessment (CCMA), 1305 East/West Highway, Silver Spring, MD 20910

²CSS-Dynamac Consolidated Safety Services, Fairfax, VA 22030, under CSS-Dynamac Contract #EA-133C-14-NC-1384

³Center for Coastal Fisheries and Habitat Research, National Centers for Coastal Ocean Science, National Ocean Service, National Oceanic and Atmospheric Administration

3.1 INTRODUCTION

The objective of this chapter is to expand on previous knowledge to provide a spatially-comprehensive characterization of fish and benthic communities within the STEER. This comprehensive assessment of the fish and benthic communities will be used: 1) as an inventory of the current resources, 2) as a baseline from which to monitor the success rate of any future management actions, and 3) to inform management decisions for the STEER, such as the locations for restoration actions. Lastly, we compared results from STEER with other locations in the U.S. Caribbean.

3.2 METHODS

Site Selection

Field surveys were conducted in June 2012 to characterize the fish communities and associated habitats in the STEER marine ecosystem. Sites were randomly selected within strata to ensure coverage of the entire study region (Figure 3.1). NOAA's existing benthic habitat map (Kendall *et al.*, 2001) was used as the basis for site stratification. Although a newly updated benthic map was being produced (Costa *et al.*, 2013) it was not yet completed at the time of this study. The strata that were chosen for this study included hardbottom, unconsolidated sediments, and mangrove. The Hardbottom comprised bedrock, pavement (i.e., flat, low-

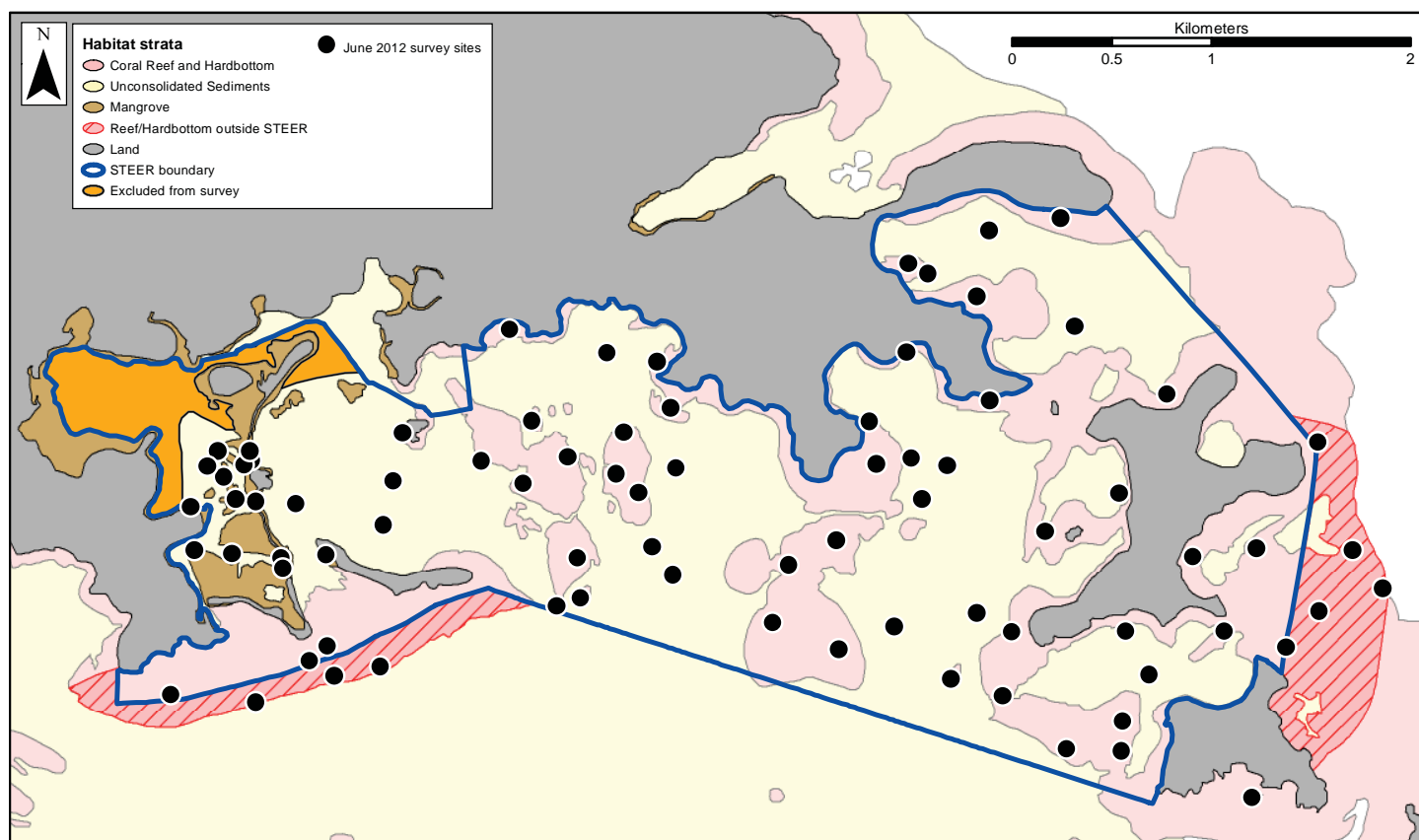


Figure 3.1. Benthic habitat strata and site locations of the June 2012 survey of benthic habitat composition, fish communities, invertebrates, and marine debris. The boundaries of the St. Thomas East End Reserves (STEER) are shown for reference.

relief hardbottom), rubble, and coral reef, while the Unconsolidated Sediments stratum comprised submerged aquatic vegetation (i.e., seagrass and macroalgae), as well as uncolonized sand and mud. The Mangrove stratum comprised the seaward edge of mangrove habitat able to be surveyed with these visual underwater survey methods. In addition, two hardbottom areas outside of STEER that were of interest to STEER's Core Planning Group were included as a separate stratum (9 sites total). One of the "outside STEER" sites south of Little St. James was not part of the random selection but a targeted location. The site was chosen with input from The Nature Conservancy (TNC) during extra time at the end of the mission, due to interest in potential effects of island development on the surrounding marine ecosystem.

Due to water quality concerns and low visibility, portions of Mangrove Lagoon and Benner Bay were excluded from the study area (Figure 3.1). In order to effectively survey fish using underwater visual methods, it is necessary that divers have a minimum of 2 meters visibility. Randomly selected alternate sites were available for each stratum if

low visibility prevented a primary site from being surveyed. Surveys for two primary sites (both located south of Benner Bay) could not be completed because they did not meet the visibility requirements. In addition, extra precautions were taken in the area where the inter-island ferries traverse through STEER; while the majority of surveys in this high traffic area were successfully completed, it was necessary to abort one site due to safety concerns. Surveys were completed at alternate sites for all low visibility and high traffic sites that were aborted.

Field Methods

The surveys of benthic habitats, fish communities, marine debris and macroinvertebrates were conducted within a 25x4 m transect (100 m²), along a random heading. Two divers performed the survey at each site (Figure 3.2a,b). One diver (fish diver) was responsible for visual counts and size estimation of fish species. The second diver (habitat diver) quantified benthic habitat composition, macroinvertebrates and marine debris. These methods have been used to monitor St. John, USVI and other locations within

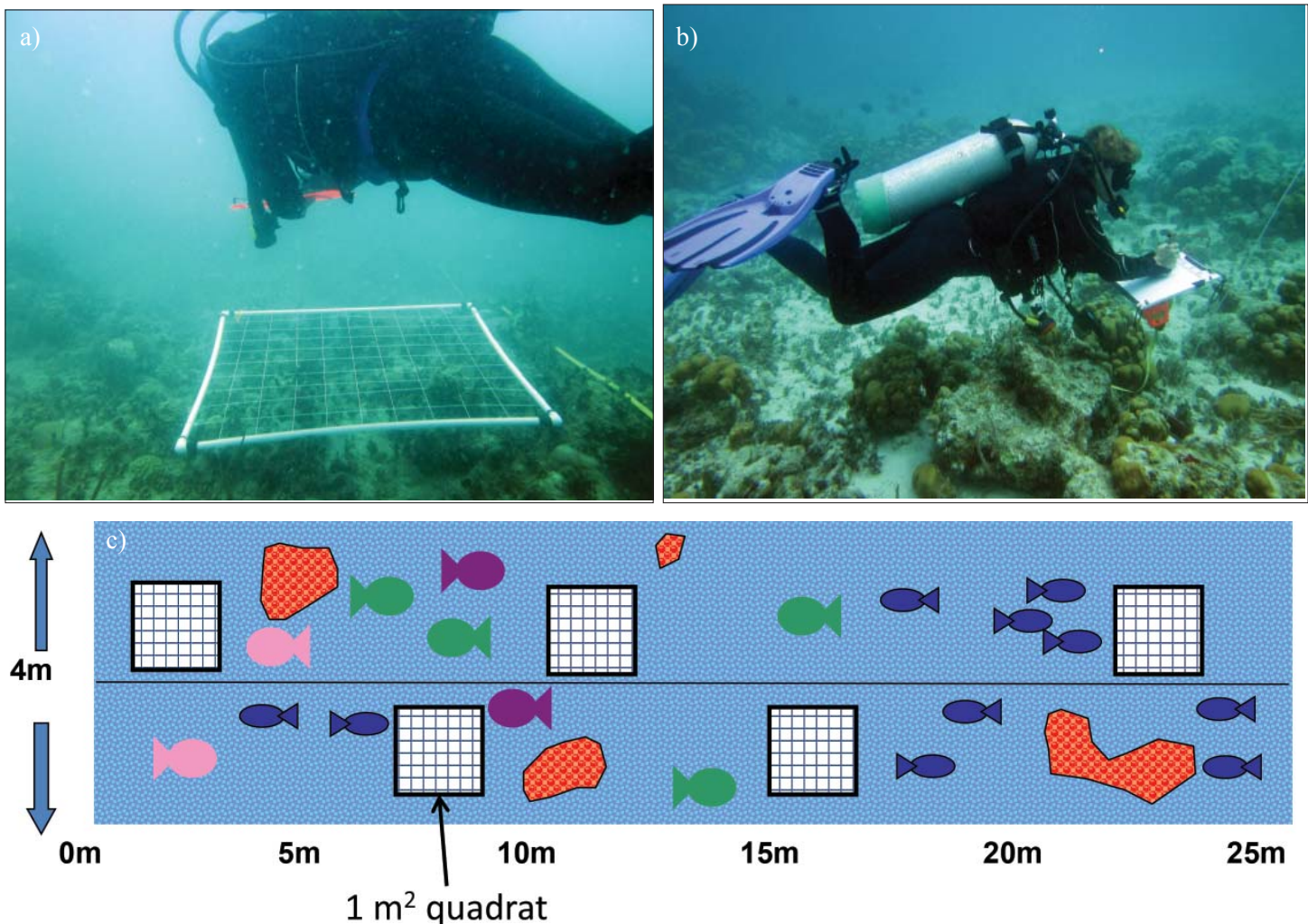


Figure 3.2. a) Diver collecting data on benthic habitat, b) diver collecting data on fish composition, and c) schematic representation of the placement of the 1m² quadrat along a 25 m transect tape during fish and benthic community surveys.

Table 3.1. Abiotic and biotic variables measured in five quadrats along fish transects.

Benthic Variables	Measurements		
	Cover (%)	Height (cm)	Abundance (#)
Abiotic			
Hardbottom	X	X	
Sand	X		
Rubble	X		
Fine sediment/silt	X		
Rugosity			
Water depth			
Biotic			
Corals (by species)	X		
Algae			
Macroalgae	X	X	
Turf Algae	X		
Crustose/coralline algae (CCA)	X		
Filamentous algae/cyanobacteria	X	X	
Seagrass (by species)	X	X	
Gorgonians			
Sea rods, whips and plumes	X	X	X
Sea fans	X	X	X
Encrusting form	X		
Sponges			
Barrel, tubes, rope, vase	X	X	X
Encrusting form	X		
Other benthic macrofauna			
Anemones and hydroids	X		X
Tunicates and zoanthids	X		
Mangroves			
Prop roots			X
Prop roots colonized by algae			X
Prop roots colonized by sponges			X
Prop roots colonized by other biota			X

NOAA's Caribbean Coral Reef Ecosystem Monitoring Project (CCREMP) (Pittman *et al.*, 2008; Pittman *et al.*, 2010; Friedlander *et al.*, 2013). The standardized protocols allow for comparisons to be made between different areas. In addition, the protocols include measurement of numerous variables that can be used to monitor and evaluate changes following the reduction in the input of land-based sources of pollution to the marine environment.

Fish Census

Fish surveys were conducted along the 25x4 m transect (100 m²) using a fixed survey duration (15 minutes) regardless of habitat type or complexity. The number of individuals per species was recorded in 5 cm size class increments up to 35 cm using visual estimation of fork length. If the individual could not be identified to species, they were identified to the extent possible (i.e., genus or family).

Individuals greater than 35 cm were recorded as an estimate of the actual fork length to the nearest centimeter. At mangrove sites, the survey was conducted along the edge of the mangrove canopy. The transect was laid out as close to the prop roots and as far into the mangroves as possible, up to 2 m, and then out to the edge of the mangrove overhang such that the total area surveyed was still 100 m².

Benthic habitat composition

The habitat diver first assigned an overall bottom type (i.e., hardbottom, unconsolidated sediments, or mangrove) to each transect based on in situ observation. Data on the percent cover of abiotic and biotic composition at each survey site were recorded within five 1 m² quadrats placed randomly along the 25x4 m transect so that one quadrat fell within every 5 m interval along the transect. The quadrat was placed at each randomly chosen meter mark and systematically alternated from side to side along the transect tape (Figure 3.2c). Several variables were measured to characterize benthic composition and structure (Table 3.1). The quadrat was divided into 100 smaller 10x10 cm squares with string (1 small square = 1% cover) to help the diver with estimation of percent cover. Percent cover was determined by looking at the quadrat from above and visually estimating percent cover in a two dimensional plane. Information was recorded for the following variables: Abiotic cover - the percent cover (to the nearest 1%) of four abiotic substrate categories (hardbottom, sand, rubble, and fine sediments/silt) was estimated within each 1 m² quadrat. The maximum vertical relief of the hardbottom was also measured.

Biotic cover - the percent cover (to the nearest 0.1%) of algae, seagrass, live corals, sponges, gorgonians (also known as "soft" corals), and other biota was estimated within each 1 m² quadrat. Taxa were identified to the following levels: stony coral (species), seagrass (species), algae (morphological group), sponge (morphological group), and gorgonians (morphological group) (Table 3.1). For stony and fire corals, the percentage of bleached coral and diseased/dead coral was estimated to the nearest 0.1 percent. In addition, the presence of *Acropora palmata* or *A. cervicornis* either within the transect area (100 m²) or the vicinity of the sample site was also noted by the divers.

Maximum canopy height - the maximum height of sponges, gorgonians, and soft algal groups was recorded to the nearest 1 cm in each quadrat.

Number of individuals - the number of individual upright sponges, gorgonians, non-encrusting anemones, and non-encrusting hydroids was recorded in each quadrat.

Rugosity – for hardbottom sites, rugosity was measured by placing a 6 m chain at two randomly selected positions, ensuring no overlap, along the 25 m belt transect. The chain was positioned along the centerline of the transect such that it followed the substrate's relief. The straight-line horizontal distance covered by the chain was measured. An index of rugosity (R) was calculated as the ratio of contoured surface distance (d) to linear distance (L = 6m) using $R=1-d/L$. Rugosity is unitless and can range from zero (no relief) to one.

Mangrove habitat data – For surveys conducted in mangrove habitat, all of the habitat variables were collected along with additional data, including: number of prop roots, number of prop roots colonized by algae, number of prop roots colonized by sponges and number of prop roots colonized by other biota (tunicates, anemones, zooanthids, etc).

Macroinvertebrate counts - The habitat diver counted the abundance of spiny lobsters (*Panulirus argus*), long-spined urchins (*Diadema antillarum*), and the abundance and sexual maturity of queen conchs (*Lobatus gigas*) within the 25x4 m transect at each site. The maturity of each conch was determined by the absence (immature) or presence (mature) of a flared lip.

Marine debris

The number and type of marine debris within the 25x4 m transect were recorded. Marine debris size and the area of habitat it affected were estimated. Any flora or fauna that were colonizing the debris item were noted.

Data Analysis

Benthic Habitat

While many benthic variables were measured during the surveys, data analyses for this report focused primarily on describing differences among major habitat types and broad-scale spatial patterns in the percent cover of the sessile biotic components as described in Table 3.1. Quadrat measurements along each transect were averaged and cumulative coral species richness (the total number of species present was calculated for each site. Average site values were used to calculate means and standard errors of measured variables for each habitat strata (mangrove, unconsolidated sediments, hardbottom inside STEER, hardbottom outside STEER). In addition, data were plotted in a geographic information system (ArcGIS v10.1, ESRI) to examine broad spatial patterns in the benthic cover variables.

Fish

Several fish metrics were summarized by stratum (mangrove, unconsolidated sediments, hardbottom inside

STEER, hardbottom outside STEER). Strata-wide means and standard errors (SE) were estimated for biological community metrics (total density, total biomass, species richness, Shannon diversity, and density and biomass of trophic groups). Means and SE calculations were computed employing methods described by Cochran (1977) in the statistical analysis software, SAS v9.3 (Proc Survey means). Trophic groups surveyed included piscivores, herbivores, invertivores, and zooplanktivores and were defined for each species based on diet information from Randall (1967). It is important to note that these groups are not mutually exclusive because many fish species can be classified into two or more of these groups based on diet. In those circumstances the trophic group was assigned based on the dominant diet component. Biomass was calculated using published length-weight relationships based on the allometric scaling law,

$$W = aL^b$$

where L is length in centimeters and W is weight in grams, a is a condition factor related to body form, and b is the scaling exponent indicating either isometric or allometric growth. The midpoint of each size class was used for L values up to 35 cm, and the actual length was used for fish >35 cm. For fish in the 0-5 cm size class, 3 cm was used as the mid-point because we do not typically observe fish <1 cm. Values for a and b by species were obtained from FishBase (Froese and Pauly, 2008). Biomass for species with no published length-weight relationships was calculated using terms for the closest congener with most similar morphology.

Species diversity was calculated using the Shannon Index (H'), a measure that incorporates both richness and evenness (relative abundances):

$$H' = \sum p_i(\log p_i)$$

where p_i is the relative abundance of each species. H' increases as both the richness and evenness of the community increase. Typical values for H' range from 1.5 – 3.5 and rarely exceed 4.

Data were plotted in ArcGIS to examine broad spatial patterns in the fish metrics. In addition, select families and species of commercial and/or ecological interest were selected for further examination. For each species/family, a summary of the species distribution, size frequency, and mean density and biomass by habitat type and study area was calculated. Age class (juveniles/sub-adults and adult) was identified based on mean length at maturity as identified by FishBase (Froese and Pauly, 2008) and

García-Cagide *et al.* (1994). Where length at maturity was unknown, 1/3 of maximum size was used as a proxy as in Pittman *et al.* (2008, 2010). Percent occurrence, mean density and biomass (per 100 m²) and corresponding SE were calculated for each species across the sites within STEER. This information was used to create a summary table of all species observed in this characterization across the STEER sampling domain (Appendix 1 in Bauer *et al.*, 2014), as well as the hardbottom sites outside STEER (Appendix 2 in Bauer *et al.*, 2014).

Differences and similarities in species composition were further examined using multivariate statistical techniques (Primer v.6, Clarke and Warwick, 2001). Density and biomass data were square-root transformed prior to analysis. Data were arranged in a species density/biomass by site data matrix, which was used to construct a triangular matrix of the percentage similarity in community composition between all pairs of sites using the Bray-Curtis Coefficient. The coefficient is a measure of how similar samples were to each other, ranging from 0% (complete dissimilarity) to 100% (complete similarity). Next, non-metric multidimensional scaling (nMDS) was used to place samples in a two-dimensional configuration such that the rank order of the distances between the samples agreed with the rank-order of the similarities from the Bray-Curtis matrix. Sites were coded by bottom type and management (Inside/Outside STEER) for examination of visual patterns of between site similarity. These factors were also used to test for significant differences in similarity using Analysis of Similarities (ANOSIM), a multivariate, non-parametric version of ANOVA. Outputs of the ANOSIM test include an R statistic and p-value. R is a difference of average rank dissimilarities between and within groups, scaled so that R ranges between 0 (no differences) and 1 (perfect division), while the p-value gives the statistical significance for a test of R = 0. Significant differences in fish community structure were examined with the similarity percentages (SIMPER) routine to identify those species that contributed most to the observed dissimilarity.

To see how fish communities in STEER compare with other parts of the U.S. Caribbean, key fish community metrics (species richness, total biomass, and biomass of groupers, snappers, grunts, and parrotfish) were compared with other locations within NOAA's Caribbean Coral Reef Ecosystem Monitoring Project (http://www8.nos.noaa.gov/biogeography_public/query_main.aspx), including St. John (2011), Eastern St. Croix (2010), which includes the Buck Island Reef National Monument and East End Marine Park, and Southwest Puerto Rico, including La Parguera/Guanica (2012). Mangrove strata comparisons could only



A school of snapper swim below an undercut mangrove stand near the false entrance to Mangrove Lagoon.

be made with SW Puerto Rico. Sites on the mid-shelf reef in St. John were removed from the analysis to allow for a more equitable depth comparison with STEER. A Shapiro-Wilks test and visual examination of the data were used to assess normality, and as data were non-normally distributed, nonparametric Wilcoxon tests and the corresponding non-parametric Dunn's multiple comparisons tests (Zar, 1999) were used to test for differences among regions (JMP v11.0). Means and standard errors of these metrics were also plotted for visualization.

3.3 RESULTS AND DISCUSSION

A total of 80 sites were surveyed during the two-week field mission: 26 on unconsolidated sediments, 10 along the mangrove fringes, 35 on hardbottom within STEER, and nine on hardbottom outside STEER boundaries. Two sites within the hardbottom stratum were identified as soft bottom habitat by the survey divers and were subsequently grouped with the unconsolidated sediment surveys for analysis. Conversely, one site within the unconsolidated sediment stratum was classified as hardbottom during the survey and subsequently included with the hardbottom surveys for analysis.

Benthic Habitat

Abiotic composition

Not surprisingly, hardbottom substrate dominated sites within the hardbottom strata with minor components of rubble and sand. No fine sediment was observed along transects conducted at these sites (Figure 3.3a). In contrast, soft bottom sites were primarily composed of sandy bottom with some fine sediment and rubble (Figure 3.3b).

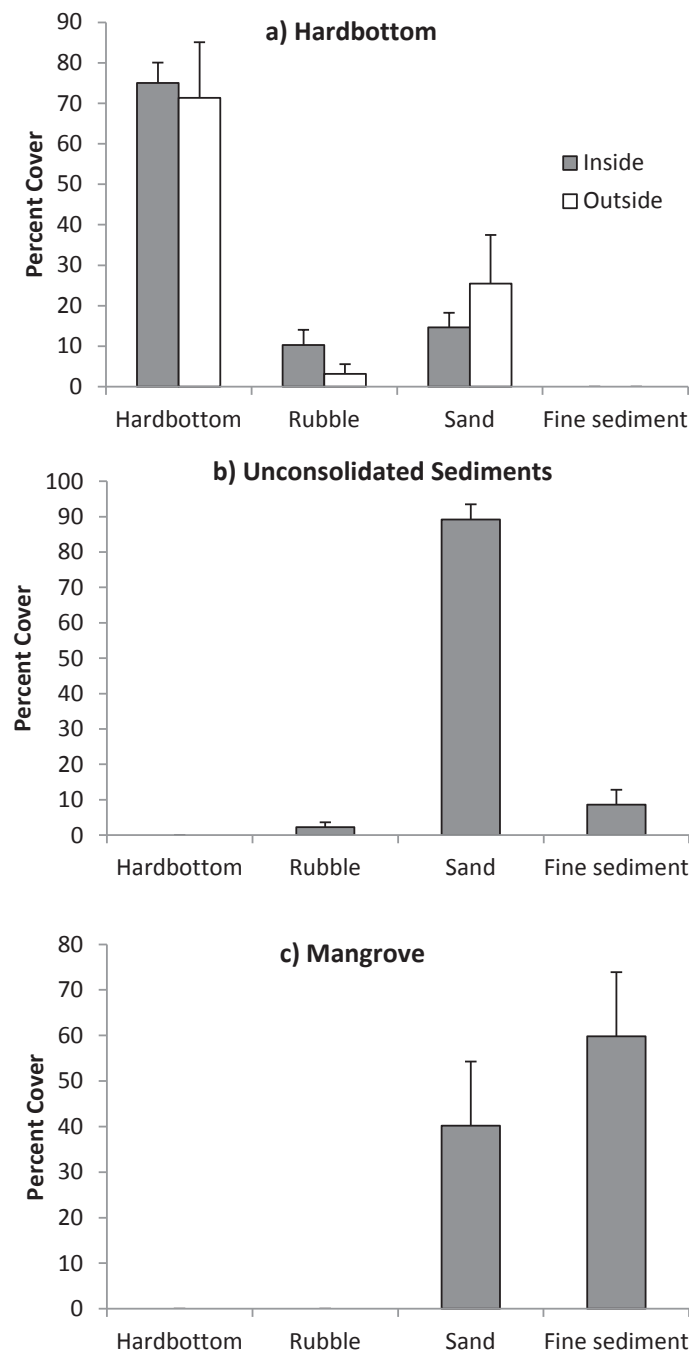


Figure 3.3. Mean (\pm SE) percent cover of abiotic substrate by bottom type: a) hardbottom inside (N=35) and outside (N=9) the STEER boundaries, b) softbottom (N=26), and c) mangrove (N=10).

The predominance of sand over fine sediments may be attributable to several factors including bottom currents, and the proximity of hardbottom habitats to the unconsolidated sediment survey locations. Portions of Benner Bay and Mangrove Lagoon that were excluded from the survey are bordered by vegetated coastline and dominated by fine sediment. Mangrove sites were characterized with fine sediments and sandy bottom (Figure 3.3c).

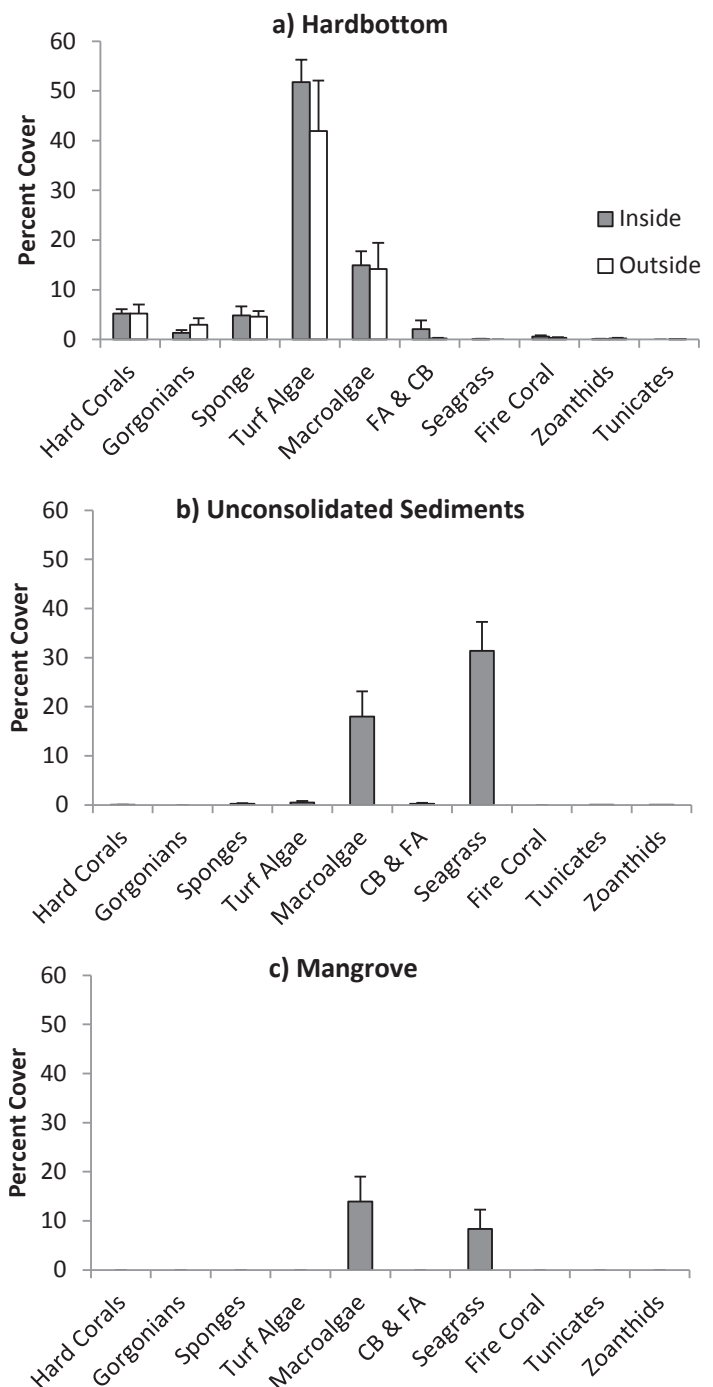


Figure 3.4. Mean (\pm SE) percent cover of major cover groups by bottom type: a) hardbottom sites inside (N=35) and outside (N=9) the STEER boundaries, b) softbottom (N=26), and c) mangrove (N=10). FA & CB = filamentous algae and cyanobacteria.

Biotic Composition Overview

Turf algae dominated the biotic composition of hardbottom sites with a mean (\pm SE) percent cover of $51.8 \pm 4.5\%$ inside/ $41.9 \pm 10.1\%$ outside, followed by macroalgae ($14.9 \pm 2.8\%$ inside/ $14.1 \pm 5.3\%$ outside), hard (scleractinian)

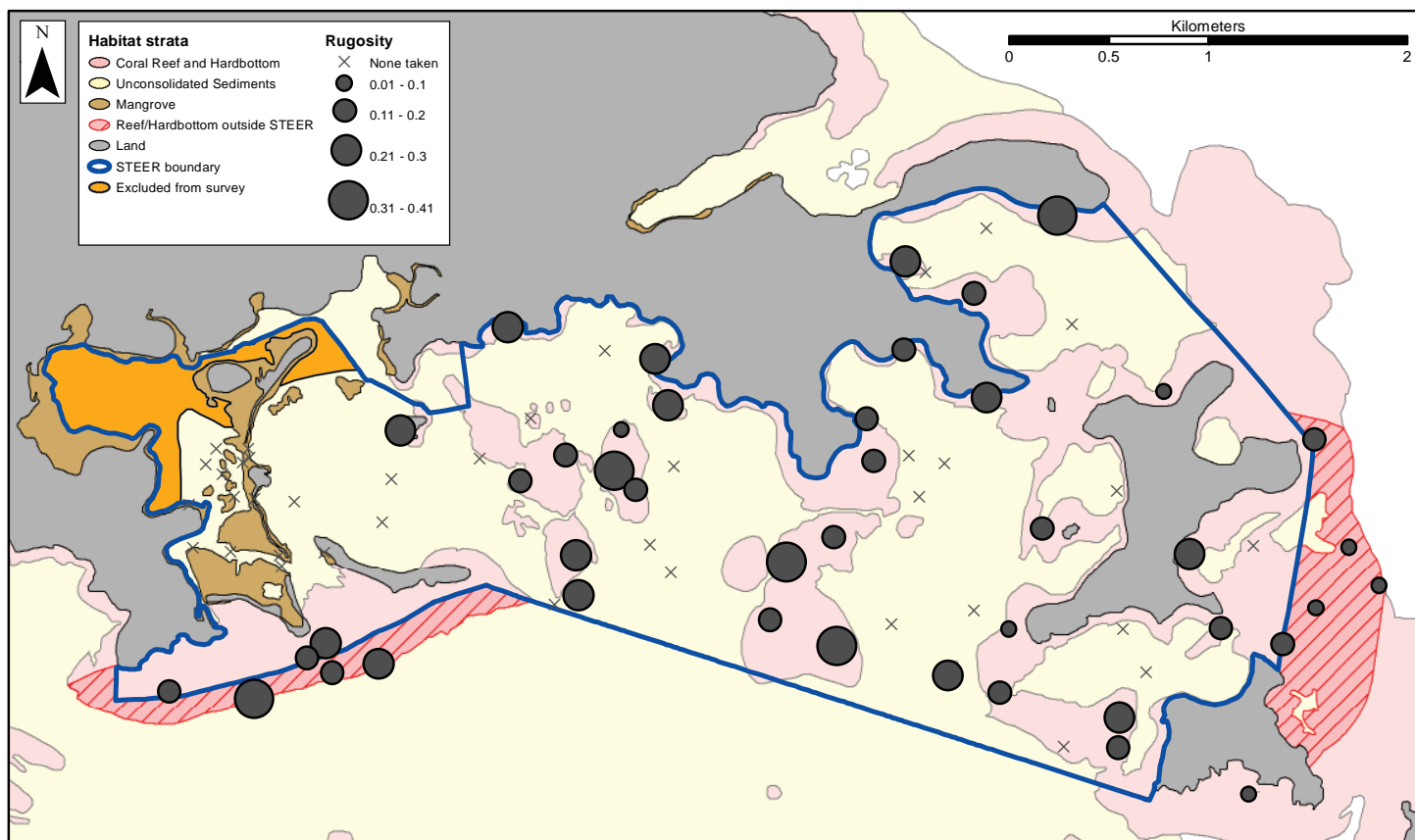


Figure 3.5. Mean rugosity (hardbottom sites only).

corals ($5.2 \pm 0.8\%$ inside/ $5.2 \pm 1.8\%$ outside), sponges ($4.8 \pm 1.8\%$ inside/ $4.6 \pm 1.1\%$ outside), cyanobacteria & filamentous algae ($2.1 \pm 0.2\%$ inside/ $1.8 \pm 0.1\%$ outside), and gorgonians ($1.3 \pm 0.5\%$ inside/ $2.9 \pm 1.3\%$ outside, Figure 3.4a). Other types of biotic cover were documented in small amounts: fire coral (*Millepora spp.*) ($0.6 \pm 0.2\%$ inside/ $0.3 \pm 0.1\%$ outside), zoanthids ($0.07 \pm 0.02\%$ inside/ $0.1 \pm 0.1\%$ outside), seagrass ($0.06 \pm 0.05\%$ inside/ 0 outside), and tunicates (0 inside/ $0.008 \pm 0.008\%$ outside). Bare, uncolonized substrate averaged $19.1 \pm 3.7\%$ inside/ $30.4 \pm 11\%$ outside. Rugosity ranged from 0.02 to 0.41 and averaged

$0.17 \pm 0.04\%$ inside/ $0.15 \pm 0.04\%$ outside (Figure 3.5). The second highest rugosity (0.37) was recorded at a site in the southwestern portion of the study area, outside the STEER boundaries.

Unconsolidated sediment site assemblages were composed of mostly seagrass ($31.4 \pm 5.9\%$) and macroalgae ($17.9 \pm 5.1\%$, Figure 3.4b). Minor components of the benthic community include: turf algae ($0.5 \pm 0.4\%$), sponge ($0.25 \pm 0.11\%$), cyanobacteria & filamentous algae ($0.23 \pm 0.23\%$), hard corals ($0.04 \pm 0.02\%$), tunicates ($0.009 \pm 0.007\%$), and zoanthids ($0.007 \pm 0.007\%$). Bare substrate averaged $49.6 (\pm 5.9\%)$ which is over twice the mean at hardbottom sites. Substrate found at mangrove sites was mostly bare, unconsolidated sediment ($77.71 \pm 6.30\%$). The benthic community at these sites was comprised of macroalgae ($13.9 \pm 5.1\%$) and seagrass ($8.4 \pm 3.9\%$, Figure 3.4c). Other algae types (turf algae, cyanobacteria and filamentous algae) were largely absent from the benthic substrate in mangrove habitats. Number of prop roots per 1 m^2 ranged from 8-50 roots/ m^2 and averaged 20 (± 4.0) across all mangrove sites. Almost 100% of prop roots had algae growing on them (99.9%) with sponges and other invertebrates (tunicates, anemones, and zoanthids) occurring on 28% and 47.4% of the prop roots, respectively (Figure 3.6).



Figure 3.6. Mangrove prop roots colonized with algae and benthic fauna.

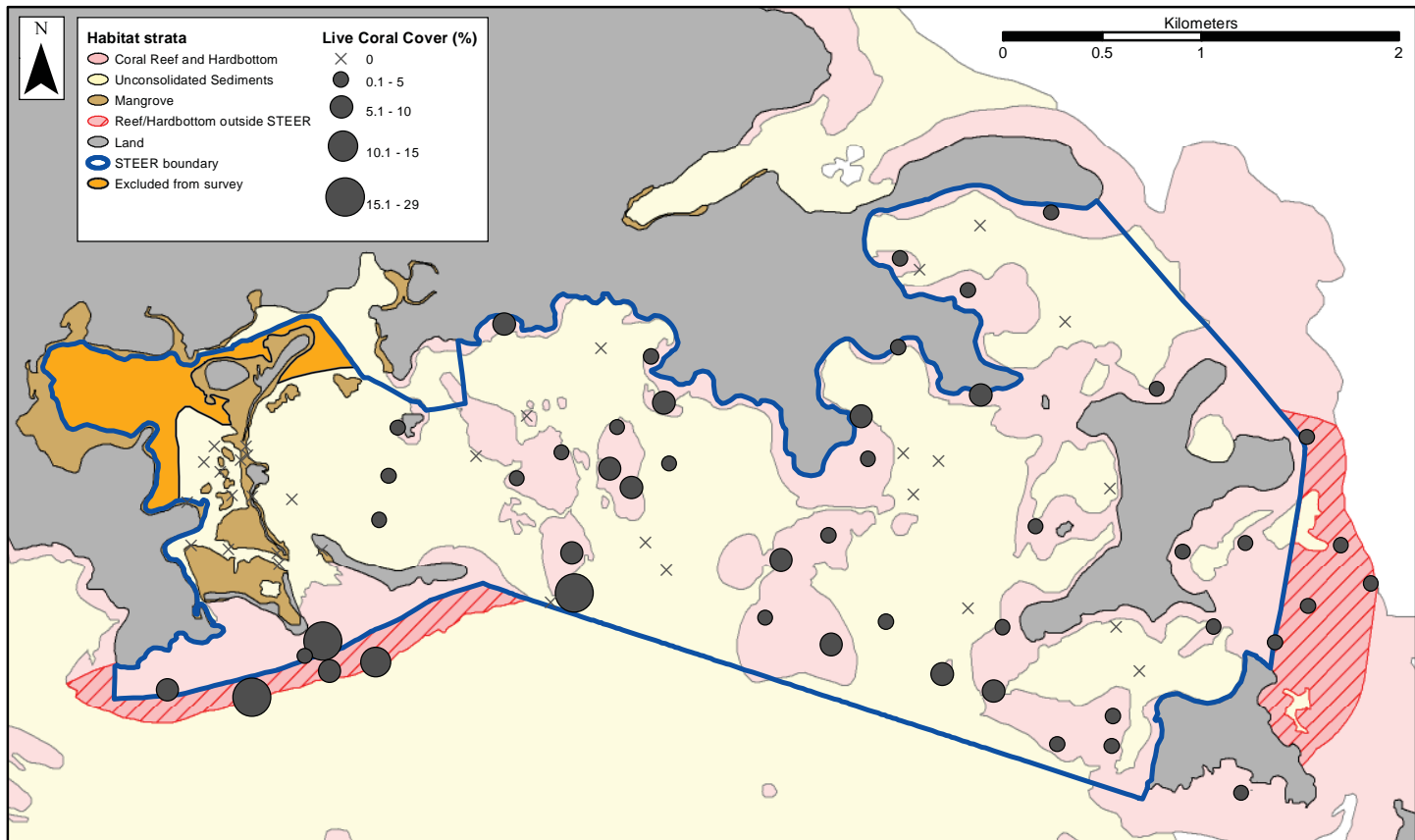


Figure 3.7. Percent live cover of hard corals.

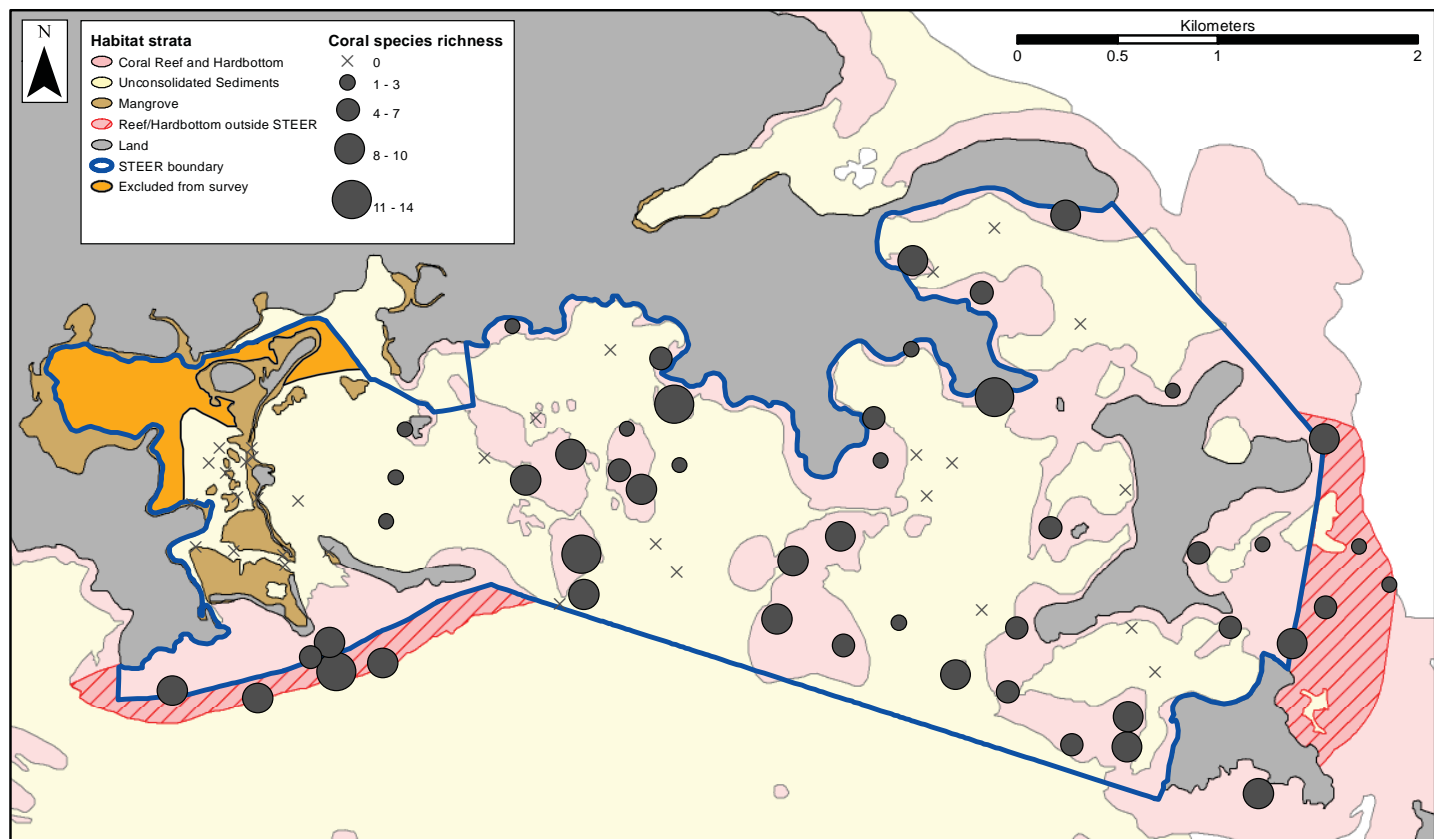


Figure 3.8. Hard coral species richness.

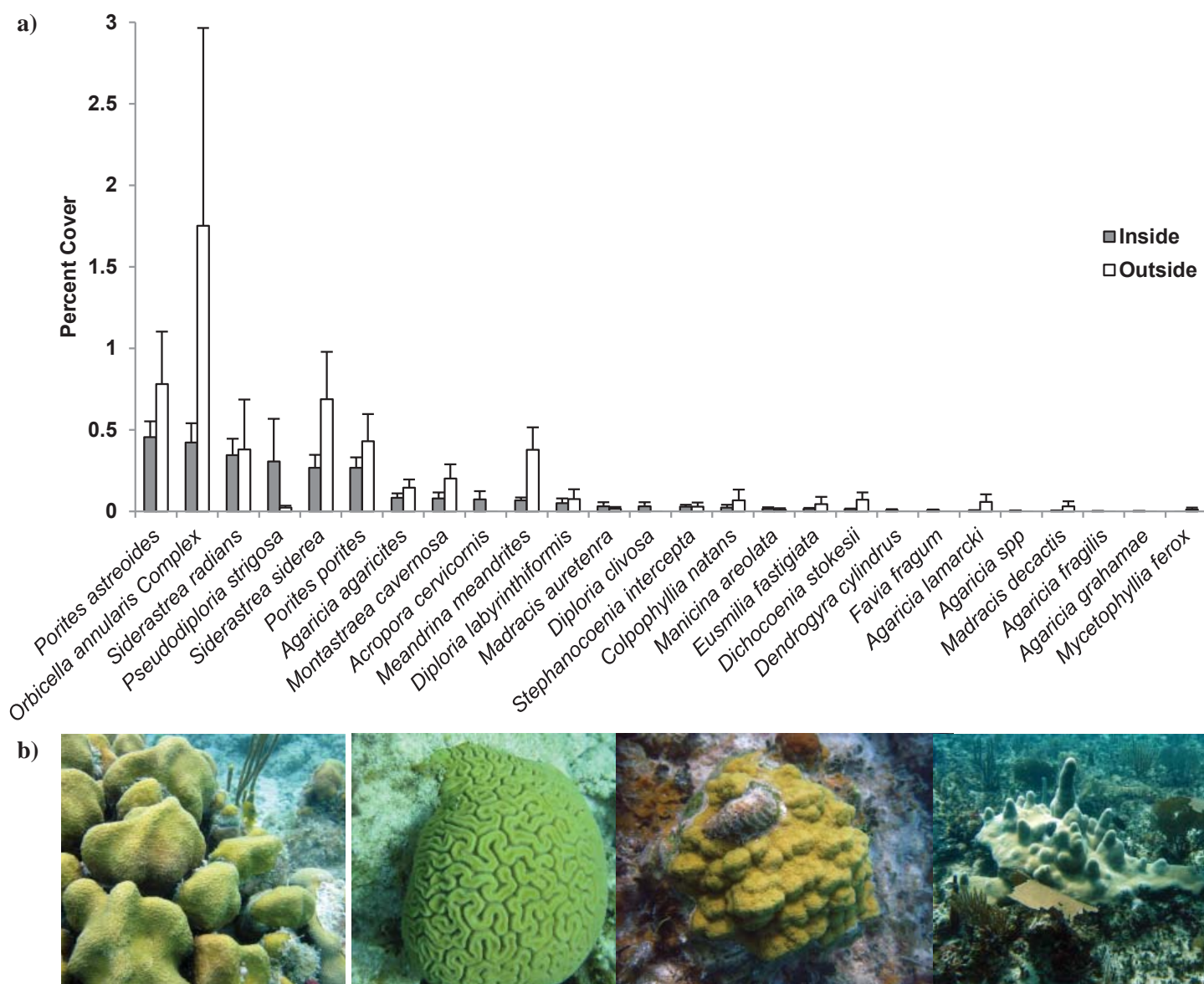


Figure 3.9. a) Mean (±SE) percent cover of hard corals by species at hardbottom sites (N=44). b) From left to right: common star coral, grooved brain coral, mustard hill coral, and pillar coral.

Hard Coral Composition

Live scleractinian coral cover ranged from 0% in mangrove and soft bottom habitats to 0.08-29% cover in hardbottom sites (Figure 3.7). The greatest coverage of hard corals was found on a reef tract that runs from Long Point to just east of Cas Cay, located in the southwest corner of the STEER. The STEER boundary passes through the middle of the reef tract so that only half of the ecosystem falls under any management plan. Three of the four sites with the highest coral cover (11-29%) are in this reef tract with two sites (16.8% and 11%) located outside the STEER boundary. The top six sites for coral cover (7.6-29%) are all in open water conditions in the southern region of the STEER and further from land features. Overall, coral cover was still low with an average of $5.2 \pm 0.8\%$ and $5.2 \pm 1.8\%$ on hardbottom habitats inside and outside STEER, respectively.

Hard coral species richness ranged from 0-14 at individual sites with an average of 7.2 ± 0.4 inside and $6.9 \pm 1.2\%$ outside (Figure 3.8). Unlike percent cover, species richness did not reflect any small scale regional patterns, and it does not appear to correlate with percent cover. The five sites with highest richness are evenly distributed throughout hardbottom habitats inside and outside of STEER: two are adjacent to consolidated reef on the shoreline, two are open water hardbottom, and one is outside the STEER boundaries on a large reef tract.

A total of 26 hard coral species were documented, with 25 of those species recorded at sites inside STEER. *Porites astreoides* (mustard hill coral) was the most abundant species, followed by *Orbicella annularis* complex (boulder star

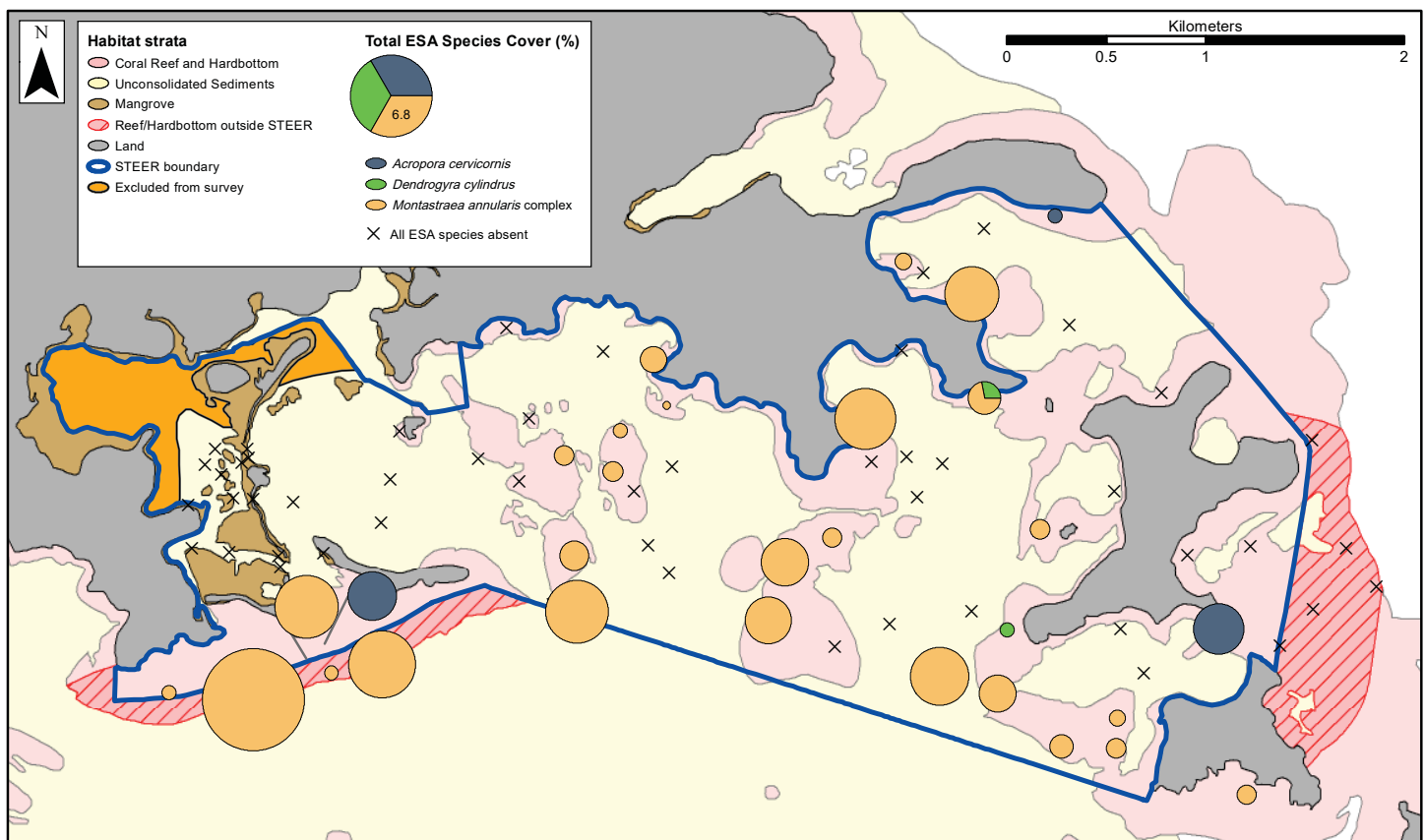


Figure 3.10. Percent cover of coral species listed as threatened under the Endangered Species Act (ESA) or proposed for future listing. The sizes of the pies are scaled by the total percent cover of the six species at that site.

coral), *Siderastrea radians* (lesser starlet coral), *Diploria strigosa* (symmetrical brain coral), *Siderastrea siderea* (massive starlet coral), and *P. porites* (finger coral) (Figure 3.9). Outside the STEER boundaries 19 species were recorded with the most abundant species being *M. annularis* complex followed by *P. astreoides*, *S. siderea*, *P. porites*, and *S. radians*.

Acropora cervicornis (staghorn coral), a species listed as threatened under the Endangered Species Act (ESA), was documented at three sites dispersed around the STEER and *Dendrogyra cylindrus* (pillar coral), a species of concern listed as threatened under the ESA, was recorded at two locations, both of which are on hardbottom close to the shoreline (Figure 3.10). One of the sites with *A. cervicornis* is located just inside the reserve on the reef tract bisected by the STEER boundary. This specific reef tract is also home to three sites out of the top four with the greatest overall hard coral cover (Figure 3.7).

The low coral cover observed in STEER reflects the record declines of coral reefs in the USVI and the Caribbean as a whole (Wilkinson, 2000; Catanzaro *et al.*, 2002; Jeffrey *et al.*, 2005; Rogers *et al.*, 2008). Rogers *et al.* (2008) reported that some reefs in the USVI had over 40% coral cover during the 1980s but was subsequently reduced

to 25% cover by the 1990s, with hurricane damage and disease cited as the main causative factors. Additional stressors contributing to decline in coral cover include sediment input from increased development (MacDonald *et al.*, 1997; Brooks *et al.*, 2007), associated nutrient enrichment (Fabricius, 2005) and land-based sources of pollution (Warne *et al.*, 2005). Additionally, climate change poses a broader regional threat to corals (Donner *et al.*, 2007). The mass bleaching event in 2005 followed by a coral disease outbreak (Bruno *et al.*, 2007) caused a 60% decline in coral cover in the USVI (Miller *et al.*, 2009).

The 2005 bleaching event was captured by multiple monitoring programs around the Caribbean. Each region in the USVI being monitored had low coral cover across reef types and years (Rothensberger *et al.*, 2008), with each island's reefs exhibiting the trend of decline (Rogers *et al.*, 2008). In St. Croix, weighted mean estimates of live coral decreased from 8.0% in February 2001 to 2.9% in October 2006, similar to reefs in St. John which were 8.4% in 2001 and declined to 4.5% by July of 2006 (Rogers *et al.*, 2008). Friedlander *et al.* (2013) reported that coral cover has continued to decline in St. John to less than 3% regardless of protective status as of 2009.

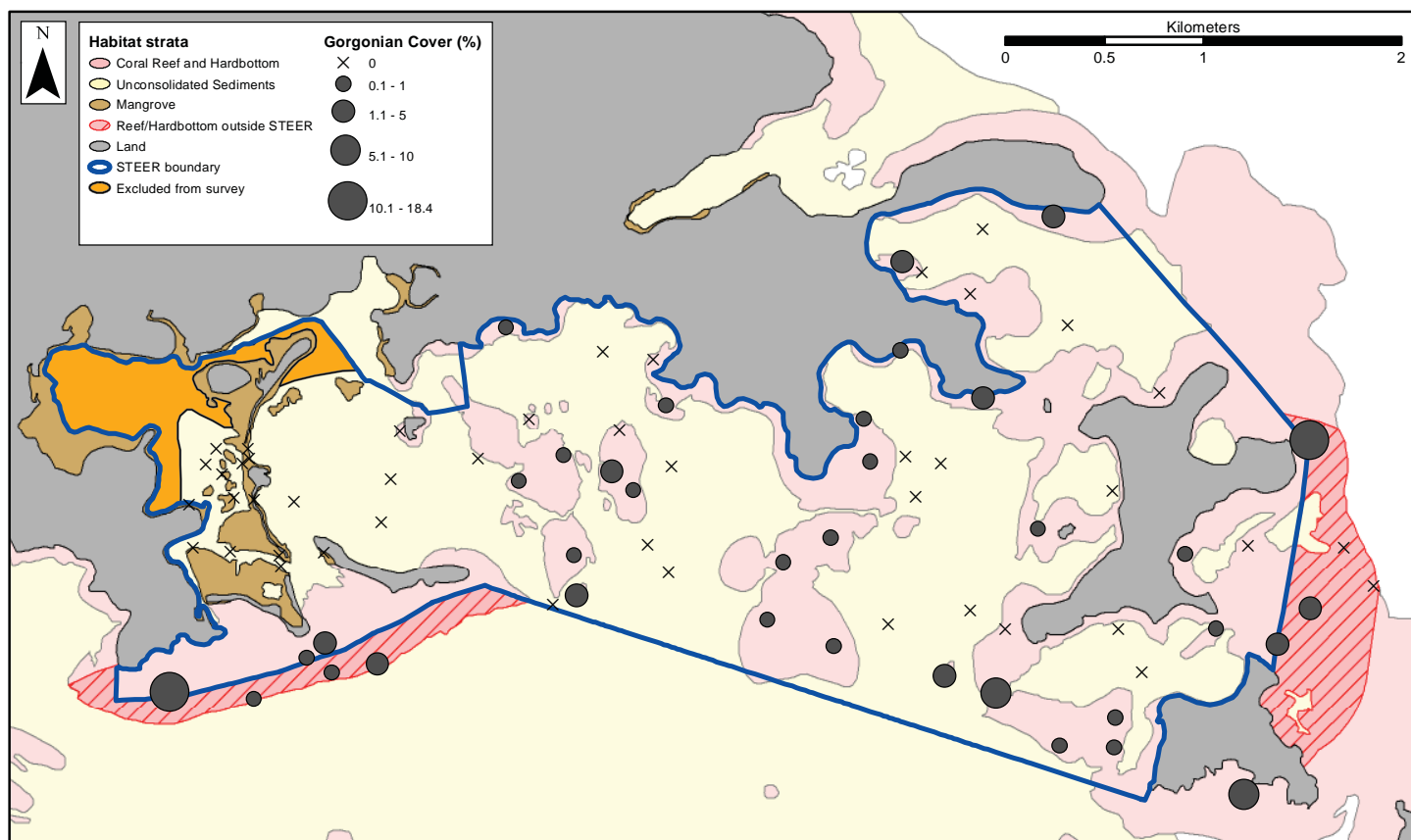


Figure 3.11. Percent gorgonian cover.

The Territorial Coral Reef Monitoring Program (TCRMP), which began in 2001, uses digital video transects to estimate benthic cover of hardbottom habitats at 33 long-term monitoring sites in the USVI, including 16 locations around St. Thomas (Smith *et al.*, 2011). This effort includes nearshore, midshelf, and outershelf reef systems in an effort to capture the diversity of reef types in the USVI. Mean live coral cover for all nearshore reefs in St. Thomas declined from 20.3% in 2002 to 9.3% in 2007 after the bleaching event and subsequent disease outbreak. This is the lowest average cover observed since the monitoring began in 2001 (unpublished data, Smith pers. comm). Since 2007, percent coral cover has made a comeback across all nearshore sites with a mean of 14.4% in 2013 (T. Smith, UVI, unpublished data).

Coral cover in STEER falls in line with values from similar studies and surveys conducted in St. Croix and St. John (Pittman *et al.*, 2008; Friedlander *et al.*, 2013), but were much lower than the data reported by TCRMP. For example, the only TCRMP site within STEER, at Cocus Rock, is reported to have over twice the coral cover than two randomly selected sites in the current study that were located within 300 m of the TCRMP site and at similar depths (11.5% TCRMP, 3.88% & 4.6% this study). Differences of overall coral cover across sites were most likely attributable to sample size within STEER, the focus of this

study in the shallow waters of STEER (35% of NOAA sites were <5m; 100% of TCRMP were >5m), differences in how sites were selected, and the distinct estimation methods. The higher overall values at TCRMP sites do call attention to other nearshore reefs around St. Thomas that should be considered for protection (Smith *et al.*, 2011). This may also reflect deeper reefs being less susceptible to bleaching (Sheppard, 2006; T. Smith, UVI, pers comm.) and subsequent disease outbreaks and therefore maintain higher coral cover overall.

Coral community structure inside and around STEER is similar to that of nearshore reefs around St. John (Friedlander *et al.*, 2013), St. Croix (Pittman *et al.*, 2008), and Puerto Rico (Bauer *et al.*, 2013). Our study sites also reflect the species composition documented by TCRMP in STEER and around St. Thomas (Smith *et al.*, 2011). The majority of reefs in the USVI have become dominated by *Montastraea*, *Porites*, *Siderastrea*, and *Diploria* genera (Jeffrey *et al.*, 2005; Herzlieb *et al.*, 2005; Rogers *et al.*, 2008), particularly after the decades long decimation of acroporids due to white band disease, hurricanes (Rogers *et al.*, 1982), and bleaching events (Miller *et al.*, 2006; Rogers *et al.*, 2008). ESA-listed species were infrequently observed in the 2012 field survey. Elkhorn coral (*Acropora palmata*), was documented at three sites in STEER during this study and ranged from 0.2% to 2.6 coverage. Stag-

horn coral (*A. cervicornis*) was not recorded in any survey quadrats, but was observed at one shallow site east of Great St. James Island.

Gorgonian Composition

Gorgonian cover ranged from 0-18.4% and primarily occurred on hardbottom habitat (Figure 3.11). Sites with the highest percent cover (7.44-18.4%) are all exposed to open ocean conditions. Each of the high cover sites are located in different quadrants of STEER: one is on the Long Point reef tract in the southwest corner, one is located outside the southeast corner of STEER Oceanside of Little St. James, and the other is outside the northeast corner of STEER on the eastern shoreline of St. James. This pattern is not surprising as all three of these sites are exposed to high wave action characteristic of open ocean and prevailing on-shore wind patterns, conditions in which gorgonian communities tend to thrive. Average gorgonian cover on hardbottom inside STEER was relatively low ($0.9 \pm 0.3\%$) and consisted of sea plumes/rods/whips ($0.49 \pm 0.14\%$), sea fans ($0.36 \pm 0.19\%$), and encrusting gorgonians (0.05 ± 0.02).

Sponge Composition

Sponge cover ranged from 0-64.6% and averaged $2.71 \pm 0.84\%$ across all sites (Figure 3.12). There was no distinctive spatial pattern exhibited by sponge communities—

sites with greater than average coverage are distributed on hardbottom habitats throughout STEER and outside the reserve. Inside STEER, mean percent cover differed greatly between hardbottom ($4.82 \pm 1.8\%$) and unconsolidated sediments ($0.25 \pm 0.11\%$) habitats. Barrel/tube/vase (BTV) sponges accounted for the majority of percent cover overall ($3.44 \pm 1.48\%$), while encrusting sponge comprised a smaller portion of the sponge community ($1.38 \pm 0.36\%$). On the nine hardbottom sites surveyed outside STEER, mean percent cover was similar to inside: overall sponge ($4.59 \pm 1.1\%$), BTV sponges ($2.72 \pm 0.82\%$), and encrusting sponge ($1.86 \pm 0.43\%$).

Algae and Seagrass Composition

Macroalgal cover ranged from 0-100% and was found throughout the STEER. Means were similar across bottom types: hardbottom ($14.77 \pm 2.44\%$), unconsolidated sediments ($17.99 \pm 5.14\%$), and mangrove sites ($13.94 \pm 5.1\%$, Figure 3.13). Percent cover across STEER averaged $15.71 \pm 2.21\%$. The two sites with the highest cover (90-100%) were located in soft bottom habitat near the Mangrove Lagoon. Turf algae ranged widely across STEER (0-91.56%) with an overall average cover of $27.53 \pm 3.57\%$ but was seen almost exclusively on hardbottom habitats ($49.78 \pm 4.11\%$, Figure 3.14). Cyanobacteria and filamentous algae (CB & FA) was documented at only 17 out of the 80 sites

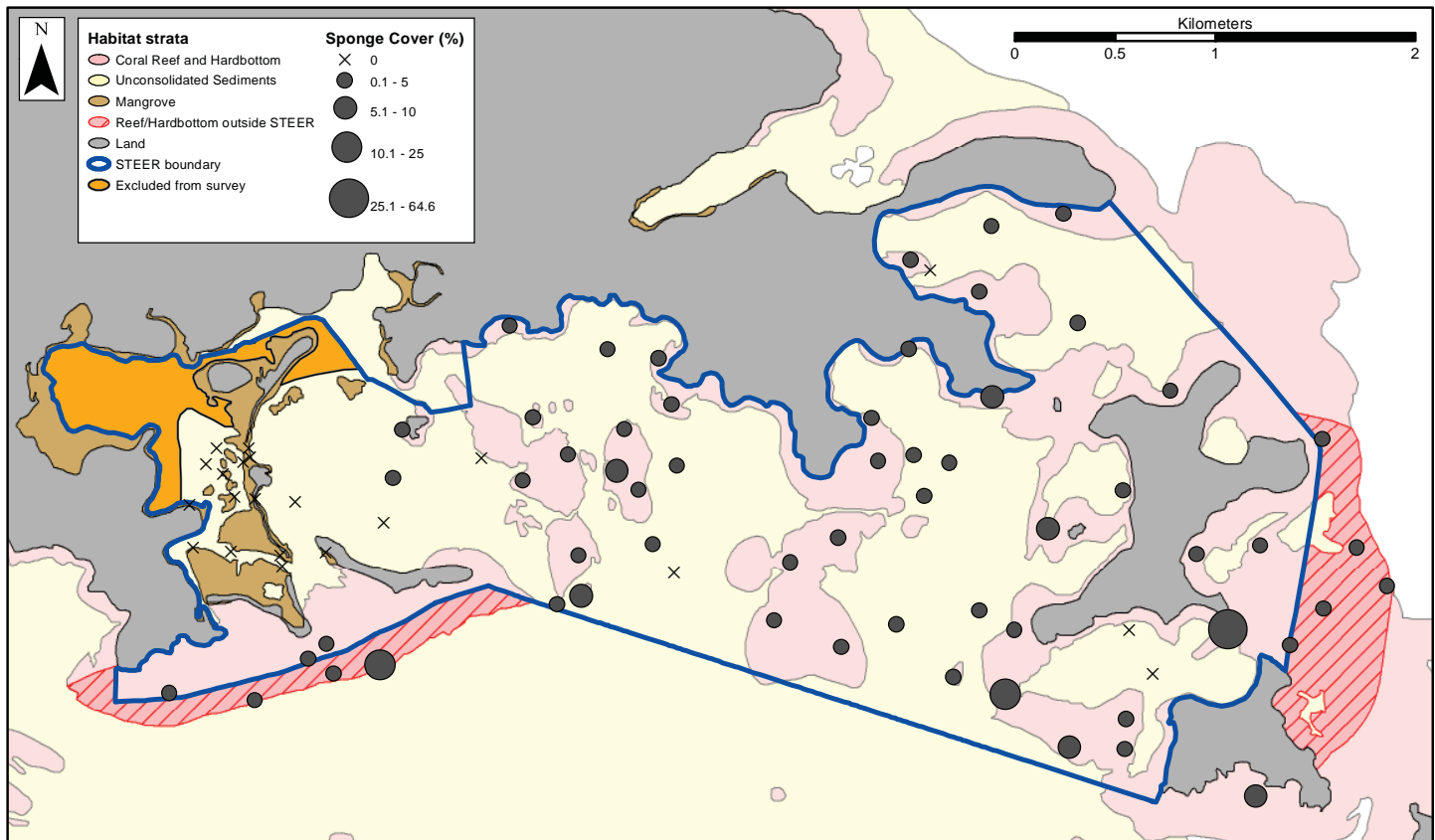


Figure 3.12. Percent sponge cover.

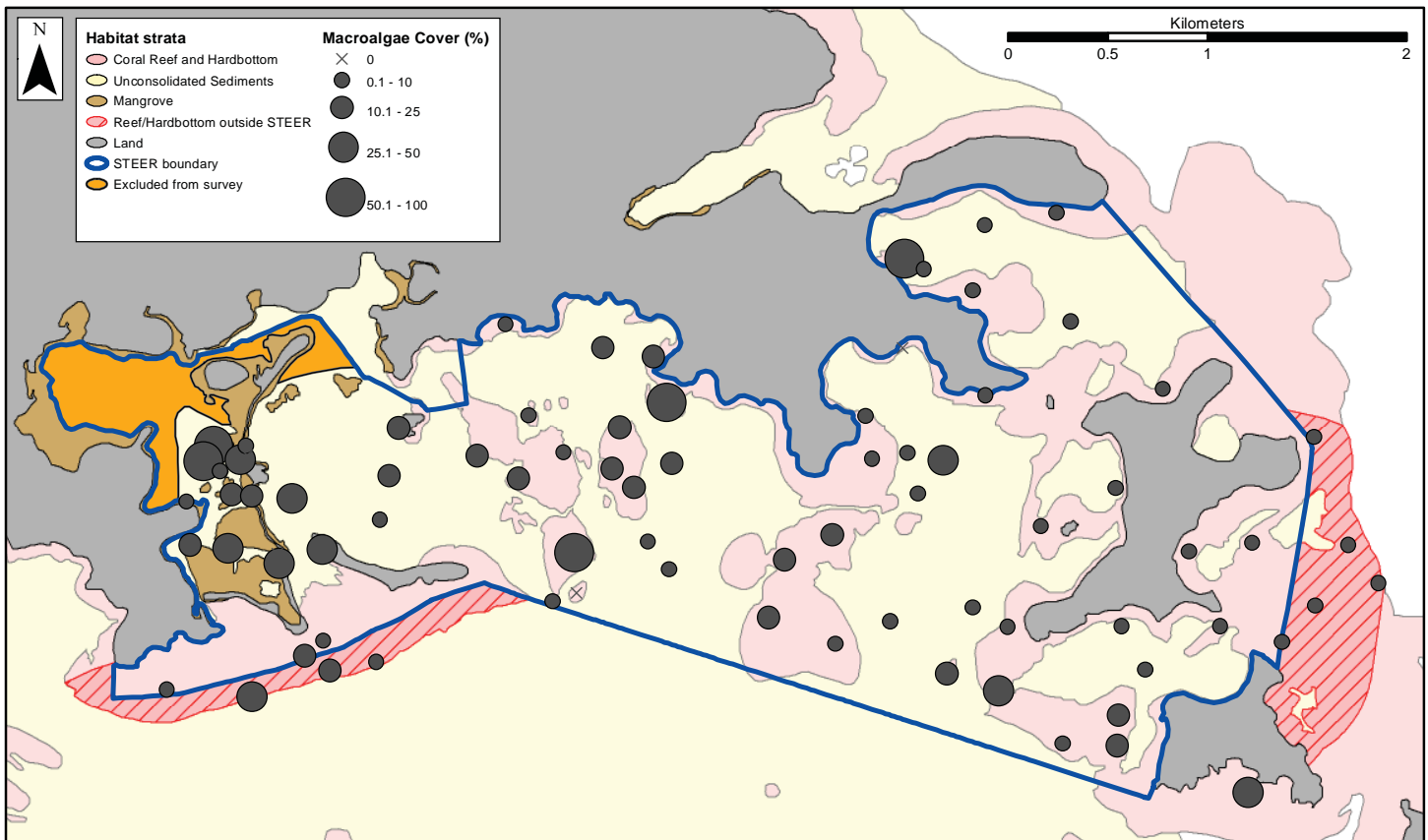


Figure 3.13. Percent macroalgae cover.

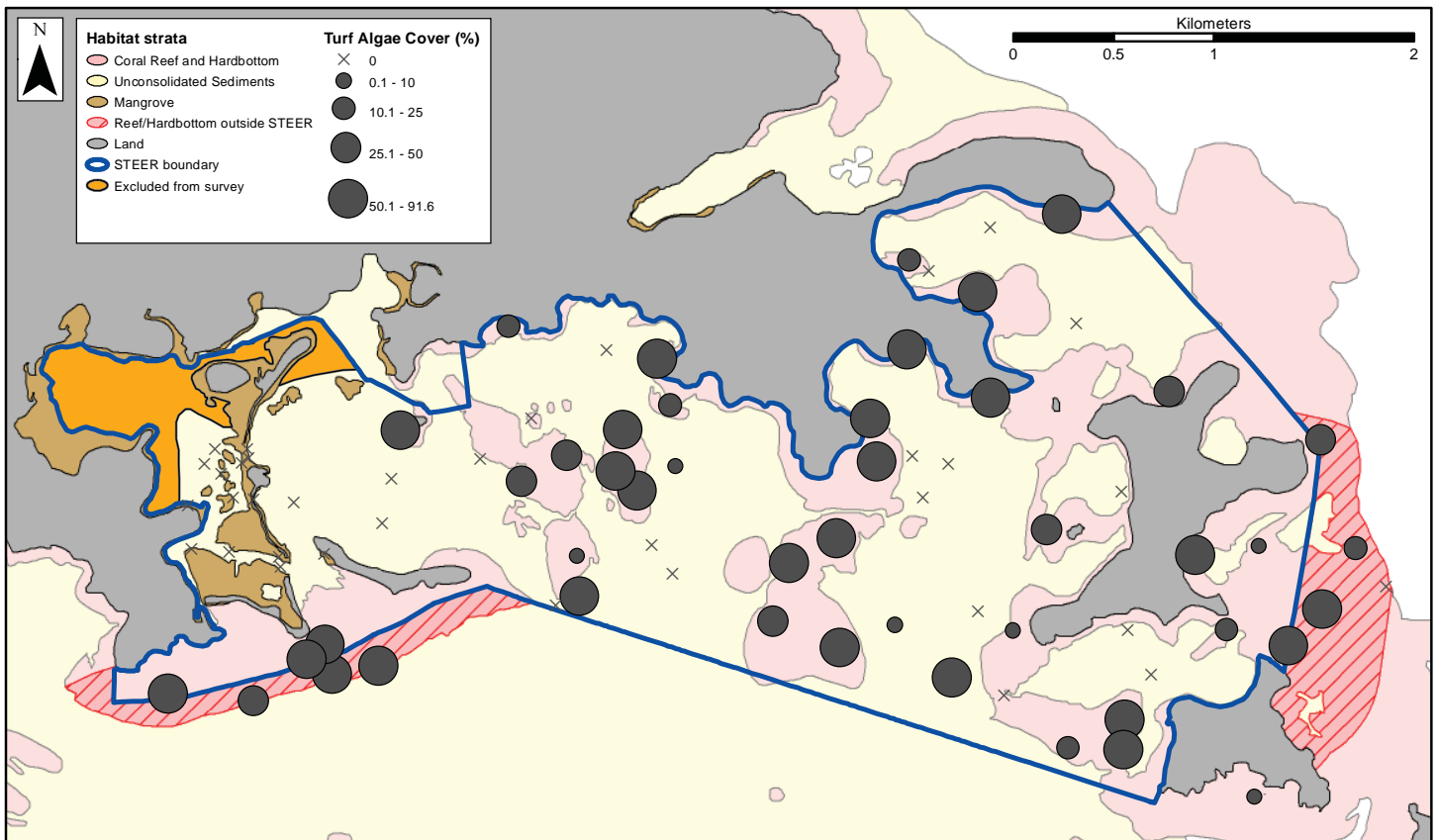


Figure 3.14. Percent turf algae cover.

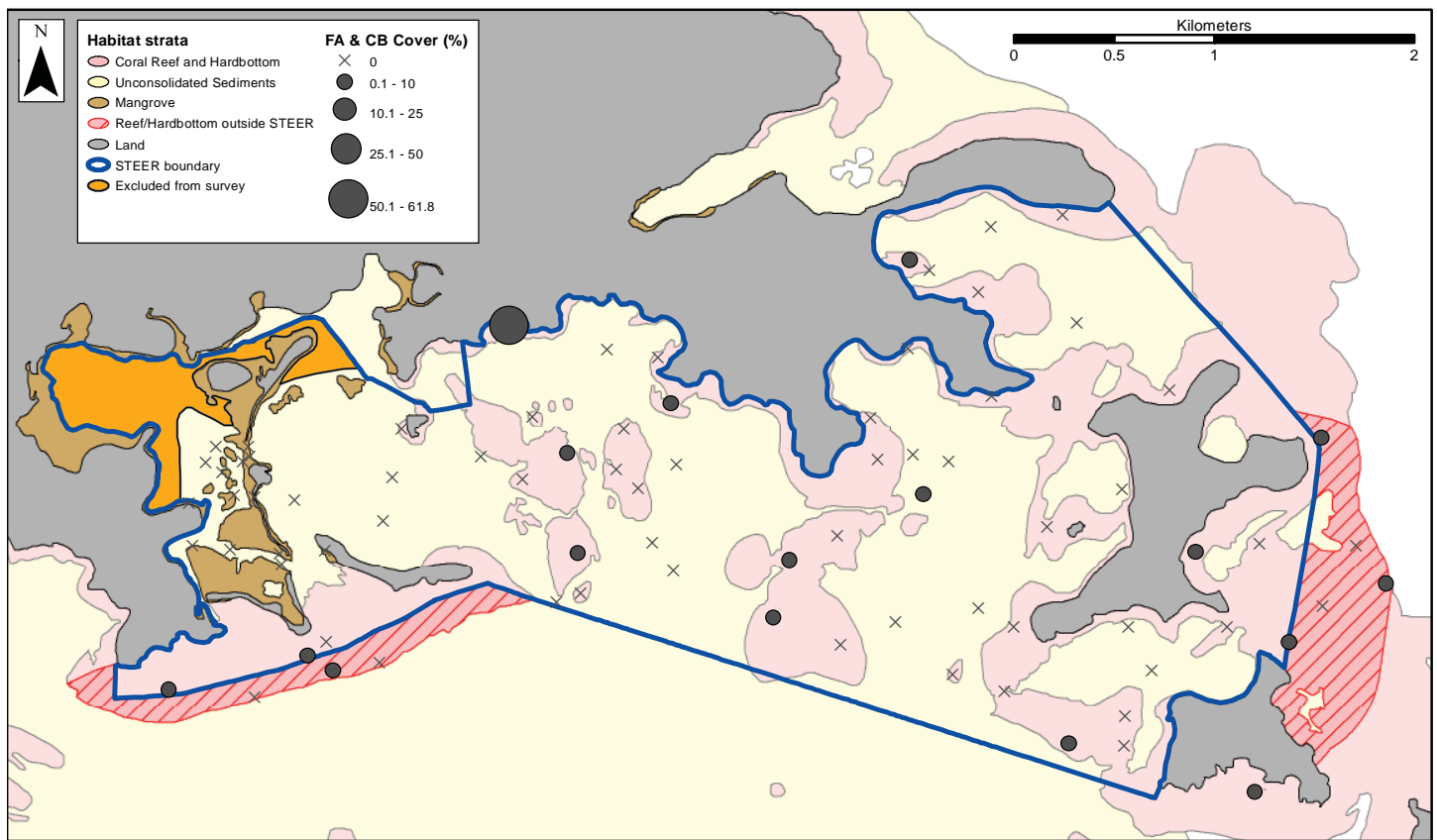


Figure 3.15. Percent filamentous algae and cyanobacteria (FA & CB) cover.

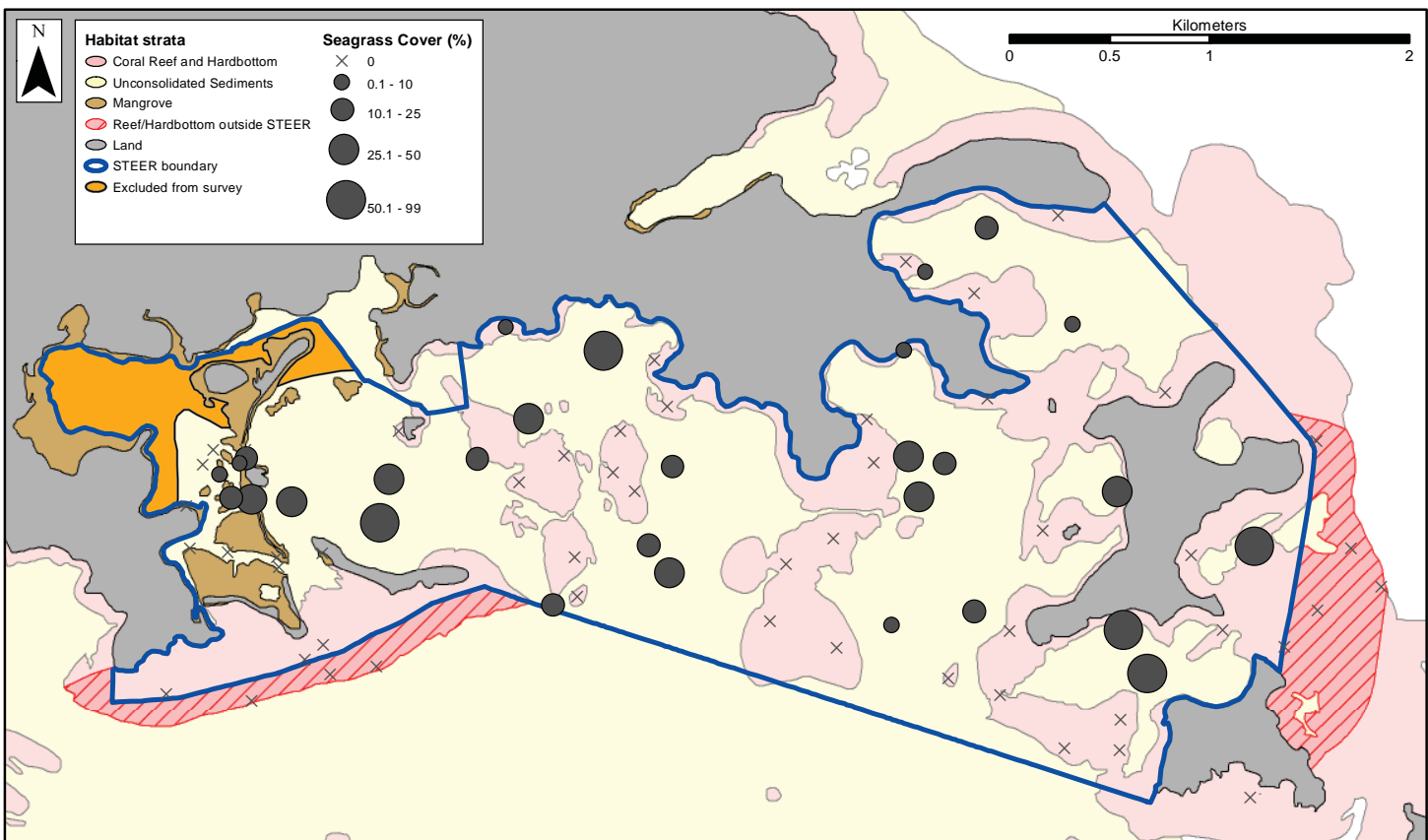


Figure 3.16. Percent seagrass cover.

surveyed with an overall average of $0.99 \pm 0.77\%$ found almost exclusively on hardbottom habitats (Figure 3.15).

Seagrass was observed on unconsolidated sediments and mangrove areas, as well as some sand gaps interspersed among hardbottom habitats. Percent cover varied from 0-99% (Figure 3.16). The absence of seagrass at only four of the 26 soft sediment sites is more informative than the pattern of seagrass presence: two sites devoid of seagrass border Mangrove lagoon and the other two sites are located in a channel through mangrove habitat that is near the Bovoni landfill. All four sites are documented as having only macroalgal cover: the two near the lagoon with macroalgal cover of 90% and 100%, the other two near the landfill had 32% and 46%. The majority of seagrass was recorded in unconsolidated sediment habitats: *Syringodium filiforme* ($18.14 \pm 4.76\%$) and *Thalassia testudinum* ($13.04 \pm 3.98\%$) were the most common species recorded with a very small amount of *Halodule wrightii* (0.21 ± 0.12 , Figure 3.17). All three species were found on unconsolidated sediments, *Halodule* and *Thalassia* were both seen in mangrove habitats, and a minor amount of *Thalassia* was documented at hardbottom habitats.

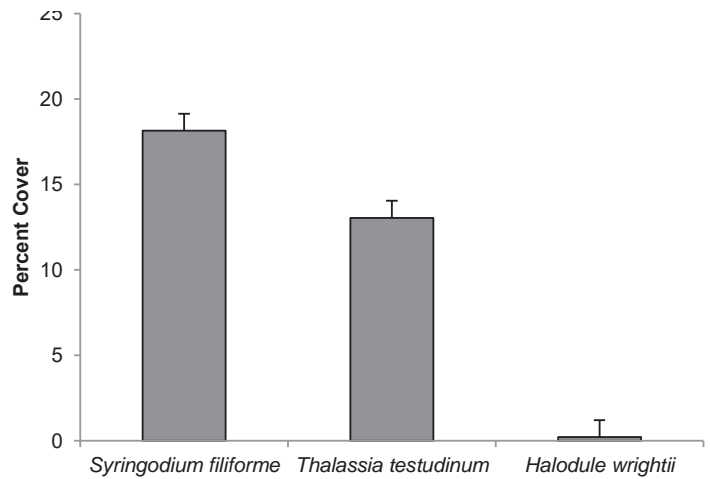


Figure 3.17. Percent cover of seagrass by species on unconsolidated sediments.

Macroinvertebrates

A total of seven spiny lobsters (*Panulirus argus*) were observed at three sites during the 80 surveys. Five lobsters were documented in a mangrove fringe near Mangrove Lagoon, one lobster was spotted outside the STEER along the reef tract located in the southwest region of the STEER, and one lobster was seen on hardbottom near Little St. James in the southeast corner of the reserve.

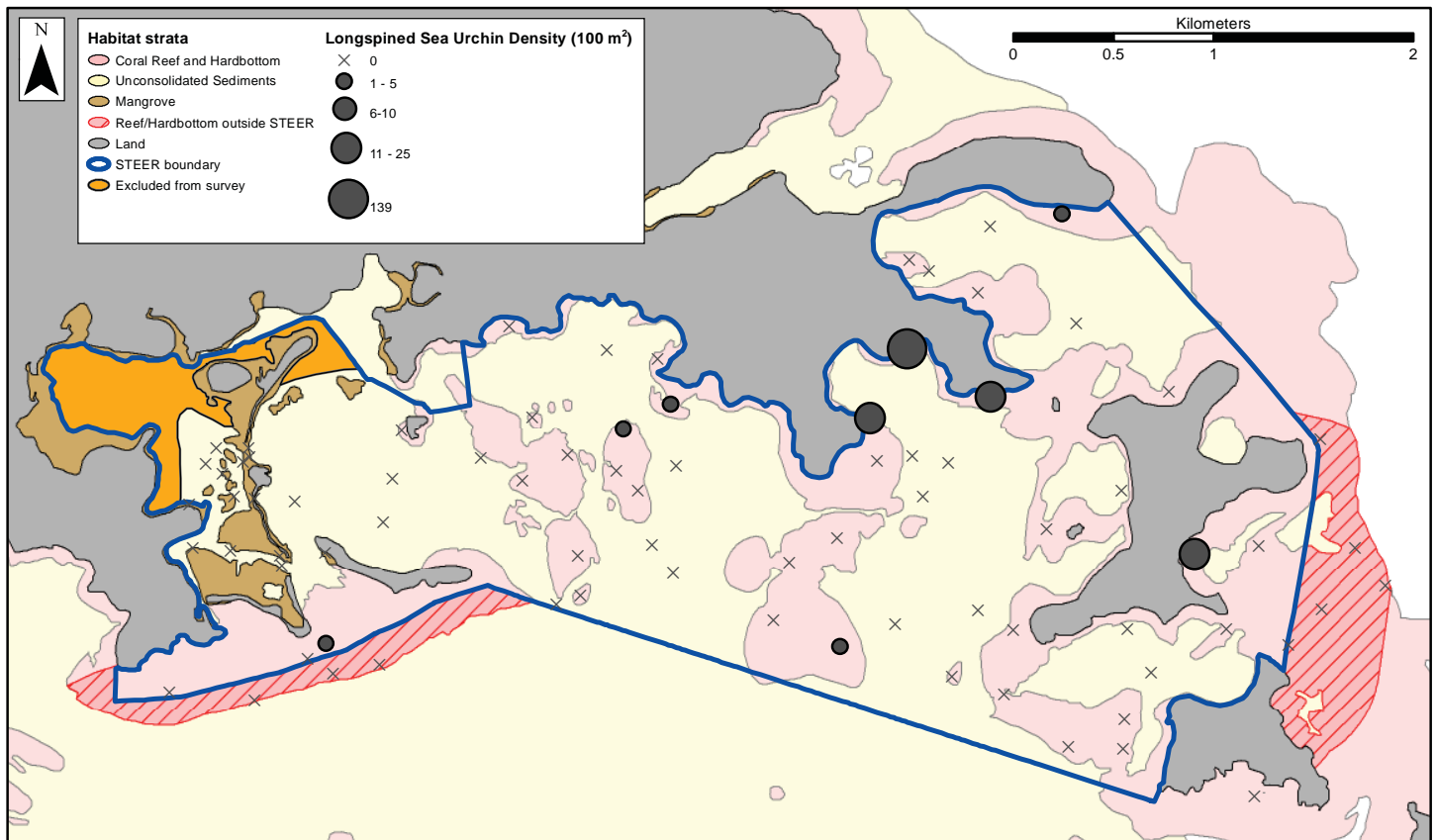


Figure 3.18. Density of longspined sea urchin (*Diadema antillarum*).

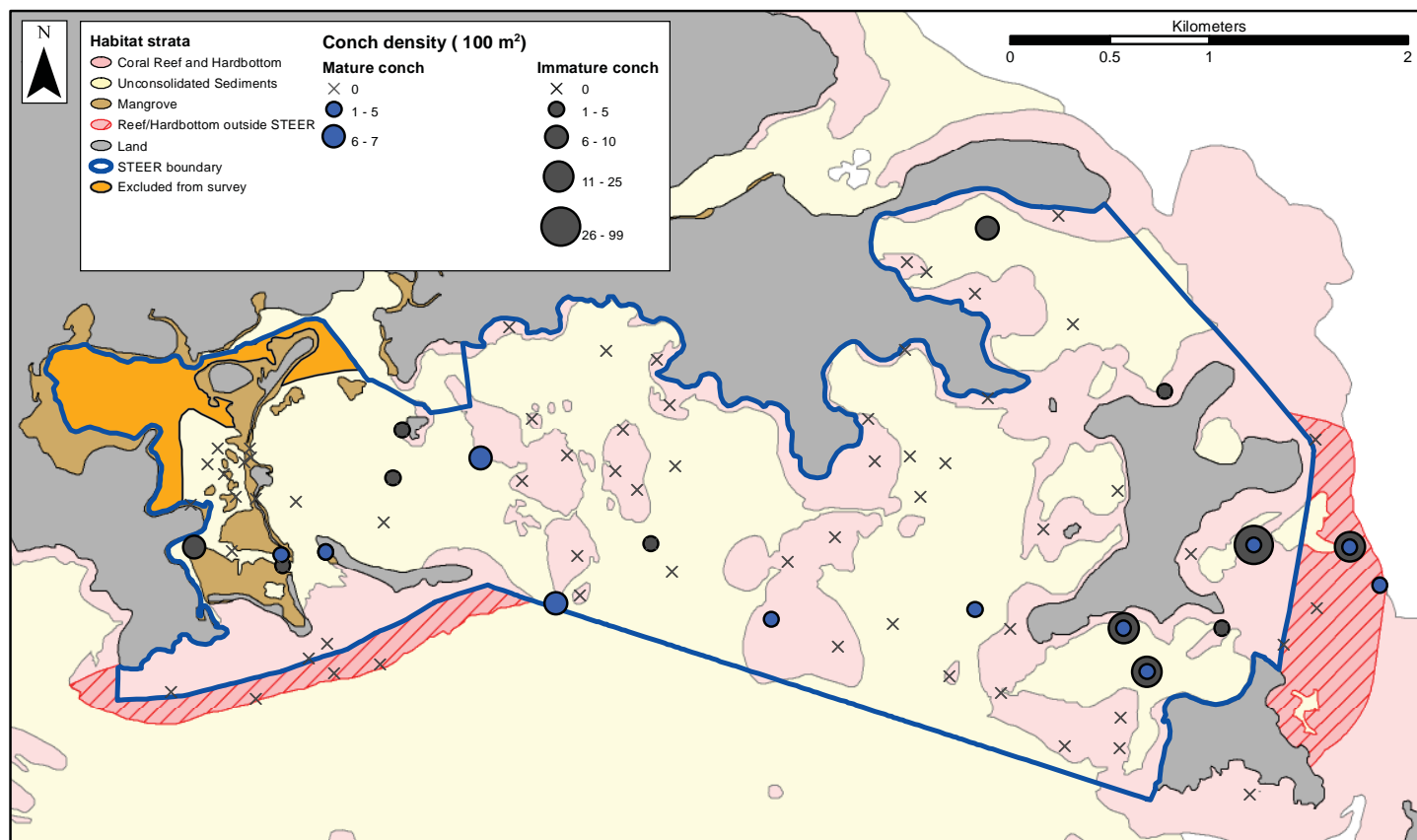


Figure 3.19. Density of mature and immature queen conch (*Lobatus gigas*).

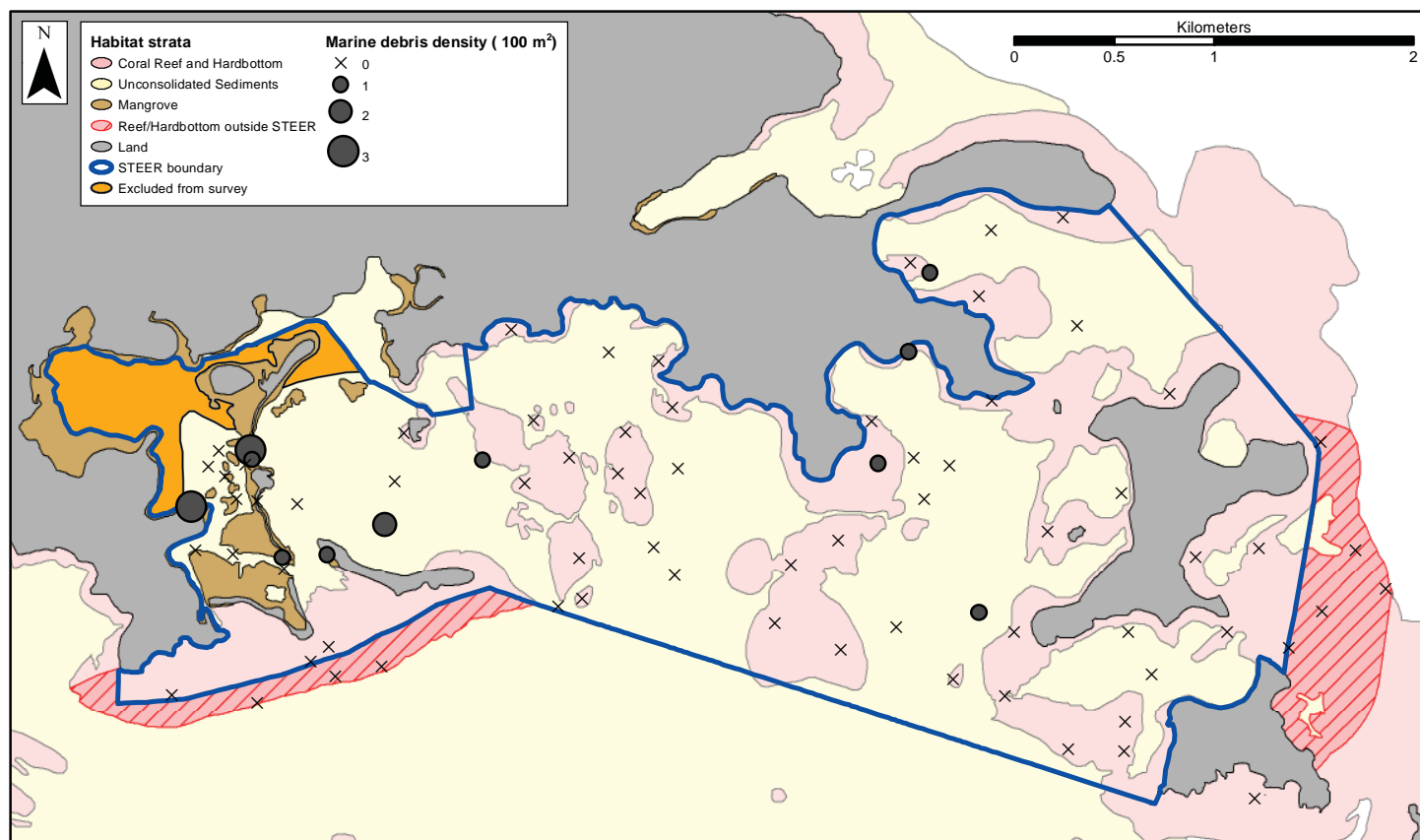


Figure 3.20. Density of total observed marine debris.

Long-spined urchin (*Diadema antillarum*) was reported at 9 sites, all hardbottom substrates, ranging from 2 to an impressive 139 individuals (Figure 3.18). The top three sites for urchin abundance making up 86.5% of the urchins documented (16-139 / 100m²) are all located on land adjacent sites in Cowpet Bay.

Immature queen conch (*Lobatus gigas*) were documented at 13 sites (7 unconsolidated sediments, 4 hardbottom, 2 mangrove, Figure 3.19) throughout the STEER and at one site outside the boundaries. Abundance ranged from 1-99 individuals and showed no distinct spatial pattern. Mature queen conch were observed in much lower numbers (0-7 individuals, Figure 3.19) at 11 sites (6 soft bottom, 3 hardbottom, 2 mangrove) dispersed around and outside the reserve. Mature conch were observed primarily at sites where immature conch were also present.

Marine Debris

Marine debris was detected at 16 sites in all three bottom types (Table 3.2). Debris items were found primarily close to shore or in the southern part of Benner Bay (Figure 3.20). Types of gear varied from minor fishing leader wire to general trash items such as bottles and plastic bags.

Fish

Community metrics

The fish community observed in the 2012 survey consisted of 36 taxonomic families and 125 species within STEER (Appendix 1 in Bauer *et al.*, 2014). An additional two families, represented by six species, were observed at the sites located in adjacent hardbottom areas outside STEER. Fish species richness ranged from 1 to 39 species per site (100 m²). Mean richness was highest on hardbottom, with similar levels inside and outside STEER, followed by mangrove (Figure 3.21b, Figure 3.22). The two surveys with 39 observed species were conducted on hardbottom near Cow and Calf Rocks and on reef outside STEER south of Patricia Cay, respectively. Shannon diversity, which is a product of richness and evenness, followed similar trends, with highest diversity on hardbottom and intermediate levels in mangrove (Figure 3.21a, Figure 3.23). Unconsolidated sediments were typified by lower species richness and diversity.

Mangrove sites exhibited the highest mean total fish density (Figure 3.21c, Figure 3.24), whereas mean levels of biomass were highest on hardbottom (Figure 3.21d, Figure 3.25). At many mangrove locations, the high density levels were largely due to the presence of schooling silversides and herring (Families Atherinidae and Clupeidae) and small juvenile grunts (Family Haemulidae). The hardbottom site

Table 3.2. Number and type of fouling organisms on marine debris items found during STEER transects.

Debris type	Total number	Colonized by
Trap float	1	Uncolonized
Fishing leader	1	Uncolonized
Wood	1	Sponge
Ladder	1	Macroalgae
Chain	1	Macroalgae
Glass bottle	5	Macroalgae, invertebrates
Paper	1	Macroalgae
Plastic bag	2	Uncolonized
Clothing	1	Sponge
Barrel	1	Uncolonized
Sunglasses	1	Uncolonized

with the highest density, located in Great Bay, was dominated by small gobies. Locations with both high density and biomass included sites on hardbottom adjacent to Cow and Calf Rocks, a patch reef southwest of Great St. James Island, a mangrove site near the false entrance to Mangrove Lagoon, and on the reef complex south of Patricia Cay, outside the STEER boundary. The survey with the third greatest biomass, located on a nearshore reef in Nazareth Bay, was characterized by the presence of several large-bodied parrotfish. Lowest density and biomass was typically observed on unconsolidated sediments, particularly at unvegetated sites.

Biomass and abundance were unevenly distributed among trophic groups (Figure 3.26). On all habitats, invertivores (I, e.g., grunts, butterfly fishes) and herbivores (H; e.g., parrotfish, damselfish) were the most numerically abundant, while piscivores (P; e.g., snappers, groupers) constituted a smaller percentage. Planktivores (PL; e.g., herring) accounted for over 20% of the abundance in mangroves, but due to their small size only 1% of the biomass. Conversely, piscivores accounted for higher proportional biomass across all habitats, particularly on unconsolidated sediments where the trophic group accounted for three-quarters of the observed biomass, largely due to the presence of several jacks and occasional barracuda.

Highest mean density and biomass of piscivores occurred in mangrove surveys, although biomass was more equitable across bottom types. Piscivores were most frequent in Mangrove Lagoon and adjacent reef south of Patricia and Cas Cay, east of Cow and Calf Rocks and in the central portion of Jersey Bay. They were notably absent from the western portion of Benner Bay and most hardbottom surveys east of Great St. James, including outside STEER boundaries (Figure 3.27).

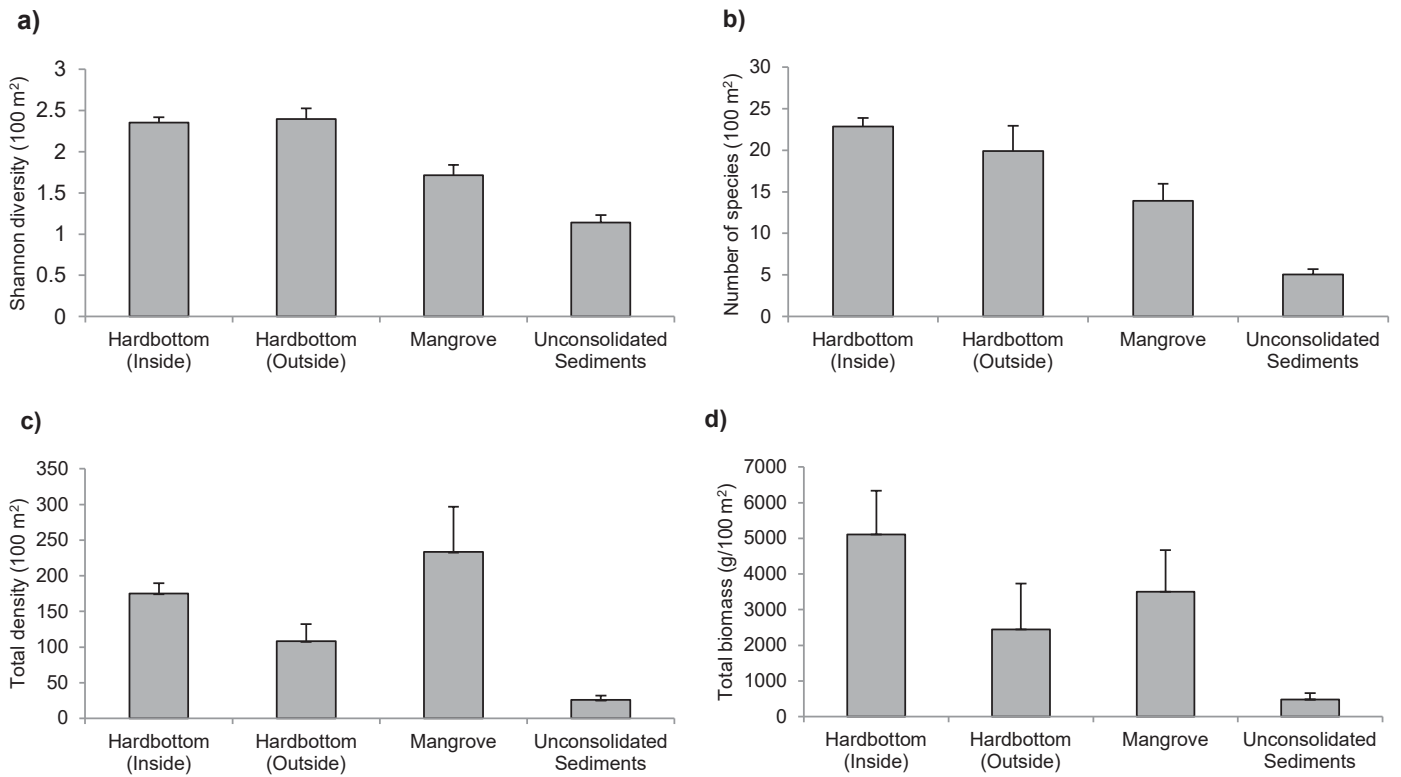


Figure 3.21. Mean (\pm SE) fish species a) Shannon diversity, b) richness, c) density, and d) biomass by habitat type.

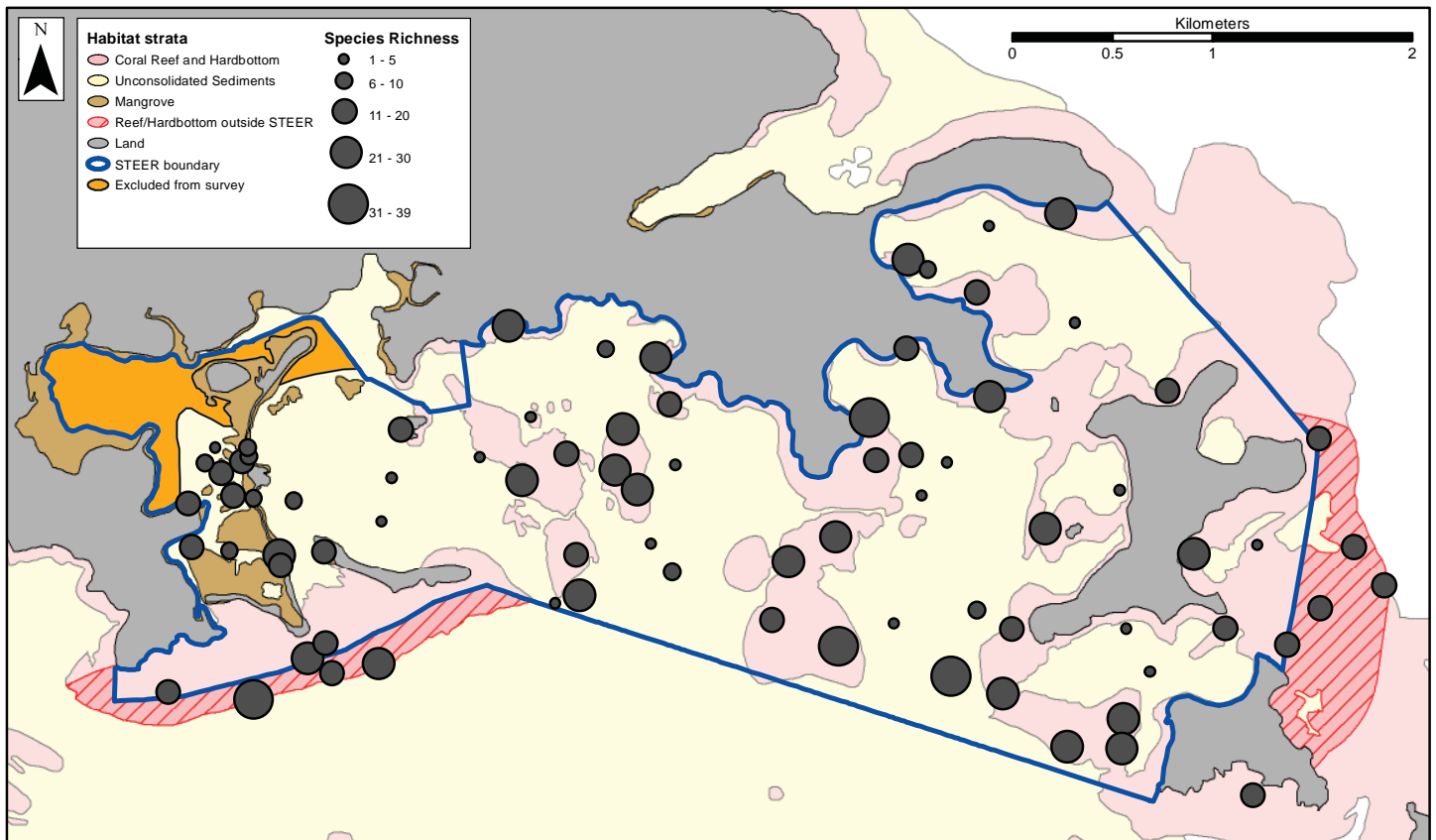


Figure 3.22. Fish species richness.

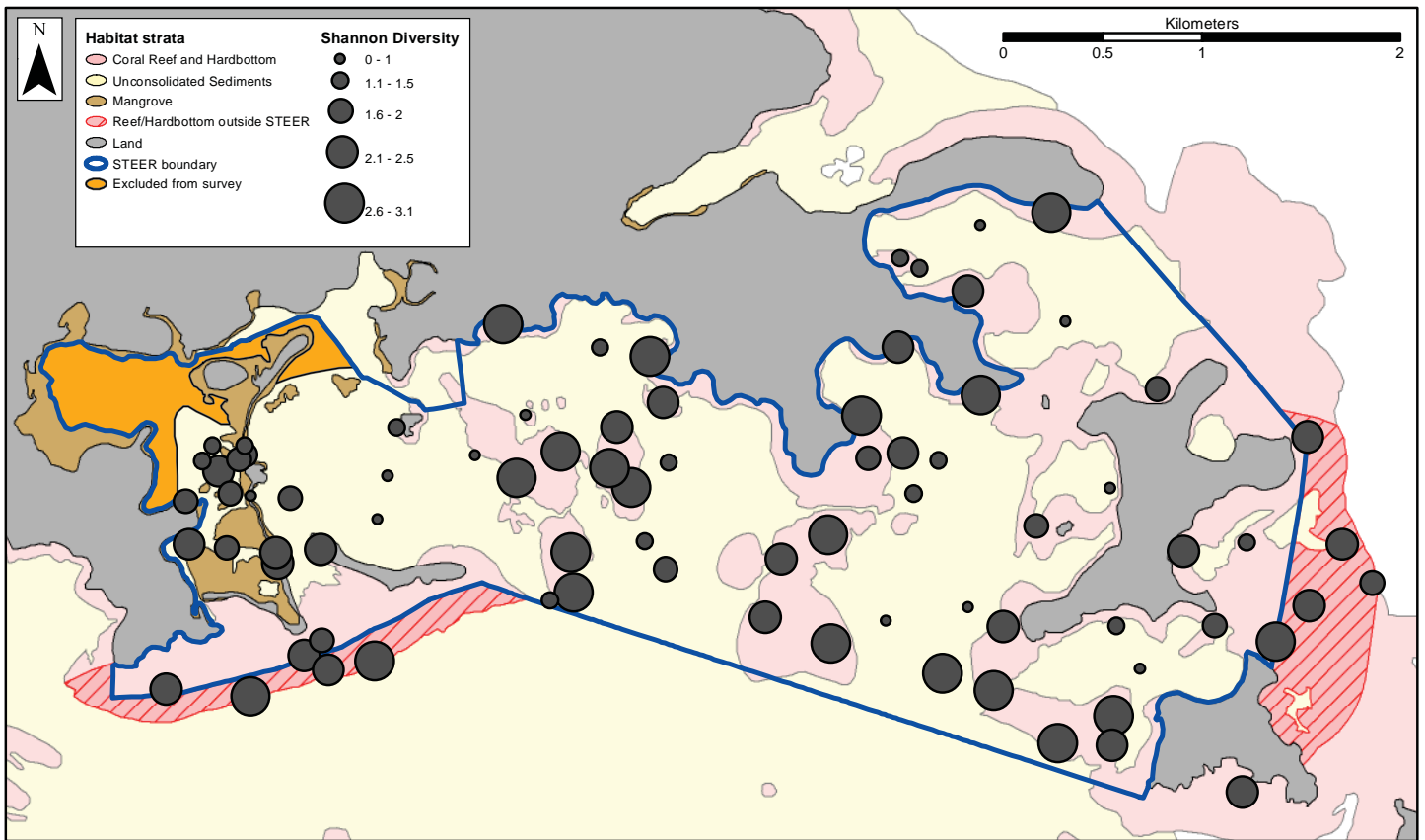


Figure 3.23. Fish species diversity.

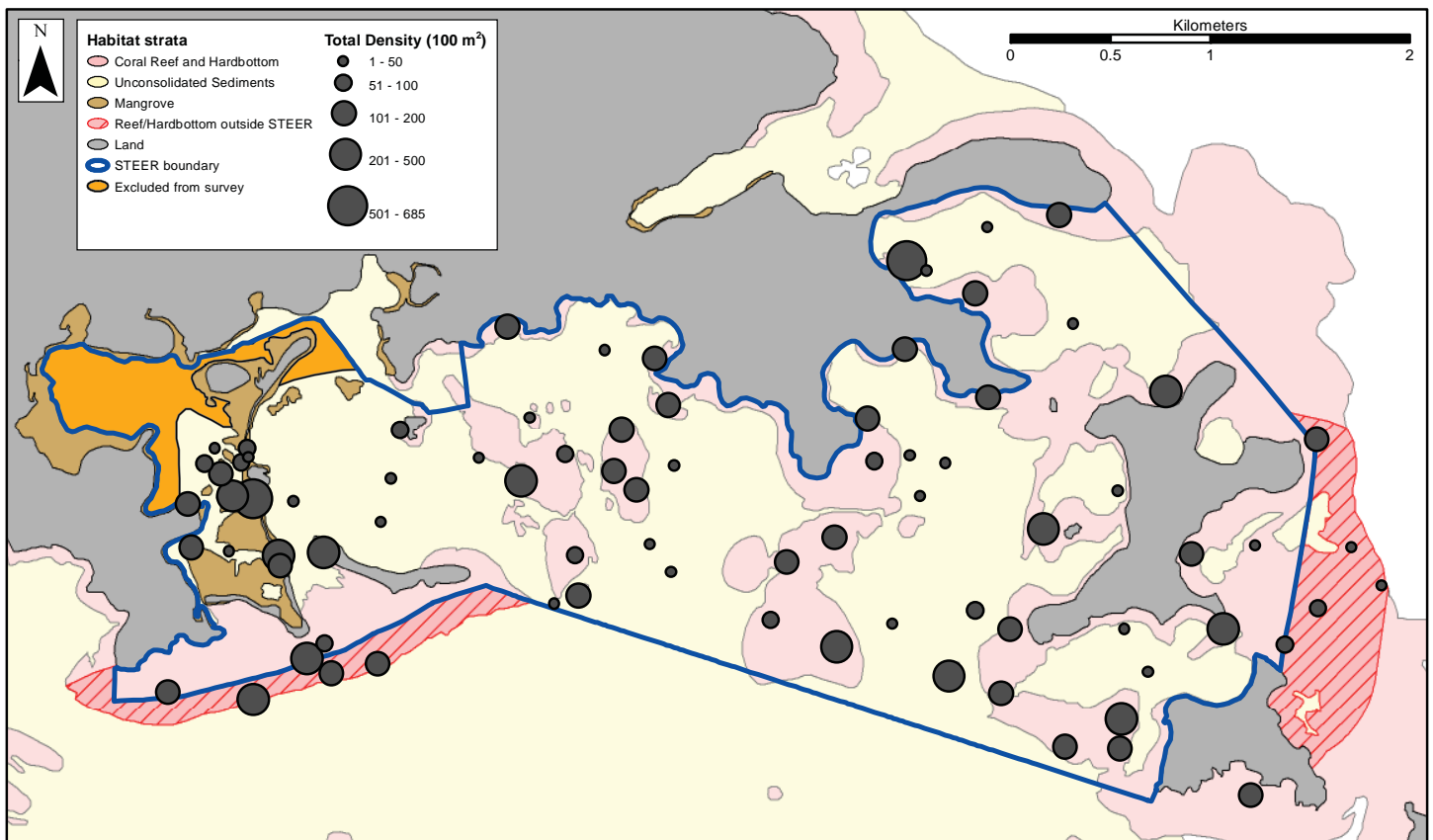


Figure 3.24. Total fish density.

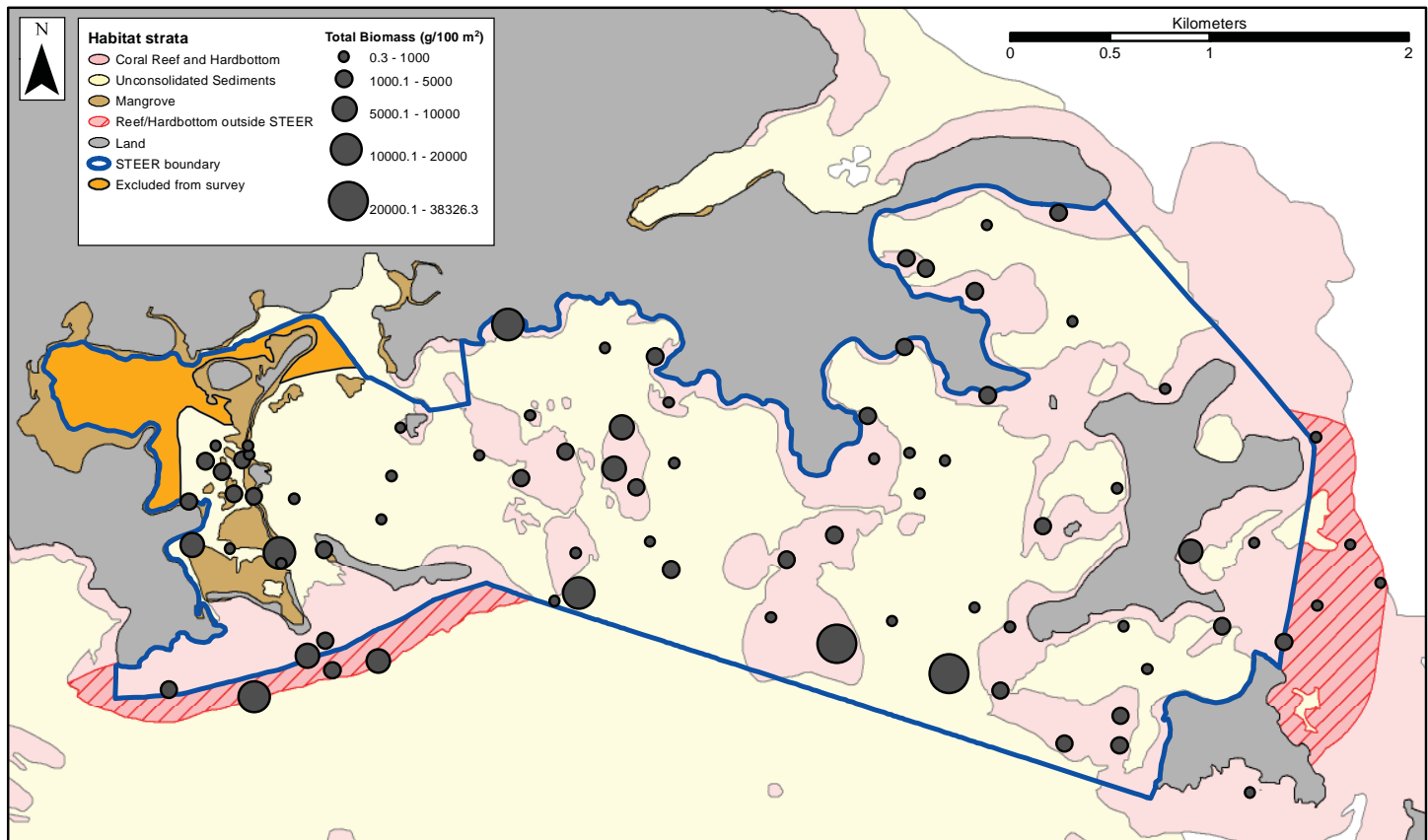


Figure 3.25. Total fish biomass.

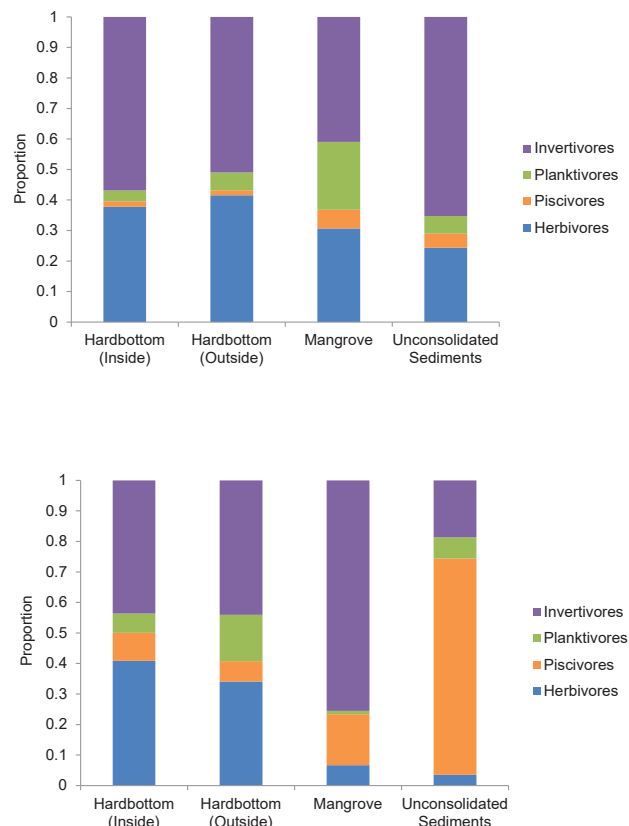


Figure 3.26. Proportional a) abundance and b) biomass of trophic groups across habitat types.

Family and species composition differed across bottom types (Table 3.3, Table 3.4). Fishes of the Family Labridae (wrasses) accounted for over a quarter of the total number of fish on hardbottom, followed by Scaridae (parrotfish), Pomacentridae (Damselfish), Gobiidae (gobies) and Acanthuridae (surgeonfishes). The most abundant species were members of these families (Table 3.3), with labrid species comprising two of the top five most abundant species on hardbottom both inside and outside STEER (*Thalassoma bifasciatum* and *Halichoeres bivittatus/garnoti*). Scaridae accounted for the highest proportion of biomass on hardbottom within STEER at 21%, followed by Acanthuridae, Haemulidae (grunts), and Lutjanidae (snappers). Two surgeonfish species, ocean surgeonfish (*Acanthurus bahianus*) and blue tang (*A. coeruleus*), ranked first and third in total biomass on hardbottom within STEER. Two snapper species, gray snapper (*Lutjanus griseus*) and yellowtail snapper (*Ocyurus chrysurus*), were also in the top five. While density proportions were similar in hardbottom surveys outside STEER, proportional biomass differed slightly, with snappers comprising 26% of the observed biomass. Yellowtail snapper accounted for the most biomass on hardbottom outside STEER, with another snapper species, lane snapper (*L. synagris*), also within the top five. The Family Serranidae (seabasses and groupers), comprised a small percent of the biomass on hardbottom both inside and

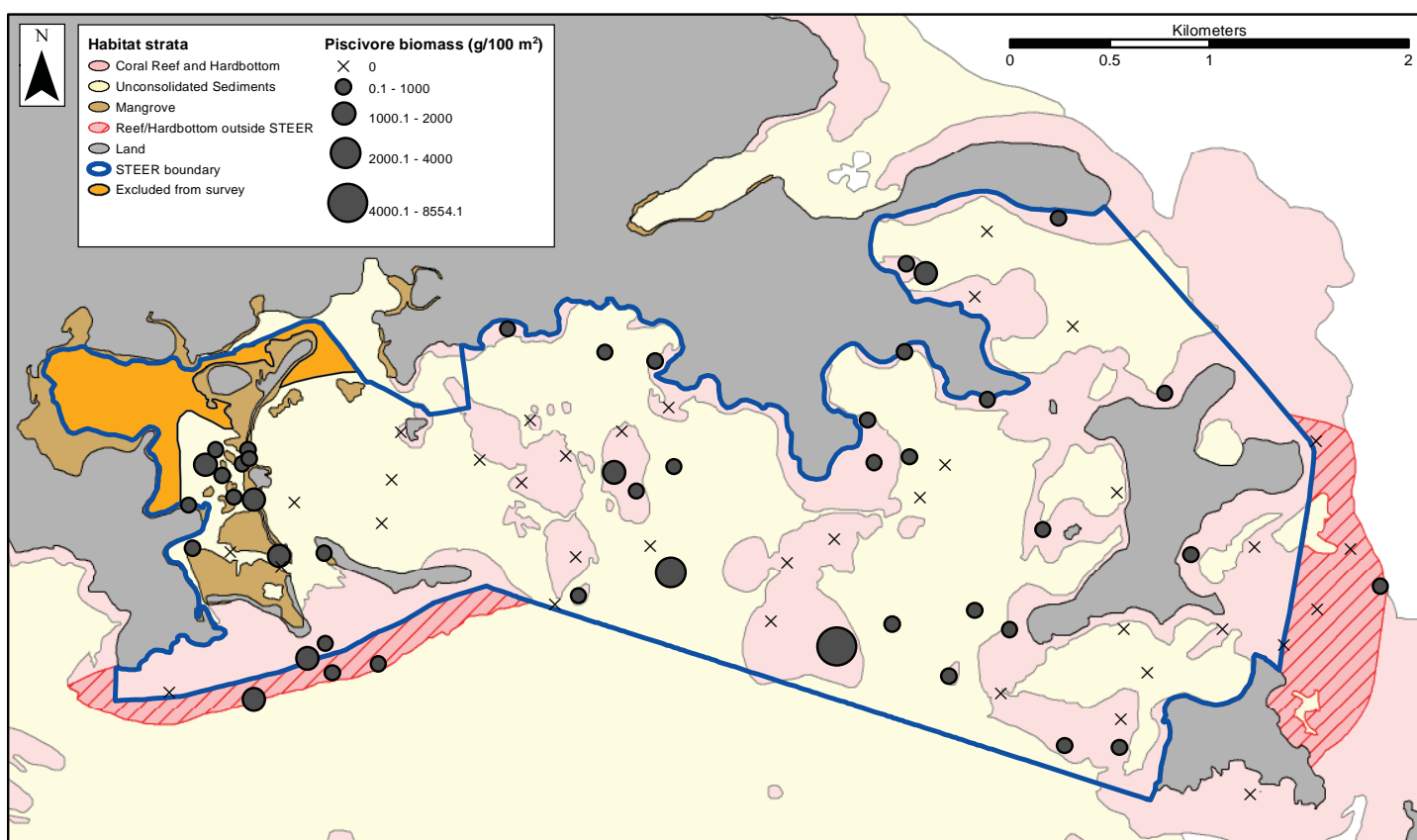


Figure 3.27. Piscivore biomass.

outside STEER (4.4% and 7.6%, respectively). Although the invasive lionfish, *Pterois volitans*, did not occur in any survey transects, one individual was anecdotally sighted near Christmas Cove.

In the mangrove habitat, over 45% of the total density was comprised of small bodied fishes of the families Atherinidae (*Atherinomorus* sp.) and Clupeidae (*Jenkinsia* sp.). The Family Haemulidae was the third most abundant, with juvenile unidentified grunts (*Haemulon* sp.), bluestriped grunt (*H. sciurus*), French grunt (*H. flavolineatum*) accounting for the remaining species within the top five. In contrast, snappers, which were the fifth most abundant family, accounted for over half of the biomass surveyed in mangroves, with gray snapper comprising nearly 40% of the total biomass. Another snapper species, schoolmaster (*L. apodus*), and two grunt species (*H. sciurus* and *H. flavolineatum*), were in the top five for biomass, as was the nurse shark *Ginglymostoma cirratum*.

On unconsolidated sediments, Labridae accounted for 37.5% of the total density, but only 7% of the biomass. The two most abundant species were the labrids slippery dick (*Halichoeres bivittatus*) and rosy razorfish (*Xyrichtys martinicensis*). Larger bodied jacks (Family Carangidae) and

barracuda (Family Sphyraenidae) accounted for 36.6% and 28.6% of biomass on unconsolidated sediments, respectively. Species within these two families accounted for the top three in proportional biomass.

Six species that were not observed in surveys within STEER were documented at hardbottom sites outside STEER. These included the redspotted hawkfish (*Amblycirrhitus pinos*), trumpetfish (*Aulostomus maculatus*), glasseye snapper (*Heteropriacanthus cruentatus*), hogfish (*Lachnolaimus maximus*), yellowmouth grouper (*Mycteroperca interstitialis*), and longjaw squirrelfish (*Neoniphon marianus*).

The nMDS and ANOSIM analyses further indicate that fish assemblages in STEER differ by bottom type. There was a clear separation of fish communities, based on fish density data, between hardbottom, unconsolidated sediment, and mangrove surveys (Figure 3.28a). Mangrove and hardbottom sites tended to be clustered within habitat type (Figure 3.28a), indicating a high degree of similarity in species composition among sites. In contrast, unconsolidated sediment sites tended to be more dispersed, indicating more dissimilarity among sites within this group. While sites within this bottom type were generally characterized

Table 3.3. Top five families in abundance and total biomass, shown as percent of total, by habitat strata.

Hardbottom (Inside)			
Family	Density	Family	Biomass
Labridae	27.0%	Scaridae	21.0%
Scaridae	15.8%	Acanthuridae	18.3%
Pomacentridae	15.3%	Haemulidae	18.0%
Gobiidae	13.8%	Lutjanidae	17.7%
Acanthuridae	13.5%	Serranidae	4.4%
Hardbottom (Outside)			
Family	Density	Family	Biomass
Labridae	27.8%	Lutjanidae	26.1%
Scaridae	20.7%	Scaridae	22.8%
Pomacentridae	15.9%	Pomacanthidae	8.6%
Acanthuridae	9.5%	Acanthuridae	8.3%
Gobiidae	9.1%	Serranidae	7.6%
Mangrove			
Family	Density	Family	Biomass
Atherinidae	23.1%	Lutjanidae	51.5%
Haemulidae	22.3%	Haemulidae	21.7%
Clupeidae	21.2%	Ginglymostomatidae	8.2%
Lutjanidae	13.1%	Scaridae	4.4%
Gerreidae	8.2%	Sphyraenidae	3.7%
Unconsolidated Sediments			
Family	Density	Family	Biomass
Labridae	37.5%	Carangidae	36.8%
Scaridae	22.2%	Sphyraenidae	28.6%
Gerreidae	10.3%	Labridae	7.4%
Lutjanidae	7.8%	Echeneidae	6.2%
Haemulidae	7.7%	Scombridae	5.2%

by low overall abundance, they often varied in their species composition. Within coral reef and hardbottom, there was not distinct separation of sites located inside versus outside the STEER. The results of the ANOSIM test also indicate that there was a statistically significant difference in community composition among the three bottom types, and that the groups were well-separated (Global $R = 0.757$, $p < 0.001$). Pairwise comparisons indicated high dissimilarity between hardbottom and unconsolidated sediments and between hardbottom and mangrove (Table 3.5). The top four species that contributed to dissimilarity between hardbottom and mangrove strata, as determined by the SIMPER analysis, included small herring (*Jenkinsia sp.*), flagfin mojara (*Eucinostomus melanopterus*), and schoolmaster (*Lutjanus apodus*), which were more common in mangroves, and bluehead wrasse (*T. bifasciatum*), and ocean surgeonfish (*A. bahianus*), which were more common on hardbottom. The top five species contributing to the

Table 3.4. Top five species in abundance and total biomass, shown as percent of total, by habitat strata.

Hardbottom (Inside)			
Species	Density	Species	Biomass
<i>Thalassoma bifasciatum</i>	15.9%	<i>Acanthurus bahianus</i>	8.2%
<i>Coryphopterus personatus/hyalinus</i>	12.5%	<i>Haemulon flavolineatum</i>	7.6%
<i>Acanthurus bahianus</i>	7.9%	<i>Acanthurus coeruleus</i>	6.0%
<i>Halichoeres bivittatus</i>	6.3%	<i>Lutjanus griseus</i>	5.9%
<i>Scarus iseri</i>	5.7%	<i>Ocyurus chrysurus</i>	5.2%
Hardbottom (Outside)			
Species	Density	Species	Biomass
<i>Thalassoma bifasciatum</i>	13.3%	<i>Ocyurus chrysurus</i>	14.0%
<i>Stegastes partitus</i>	9.3%	<i>Sparisoma viride</i>	11.7%
<i>Halichoeres garnoti</i>	6.9%	<i>Lutjanus synagris</i>	9.2%
<i>Coryphopterus personatus/hyalinus</i>	6.8%	<i>Pomacanthus arcuatus</i>	5.9%
<i>Sparisoma aurofrenatum</i>	6.5%	<i>Mulloidichthys martinicus</i>	5.4%
Mangrove			
Species	Density	Species	Biomass
<i>Atherinomorus species</i>	23.1%	<i>Lutjanus griseus</i>	39.3%
<i>Jenkinsia species</i>	21.2%	<i>Haemulon sciurus</i>	13.6%
<i>Haemulon species</i>	8.0%	<i>Ginglymostoma cirratum</i>	8.2%
<i>Haemulon sciurus</i>	7.1%	<i>Lutjanus apodus</i>	7.5%
<i>Haemulon flavolineatum</i>	6.8%	<i>Haemulon flavolineatum</i>	4.3%
Unconsolidated Sediments			
Species	Density	Species	Biomass
<i>Xyrichtys martinicensis</i>	18.1%	<i>Sphyraena barracuda</i>	28.6%
<i>Halichoeres bivittatus</i>	16.2%	<i>Caranx crysos</i>	24.1%
<i>Gerres cinereus</i>	7.5%	<i>Carangoides bartholomaei</i>	10.2%
<i>Sparisoma radians</i>	6.7%	<i>Echeneis naucrates</i>	6.2%
<i>Ocyurus chrysurus</i>	4.6%	<i>Scomberomorus regalis</i>	5.2%

dissimilarity between hardbottom and unconsolidated sediments strata included wrasse, surgeonfish, parrotfish, and damselfish species that were all more abundant on hardbottom than unconsolidated sediments (Table 3.5).

One of the hardbottom surveys outside STEER showed similarity with the surveys on unconsolidated sediments

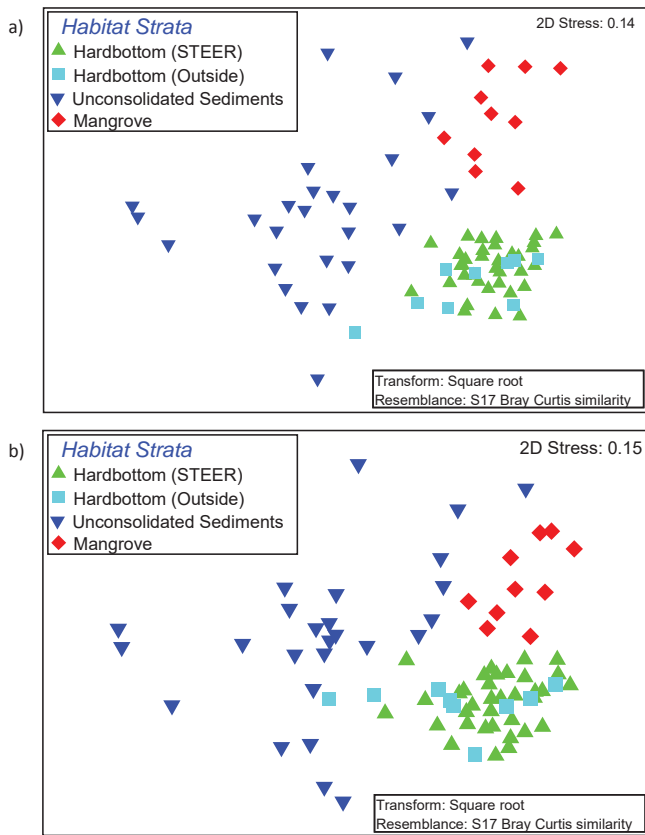


Figure 3.28. Non-metric multidimensional (nMDS) scaling ordination based on between site similarity composition using species a) density and b) biomass data. Sites are color-coded by habitat type and study area.

by its location on the nMDS plot. This site, located east of Great St. James Island, was characterized by rubble habitat and low overall abundance. The R statistic for the pair of mangrove and unconsolidated sediments was lower indicating while the two groups were still clearly different, there was some overlap in species composition. The top species contributing to the dissimilarity between the two strata were all more abundant on mangrove and included small herring (*Jenkinsia sp.*), schoolmaster (*L. apodus*), gray snapper (*L. griseus*), bluestriped grunt (*H. sciurus*), and flagfin mojarra (*E. melanopterus*). Using species biomass resulted in a similar MDS configuration (Figure 3.28b) and ANOSIM results as for species density, albeit with a slightly lower Global R ($R = 0.661$, $p < 0.001$). Pairwise comparisons showed hardbottom and unconsolidated sediments were well separated (Table 3.5). The results of the SIMPER analysis indicated that the top species contributing to the dissimilarity between these two strata included yellowtail snapper and two species of surgeonfish and parrotfish. The R value for the hardbottom-mangrove pairwise comparison was slightly lower than with the abundance data ($R = 0.662$) indicating a slightly higher degree of similarity but still overall clearly different in terms of species biomass assemblage. Similar to the ANOSIM analysis using density data, mangrove and unconsolidated sediments showed less separation than the other paired habitats.

Table 3.5. Pairwise Analysis of Similarities (ANOSIM) comparisons between habitat types based on fish species density and biomass data, and list of top five species contributing to the dissimilarity from the SIMPER analysis. Species are listed in decreasing order of percent contribution to the average dissimilarity. The R statistic ranges between 0 and 1 and represents whether pairs of habitats are well separated (closer to 1) or barely separable (closer to 0). Species are listed in decreasing order of percent contribution to the average dissimilarity.

Group	Species Density			Species Biomass		
	R	p-value	Top species (% contribution to dissimilarity)	R	p-value	Top species (% contribution to dissimilarity)
Hardbottom, Mangrove	0.894	0.001	<i>Jenkinsia sp.</i> (6.02%)	0.662	0.001	<i>Lutjanus griseus</i> (9.96%)
			<i>Thalassoma bifasciatum</i> (5.44%)			<i>Haemulon sciurus</i> (5.89%)
			<i>Eucinostomus melanopterus</i> (4.15%)			<i>Lutjanus apodus</i> (5.18%)
			<i>Acanthurus bahianus</i> (3.92%)			<i>Acanthurus bahianus</i> (5.03%)
			<i>Lutjanus apodus</i> (3.92%)			<i>Acanthurus coeruleus</i> (3.88%)
Hardbottom, Unconsolidated Sediments	0.772	0.001	<i>Thalassoma bifasciatum</i> (8.42%)	0.724	0.001	<i>Acanthurus bahianus</i> (8.07%)
			<i>Acanthurus bahianus</i> (5.88%)			<i>Acanthurus coeruleus</i> (5.73%)
			<i>Halichoeres bivittatus</i> (4.51%)			<i>Sparisoma aurofrenatum</i> (4.88%)
			<i>Sparisoma aurofrenatum</i> (4.25%)			<i>Ocyurus chrysurus</i> (3.86%)
			<i>Stegastes partitus</i> (4.17%)			<i>Sparisoma viride</i> (3.46%)
Unconsolidated Sediments, Mangrove	0.382	0.001	<i>Jenkinsia sp.</i> (9.55%)	0.327	0.001	<i>Lutjanus griseus</i> (16.93%)
			<i>Lutjanus apodus</i> (7.61%)			<i>Haemulon sciurus</i> (10.06%)
			<i>Eucinostomus melanopterus</i> (7.45%)			<i>Lutjanus apodus</i> (9.81%)
			<i>Haemulon sciurus</i> (6.57%)			<i>Sphyræna barracuda</i> (6.80%)
			<i>Lutjanus griseus</i> (6.25%)			<i>Haemulon sp.</i> (4.99%)

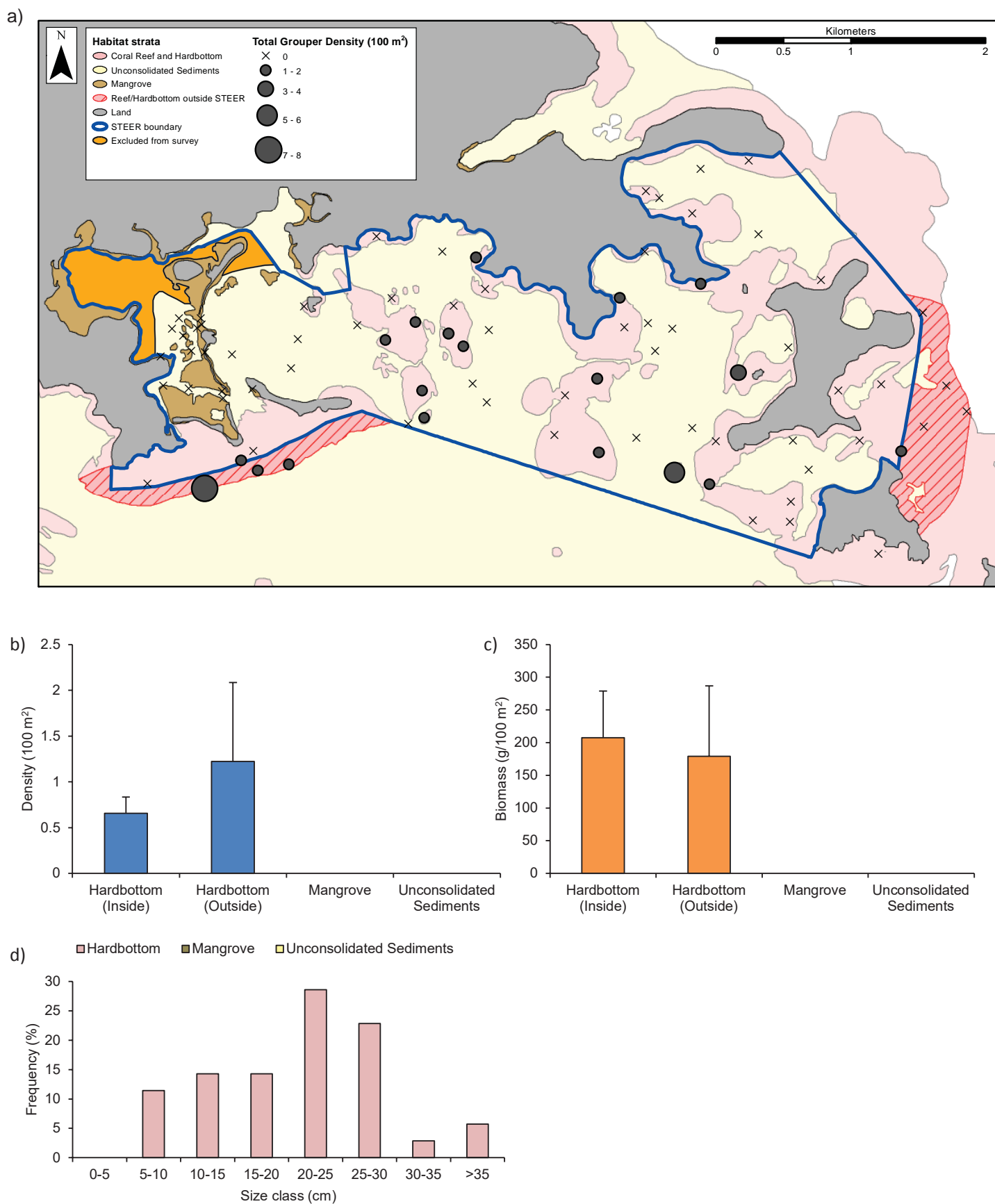


Figure 3.29. Grouper (Family Serranidae) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency.

Select Families and Species

Grouper (*Serranidae*)

Grouper (*Cephalopholis* and *Epinephelus spp.*), rarely seen, were primarily small in size, and were exclusively associated with hardbottom (Figure 3.29). Most grouper individuals belonged to two species: graysby (*Cephalopholis cruentata*), and red hind (*Epinephelus guttatus*). Notably, only one coney (*Cephalopholis fulva*), typically a common grouper species in the Caribbean, was observed. Species in the larger-bodied genus *Mycteroperca* were largely absent from the survey, with the exception of one yellowmouth grouper (*Mycteroperca interstitialis*) documented at a site outside STEER on the reef complex south of Patricia Cay. Red hind was the most abundant grouper species observed in the study, occurring in 13% of surveys within STEER. The species was also sighted in three surveys outside of STEER, east of Great St. James Island and south of Patricia Cay/Mangrove Lagoon (Figure 3.30). The species was found exclusively on hardbottom, including patch reefs, nearshore rocky areas, and high aggregate reef. The site with the highest density was a patch reef southwest of Great St. James Island. The majority of observed individuals were small adults, although two 40 cm fish were also seen (Figure 3.30d).

Graysby showed a similar distribution pattern and habitat affiliation as red hind. The species occurred in 11% of transects within STEER and in two surveys outside STEER. Mean density was higher, but more variable, on hardbottom outside STEER compared to inside, primarily due to the presence of six individuals at one site (Figure 3.31). The majority of individuals were small adults 15-30 cm in length (Figure 3.31d).

Snappers (*Lutjanidae*)

Snappers were detected across the shelf in all investigated habitats but were most abundant in mangroves (Figure 3.32). However, as described below, distribution varied by species and lifestage. The lowest mean density and biomass was observed over unconsolidated sediments. A total of seven Lutjanid species were documented, with schoolmaster (*Lutjanus apodus*), yellowtail (*Ocyurus chrysurus*), and gray snappers (*L. griseus*) accounting for the majority of observations. The remaining species, mahogany (*L. mahogoni*), lane (*L. synagris*), dog (*L. jocu*) and mutton (*L. analis*) were less frequently sighted (Appendix 1 in Bauer *et al.*, 2014). Lutjanidae size frequency was skewed toward smaller size classes (Figure 3.32d).

Gray snapper were observed in 23% of survey transects within STEER and were almost exclusively associated with mangrove fringes and cays (Figure 3.33). However,

the second highest observed density, and second highest biomass, was observed on a patch reef in St. James Bay. The site with the highest density and biomass was located near the false entrance to Mangrove Lagoon. About 70% of observed individuals were juveniles/subadults, while the remaining were mostly small adults (Figure 3.33d). Two individuals >35 cm were observed in Mangrove Lagoon. Schoolmaster were observed at 24% of sites within STEER and also at two additional reef sites outside the reserve. Similar to gray snapper, the species was most abundant in nearshore mangrove fringes and cays (Figure 3.34). A few individuals were observed on hardbottom habitat across the study area, while none were observed on unconsolidated sediments. The majority of schoolmaster (>90%) were juveniles/subadults (Figure 3.34d). All of the adult-sized individuals, about 9% of the total, were located on hardbottom.

Yellowtail snapper was the most frequently sighted Lutjanid species, occurring in over 50% of survey transects within STEER. Mean density of yellowtail snapper was similar in both mangrove and hardbottom habitats, with lower levels on unconsolidated sediments (Figure 3.35). The sites with the highest abundance and biomass were mostly located in the western half of the study area, and were less frequently sighted in the east (Figure 3.35). Highest biomass levels were observed on reefs south and east of Patricia and Cas Cays. The majority (~70%) of individuals were juveniles/subadults, particularly those associated with unconsolidated sediments and mangrove (Figure 3.35d). The majority of observed adults were located on hardbottom, leading to higher mean biomass compared to the other bottom types. Smaller sized adults were most common, with no individuals >35 cm length observed.

Mahogany snapper were not commonly observed, occurring at 7% of sites within STEER, but notably larger densities were present in survey transects at Cow and Calf Rocks (20 /100 m²) and a nearby patch reef (10 /100 m²) (Figure 3.36). This species was absent from unconsolidated sediments and from hardbottom surveys outside the reserve boundary. Adult-sized individuals were only observed on hardbottom.

Grunts (*Haemulidae*)

Fishes in the grunt family were present in all habitat types. Mangrove habitat had the highest mean density, while hardbottom and unconsolidated sediment habitats exhibited similar density levels (Figure 3.37). Mean biomass was highly variable on hardbottom habitat due to a large concentration of biomass near Cow and Calf Rocks (>23

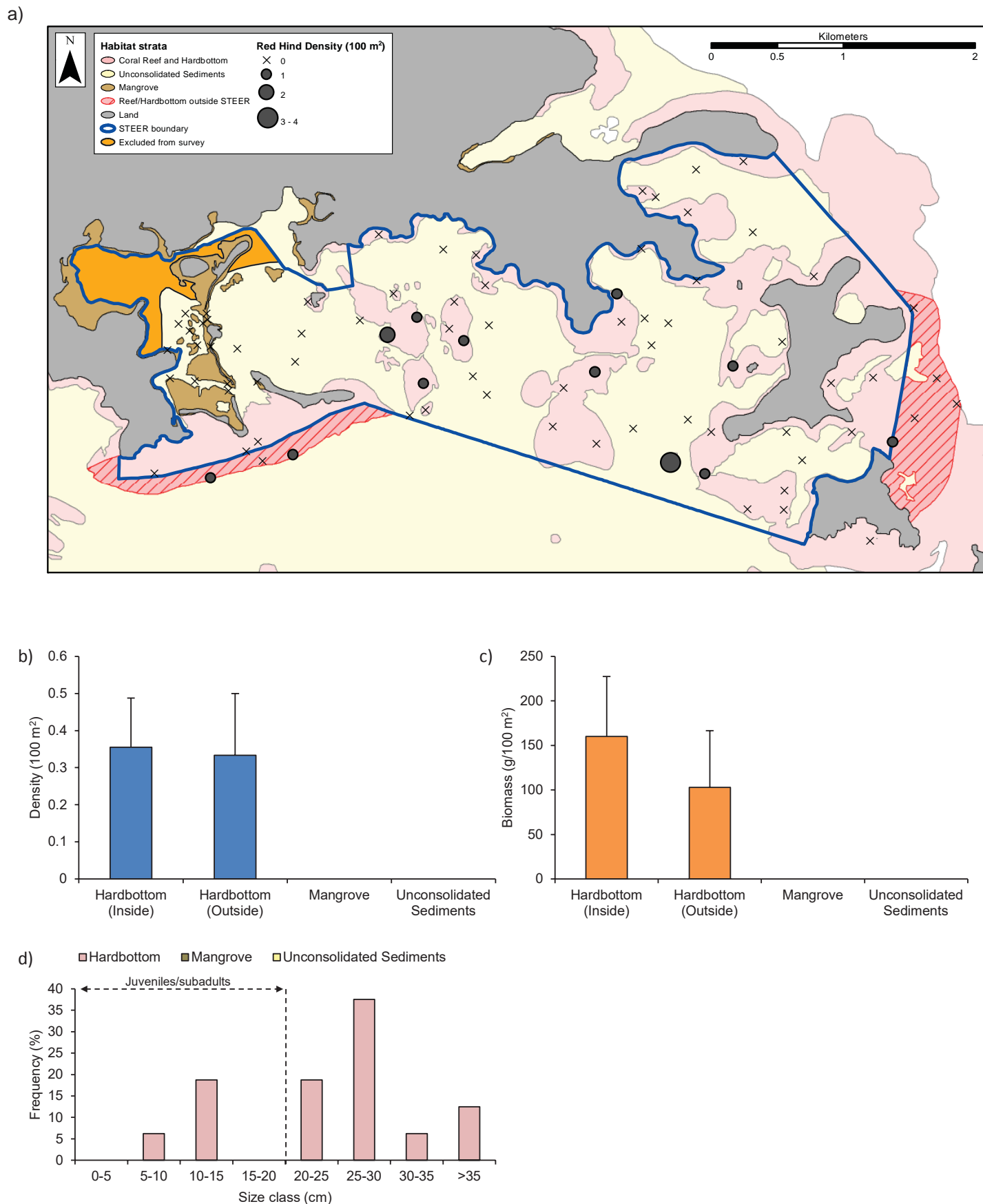


Figure 3.30. Red hind (*Epinephelus guttatus*) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency.

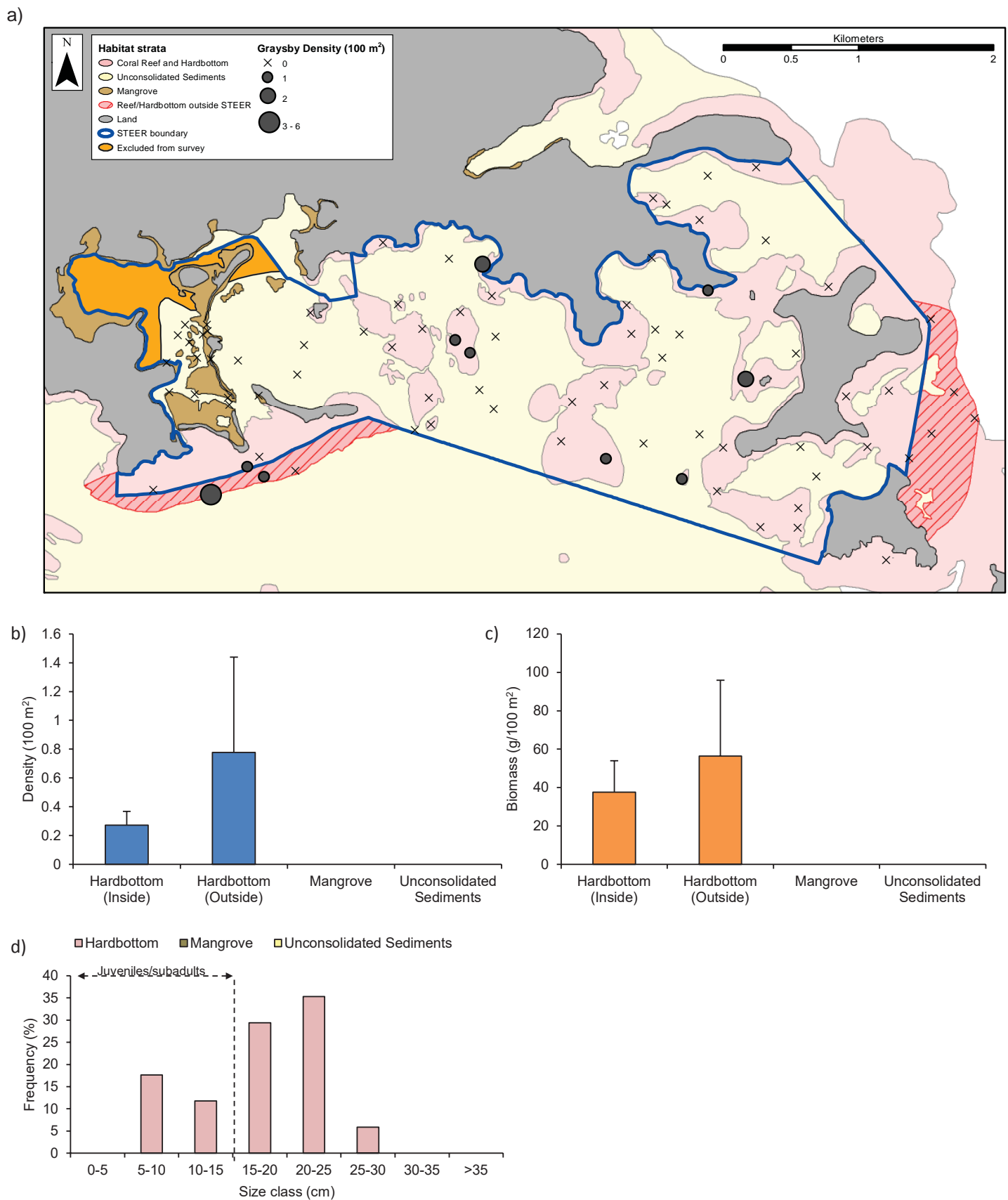


Figure 3.31. Graysby (*Cephalopholis cruentata*) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency.

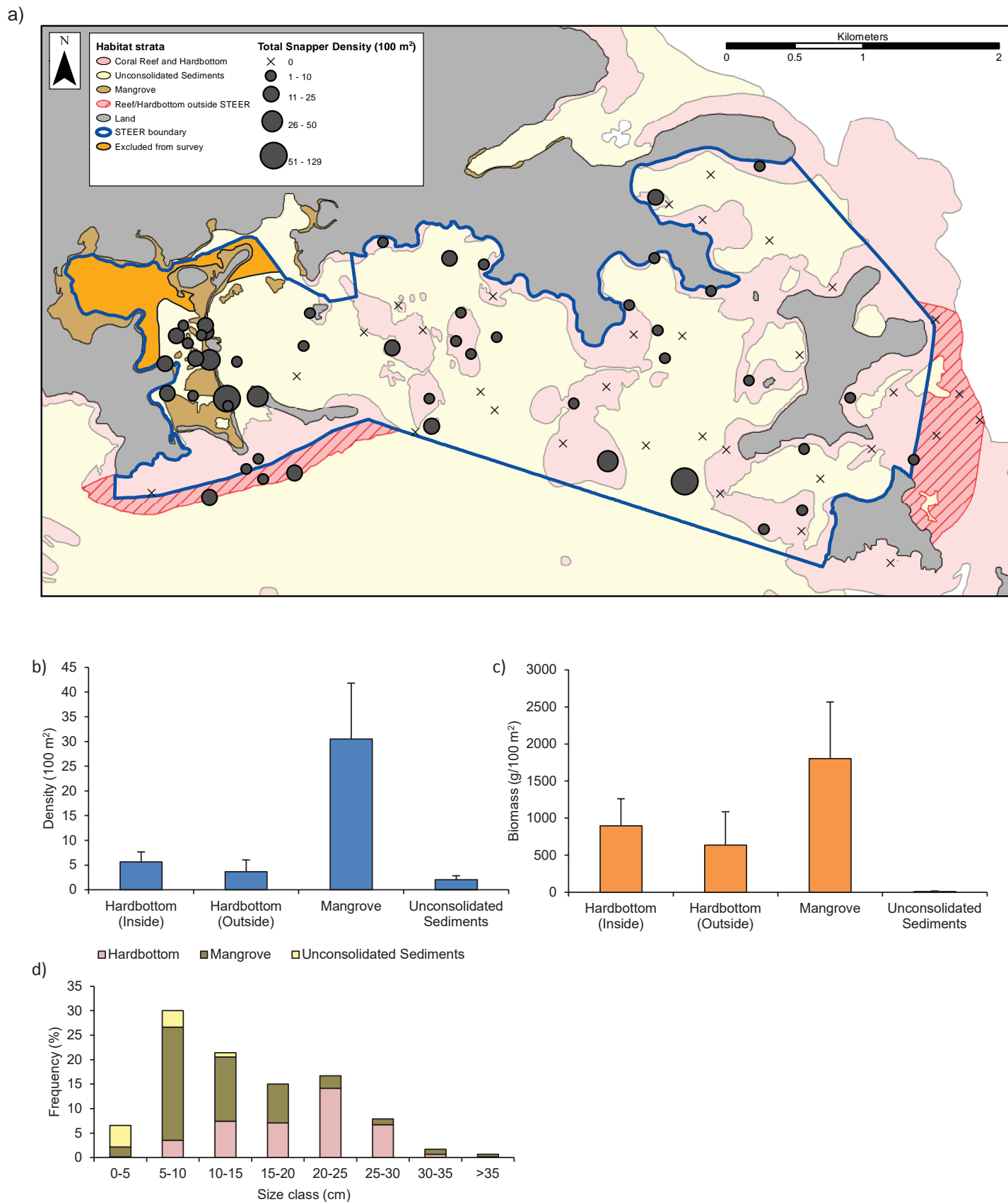


Figure 3.32. Snapper (Family Lutjanidae a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency.

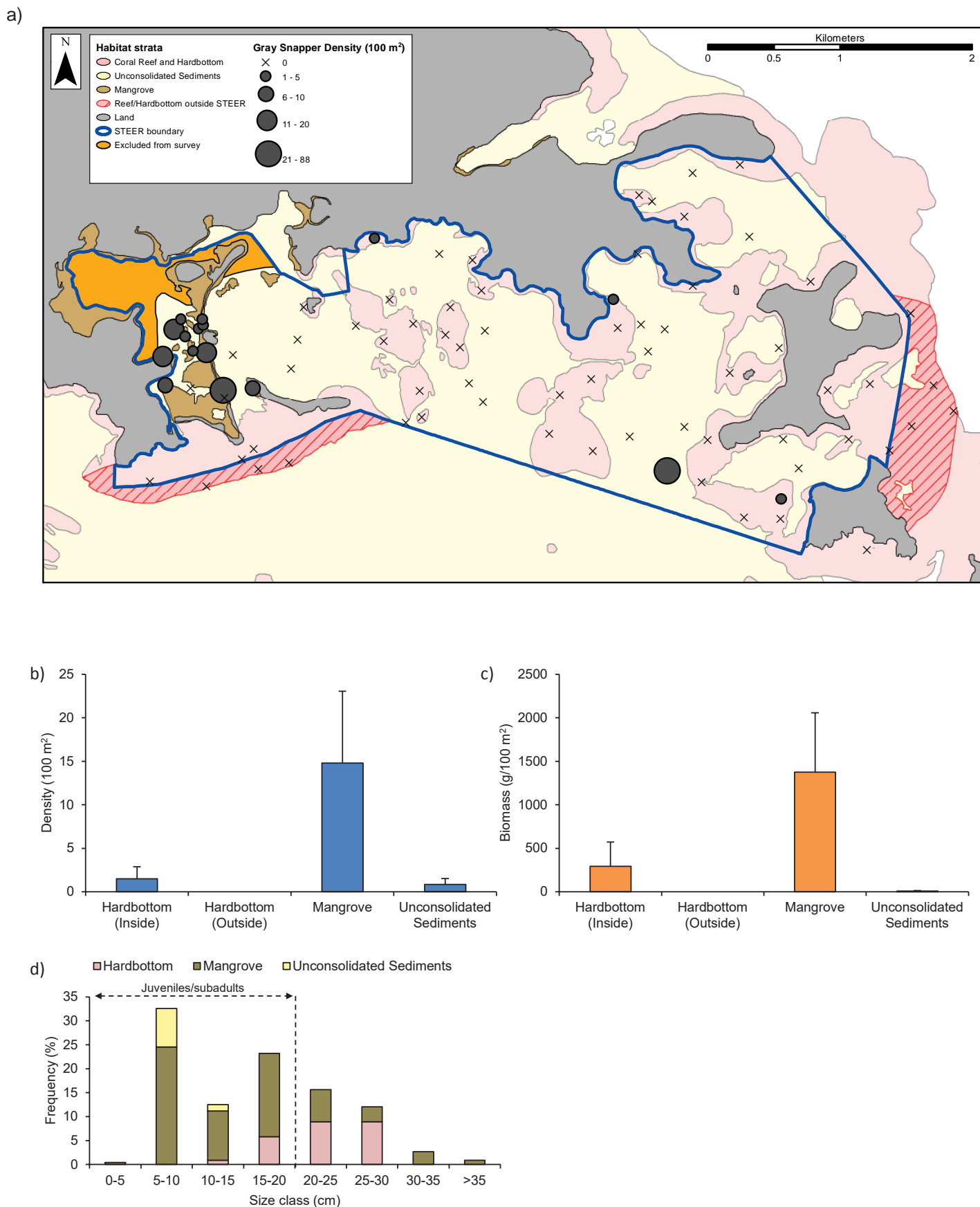


Figure 3.33. Gray snapper (*Lutjanus griseus*) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency.

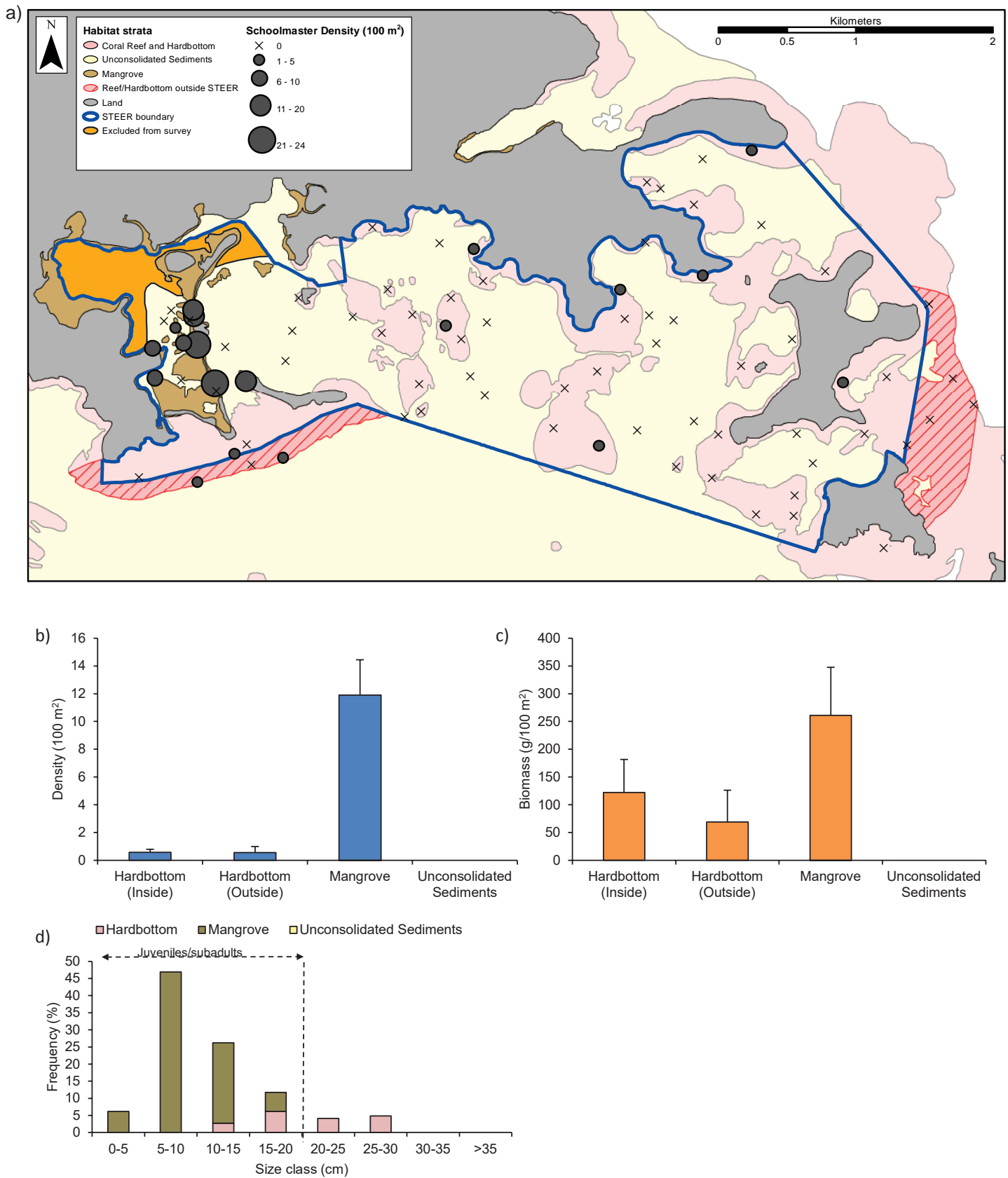


Figure 3.34. Schoolmaster (*Lutjanus apodus*) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency.

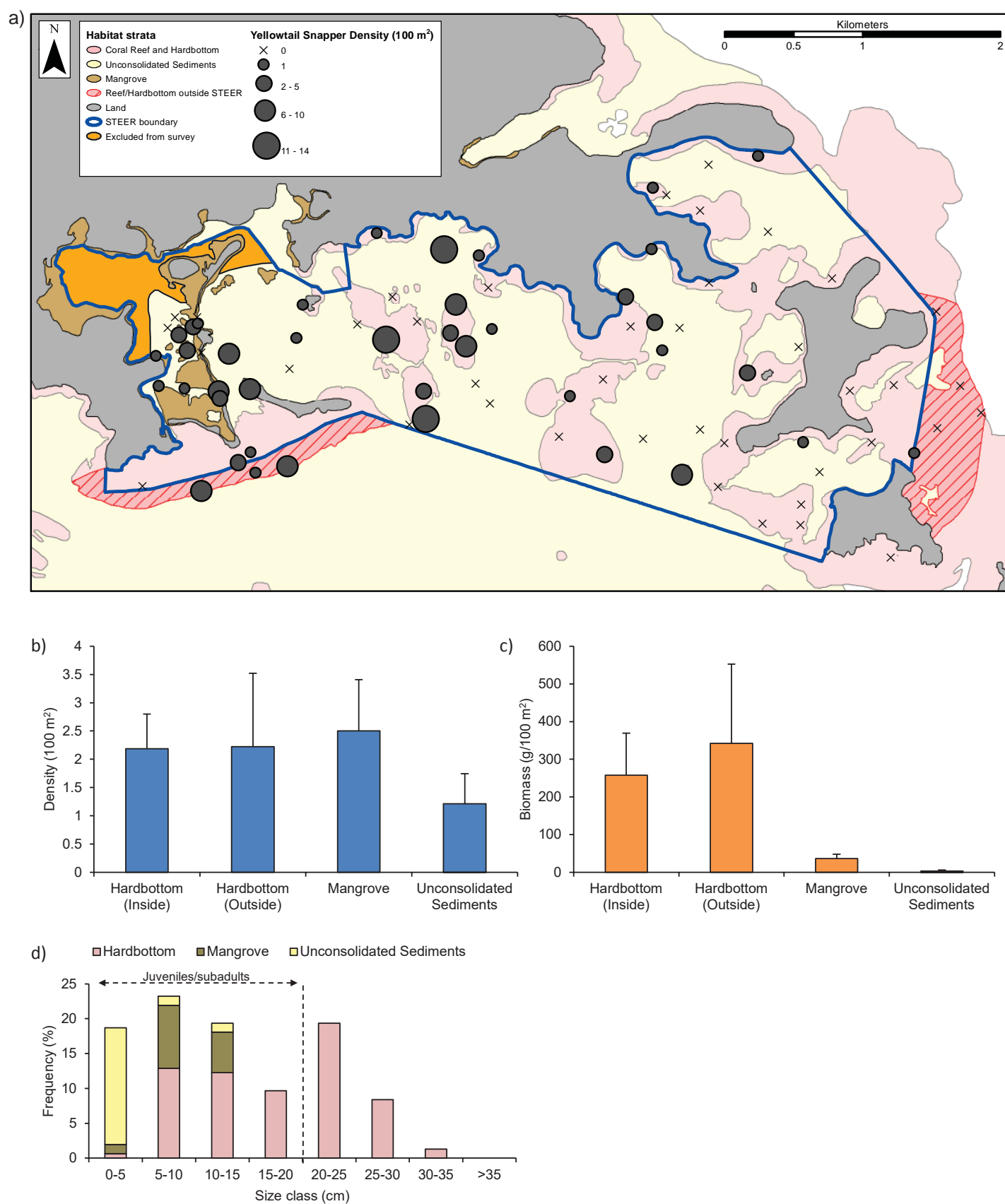


Figure 3.35. Yellowtail snapper (*Ocyurus chrysurus*) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency.

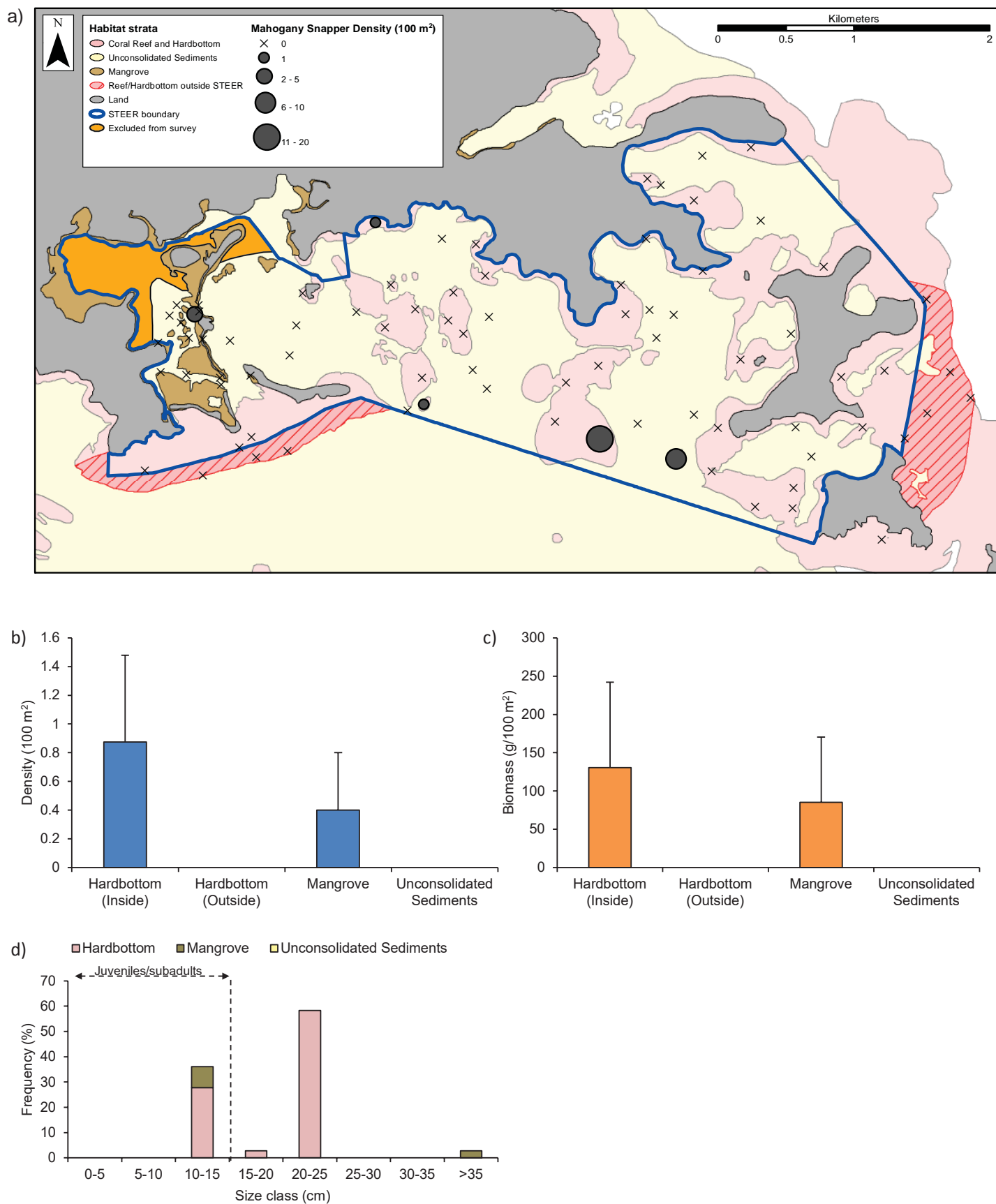


Figure 3.36. Mahogany snapper (*Lutjanus mahogoni*) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency.

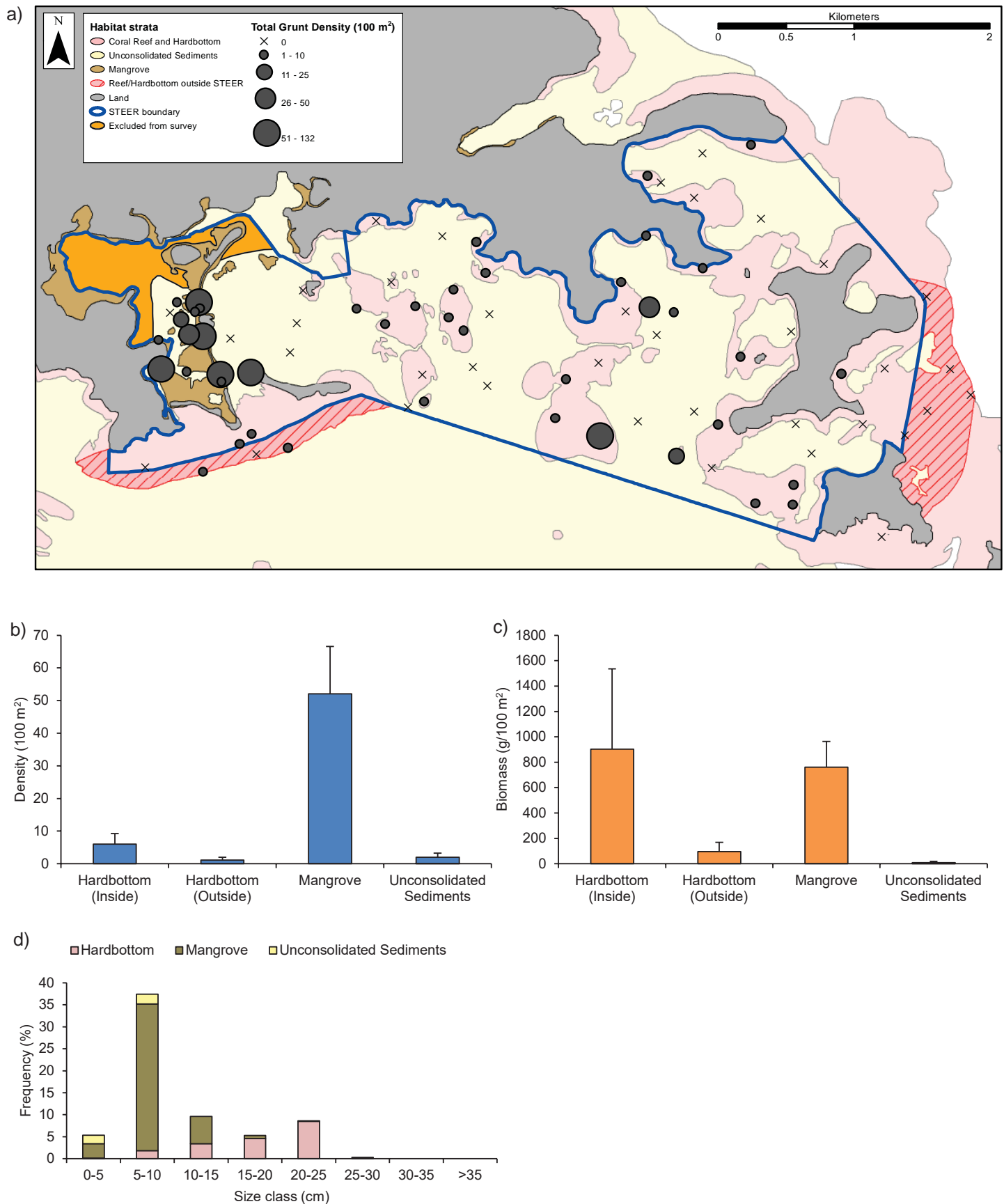


Figure 3.37. Grunt (Family Haemulidae) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency.

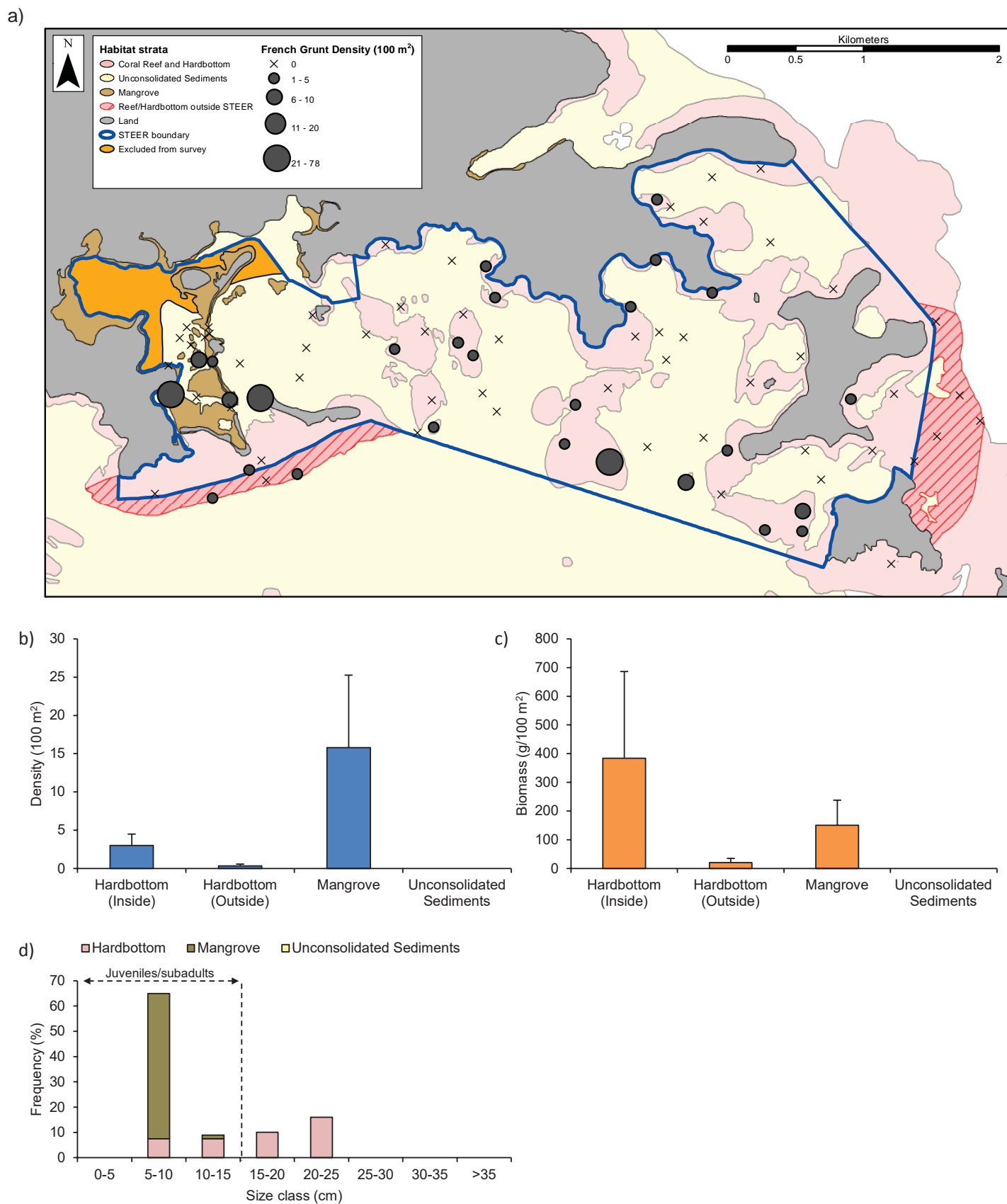


Figure 3.38. French grunt (*Haemulon flavolineatum*) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency.

kg/100 m²). The family was represented by nine species (Appendix 1 in Bauer *et al.*, 2014). French grunt (*Haemulon flavolineatum*), bluestriped grunt (*H. sciurus*), white grunt (*H. plumieri*), and tomtates (*H. aurolineatum*) were most frequently sighted and had the highest mean abundance and biomass of the grunt species. Porkfish (*Anisotremus virginicus*), black margate (*A. surinamensis*), and the remaining species were observed less frequently (Appendix 1 in Bauer *et al.*, 2014). Over one-third of observed grunts in mangrove habitat were small juveniles that could not be identified to the species level. These juveniles were also associated with unconsolidated sediments.

French grunt were present within 35% of survey transects in STEER, as well as two hardbottom sites outside the STEER boundary. The species was commonly observed on mangrove hardbottom habitats across the study area but absent from surveys on unconsolidated sediments (Figure 3.38). While mean density was highest in mangroves, variability was also relatively high and the majority of individuals were small juveniles. In contrast, a larger range of sizes were present on hardbottom habitat, although sub-adults and small adults were most common.

Bluestriped grunt were sighted in 21% of surveys within STEER and one site outside the STEER boundary. The species was most abundant in mangrove habitat, with densities up to 50 / 100m² (Figure 3.39). Mean biomass was highest on hardbottom but varied across space; when present, the species generally occurred as singular individuals or occasional larger aggregations. While all size classes were present in mangroves, juveniles/sub-adults were most common (Figure 3.39d). Hardbottom was typically inhabited by the larger size classes.

Tomtate were found across all habitats but was less frequently sighted than the previously described grunt species, occurring in only 8% of survey transects within STEER. However the species was observed in densities >10 / 100m² at a few locations, including Cow and Calf Rocks and a soft sediment site outside Cowpet Bay (Figure 3.40). Approximately 60% of observed individuals were juveniles/sub-adults, with the remaining being in the size class just above the average size at maturity (Figure 3.40d).

Surgeonfish (*Acanthuridae*)

Acanthurids (surgeonfish) were present in all three bottom types and across all depths throughout the STEER, but the family was most abundant in hardbottom habitats (Figure 3.41). Fewer sightings were observed in Benner Bay and Mangrove Lagoon, with the exception of the survey near the false entrance. Ocean surgeonfish (*Acanthurus bahia-*

nus) and blue tang (*A. coeruleus*) were the most frequently observed species in both study areas, while doctorfish (*A. chirirgus*) was relatively less common (Appendix 1 in Bauer *et al.*, 2014).

Ocean surgeonfish were observed at multiple sites spanning the shelf and bottom types, including nearshore and lagoon areas (Figure 3.42). The species was present in over half of transects within STEER and all but two of the hardbottom sites outside STEER. A cluster of particularly high density sites were located on spur and groove and pavement with sand channels habitat in Jersey Bay. Mean density and biomass were higher on hardbottom inside STEER compared to outside the reserve. Size frequencies tended towards the smaller size classes, with >40% of observed individuals under 5 cm in length (Figure 3.42d).

Blue tang were documented in 42% of survey transects within STEER, primarily on hardbottom. The species was only present at one mangrove location and was absent from unconsolidated sediment surveys (Figure 3.43). Blue tang were also documented at the majority of hardbottom sites outside the reserve boundary, although mean density and biomass were lower compared to inside STEER. The species occurred across most hardbottom types but the two locations with highest observed densities were located on shallow nearshore rock/boulder habitat in Great Bay. Similar to ocean surgeonfish, smaller size classes were most frequent and all observed individuals were <20 cm (Figure 3.43d).

Parrotfish (*Scaridae*)

Parrotfishes (Family Scaridae) were a common component of the STEER fish community. The family was represented by 11 species (Appendix 1 in Bauer *et al.*, 2014). The species with the highest site frequency, density and biomass were striped parrotfish (*Scarus iseri*), redband parrotfish (*Sparisoma aurofrenatum*), princess parrotfish (*Sc. taeniopterus*), and stoplight parrotfish (*Sp. viride*). Most larger-bodied species were absent from the study areas, however rainbow parrotfish (*Sc. guacamaia*) occurred in one survey transect in Nazareth Bay. The site was also characterized by the largest observed parrotfish biomass in STEER and by the presence of several adult-sized yellowtail parrotfish (*Sp. Rubripinne*). A spawning aggregation of yellowtail parrotfish has previously been noted at nearby Coculus Rock (Smith *et al.*, 2011). Overall, highest levels of density and biomass were generally observed on hardbottom habitat, with intermediate and lowest levels in mangroves and unconsolidated sediments, respectively (Figure 3.44). Species composition varied across bottom types. All eleven species were observed on hardbottom, while mangrove was

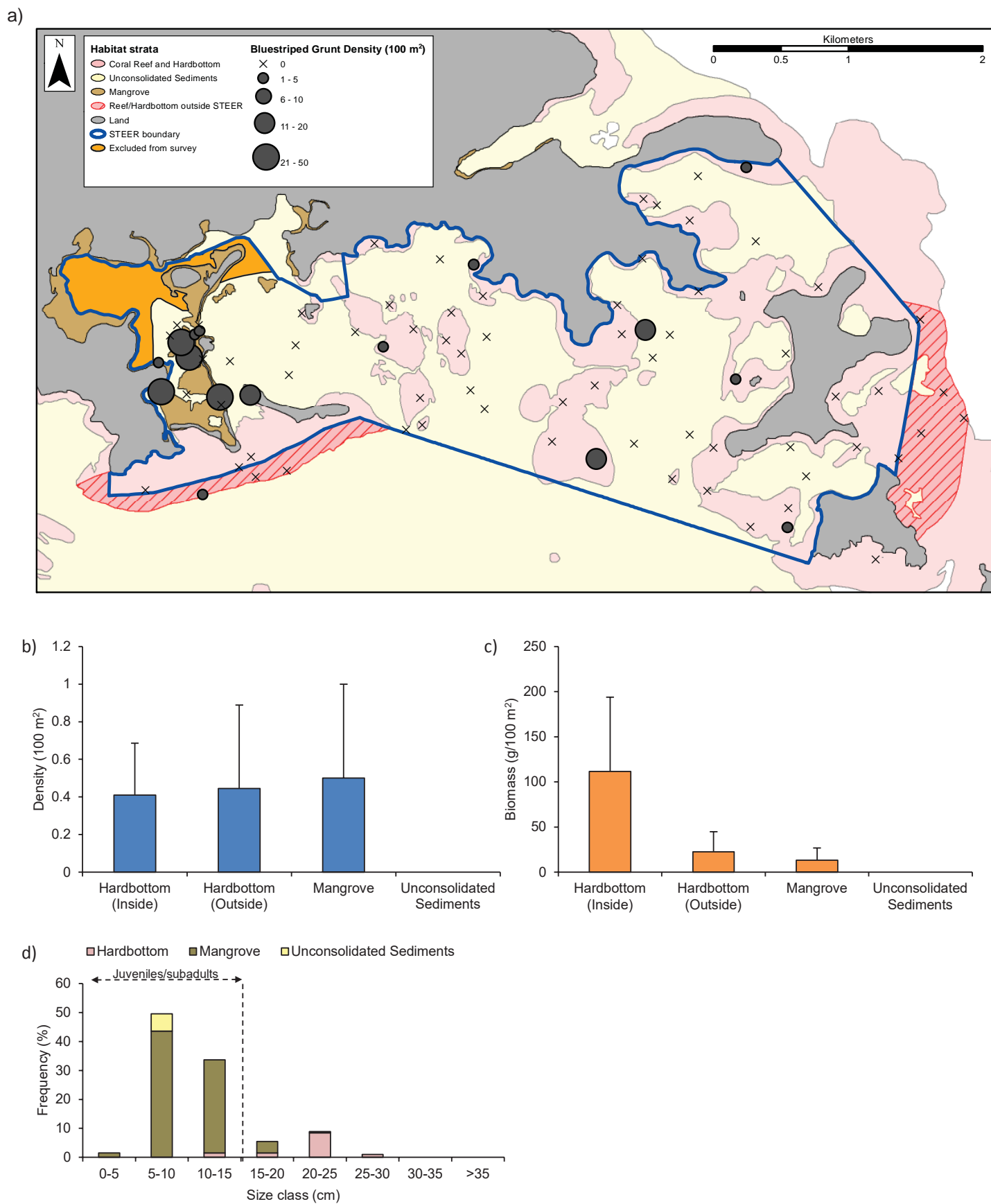


Figure 3.39. Bluestriped grunt (*Haemulon sciurus*) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency.

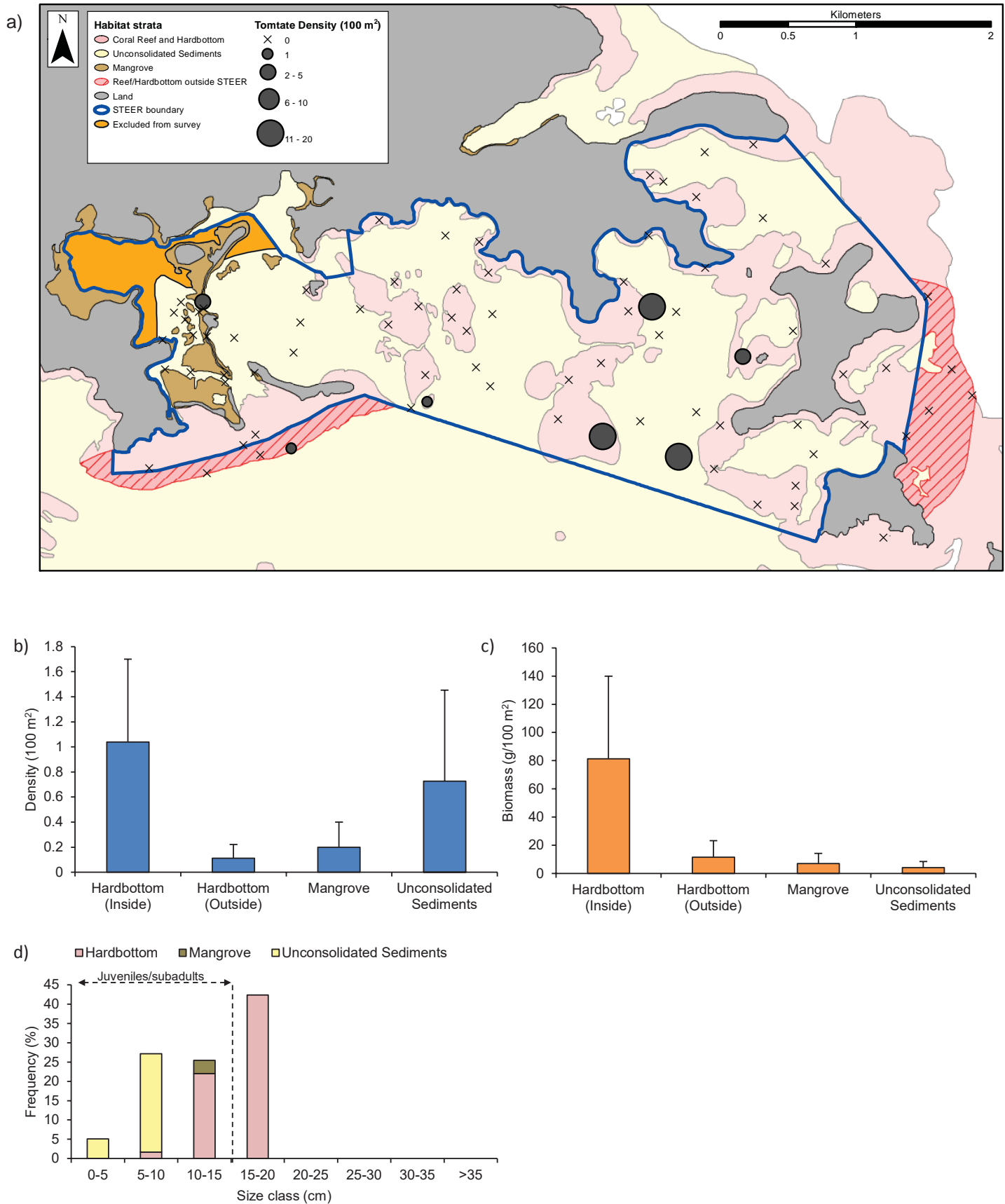


Figure 3.40. Tomate (*Haemulon aurolineatum*) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency.

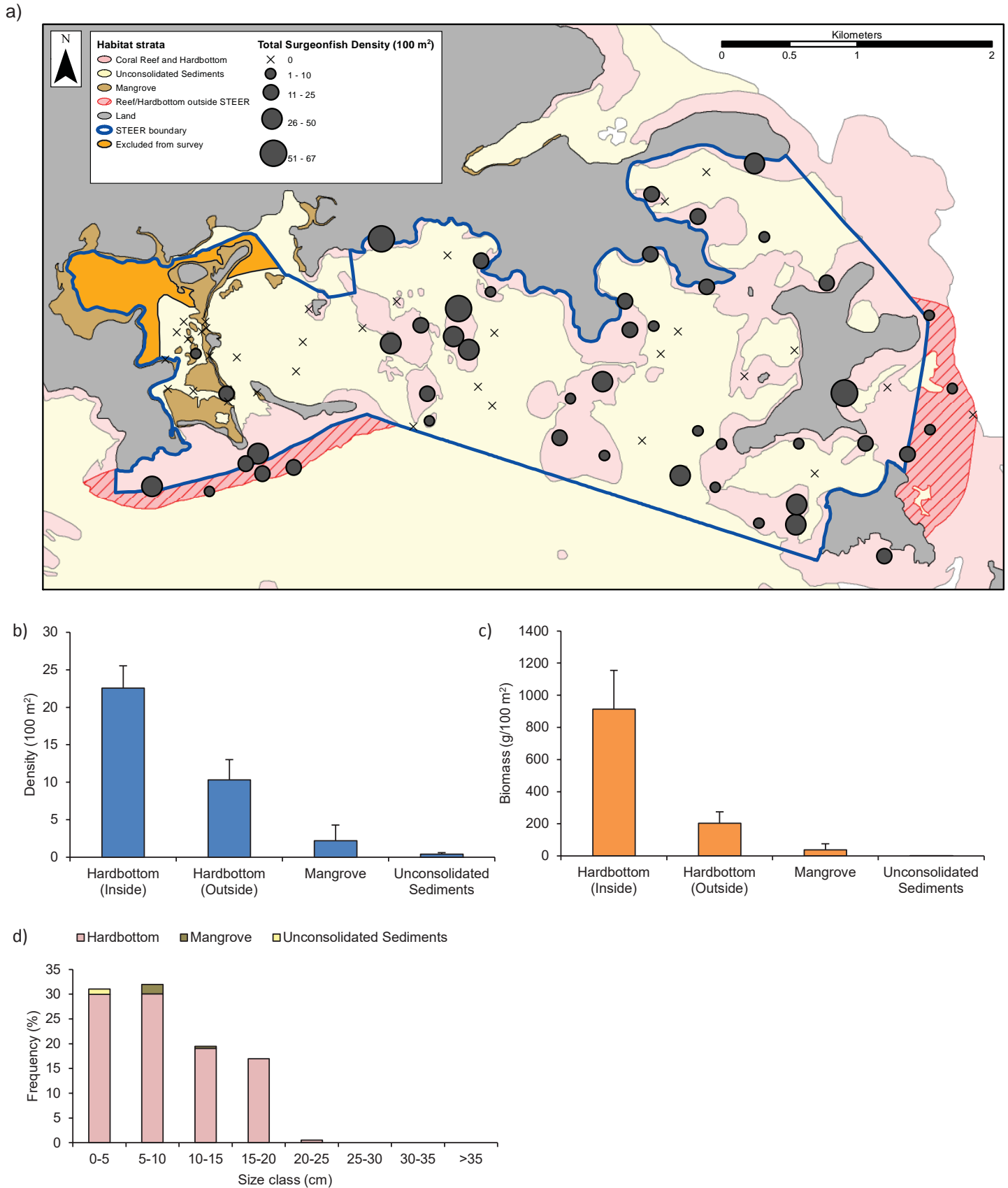


Figure 3.41. Surgeonfish (Family Acanthuridae) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency.

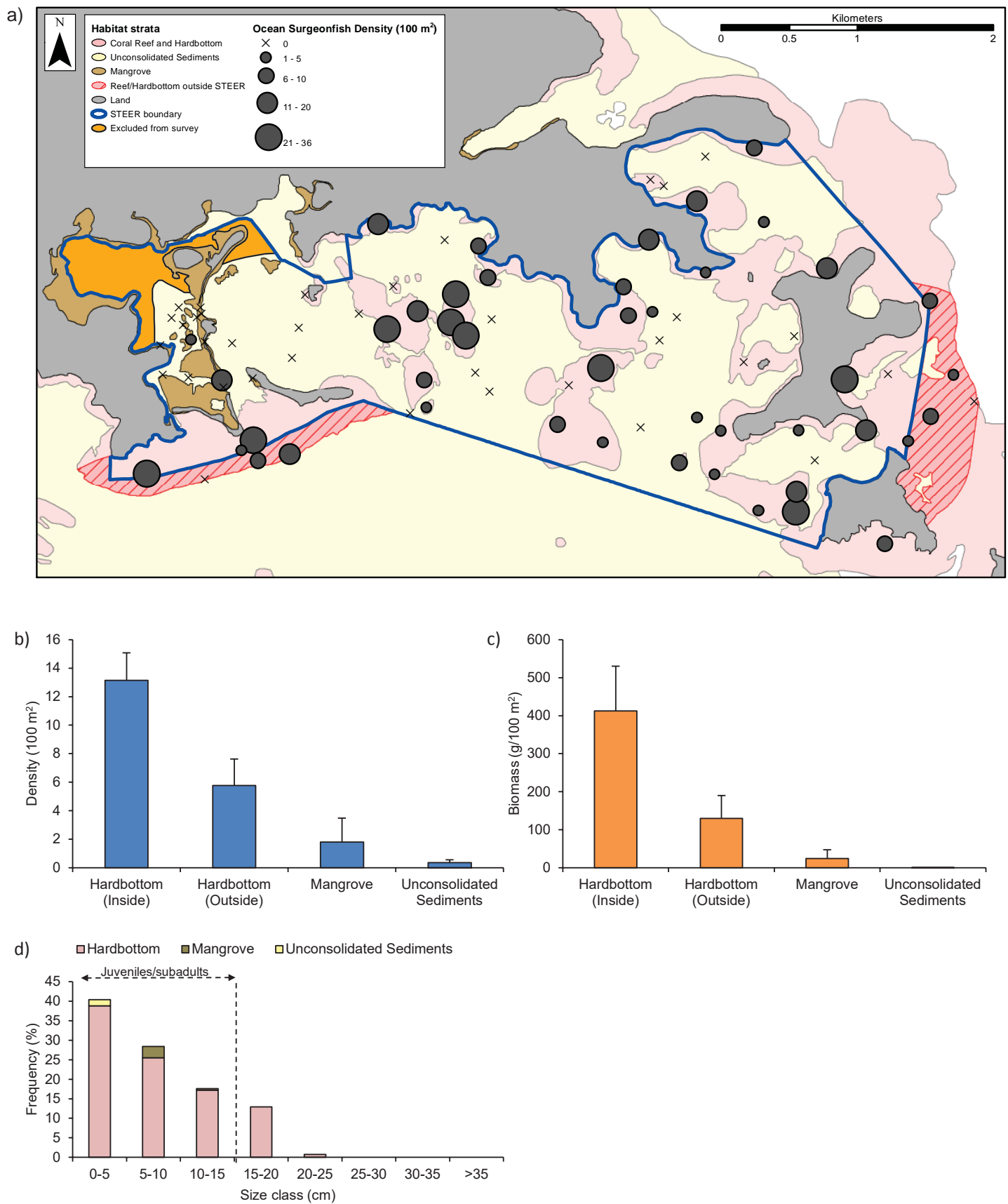


Figure 3.42. Ocean surgeonfish (*Acanthurus bahianus*) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency.

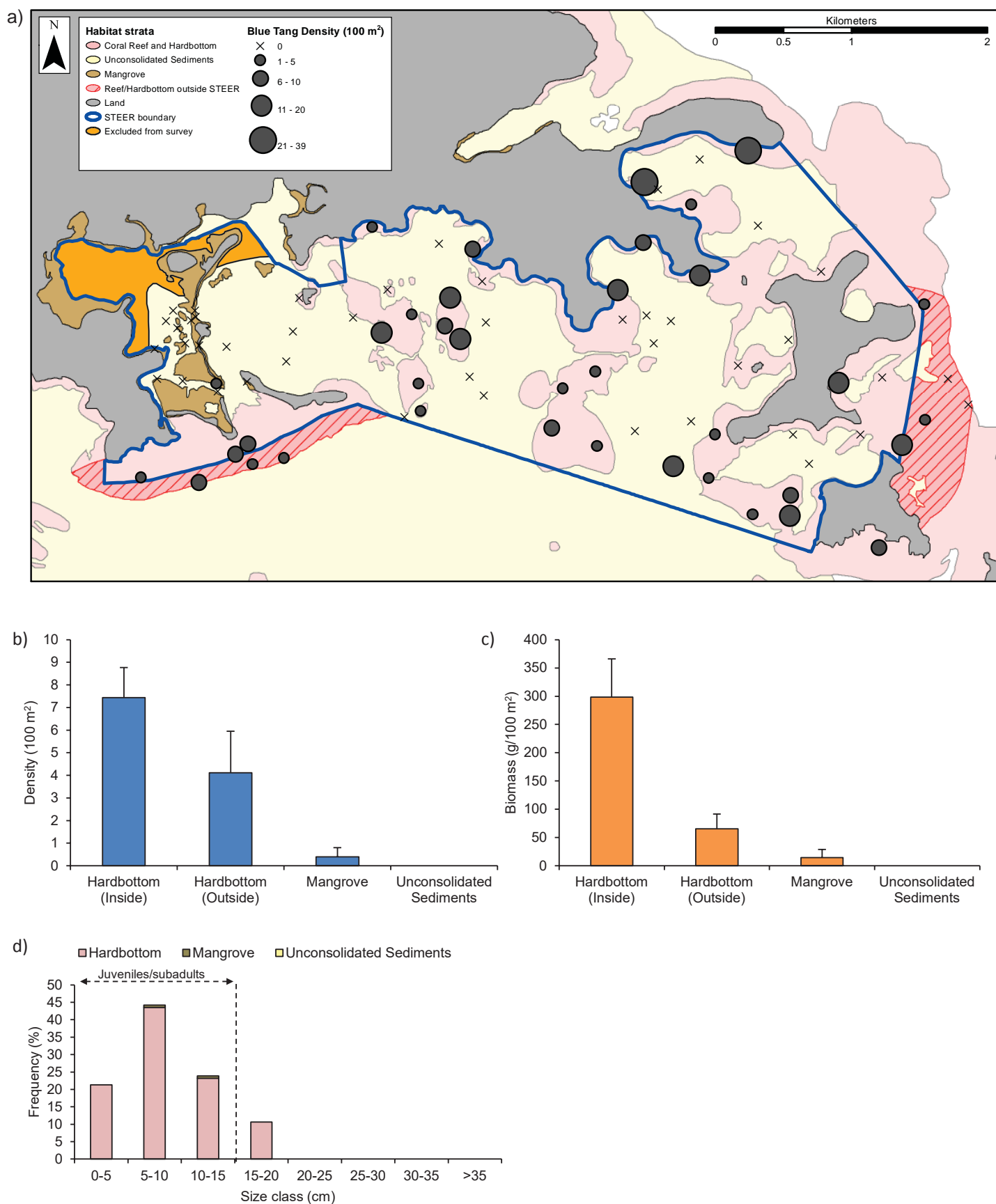


Figure 3.43. Blue tang (*Acanthurus coeruleus*) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency.

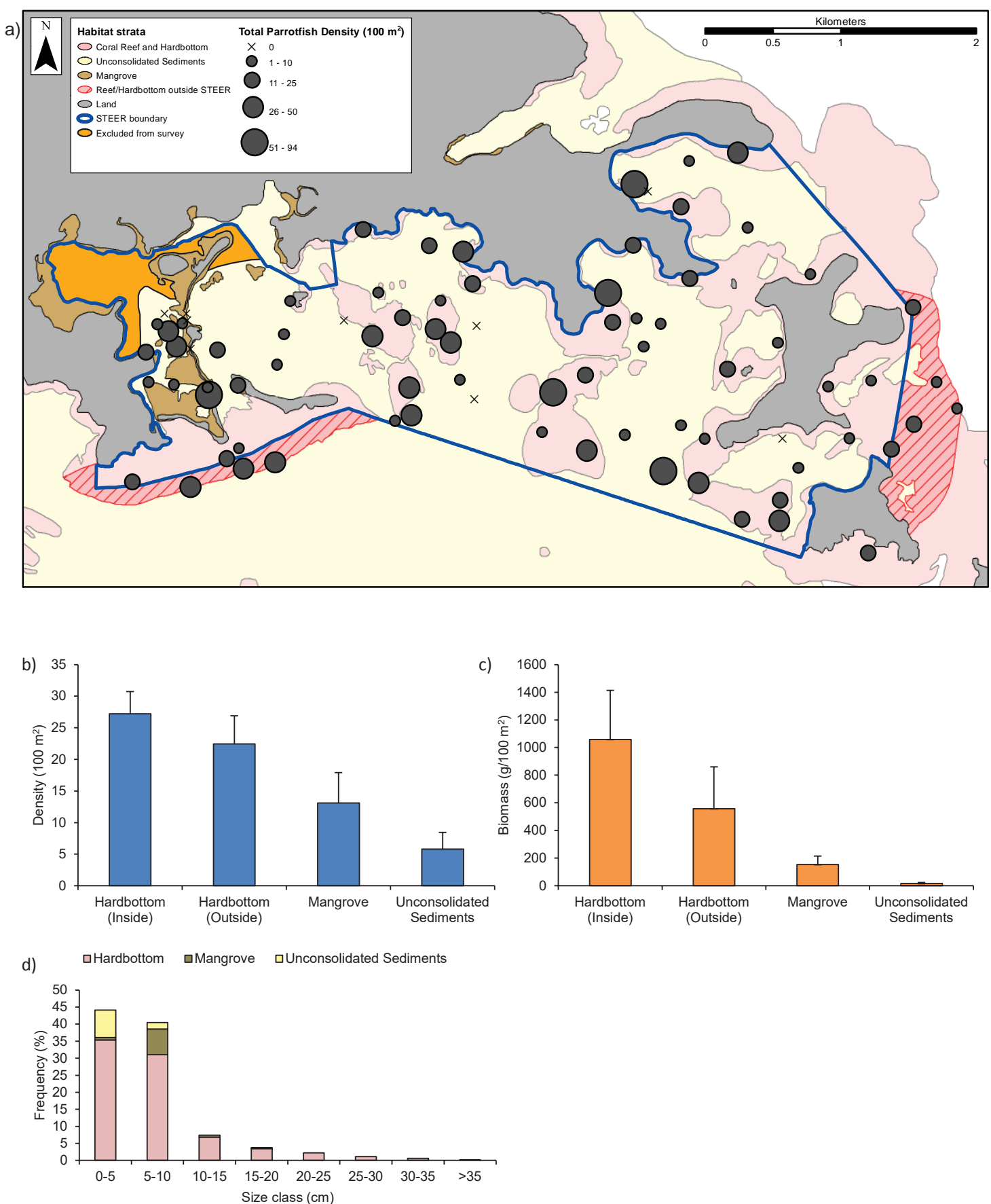


Figure 3.44. Parrotfish (Family Scaridae) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency.

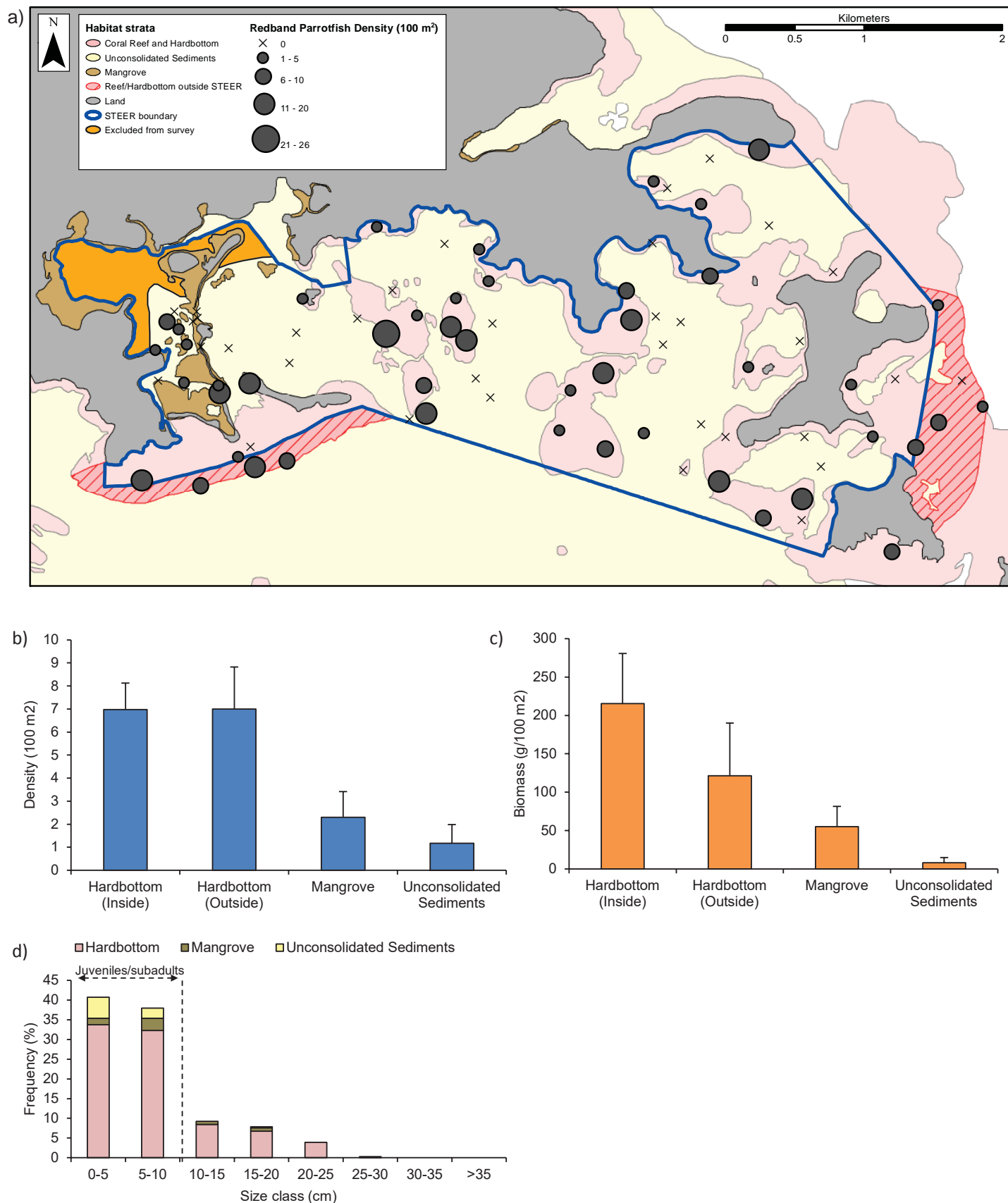


Figure 3.45. Redband parrotfish (*Sparisoma aurofrenatum*) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency.

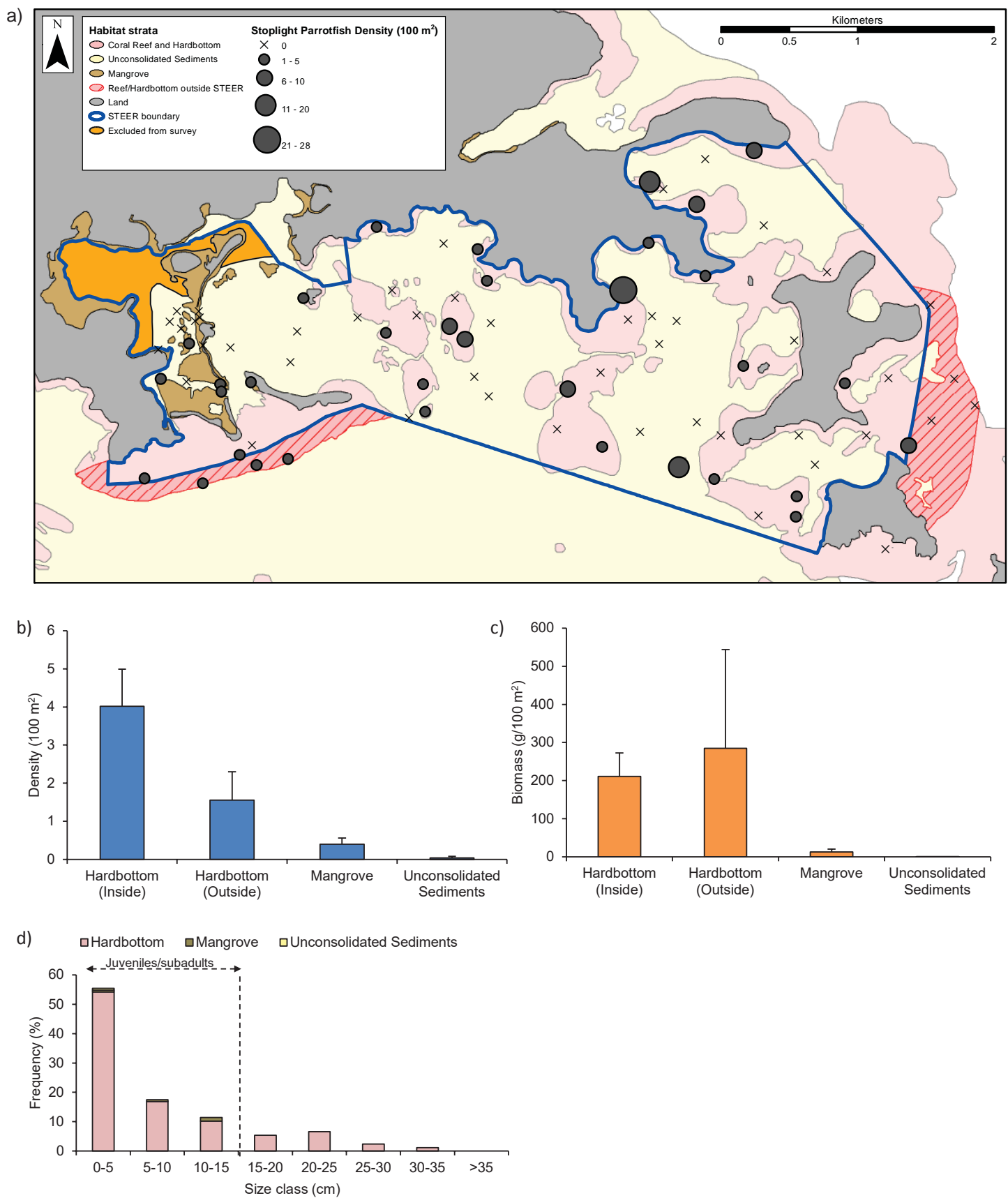


Figure 3.46. Stoplight parrotfish (*Sparisoma viride*) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency.

only occupied by striped, redband, stoplight, and bucktooth parrotfishes. Unconsolidated sediments were also typified by juveniles of the aforementioned species, as well as the smaller bodied bluelip parrotfish (*Cryptotomus roseus*).

Redband parrotfish were sighted in 54% of survey transects within STEER. Mean density was similar on hardbottom inside and outside STEER, with slightly higher mean biomass inside STEER (Figure 3.45). The species was common across all hardbottom types, including nearshore rock/boulder, patch reefs, and pavement. Lower densities were typically observed over unconsolidated sediments and mangrove, with the notable exception of mangrove sites near the false entrance to Mangrove Lagoon and Cas Cay. The size distribution was skewed towards the smaller size classes (0-10 cm TL) while ~20% of the observed individuals were above the mean size at maturity (Figure 3.45d).

Stopsight parrotfish were observed in all three habitats with STEER but exhibited highest abundance and biomass on hardbottom (Figure 3.46). The species was present across all hardbottom types but sites with higher values occurred on nearshore rocky habitat in Cowpet and Great Bays and a patch reef in St. James Bay. Biomass at hardbottom sites outside STEER exhibited a relatively higher degree of variability due to the presence of a few larger sized individuals at one location. Over 50% of the observed individuals were in the smallest size class (0-5 cm), while 15% were small adults (Figure 3.46d).

Striped parrotfish were present in 59% of survey transects within STEER and were common on both hardbottom and in mangroves (Figure 3.47). The site with the highest observed density, located in Cowpet Bay, was also characterized by the highest density of the spotlight parrotfish. The species was less frequently sighted in the eastern portion of the study area and was absent at several of the surveys east of Great St. James Island. Small juveniles were dominant, particularly in mangroves and on soft sediments, and overall only 5% of observed individuals were larger than the mean size at maturity (Figure 3.47d).

Princess parrotfish occurred in 23% of survey transects within STEER and in over half of the hardbottom surveys outside the reserve. Highest mean density and biomass occurred on hardbottom, while the species was absent from surveys in mangrove (Figure 3.48). The densest aggregation occurred on hardbottom in Great Bay, while the location with the second highest observed density was on unconsolidated sediments near the false entrance to Mangrove Lagoon. The overwhelming majority (95%) of observed individuals were juveniles/subadults (Figure 3.48d).

Other species

Wrasses (*Labridae*)

Fishes of the Family Labridae were ubiquitous members of the STEER fish community, occurring in 85% of all surveys within STEER and all but one hardbottom survey. Wrasses were present in all habitats and across the geographic area, but with fewer sighting frequencies in Mangrove Lagoon (Figure 3.49). Mean density and biomass were highest on hardbottom. The family was represented by 12 species (Appendix 1 in Bauer *et al.*, 2014), with some variation by habitat. Bluehead (*Thalassoma bifasciatum*), slippery dick (*Halichoeres bivittatus*), yellowhead wrasse (*H. garnoti*), and clown wrasse (*H. maculipinna*) the most frequently encountered species on hardbottom, while razorfish species, particularly rosey razorfish (*Xyrichtys martinicensis*), were more frequent on unconsolidated sediments. Hogfish (*Lachnolaimus maximus*) was absent from surveys within STEER but one individual was sighted within a transect on the reef complex south of the reserve boundary. The size distribution skewed towards the smaller size classes (Figure 3.49d) because small-bodied Labrid species were most abundant.

Goatfishes (*Mullidae*)

Goatfish were present in 20% of survey transects within STEER, in addition to several of the hardbottom surveys outside STEER. The family was represented by two species, spotted goatfish (*Pseudupeneus maculatus*) and yellow goatfish (*Mulloidichthys martinicus*). Yellow goatfish was observed exclusively on hardbottom while spotted goatfish was documented within a couple of unconsolidated sediment surveys. Goatfishes were typically observed in low densities (1-5/ 100 m²) but one larger cluster (27/ 100 m²) was recorded on an area of pavement with sand channels in Jersey Bay (Figure 3.50).

Damselfishes (*Pomacentridae*)

Damselfishes were present across all bottom types, occurring in 62% of surveys within STEER and all but one hardbottom site outside the reserve. Highest densities and biomass were observed on hardbottom, with intermediate levels in mangrove habitat and low frequency of occurrence on unconsolidated sediments (Figure 3.51). Damselfish were prevalent across hardbottom types but highest densities were observed at sites in the southern portion of the study area, including surveys near Cow and Calf Rocks, southeast of Great St. James Island, and outside the STEER boundary south of Patricia Cay. The family was represented by 10 species (Appendix 1 in Bauer *et al.*, 2014). Frequently observed species included the bicolor damselfish (*Stegastes partitus*), cocoa damselfish (*S. variabilis*), longfin damselfish (*S. dieneaeus*), and beaugregory (*S. leucostictus*).

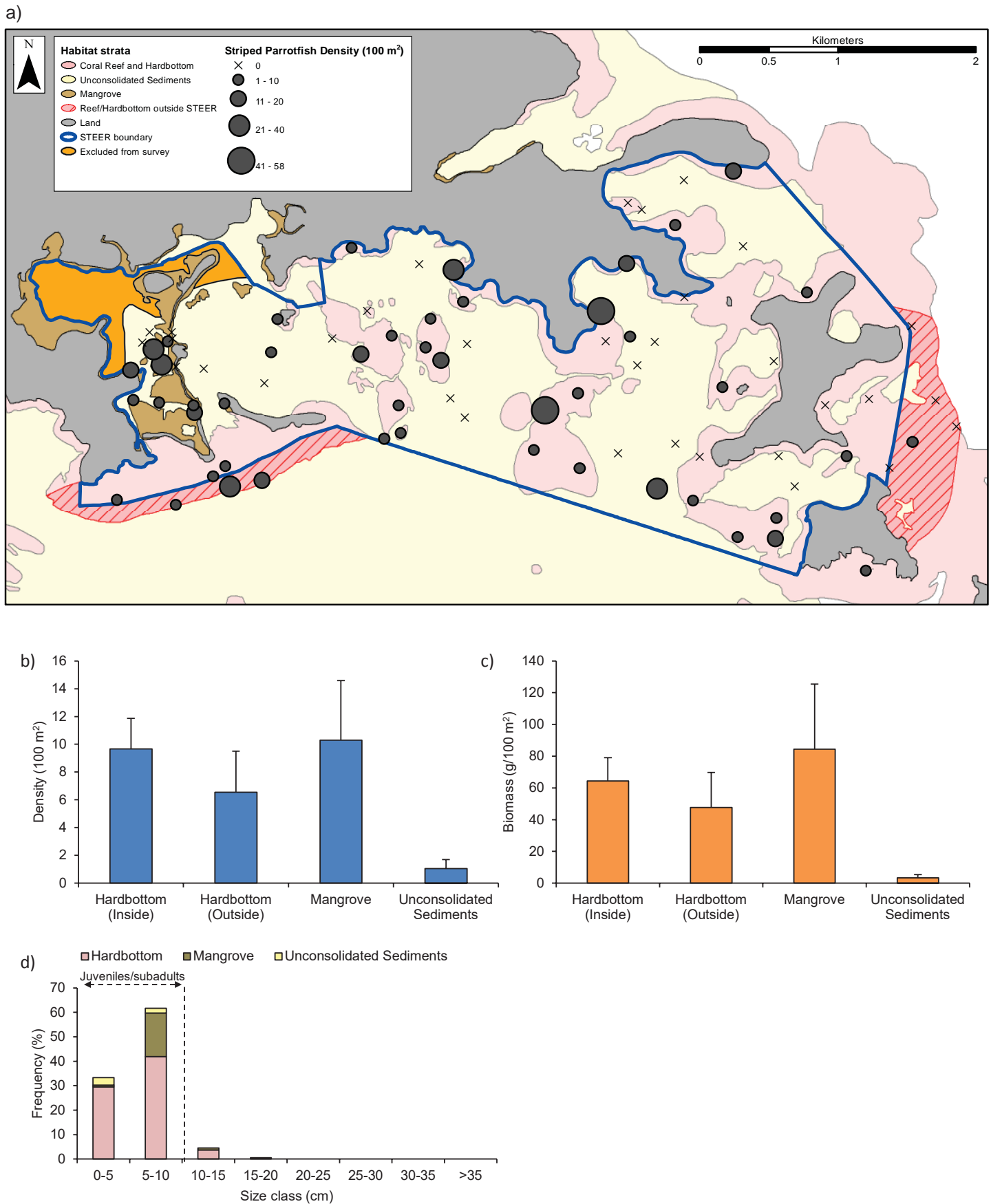


Figure 3.47. Striped parrotfish (*Scarus iseri*) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency.

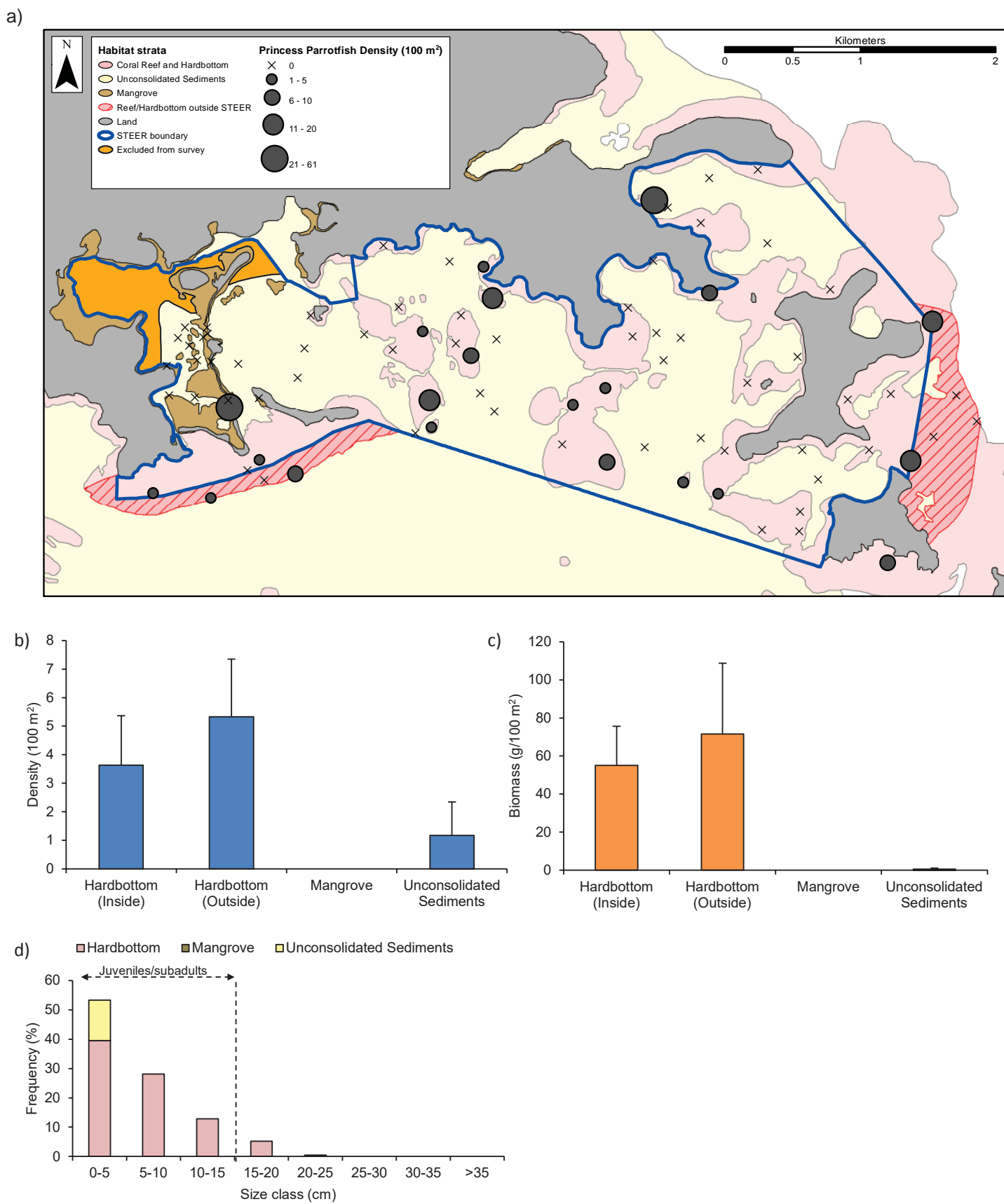


Figure 3.48. Princess parrotfish (*Scarus taeniopterus*) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency.

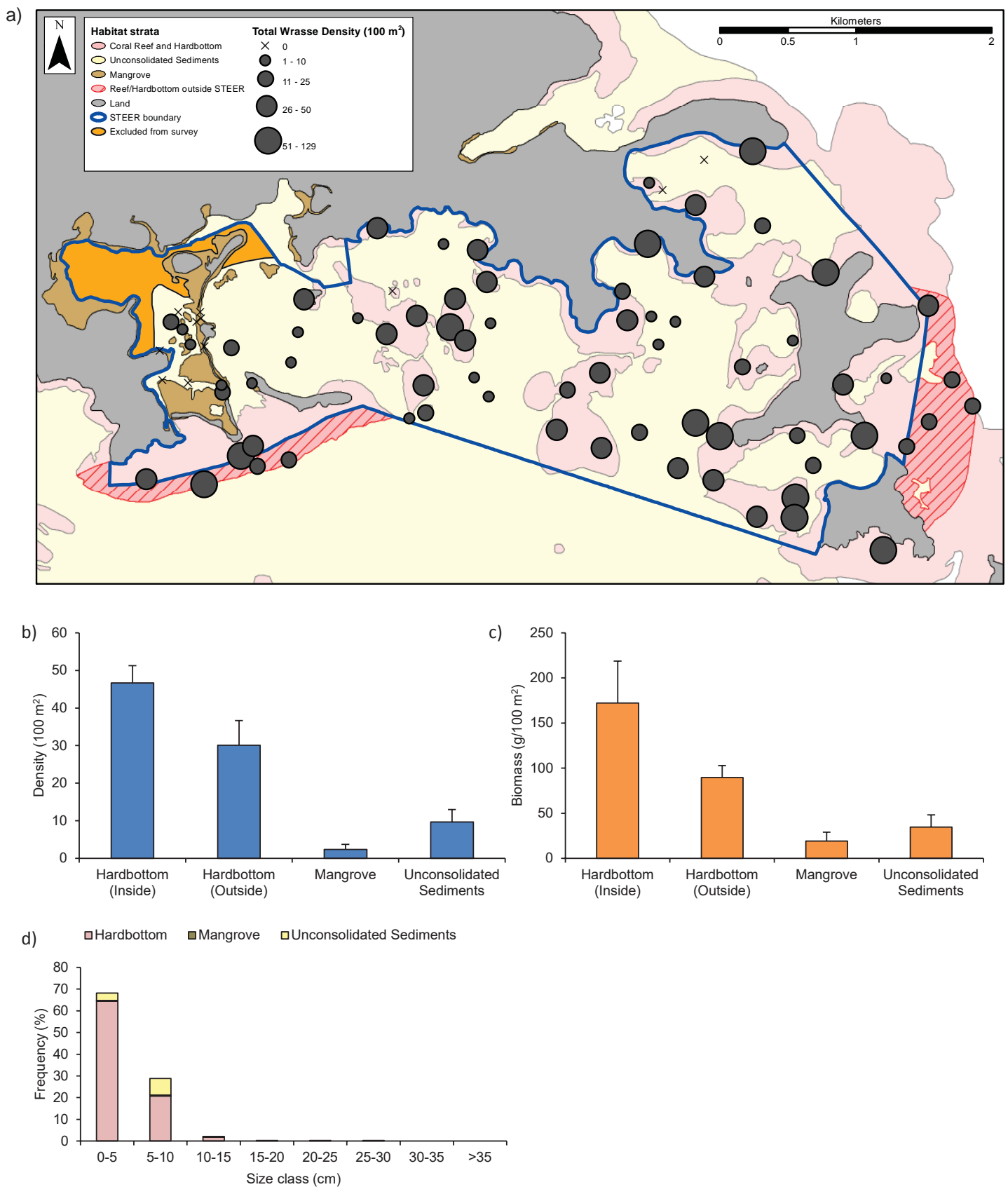


Figure 3.49. Wrasse (Family Labridae) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency.

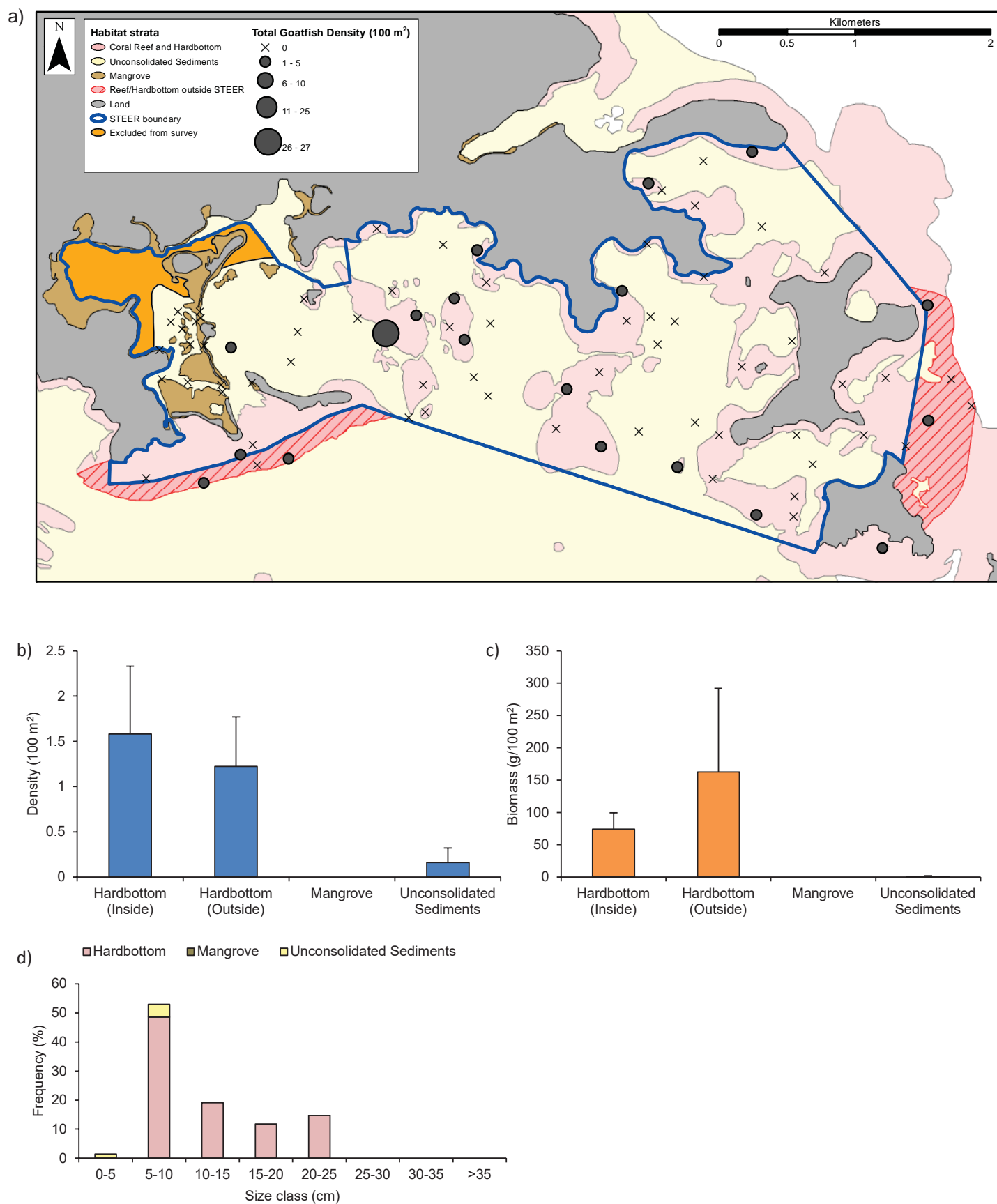


Figure 3.50. Goatfish (Family Mullidae) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency.

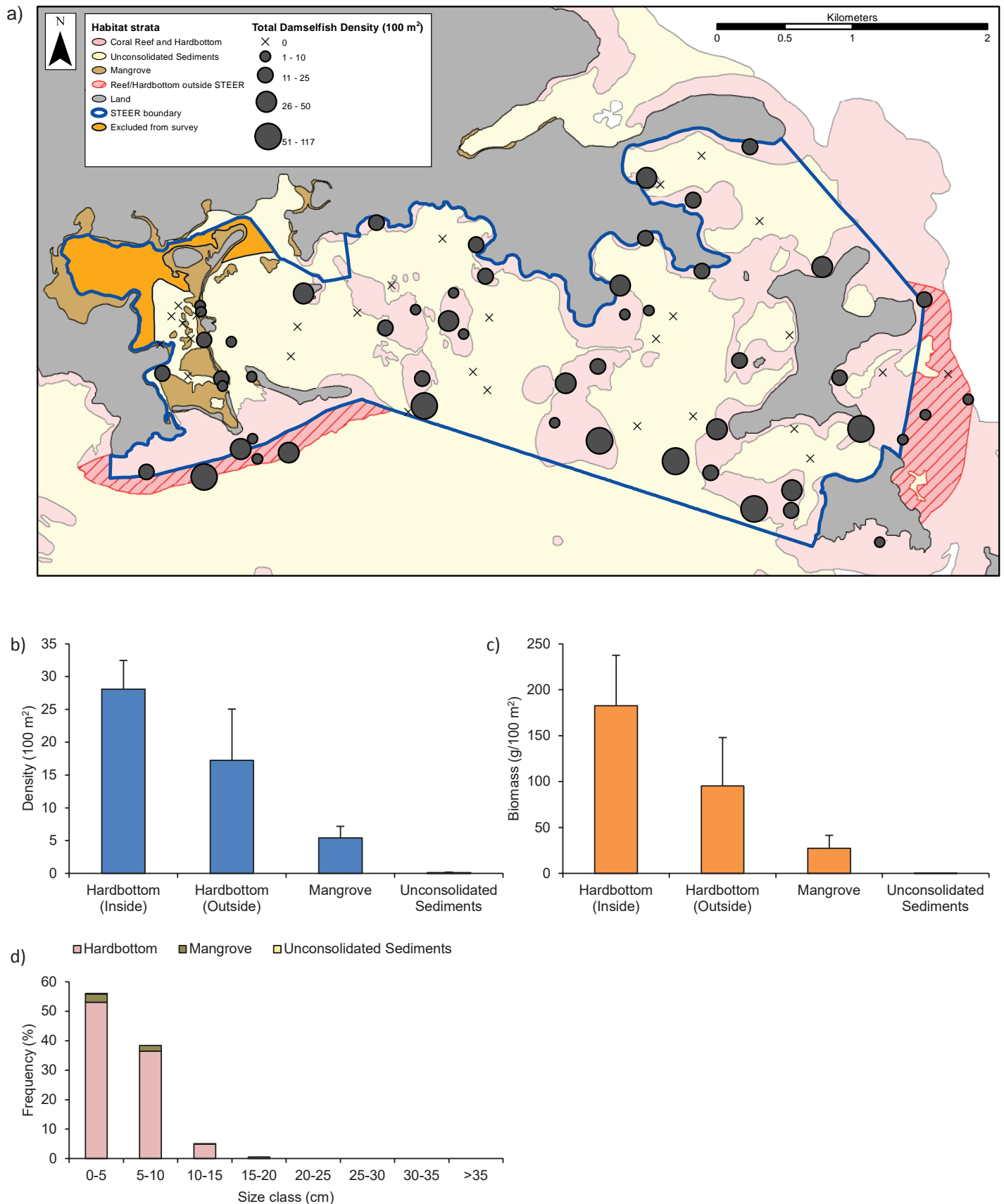


Figure 3.51. Damselfish (Family Pomacentridae) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency.

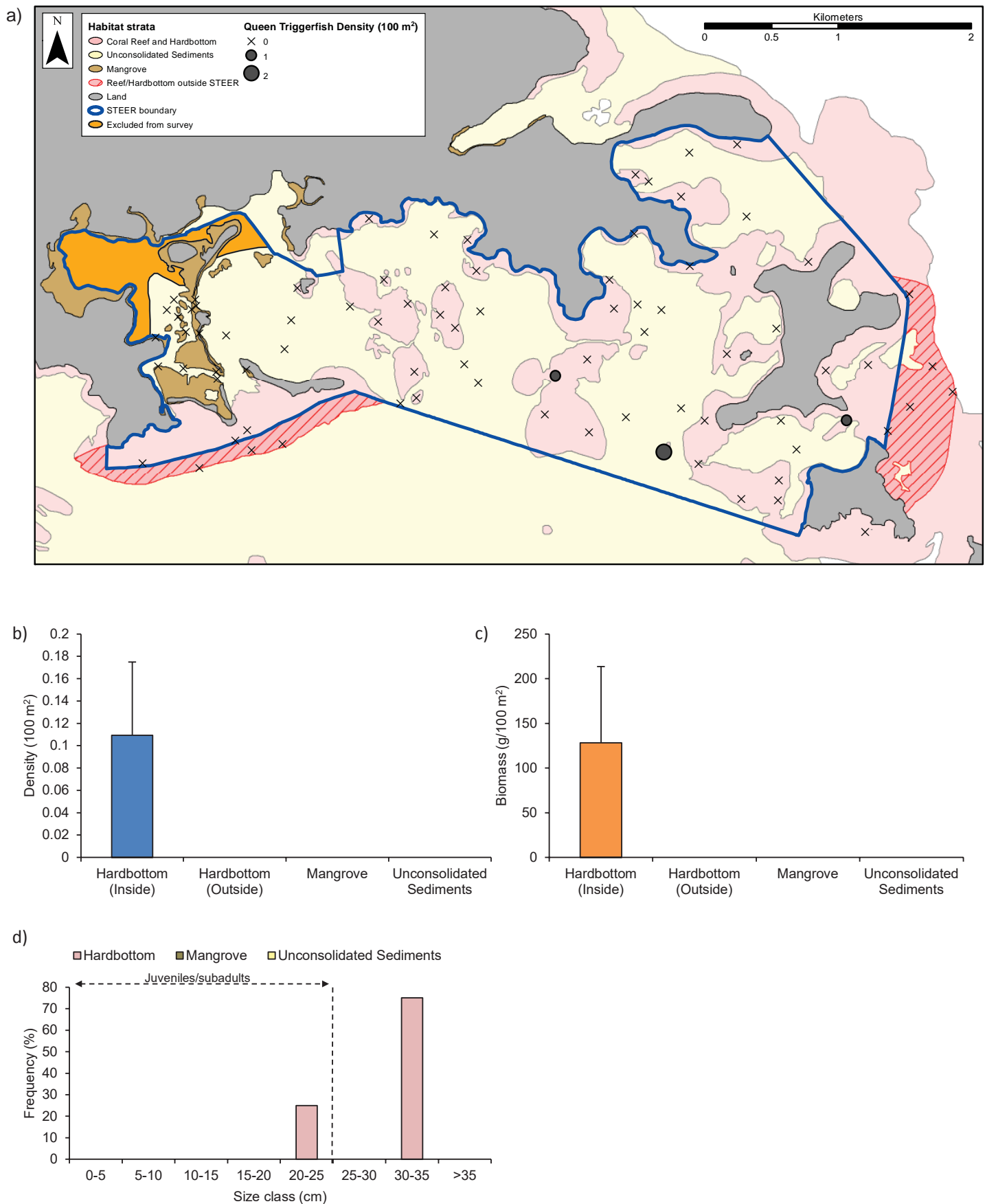


Figure 3.52. Queen triggerfish (*Balistes vetula*) a) spatial distribution, b) mean density (\pm SE) by habitat, c) mean biomass (\pm SE) by habitat, and d) size frequency.

Triggerfish (*Balistidae*)

Queen triggerfish (*Balistes vetula*, Family Balistidae) was uncommonly encountered in the survey, occurring at only three sites within STEER and none outside (Figure 3.52). The species was observed exclusively on hardbottom in the central and southeastern areas of STEER and in densities of 1-2/ 100m². Three of the four observed individuals were adult-sized (Figure 3.52d).

Comparison with Other U.S. Caribbean Monitoring Locations

The benthic community found on hardbottom sites in STEER are similar to those described by long term monitoring programs using the same methods as this study in St. Croix, around Buck Island Reserve National Monument and the northeastern St. Croix shore including the East End Marine Park from 2001-2006 (Pittman *et al.*, 2008), and around the island of St. John from 2001-2009 (Friedlander *et al.*, 2013). Overall community structure was similar: hardbottom habitats in all three locations were dominated by turf and macroalgae with low overall coral cover ($5.6 \pm 0.5\%$ in St. Croix; $4.8 \pm 0.5\%$ St. John) (Figure 3.53). However, turf algae cover averaged $49.7\% (\pm 4.11)$ in STEER compared to only 33% in St. John (Friedlander *et al.*, 2013) and $36\% (\pm 1.6)$ in St. Croix (Pittman *et al.*, 2008). Due to the ephemeral nature of turf algae, percent cover values can be influenced by seasonal and interannual variation.

Unconsolidated sediments habitats in all three study regions were dominated by seagrass with STEER hosting the highest percent cover ($31.4 \pm 5.9\%$ STEER; St. John $22.2 \pm 1.6\%$; St. Croix $19.84 \pm 2.9\%$). St. John and STEER data show similar macroalgal cover ($17.9 \pm 5.1\%$ STEER; $16.2 \pm 1.2\%$ St. John). The unconsolidated sediments sites of St. Croix had much less macroalgae ($6.92 \pm 1.1\%$). Seagrass beds of all three studies were composed of mostly *Syringodium filiforme* and *Thalassia testudinum* in which smaller amounts of sponges, gorgonians, living corals and other benthic invertebrates were also documented. *Syringodium* was the dominant seagrass species found at STEER sites as opposed to *Thalassia* which was the greatest component in beds around St. Croix and St. John.

Nonparametric Wilcoxon tests and the corresponding non-parametric Dunn's multiple comparisons tests (Zar, 1999) were used to test differences among regions (JMP v11.0). On hardbottom habitat, fish species richness in STEER was similar to St. John and was significantly greater compared to St. Croix ($p=0.001$) and Southwest Puerto Rico (SWPR) ($p<0.001$) (Figure 3.54). Total biomass on hardbottom in SWPR was significantly lower than in St. Croix and St.

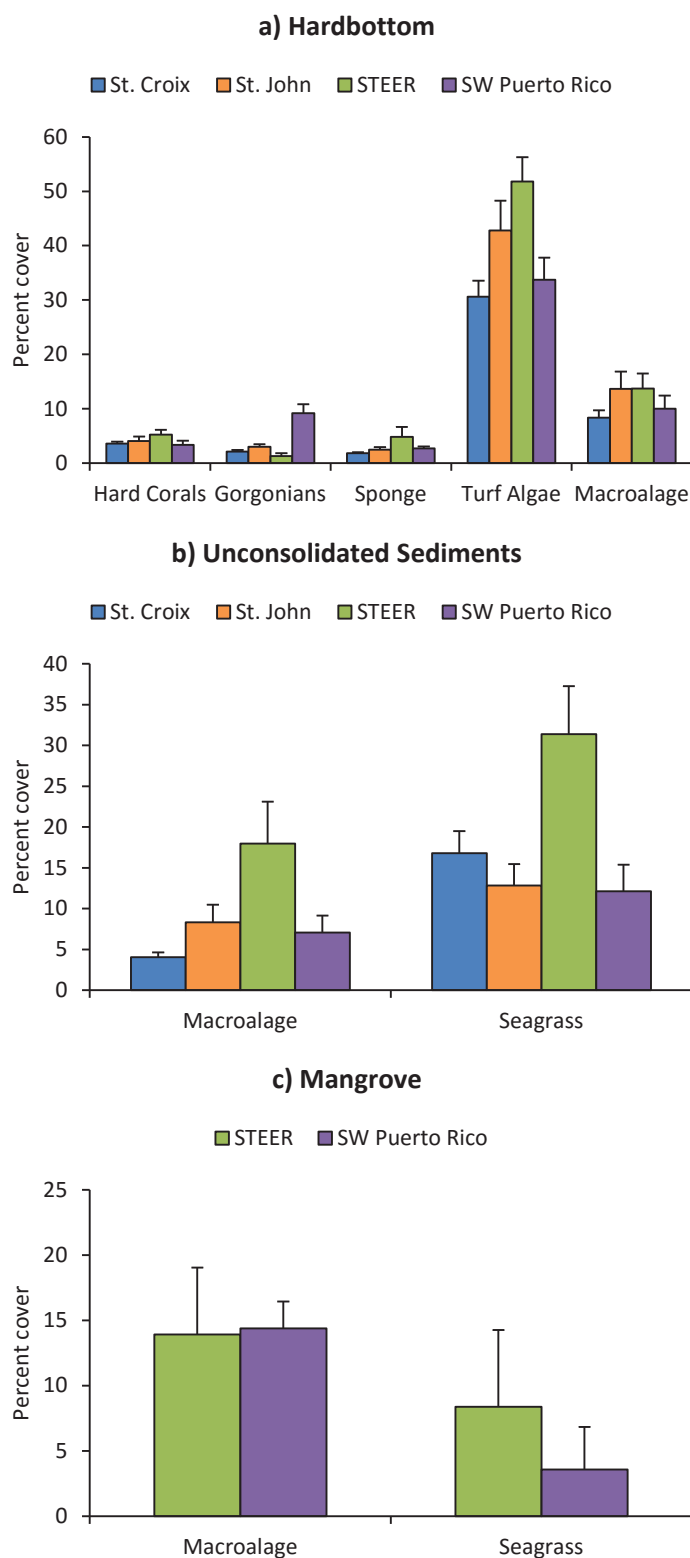


Figure 3.53. Comparison of benthic cover groups between STEER and other locations within NOAA's Caribbean Coral Reef Ecosystem Monitoring Program. Estimated mean (\pm SE) percent cover on a) hardbottom, b) unconsolidated sediments, and c) mangrove.

John, but STEER did not differ significantly from any of the other study areas (i.e., $p > 0.05$). At the family level, biomass levels were not consistent across regions. Grouper biomass did not vary significantly among regions, while grunt biomass was significantly greater on hardbottom in STEER compared to St. Croix ($p = 0.009$ and $p = 0.002$, respectively). Similarly, snapper biomass was significantly lower in St. Croix compared to STEER, SWPR, and St. John ($p < 0.01$ for all comparisons). Parrotfish biomass was

similar between all four regions with no significant differences detected.

On unconsolidated sediments, total fish biomass was significantly lower in STEER and St. John compared to NE St. Croix, due primarily to the occurrence of several southern stingrays in St. Croix in 2010. Grouper biomass did not vary significantly among regions. While grunt biomass was significantly greater in SWPR compared to St. Croix and St. John, no pairwise comparisons with STEER were signifi-

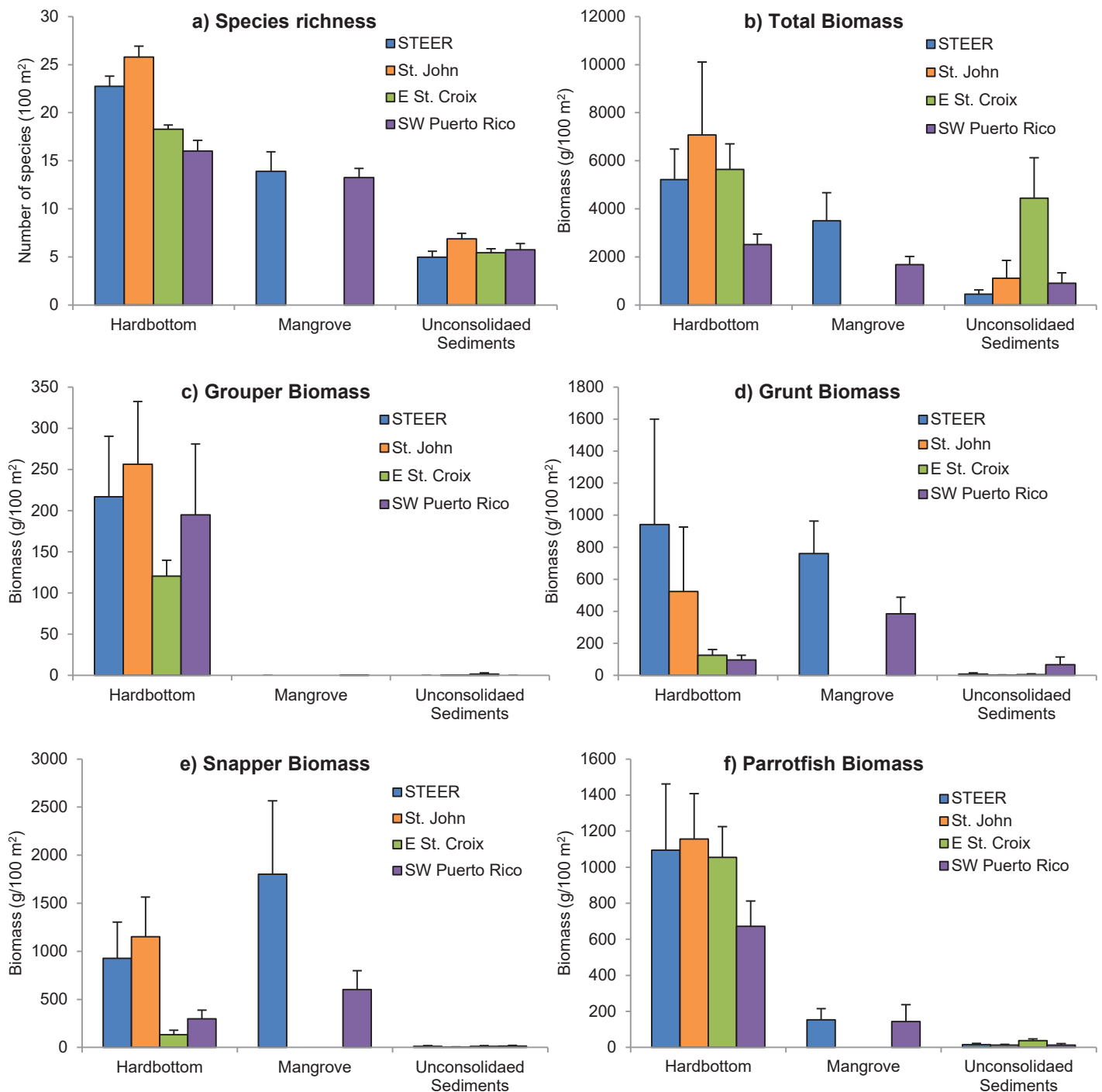


Figure 3.54. Comparison of community metrics between STEER and other locations within NOAA's Caribbean Coral Reef Ecosystem Monitoring Program. Estimated mean (\pm SE) a) species richness, b) total biomass, c) grouper biomass, d) grunt biomass, e) snapper biomass, and f) parrotfish biomass.

icant. Snapper and parrotfish biomass were similar across all study areas with no statistical differences detected. Mangroves in STEER had similar fish community metrics compared to SWPR. Although mean total biomass was larger in STEER than SWPR, there was high variance among STEER sites and results of the non-parametric tests indicated no significant difference between locations. Species richness and biomass of the other groups also did not vary significantly between STEER and SWPR.

Despite differences in fishing regulations, fish community metrics in STEER were similar to other U.S. Caribbean monitoring locations sampled with the same methodology. With a few exceptions for baitfishing and hook and line with permit, STEER is primarily a no-take reserve. Portions of the St. John (Virgin Islands National Park, Virgin Islands National Reef Monument) and St. Croix (Buck Island National Reef Monument, East End Marine Park) study areas are also no-take, while the SWPR study area is open to fishing. In addition, the maximum depth in STEER is much shallower than the other locations. On hardbottom habitat, STEER differed only from SWPR (higher species richness) and St. Croix (higher grunt and snapper biomass). The difference from St. Croix may be partially due to differences in habitat as eastern St. Croix lacks the extensive mangroves that in STEER appear to serve as important nursery areas for juvenile fishes in these families. No metrics were significantly different between STEER and nearby St. John. Although this study focused on broad community metrics for this assessment, more in-depth analysis could be conducted to look for differences at the species level, or to examine how factors such as fishing pressure and other anthropogenic factors, depth, and habitat complexity affect community composition across the USVI and Puerto

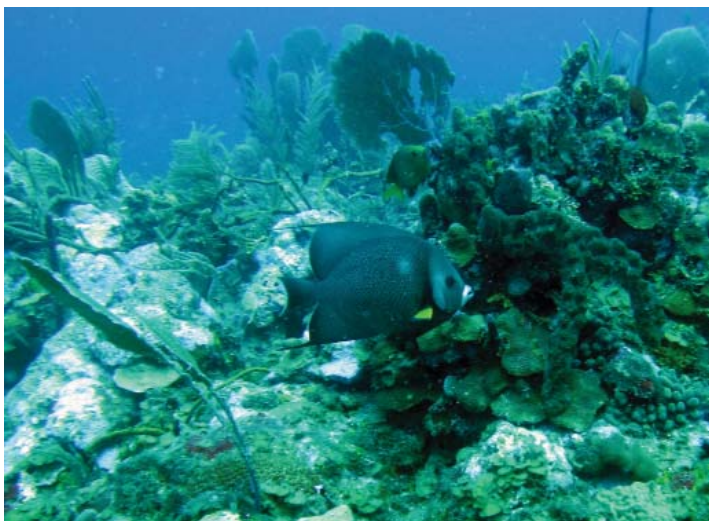
Rico. For instance, although fishing is greatly restricted in STEER, poaching may still occur (Dillard and D'lorio, 2012). At the same time, fewer snapper are generally taken on the south shore of St. Thomas and St. John due to concerns about ciguatera poisoning (Smith *et al.*, 2011).

3.4. CONCLUSIONS

This study provides a baseline characterization of the fish and benthic communities of STEER necessary to implement and assess effectiveness of the comprehensive management plan (STEER, 2011) and potential watershed restoration activities (Horsley Witten, 2013a). STEER includes an interconnected mosaic of marine habitats, including the largest remaining mangrove system in St. Thomas (IRF, 1993), extensive beds of submerged aquatic vegetation, and coral reef communities. The establishment of STEER enables this complex area to be managed as one comprehensive unit rather than the existing individual reserves (STEER, 2011).

In general, both benthic and fish community metrics in the STEER were similar to other U.S. Caribbean monitoring locations sampled with the same methodology. The low coral cover observed in the STEER reflects the significant decline in corals seen throughout the Caribbean, likely due to a number of factors including disease and hurricane damage. Surveys conducted in the southern portion of Mangrove Lagoon and adjacent coral reefs, however, ranked higher in regards to hard coral cover, fish species richness, total fish density and total fish biomass. Further research is needed to fully examine the current and potential nursery function of the entire lagoon.

The STEER encompasses a unique seascape in the USVI while possibly being one of the most impacted due to the high population density in the watershed. In addition to changes in the coral reefs as previously noted, historical studies also indicate changes in the mangrove and seagrass communities. Grigg *et al.* (1971) noted an increase in shoreline development and boating activity and associated poor water quality in Benner Bay. The authors also expressed concern about increasing anthropogenic stress on Mangrove Lagoon and Benner Bay, where turbidity and other water quality parameters varied with fluctuations in storm runoff and tidal cycles. In addition to the increasing development and maritime activities, at that time, a sewage treatment plant also discharged effluent directly into the inner Mangrove Lagoon, which receives little flushing (Grigg *et al.*, 1971). The outfall continued to be operational until wastewater was diverted to the new Mangrove Lagoon Wastewater Treatment Plant (MLWTP) in 2003, which discharges into the ocean off Long Point (DPNR, 2003). There



Gray Angelfish (*Pomacanthus arcuatus*) in STEER.

have been notable changes in the seascape of the Mangrove Lagoon complex over time. Whereas seagrass covered much of the seafloor in the lagoon in the 1970s, macroalgae presently dominates the benthos (Colletti, 2011).

In a recent assessment of chemical contaminants in sediments within STEER, higher levels of chemical contaminants were found in Mangrove Lagoon and Benner Bay, as well as significant sediment toxicity (Pait *et al.*, 2013). The areas where highest contaminant levels were observed were not sampled in this survey due to low visibility and water quality concerns for diver health. However, previous research indicated that the fish community in the inner Mangrove Lagoon differs from the middle and outer portions that were included in this study. In a recent trapping study, researchers from DPNR and UVI found significant differences in species diversity, species composition, and abundance of fishes between the degraded inner lagoon section and the middle/outer lagoon sections (Murray, 2009; Colletti, 2011). In particular, Colletti (2011) found that mangrove fringes in close proximity to dense seagrass/macroalgal beds and coral reefs in the southern areas had significantly higher species richness and abundance of juvenile fish than the inner bay, which was characterized by high cyanobacterial cover. Similar trends of lower fish species richness and density in the inner lagoon in comparison to middle/outer sections were found in a previous trapping study conducted from 1986-1988 (Boulon, 1992). Overall, Colletti (2011) found similar patterns in juvenile fish abundance in comparison to Boulon (1992), but fewer species were observed in comparison to the earlier study. The differences in fish community between the different sections of the lagoon may be influenced by additional factors, including contaminated water/sediments and low dissolved oxygen. Murray (2009) observed that occasionally fish caught in the inner lagoon were dead when traps were retrieved after being deployed for one day, and it was presumed that these fish died of suffocation due to the extremely low dissolved oxygen present. Insufficient food could also be a factor for species that are primarily benthic feeders (e.g., grunts) as Pait *et al.* (2013) observed diminished infaunal community in this area. The outer portion of the lagoon experiences more flushing from the false entrance to Mangrove Lagoon between Patricia and Bovoni Cays. Shoals have built up at the mouths of additional inlets (DPNR, 2003). Overall, the DPNR/UVI work and the current assessment demonstrate the importance of Mangrove Lagoon and Benner Bay as nursery habitat for snappers, grunts, parrotfish, and other species.

Protected status and water quality improvement are two approaches that may help improve coral, mangrove and



Elkhorn coral (*Acropora palmata*) and other species colonize a coral reef in STEER.

seagrass ecosystem health, but cannot mitigate the effects of the greatest global threat to coral reefs: high thermal stress (Hughes *et al.*, 2003; Bruno *et al.*, 2007; Baker *et al.*, 2008). Sustained high water temperatures in the summer often lead to bleaching events that can be followed by disease outbreaks due to the compromised health of bleached corals (Miller *et al.*, 2009). Contributing to the disease outbreaks are warmer winter temperatures that may reduce the mortality rates of coral pathogens and increase the amount of coral disease (Bruno *et al.*, 2007). Adopting a seascape-wide coral reef management approach includes protecting mesophotic and mid-shelf reefs, which are not as susceptible to bleaching or disease and could serve as “reef refuges” to maintain healthy stocks of some benthic and fish communities. These stocks would then in turn provide source populations for nearshore systems like STEER (Smith *et al.*, 2001). As part of this assessment, we also surveyed several locations just outside of the STEER boundaries that were of interest to managers. While the area to the east of STEER primarily consisted of pavement/rubble, sites on the reef complex south of Patricia Cay/Mangrove Lagoon ranked high in terms of coral cover and fish biomass. Although the shoreward portion of this reef complex is already included within STEER, this area could also be considered for inclusion in the reserve due to its ecological importance.

This study provides just one spatially comprehensive snapshot of the current conditions within STEER, so it is important that fish and benthic communities within the reserves continue to be monitored over time to meet management objectives (STEER, 2011). In addition to yearly monitoring of TCRMP permanent sites, the coral reef and hardbottom habitats in St. Thomas and STEER will contin-

ue to be surveyed every two years through NOAA's National Coral Reef Monitoring Program (NCRMP). At each survey location, fish belt transects, as used in this study, will be conducted, while line point intersect (LPI) and coral demographics transects will be used to characterize the benthic communities (NOAA, 2013 a,b). Mangroves, seagrasses, and other soft sediments will not be included in the NCRMP monitoring; however these habitats provide critical ecosystem services, including nutrient cycling, carbon sequestration, shoreline stabilization, nursery habitat for fish, and protection from wind/waves. Hence, further research and monitoring in these areas are warranted. Due to the limitations in conducting visual surveys in much of Mangrove Lagoon and Benner Bay, a repeat of the trapping study (Murray, 2009; Colletti, 2011) or alternative methods could be explored to cover areas that cannot be surveyed by visual census. In addition, periodic re-mapping of benthic habitats will capture large-scale changes in the reserve and the habitats that support benthic invertebrate and fish communities.



Seagrass bed in STEER, including turtle grass (*Thalassia testudinum*) and manatee grass (*Syringodium filiforme*).

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CHAPTER 4: AN ASSESSMENT OF CHEMICAL CONTAMINANTS, TOXICITY AND BENTHIC INFAUNA IN SEDIMENTS FROM THE ST. THOMAS EAST END RESERVES (STEER)

Anthony S. Pait¹, S. Ian Hartwell¹, Andrew L. Mason¹, Robert A. Warner¹, Christopher F. G. Jeffrey^{1,2}, Anne M. Hoffman³, Dennis A. Apeti¹, Francis R. Galdo Jr.⁴, and Simon J. Pittman^{1,5}

¹NOAA/NOS/National Centers for Coastal Ocean Science, Center for Coastal Monitoring and Assessment (CCMA), 1305 East/West Highway, Silver Spring, MD 20910

²CSS-Dynamac Consolidated Safety Services, Fairfax, VA 22030, under CSS-Dynamac Contract #EA-133C-14-NC-1384

³The Nature Conservancy, St. Thomas, USVI

⁴The University of the Virgin Islands, St. Thomas, USVI

⁵The Marine Institute, Plymouth University, United Kingdom

4.1 INTRODUCTION

This chapter covers the results of the chemical contaminant analysis in sediments, sediment bioassays, and benthic infaunal analysis in the STEER. In this phase of the project, 185 chemical contaminants, including a number of organic (e.g., hydrocarbons and pesticides) and inorganic (e.g., metals) compounds, were analyzed in sediments from 24 sites in the STEER. Sediment toxicity was characterized using a series of bioassays including amphipod mortality, sea urchin fertilization impairment, and the cytochrome P450 Human Reporter Gene System (HRGS), along with a characterization of the benthic infaunal community.

The assessment of contaminants throughout the STEER was one of the assessment priorities voiced by environmental managers in the STEER, during initial discussions on the proposed project. Because of the presence of a number of potential sources of pollution including the Bovoni Landfill, marinas and boatyards, commercial/industrial operations, an EPA Superfund site, and residential areas served primarily by septic systems, it was thought that chemical contaminants and associated toxicity on resident biota might be an issue impacting the health and productivity of the STEER, including fisheries nursery areas. Additional background information along with data (e.g., appendices) from this part of the project can be found in Pait *et al.* (2013), and online at: <http://coastalscience.noaa.gov/projects/detail?key=19>.



The Bovoni Landfill on the western end of the STEER receives solid waste from both St. Thomas and St. John.

The SQT has been developed to assess the presence and impacts of chemical contaminants in benthic habitats (Chapman *et al.*, 1987). Additional information on each of these three components follows.

Overview of the Chemical Contaminants

The quantification of chemical contaminants in sediments provides the opportunity for understanding what chemical stressors are present, their concentrations, how these concentrations compare to established sediment quality guidelines, along with providing input to the other two components of the triad. Each of the contaminant classes

analyzed for this project are discussed below.

Polycyclic Aromatic Hydrocarbons. Also referred to as PAHs, polycyclic aromatic hydrocarbons are associated with the use and combustion of fossil fuels (e.g., oil and gas) and other organic materials (e.g., wood and trash). Natural sources of PAHs include forest fires, and the decay of vegetation. The PAHs analyzed are two to six ring aromatic compounds. A number of PAHs bioaccumulate in aquatic and ter-

restrial organisms, are toxic, and some including benzo[a]pyrene, benz[a]anthracene, chrysene, benzo[b]fluoranthene, benzo[k]fluoranthene, dibenzo[a,h]anthracene, and indeno[1,2,3-c,d]pyrene, are likely carcinogens (USDHHS, 1995).

Aliphatic Hydrocarbons. In addition to the PAHs, another group of hydrocarbons, the aliphatics were analyzed in the sediments. Aliphatic hydrocarbons are characterized by straight chain or branched nonaromatic structures. Ali-

Sediment Quality Triad

Chemical contaminant analysis in sediments, along with sediment bioassays and benthic infaunal analysis make up what is referred to as the Sediment Quality Triad (SQT).

phatic hydrocarbons are often associated with uncombusted fuels such as gasoline, diesel or oil.

Polychlorinated Biphenyls. Commonly referred to as PCBs, polychlorinated biphenyls are synthetic compounds that have been used in numerous applications ranging from electrical transformers and capacitors, to hydraulic and heat transfer fluids, to pesticides and in paints. Approximately 60 percent of PCBs manufactured in the U.S. were used in electrical applications (EPA, 1997). PCBs have a biphenyl ring structure (two benzene rings with a carbon to carbon bond) and a varying number of chlorine atoms. There are 209 PCB congeners.

PCBs were manufactured in the U.S. between 1929 and 1977. In the United States, all PCBs were produced by a single manufacturer, and the commercial products were referred to as Aroclors. Aroclors are mixtures of PCB congeners. The manufacture of PCBs in the U.S. was banned in 1979 due to their toxicity. Because PCBs bioaccumulate and degradation in the environment proceeds only slowly, they are now ubiquitous contaminants. Exposure to PCBs in fish has been linked to reduced growth, reproductive impairment and vertebral abnormalities (EPA, 1997).

Organochlorine Pesticides. Beginning in the 1950s and continuing in to the early 1970s, a series of chlorine containing hydrocarbon insecticides were used to control mosquitoes and agricultural pests. One of the best known of the organochlorine pesticides was the insecticide DDT (dichlorodiphenyltrichloroethane).

The use of many of the organochlorine pesticides, including DDT, was banned due to their environmental persistence, potential to bioaccumulate, and in particular chronic effects on nontarget organisms. Organochlorine pesticides are typically neurotoxins, and DDT along with PCBs have also been shown to interfere with the endocrine system (Rogen and Chen, 2005). DDT and its metabolite DDE, for example, were specifically linked to eggshell thinning in birds, particularly raptors, but also in pelicans (Lincer, 1975). A number of organochlorine pesticides are toxic to nontarget aquatic life as well, including crayfish, shrimp and some species of fish. While DDT was banned by the EPA for most uses in the U.S. in 1972, it is still effectively used in some developing countries, particularly the inside of living areas, to help control mosquitos that can transmit malaria.

Most uses of the organochlorine insecticide chlordane were banned in 1978, and all uses were banned by 1988. A primary non-agricultural use of chlordane was in the treatment of wooden structures to prevent damage by termites.

Because of their persistence and heavy use in the past, residues of organochlorine pesticides can be found as well in the environment, including in biota. The persistence of these compounds and toxicity to nontarget organisms continues to be an environmental concern.

Butyltins. This compound class has a range of uses, from biocides to catalysts to glass coatings. In the 1950s, tributyltin, or TBT, was first shown to have biocidal properties (Evans, 1970; Bennett, 1996). In the late 1960s, TBT was incorporated into an antifoulant paint system, quickly becoming one of the most effective paints ever used on boat hulls (Birchenough *et al.*, 2002). TBT was incorporated into a polymer paint system that released the biocide at a constant and minimal rate, to control fouling organisms such as barnacles, mussels, weeds, and algae (Bennett, 1996).

TBT was linked to endocrine disruption, specifically an imposex (females developing male characteristics) condition in marine gastropods, and in other mollusks (e.g., oysters), abnormal shell development, and poor weight gain (Batley, 1996). Beginning in 1989, the use of TBT as an antifouling agent was banned in the U.S. on non-aluminum vessels smaller than 25 meters in length (Gibbs and Bryan, 1996). In a survey of TBT in the USVI, Strand *et al.* (2009) found evidence of elevated levels of TBT and its degradation products in gastropod species, as well as imposex organisms at several locations, including the harbor in Charlotte Amalie Bay, St. Thomas.

In the aquatic environment, TBT is degraded by microorganisms and sunlight (Bennett, 1996). The transformation involves sequential debutylization resulting in dibutyltin, monobutyltin, and finally inorganic tin (Batley, 1996).

Major and Trace Elements. All the major and trace elements occur naturally to some extent in the environment. Aluminum, iron, and silicon are major elements in the Earth's crust. As their name implies, trace elements occur at lower concentrations in crustal material, however, mining and manufacturing processes along with the use and disposal of products containing trace elements can lead to elevated concentrations in the environment. Some trace and major elements are essential micronutrients, however, a number of trace elements are toxic at low concentrations.

Cadmium is used in a number of applications, including nickel-cadmium (Ni-Cd) batteries, paint pigments and in electroplating (Ellor and Stemniski, 2007). Cadmium has been shown to impair development and reproduction in several invertebrate species, and osmoregulation in herring larvae (USDHHS, 1999; Eisler, 1985). Mercury is volatile

and can enter the atmosphere through processes including mining, manufacturing, combustion of coal and volcanic eruptions (Eisler, 1987). Mercury is currently used in compact and other fluorescent light bulbs, electrical switches and relays, thermostats and in some dental amalgams. Effects of mercury on copepods include reduced growth and rates of reproduction (Eisler, 1987).

Chromium is used in stainless steel production, chromium plating, and as a pigment, and has been shown to reduce survival and fecundity in the cladoceran *Daphnia magna*, and reduced growth in fingerling chinook salmon (*Oncorhynchus tshawytscha*) (Eisler, 1986).

Copper has many applications including use in wire, electronic circuits, antifouling paints for boat hulls, copper plumbing, industrial catalysts, and in a number of alloys (e.g., brass). Copper sulfate is used in agriculture and as an anti-algal agent, although it is probably unlikely copper sulfate is used to any great extent in St. Thomas, as there appears to be little agriculture in the watershed. While an essential biological element, elevated levels of copper can impact aquatic organisms, including the functioning of gills along with reproduction and development (Eisler, 1998).



Marina area in Benner Bay.

Most of the current uses of lead appear to be in lead-acid batteries, although other uses include oxides in glass and ceramics. In the past, lead was used in paints and also in gasoline, however, these uses have ended due to environmental and human health concerns. Nickel has many applications in both industrial and consumer products. Approximately 65% of the nickel in the U.S. is used to make stainless steel. Other uses include its incorporation into a series of alloys, in rechargeable batteries (Ni-Cd), catalysts, coins, plating, and in foundry products. Corrosion-resistant zinc plating of steel (hot-dip galvanization) is an important application, accounting for roughly 50% of zinc use. In the marine industry, zinc anodes are used to protect vital engine and boat parts (e.g., propellers, struts and rudders, along with outboard and inboard engines), and is a component in some antifoulant paint formulations. Zinc is also used in batteries, and in alloys such as brass. Lead, nickel and zinc have all been shown to impact fertilization success in corals, some effects being observed in the parts per billion range (Reichelt-Brushett and Harrison, 1999; Reichelt-Brushett and Harrison, 2005). If not included

in this document, results from the analysis of additional trace and major elements, can be found in Pait *et al* (2013), and online at: <http://coastalscience.noaa.gov/projects/detail?key=19>.

Bacterial Indicator: Although not a chemical contaminant, the bacterium *Clostridium perfringens* has been used as an indicator of fecal pollution and was analyzed in the sediment samples from the STEER. This bacterium occurs in the intestines of humans and in some domestic and feral animals, and is a common cause of food poisoning.

Sediment Toxicity Tests

NOAA's NCCOS, National Status and Trends (NS&T)

Bioeffects Program routinely uses a suite of tests to assess sediment toxicity through different modes of contaminant exposure (bulk sediment, sediment porewater, and chemical extracts of contaminants from sediment) to a variety of species (invertebrates, vertebrate cells and bacteria) and different assessment end-points (i.e., mortality, impaired reproduction, physiological stress, and enzymatic response). Since the test results are not necessarily axiomatic and biological effects of contaminants occur at different levels of biological organization, i.e., from cells to ecosystems, results from a suite of toxicity tests are used in the "weight of evidence" context to infer the incidence and severity of environmental toxicity (Chapman, 1996). The toxicity bioassays used in this project included amphipod (*Ampelisca abdita*) mortality, sea urchin (*Arbacia punctulata*) fertilization impairment, and cytochrome P450 Human Reporter Gene System (HRGS).

Benthic Infaunal Analysis. Mixtures of synthetic organic compounds (e.g., PCBs and DDT), metals, PAHs, excess nutrients, and various inorganic chemicals are released into the ocean from municipal and industrial point sources, atmospheric deposition, stormwater runoff, spills, and groundwater. These anthropogenic contaminants may accumulate in the sediment in coastal bays, estuaries, and nearshore coastal zones.

Two of the most influential parameters in the distribution of benthic communities are salinity and sediment grain size. Environmental concentrations of organic enrichment and toxicants are often confounded in space and time with

gradients of salinity and grain size, making their separate and combined biological effects difficult to detect, especially at the levels of population and community. However, understanding toxic hazard due to sediment contamination by means of community assessment is valuable. Biological systems integrate the complexity of natural habitat stressors and ambient pollutant mixtures, through physical contact with sediments, ingestion of sediment, and the bioaccumulation of contaminants via food webs, along with the synergetic effects of exposure to multiple toxic chemicals.

Many examples exist in which marine benthic communities' response to contaminant and physical stressors have been documented (Hartwell and Claffin, 2005; Hartwell and Hameedi, 2007; Hartwell *et al.*, 2009; Oliver *et al.*, 2011; Wlodarska-Kowalczyk *et al.*, 2005). Impacts of contamination on marine benthos have shown that total biomass, relative proportion of deposit feeders, and abundance of species with 'opportunistic' life histories (e.g., high fecundity, short generation time, and rapid dispersal) increase with increasing organic enrichment. Some opportunistic taxonomic groups are known to be tolerant of chemical toxicants. Others are capable of thriving in physically disturbed habitats (e.g., high sedimentation, dredging operations, etc), but not necessarily in contaminated areas.

In areas impacted by excessive sedimentation from terrestrial runoff, dominant organisms tend toward surface suspension feeding modes and high reproductive potential regardless of taxonomic relationship, whereas away from the sedimentation stress, feeding modes shift to species that are deep deposit feeders along with the emergence of filter feeders. Experimental manipulation of habitats have shown that polychaete worms, in specific taxonomic lines, with opportunistic life history strategies respond positively to organic enrichment (Fleeger *et al.*, 2003). Infaunal arthropods respond negatively to toxicants and organic enrichment. The response of specific arthropod and echinoderm species to organic and toxic contamination is mediated by life history and feeding mode characteristics. Finally, the benthic community will respond to management actions that affect physical and chemical stressors in vastly shorter time frames than will coral reefs.

4.2 METHODS

The sampling strategy for sediments was developed in meetings with the STEER Core Planning Group. A strati-

fied random sampling design was selected, which is a standard benthic assessment technique. The STEER was first subdivided into five strata based on habitat and geography (e.g., hard bottom areas, seagrass beds, mangroves, etc) (Figure 4.1). Five sampling points on soft bottom sediments were then randomly selected using ArcGIS®. Both primary and alternate sites were identified throughout the STEER. Alternate sites were sampled in the event that a primary site was unsuitable due to hard bottom, obstructions, etc.

The stratified random sampling design was used in order to characterize the spatial distribution of chemical contaminants, toxicity and the benthic infaunal community (organisms living within the sediment) throughout the STEER. Using this design, the extent and concentration of chemical

contaminants and bioeffects can be compared between strata. The 2011 collection of sediment samples in the STEER occurred 14 - 17 June. The samples were collected under DPNR Permit STX-032-11.

All sediment sites were located using a GPS programmed with the site coordinates. Most of the sediment samples were collected from the charter vessel *Bright Star*, however, Mangrove Lagoon was too shallow for this vessel. To enable the collection of samples

in that stratum, personnel from the DPNR Division of Fish and Wildlife brought in a shallow draft motor boat to help NOAA personnel collect the sediment samples in Mangrove Lagoon.

In addition to the work in 2011, preliminary field work took place in the STEER in May 2010. From that effort, a total of 13 sediment samples were taken, and enough funding was available at the time to analyze four of the samples collected. The results of the analysis of these samples is also included in this report. However, as the four samples were collected from targeted sites and were not selected randomly, they could not be included in the statistical comparisons between strata, nor were they included in the calculation of the mean (average) concentration of sediment contaminants in the STEER.

Sampling Protocols

A PONAR grab (see inset) was deployed to collect the samples using a pulley and davit, and retrieved by hand. Rocks and bits of seagrass were removed. If a particular grab did not result in 200-300 g of sediment, a second grab was made and composited with material from the first. If



The PONAR grab used to take sediment samples in the STEER.

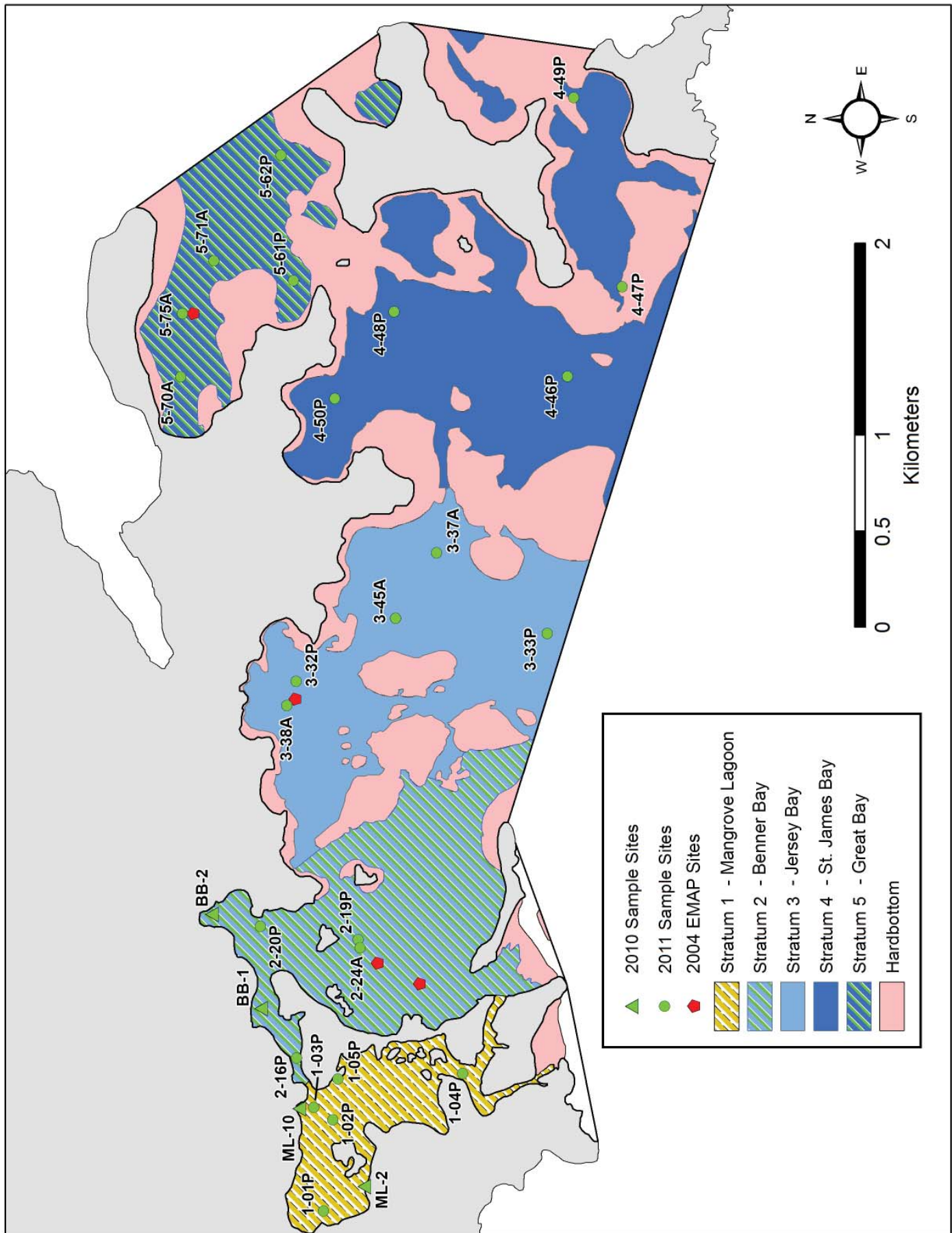


Figure 4.1. Strata and sediment sites sampled in the St. Thomas East End Reserves (STEER) in June 2011.

enough sediment had not been collected after three deployments of the grab, the site was abandoned and the boat moved on to an alternate site.

A series of protocols (Apeti *et al.*, 2012a) were used to avoid contamination of the sediment samples by equipment and cross contamination between samples and sites. All equipment was rinsed with acetone and then distilled water just prior to use at a site. Personnel handling the samples also wore disposable nitrile gloves. The top 3 cm of sediment were collected from the grab using a stainless steel sediment scoop. This top layer of sediment is referred to as surficial sediment, and is typically indicative of recent deposition.

Sediments were placed into two certified clean (I-Chem®) 250 ml labeled jars, one for organic chemical analysis, the other for major and trace element analysis, capped and then placed on ice in a cooler. Sediments for grain size analysis were placed in a WhirlPack® bag, sealed and placed on ice in a cooler. At the end of each day, sediment samples for contaminant analysis were placed in a freezer. The Whirl-Pack® bags for the grain size analysis were placed in a refrigerator rather than frozen, to avoid altering the grain size structure of the sediment.

A series of water parameters (dissolved oxygen, temperature, salinity, and conductivity) were also measured at each site, using a YSI® salinity/conductivity/temperature meter. The instrument probe was submerged to a depth of approximately 0.5 meter for the surface measurement, and within a meter of the sediment for the bottom measurement. Secchi depth was also measured at each site.

The sediment samples collected were analyzed for a suite of 185 organic (e.g., hydrocarbons and pesticides) and inorganic (e.g., metals) contaminants by TDI-Brooks International, using protocols from the NS&T Program. The list of chemical contaminants analyzed in the sediments is shown in Table 4.1. The 59 polycyclic aromatic hydrocarbons (PAHs) were analyzed using gas chromatography/mass spectrometry in the selected ion monitoring mode. The 37 aliphatic hydrocarbons were analyzed by gas chromatography/flame ionization detection. The 31 organochlorine pesticides and 38 polychlorinated biphenyls (PCBs) were analyzed using gas chromatography/electron capture detection. The four butyltins were analyzed using gas chromatography/flame photometric detection after derivatization. A subset of the sediment samples were subsequently reanalyzed using gas chromatography/mass spectrometry for confirmation of the TBT concentrations detected at certain sites from the initial analysis. Seventeen

major and trace elements were analyzed. The major and trace elements were analyzed using inductively coupled plasma mass spectrometry and atomic-fluorescence spectroscopy. Detailed descriptions of the NS&T protocols, including quality assurance/quality control (QA/QC) used in the analysis of the organic contaminants, can be found in Kimbrough *et al.* (2006); for inorganic analyses, Kimbrough and Lauenstein (2006).

Statistical Analysis for Chemical Contaminants

The sediment contaminant data were analyzed using JMP® statistical software. A Shapiro-Wilk test was first run on individual parameters to see if the data were normally distributed. When data were normally distributed, an Analysis of Variance (ANOVA) was run followed by pairwise (Tukey HSD) comparisons. If the data were not normally distributed and a log10 transformation was not effective, Spearman's nonparametric multivariate correlation was used. Some of the data were also subsequently ranked, followed by a Kruskal-Wallis test and pairwise comparisons. The statistical analyses were used, for example, to compare differences in contaminant levels between strata.

NOAA numerical sediment quality guidelines (SQG) developed by Long and Morgan (1990) and Long *et al.* (1995), known as ERM (Effects Range-Median), and ERL (Effects Range-Low), express statistically derived levels of contamination, above which toxic effects would be expected with at least a 50% frequency (ERM), and below which effects were rarely (<10 %) expected (ERL). The ratio of the ERM value to the sediment concentration for each chemical is called the ERM quotient or ERMq (Long *et al.*, 1998). The quotient expresses how close measured concentrations are to the ERM level on a zero to one scale. A quotient of one or greater means the concentrations are at or above the ERM. This also normalizes the ERMs for different chemicals to a common scale. The mean ERMq of all the contaminants averaged together expresses a measure of contamination across the entire spectrum of measured contaminants. Field research suggests that a mean ERMq value of 0.1 is a threshold where degraded communities begin to be seen, as observed in the southeast U.S. (Hyland *et al.*, 1999). The mean quotient of the ERMs and observed contaminant concentrations were calculated on a site by site basis.

Sediment Toxicity Bioassays

The protocols for the bioassays were based on standard methods, as outlined by the U.S. EPA (1999, 2002a) and ASTM (2008). Sediment samples for the bioassays were collected into three containers. Samples for the amphipod toxicity tests (bulk sediment) were collected in 1 liter (L)

Table 4.1. Compounds analyzed in sediments from the St. Thomas East End Reserves.

PAHs - Low MW	PAHs - High MW	Aliphatics	Organochlorine Pesticides	PCBs	Major and Trace Elements
Naphthalene	Fluoranthene	n-C9	Aldrin	PCB8/5	Aluminum (Al)
1-Methylnaphthalene	Pyrene	n-C10	Dieldrin	PCB18	Antimony (Sb)
2-Methylnaphthalene	C1-Fluoranthenes/Pyrenes	n-C11	Endrin	PCB28	Arsenic (As)
2,6-Dimethylnaphthalene	C2-Fluoranthenes/Pyrenes	n-C12	Heptachlor	PCB29	Cadmium (Cd)
1,6,7-Trimethylnaphthalene	C3-Fluoranthenes/Pyrenes	n-C13	Heptachlor-Epoxide	PCB31	Chromium (Cr)
C1-Naphthalenes	Naphthobenzothiophene	i-C15	Oxychlorane	PCB44	Copper (Cu)
C2-Naphthalenes	C1-Naphthobenzothiophenes	n-C14	Alpha-Chlordane	PCB45	Iron (Fe)
C3-Naphthalenes	C2-Naphthobenzothiophenes	i-C16	Gamma-Chlordane	PCB49	Lead (Pb)
C4-Naphthalenes	C3-Naphthobenzothiophenes	n-C15	Trans-Nonachlor	PCB52	Manganese (Mn)
Benzothiophene	Benz[a]anthracene	n-C16	Cis-Nonachlor	PCB56/60	Mercury (Hg)
C1-Benzothiophenes	Chrysene	i-C18	Alpha-HCH	PCB66	Nickel (Ni)
C2-Benzothiophenes	C1-Chrysenes	n-C17	Beta-HCH	PCB70	Selenium (Se)
C3-Benzothiophenes	C2-Chrysenes	Pristane	Delta-HCH	PCB74/61	Silicon (Si)
Biphenyl	C3-Chrysenes	n-C18	Gamma-HCH	PCB87/115	Silver (Ag)
Acenaphthylene	C4-Chrysenes	Phytane	DDMU	PCB95	Tin (Sn)
Acenaphthene	Benzo[b]fluoranthene	n-C19	2,4'-DDD	PCB99	Zinc (Zn)
Dibenzofuran	Benzo[k]fluoranthene	n-C20	4,4'-DDD	PCB101/90	
Fluorene	Benzo[e]pyrene	n-C21	2,4'-DDE	PCB105	
C1-Fluorenes	Benzo[a]pyrene	n-C22	4,4'-DDE	PCB110/77	
C2-Fluorenes	Perylene	n-C23	2,4'-DDT	PCB118	
C3-Fluorenes	Indeno[1,2,3-c,d]pyrene	n-C24	4,4'-DDT	PCB128	
Carbazole	Dibenzo[a,h]anthracene	n-C25	1,2,3,4-Tetrachlorobenzene	PCB138/160	
Anthracene	C1-Dibenzo[a,h]anthracenes	n-C26	1,2,4,5-Tetrachlorobenzene	PCB146	
Phenanthrene	C2-Dibenzo[a,h]anthracenes	n-C27	Hexachlorobenzene	PCB149/123	
1-Methylphenanthrene	C3-Dibenzo[a,h]anthracenes	n-C28	Pentachlorobenzene	PCB151	
C1-Phenanthrene/Anthracenes	Benzo[g,h,i]perylene	n-C29	Pentachlorobenzene	PCB153/132	
C2-Phenanthrene/Anthracenes		n-C30	Endosulfan II	PCB156/171/202	
C3-Phenanthrene/Anthracenes		n-C31	Endosulfan I	PCB158	
C4-Phenanthrene/Anthracenes		n-C32	Endosulfan Sulfate	PCB170/190	
Dibenzothiophene		n-C33	Mirex	PCB174	
C1-Dibenzothiophenes		n-C34	Chlorpyrifos	PCB180	
C2-Dibenzothiophenes		n-C35		PCB183	
C3-Dibenzothiophenes		n-C36	Butyltins	PCB187	
		n-C37	Monobutyltin	PCB194	
		n-C38	Dibutyltin	PCB195/208	
		n-C39	Tributyltin	PCB201/157/173	
		n-C40	Tetrabutyltin	PCB206	
				PCB209	

Abbreviations: MW, molecular weight; PAH, polycyclic aromatic hydrocarbons; HCH, hexachlorocyclohexane; DDMU, 1-chloro-2,2- (p-chlorophenyl)ethylene, DDT, dichlorodiphenyltrichloroethane; DDD, dichlorodiphenyldichloroethane; DDE, dichlorodiphenyldichloroethylene; PCB, polychlorinated biphenyl

jars; for the sea urchin fertilization tests (porewater extraction), a 3.79 liter (one gallon) sample of sediment was collected; and for the P450 test, a sample of sediment was taken out of the 250 ml organics jar and extracted.

Amphipod Toxicity Test

The whole sediment toxicity bioassay test is commonly used in North America for assessing sediment quality, in part because the test integrates the effects of complex contaminant mixtures in relatively unaltered sediment, and also because amphipods are fairly common and an ecologically important species in coastal waters. The organisms are standard test species with known ranges of sensitivity and their presence or absence in a particular habitat is not relevant because they are tested under standardized conditions. Results of increased mortality, significantly different from controls, is considered an indicator of marginal toxicity. Results that are significantly different from controls and greater than 20% mortality is indicative of highly toxic conditions (Thursby *et al.*, 1997).

Sea Urchin Fertilization Test

The sea urchin (*A. punctulata*) fertilization toxicity test (also known as the sperm cell test) involves exposing sea urchin sperm to sediment pore water (interstitial water), followed by the addition of eggs. This test is used extensively in assessments of ambient water quality, toxicity of industrial and municipal effluents, and sediment toxicity in coastal waters. It combines the features of testing sediment pore waters (the phase of sediments in which dissolved toxicants may be bioavailable) and exposures of gametes which often are more sensitive than adult organisms. Increased fertilization failure which is significantly different from controls is considered symptomatic of marginal toxicity. Results that are significantly different from controls and greater than 20% below control fertilization is indicative of highly toxic conditions (Carr and Bidenbach, 1999).

P450 Test

The Human Reporter Gene System (HRGS) P450 test was used to determine the presence of toxic organic compounds in the sediments. Cytochrome P450s are a family of membrane-bound enzymes that metabolize a diverse number of compounds, including natural substrates, drugs, hormones, and many toxic compounds. They are present in a wide variety of animals, plants and other organisms. P450 is shorthand for Pigment and 450 is the wavelength at which they most strongly absorb light. In this case, the HRGS reporter gene is a DNA sequence in a human cancer cell line that has been genetically engineered to include a gene (the reporter gene) from the firefly that produces luciferase, the chemical that produces light in the insect when

presented with the proper substrate. The gene is spliced into the region of the DNA strand that is activated to produce P450 enzymes when the cell is exposed to chemicals that stimulate metabolic activity. The more stimulated the cell is to metabolize a foreign compound, the more the reporter gene produces luciferase, which can be measured by increased light output.

Different compounds stimulate P450 production to differing degrees, which can be calibrated. PCBs and PAHs stimulate certain Cytochrome P450 enzymes (e.g., CYP1A), but each individual compound exhibits its own level of stimulation. Heavy metals do not stimulate P450 at all. Under appropriate test conditions, induction of CYP1A is evidence that the cells have been exposed to one or more xenobiotic organic compounds, including dioxins, furans, planar PCBs, and several PAHs. When run in parallel with a serial dilution of standard PAH toxicant benzo[a]pyrene (BaP), or TCDD (dioxin), test results can be expressed in terms of standard toxicant equivalents based on the relative reporter gene response. Samples that exhibited a response greater than 50% of a standard 10 nM TCDD threshold control were again tested against a B[a]P serial dilution to calculate responses normalized to the B[a]P EC50 (effective concentration for 50% of the test cells) or the B[a]P equivalents (B[a]P eq).

Anderson *et al.* (1999a) calculated the mean and 95% confidence interval of HRGS values from 527 sampling points in the NOAA biological effects database to be 22.7 ± 10.1 (CI=12.6-32.8) mg B[a]P Eq/kg. Hence, values less than 12.6, forming the tail of the distribution in the direction of low induction (or impact) could be interpreted as a minimal (background) level. This is consistent with data from pristine sites in Alaska and California where HRGS values did not exceed 10.4 mg B[a]P Eq/kg (Anderson *et al.*, 1999b; Fairey *et al.*, 1996). Fairey *et al.* (1996) also demonstrated that HRGS values above 60 mg B[a]P Eq/kg were highly correlated with degraded benthic communities in San Diego and Mission Bays, and with PAH concentrations above the 9,600 $\mu\text{g/kg}$ Probable Effects Level (PEL) guideline (MacDonald, 1993), which are similar to the ERLs. Based on these data, HRGS values greater than 10 and 60 mg B[a]P Eq/kg were considered to represent marginal and highly contaminated thresholds, respectively.

Differences in the ability of the P450 enzyme system to metabolize chlorinated and non-chlorinated compounds allows for differentiation between these classes of compounds in environmental samples. Since most PAHs are metabolized, they exhibit a maximum response in 6 hours, at which point the response begins to fade. Chlorinated

hydrocarbons (dioxins, furans, and certain PCBs), on the other hand, are not degraded and continue to induce CYP1A, resulting in increasing responses after 24 hours following exposure.

Benthic Infaunal Analysis

A benthic community sample was taken with the PONAR grab sampler, in addition to the samples for chemical analysis and toxicity testing. The entire contents of an acceptable grab (at least 5 cm deep) was sieved on site through a 0.5 mm mesh. In coarse sediments, nested sieves of 1.0 mm and 0.5 mm were sometimes necessary to reduce clogging of the screens and damage to the organisms. All organisms were retained in plastic containers and preserved in buffered 10% formalin containing Rose Bengal stain and sodium borate buffer.

The following data and information were recorded at each site: stratum, site, alternate (if applicable), date, water depth, time, latitude, longitude, and depth of sediment in the grab. Also included was a written description of each sampling site including digital color photographs of the site, a physical description of sediment characteristics (texture, color, odor, benthos, sheen) and photographs of the undisturbed sediment.

In the laboratory, all animals were carefully segregated into major groups (e.g., worms, clams, shrimp and crabs). They were then identified to species unless the specimen was a juvenile or damaged. At a minimum, 10% of all samples were re-sorted and re-counted on a regular basis. Also, 10% of samples were randomly selected and re-identified. The minimum acceptable sorting and taxonomic efficiency was 95%. A voucher collection composed of representative individuals of each species encountered in the project was accumulated and retained.

The benthic communities were characterized by abundance (number of animals), number of species, and diversity. Abundance was calculated as the total number of individuals per grab; species richness as the total number of species represented at a given site; and diversity was calculated

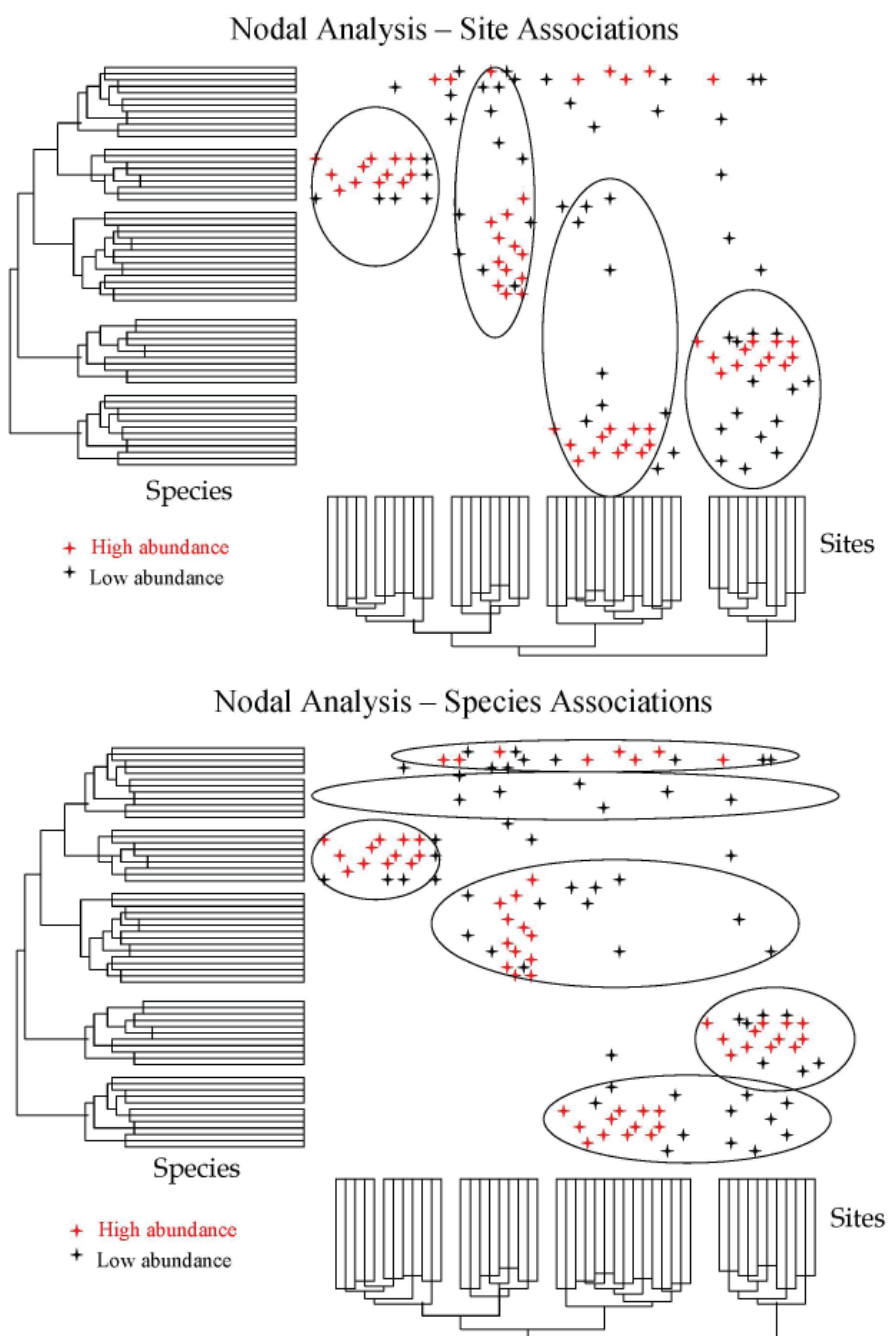


Figure 4.2. Combined cluster analysis overlays of species clusters and site clusters. The top figure illustrates the dominant species communities found in different site clusters. The lower figure illustrates how different species assemblages distribute themselves between different habitats.

with the Shannon-Weiner Index (Shannon and Weaver, 1949), using the following formula:

$$H' = -\sum_{i=1}^S p_i (\ln p_i)$$

where, S = is the number of species in the sample, i is the ith species in the sample, and

Site Cluster Characteristics

Tox	Metals	DDT	PCB	PAH	# Taxa	Abundance	TOC	GrainSize	Depth	Salinity
x	100	2	150	1050	3	300	2.5	80	2	0
x	210	0	55	3100	8	100	3	80	10	0
0	100	2	50	1080	8	400	3.5	90	12	0
x	105	1	58	2000	2	50	3	80	5	0
0	200	2	50	1900	8	250	2	93	12	0
x	100	2	50	1000	8	500	3	80	8	0

0	19	1	15	150	4	38	2.5	60	12	5
x	110	0	55	310	3	500	3	80	10	8
0	100	2	50	180	10	40	1.5	70	22	5
x	15	1	58	700	5	500	3	80	5	2
0	20	1	20	190	3	250	2	53	12	10

0	10	1	15	15	24	1380	0.5	15	32	15
0	10	0	5	20	31	500	1	10	50	18
0	10	0	2.5	18	15	4000	0.5	15	21	25
0	5	1	8	70	25	590	0.1	10	35	18

0	20	0	20	190	3	250	0.5	5	32	29
x	10	0	15	15	24	138	0.5	5	32	15
0	15	0	5	20	21	500	1	10	40	18
0	10	0	25	18	10	200	0.5	5	25	25
0	5	1	8	70	25	590	0.1	10	35	18

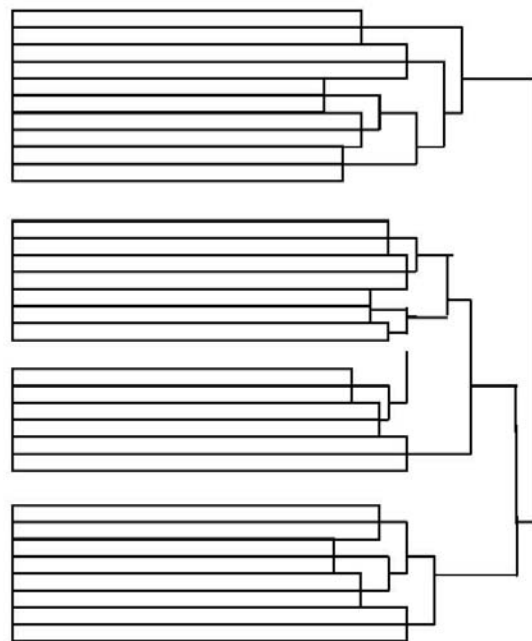


Figure 4.3. Hypothetical representation of the distribution of physicochemical habitat parameters, contaminant concentrations, and other site-specific data used to characterize site and species clusters.

p_i is the number of individuals of the i th species divided by the total number of individuals in the sample.

Nonparametric Spearman rank correlation coefficients were calculated for all parameters to assess relationships between the physical, chemical, toxicological and biological variables. Multivariate cluster analysis was employed to group site and species data. The objective was to produce a coherent pattern of association between sites and species (Figure 4.2). Cluster analysis is a two-step process including: 1) creation of a resemblance data matrix from the raw data, and 2) clustering the resemblance coefficients in the matrix. The input resemblance (similarity or dissimilarity) matrix can be created by a number of methods. Input data may or may not be standardized or transformed depending on the requirements of the method (e.g., Bray Curtis). Based on previous research (Hartwell and Clafin, 2005), the Jaccard method (Goodall, 1973) was used to generate the similarity matrix. The Jaccard method is a binary method based only on presence/absence data, and thus ignores abundance values.

Cluster analyses were calculated from the matrices using the Unweighted Pair-Group Method Using Arithmetic

Averages (UPGMA) procedure which clusters coefficients based on arithmetic mean distance calculations (Sneath and Sokal, 1973). To optimize the cluster analysis results, several manipulations of the input data were performed to remove confounding effects and bias:

- 1- Epibenthic species, such as sea anemones and tunicates, were eliminated from the data set as they are not truly infauna.
- 2- 'Artificial species' (resulting from failure to identify some specimens all the way down to species) were identified as a data bias. For example, if specimens of two to three species were identified in genus A, and other specimens were identified only to genus A, this tends to artificially increase species richness and diversity of the sample when in fact that diversity is an artifact of imperfect taxonomic identification. In some instances, specimens were only identifiable to family, order or class. To address this problem, specimens not identified to species level were eliminated, unless they were identified to a taxonomic level below which no other specimens in the collection belonged. That is, even though they were not identified to species, they were the only representative

of that taxonomic line and did represent a non-redundant taxon. In other cases, where a specimen was identified to genus and there was only one species identified in that genus, they were combined at the genus level.

3- Rare and unique species were defined as those species that were found at no more than two sites. Although they do contribute to the overall assessment of biodiversity, they were eliminated from the cluster analysis data set. Because of their limited distribution, by definition, they do not provide information on the impact of contaminant or other stressor gradients in the environment because they do not occur across a gradient.

After the data set had been finalized, a nodal analysis routine was applied to the data (Lambert and Williams, 1962). This consisted of combining independent cluster analyses in a graphical array. The first analysis clustered sites using species occurrence data. The second calculation clustered species together into groups. The intersection of site clusters on the abscissa and species clusters on the ordinate axis yields a pattern of species associations with site clusters, termed nodes (Figure 4.2). The site and species clusters were also characterized by physicochemical habitat parameters, contaminant concentrations, and other site-specific data. Plotting the tabulated values in parallel to the cluster output (Figure 4.3), allows an empirical evaluation of similarities in habitat characteristics within and between species and site clusters (i.e., what they do or do not have in common) to guide interpretation of subsequent statistical contrasts. For each species, the parameters were normalized to their abundance at each site.

Once the nodes were defined, the species data within each node were further assessed with methods developed by Clark and Warwick (2001) to assess the relative importance of a species in characterizing a set of sites in a quantitative way, called a Similar-

ity Index. Average similarity for a species is the contribution of the species to the Bray-Curtis similarity within a site group (node). This value indicates how typical the species is for the group of sites and ranges from 0 to 100%.

4.3. RESULTS AND DISCUSSION

Field Data

The collection of 25 sediment samples was planned for the June 2011 sampling mission. Twenty-nine sites were visited, however, at five sites the field team was unsuccessful in collecting sediments, resulting in sediment samples from 24 sites.

The average water depth at the sites sampled was 7.2 meters (m). As might be expected, the shallower sites were in Mangrove Lagoon. The shallowest water depth was 0.6 m at Site 1-5P in Mangrove Lagoon. The average salinity encountered on the surface of the sites was 33.2‰, the average bottom salinity 33.7‰, indicating fairly well mixed waters. The highest salinity encountered during the mission was 34.6‰, a bottom salinity reading taken at Site 5-61P; the lowest salinity encountered (28.3‰) was in Mangrove Lagoon on the surface at Site 1-1P. Additional information on field data from this phase of the STEER project by site can be found in the appendices in Pait *et al.* (2013). The average water surface temperature at the sites in the STEER

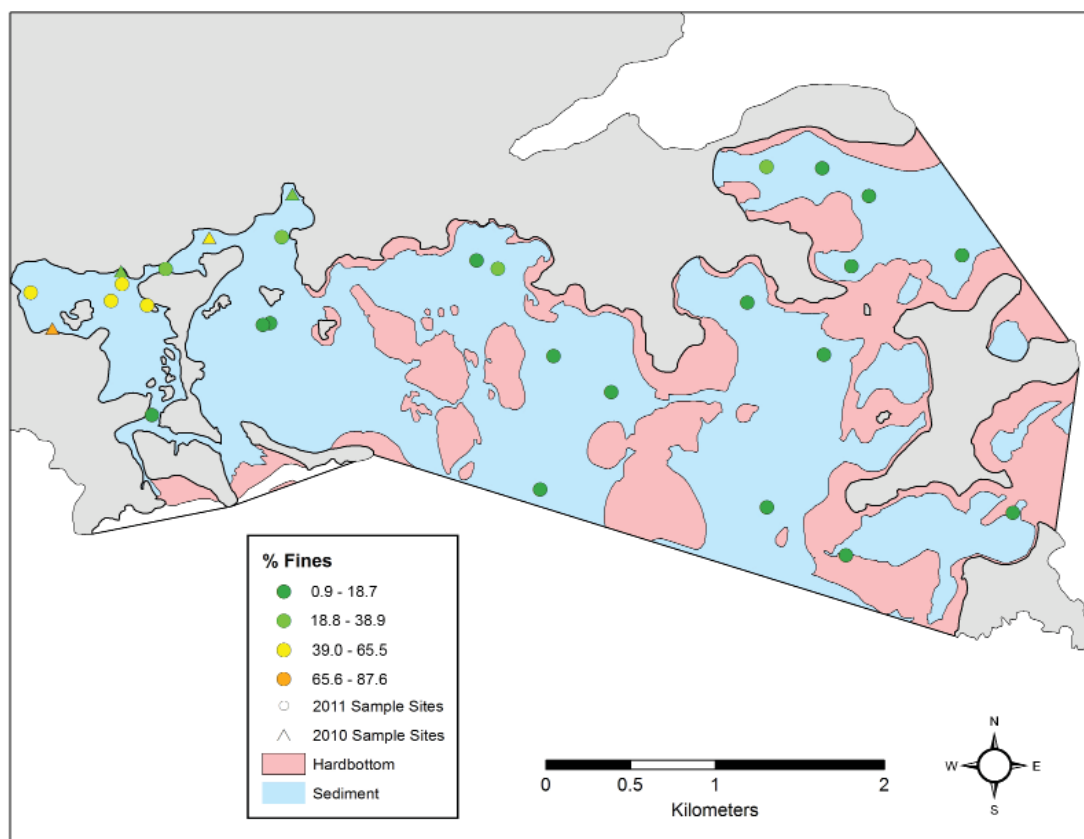


Figure 4.4. Percent fines (percent silt plus percent clay fractions) in sediments from the STEER.

was 30.1°C, the average bottom temperature was 29.5°C. A Kruskal-Wallis test run on the ranked data indicated that salinity ($p < 0.0001$) and temperature ($p < 0.0001$) varied significantly by stratum, with lower salinities and higher temperatures being found in Strata 1 and 2.

Total Organic Carbon and Grain Size

The average percent total organic carbon (TOC) in the sediments collected was 2.49%, and ranged from a low of 0.44% to a high of 5.44%. A Kruskal-Wallis test run on the ranked TOC values indicated no differences in percent TOC in the STEER by stratum ($p = 0.0645$).

Chemical contaminants, particularly organic (carbon-containing) contaminants, tend to accumulate in sediments with higher TOC values. Chemical contaminants also tend to accumulate in sediments that have a higher proportion of the smaller grain sizes (i.e., silt and clay). Smaller grain size sediments have more surface area per unit volume available for the adsorption of contaminants, and are typically found in depositional habitats. In addition, metals are attracted to sediments with higher clay content due to the charge structure on the surface of the clay particles. For this report, the percent silt and percent clay fractions are combined and referred to as percent fines (sum of the percent silt and percent clay sediment fractions). Figure 4.4 shows the percent fines in the sediments sampled in the STEER; the average content was $22.3 \pm 4.03\%$. It can be seen that areas in Mangrove Lagoon and to a certain extent in Benner Bay, had a higher proportion of percent fines in the sediments. However, the major sediment size class in the STEER was sand ($68 \pm 4.35\%$).

Chemical Contaminants

The results from the analysis of chemical contaminants in the sediments are discussed below. Additional information, including results from individual sites can be found in the appendices section in Pait *et al* (2013).

Polycyclic Aromatic Hydrocarbons

Total PAHs as used in this report refers to the sum of the 59 PAH compounds and compound classes (e.g., anthracene, C1-naphthalenes) analyzed in the STEER sediment samples. It can be seen in Figure 4.5 that there were a number of higher PAH concentrations, mainly in Mangrove Lagoon and Benner Bay, compared to the other strata and sites further offshore. As will be seen, the pattern of higher concentrations of chemical contaminants in the Mangrove Lagoon and northern Benner Bay areas was repeated for a number of the other contaminant classes analyzed.

The mean concentration of total PAHs in the sediment from the STEER was 142 ± 58 ng/g. The units ng/g (nanogram/

gram) are also referred to as parts per billion (ppb). The median total PAH concentration was 6.02 ng/g. The highest total PAH concentration was at site 1-3P, with 1,131 ng/g. In Mangrove Lagoon, the mean concentration of total PAHs in the sediments was 425 ± 214 ng/g, with the standard error indicating a wide range in the concentrations in this stratum (Stratum 1). The lowest concentration of total PAHs was found in Stratum 4, at site 4-49P (0.93 ng/g) near St. James Island. From the preliminary sampling for this project in 2010, the highest concentration of total PAHs was found at the ML-10 site in Mangrove Lagoon, at 951 ng/g.

Variation of Total PAHs Across Strata. A Kruskal-Wallis test run on the ranked data indicated a significant difference in total PAHs by stratum ($p = 0.0210$). A Tukey's HSD (Highly Significant Difference) analysis indicated that only Stratum 1 and Stratum 4 were significantly different, due in part to the large standard errors in total PAHs within each stratum. Note that the targeted samples collected in 2010 were not included in the statistical analysis by stratum.

Comparison with Other Data. EPA's Environmental Monitoring and Assessment Program (EMAP) was developed to monitor and assess the status and trends of national ecological resources (EPA, 2000). In 2004, sediment samples were collected and analyzed by EMAP at a number of locations around St. Thomas. Four of those sites were in the STEER; two sites were in Benner Bay, and one site each in Jersey Bay and Great Bay. The four EMAP sites are also included in Figure 4.1. A suite of 23 PAHs were analyzed in the sediments at each site. The detection limit for each PAH analyzed by EMAP was 10 ng/g. None of the sediments sampled within the STEER boundaries by EPA's EMAP had a detectable level of the PAHs analyzed.

Recent chemical contaminant studies have also been conducted in Puerto Rico and can be used to compare with the results found in the STEER. In Vieques, Puerto Rico, the mean concentration of total PAHs was 52.3 ± 8.7 ng/g, somewhat below total PAHs found in the STEER sediments (Pait *et al.*, 2010). In southwest Puerto Rico, Pait *et al.* (2008) reported a mean total PAH concentration in the sediments of 80.6 ± 25.5 ng/g, also lower than the mean found in the STEER.

Because of the national-level contaminant monitoring carried out by NOAA's NS&T Program, data from the STEER can be compared with data from the rest of the Nation's coastal waters. For this report, data from the STEER sediments are compared with the most recent (2006/2007) nationwide analysis of sediments from the NS&T Program. The most recent NS&T nationwide sediment analysis data

were used, as the number of compounds in some of the chemical contaminant classes (e.g., total PAHs, total PCBs and total DDT) that NS&T analyzes, have increased over time, and the 2006/2007 NS&T sediment data contains the most comparable list of analytes.

The NS&T 2006/2007 median (NS&T median) for total PAHs is 395 ng/g. Three sites, two of which were in Mangrove Lagoon (1-3P, 1-2P, and 3-32P), were above the NS&T median for total PAHs. None of the sites sampled in the STEER approached the NS&T 2006/2007 85th percentile of 2,883 ng/g. The median (i.e., the “middle value”) and 85th percentile values (elevated contaminant concentrations), are used to show how the results from the STEER sediments compare with NOAA’s national results.

NOAA Sediment Quality Guidelines for Total PAHs. As noted earlier, the NS&T Program developed effects-based, numeric guidelines to estimate the toxicological relevance of certain sediment chemical contaminants (Long *et al.*, 1998). These guidelines, the Effects Range-Low (ERL) and the Effects Range-Median (ERM) define sediment contaminant concentration ranges that are rarely (<ERL), occasionally (ERL to ERM) or frequently (>ERM) asso-

ciated with toxic effects in aquatic biota (NOAA, 1998). The ERL and ERM values for total PAHs are also shown in Figure 4.5. As can be seen, total PAHs in the sediments were below the ERM and ERL as well.

NOAA’s NS&T Program has also developed ERL and ERM guidelines for a number of individual PAHs, which are shown in Table 4.2. None of the sediments collected in the STEER exceeded the ERL, ERM or the NS&T 85th percentile for any of the individual PAHs listed. However, from the 2011 stratified random sampling, Sites 1-2P, 1-3P and 3-32 exceeded the NS&T median for a number

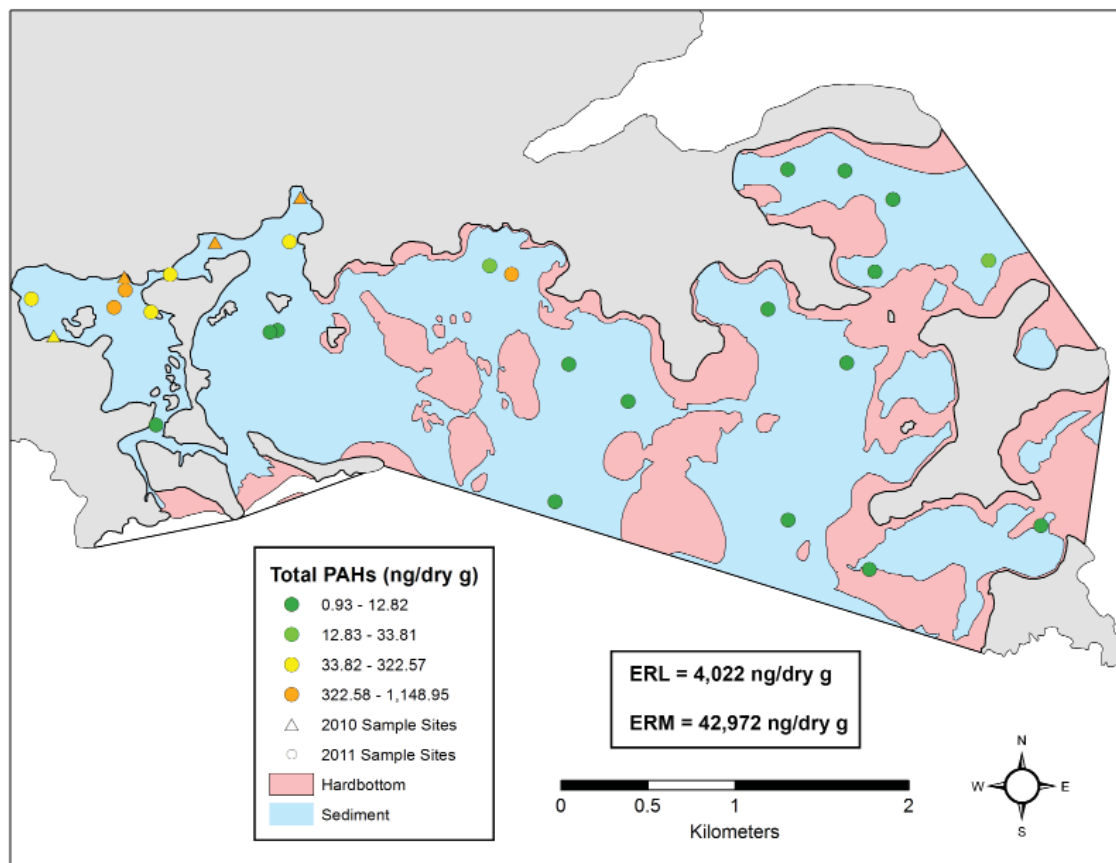


Figure 4.5 Total polycyclic aromatic hydrocarbons (PAHs) detected in sediments from the St. Thomas East End Reserves.

Table 4.2. Comparison of higher concentrations of individual PAHs at STEER sites with NOAA NS&T data.

Compound	Sites						NS&T Statistics and Guidelines			
	1-2P	1-3P	3-32P	BB-1	BB-2	ML-10	Median	85th Percentile	ERL	ERM
Acenaphthylene	2.2	3.4	0.2	2.8	4.0	2.4	2.1	15.1	44	640
Anthracene	3.2	5.6	1.3	7.4	9.2	5.7	3.4	38.7	85.3	1,100
Napthalene	7.3	9.2	0.9	5.4	6.4	7.3	3.7	27.6	160	2,100
Benzo-a-pyrene	20.3	35.0	35.1	9.6	17.5	15.6	14.7	127	430	1,600
Dibenzo(a,h)anthracene	3.4	6.4	8.2	7.4	6.9	11.6	5.0	23.8	63.4	260

All concentrations are in ng/g.

of the individual PAHs listed (Table 4.2). From the 2010 targeted sampling, BB-1, BB-2 and ML-10 also exceeded the median for a number of the listed PAHs. As will be seen later, results from the P450 test indicated a response to PAHs in Mangrove Lagoon and northern Benner Bay.

The ratios of phenanthrene-to-anthracene (P/A) and fluoranthene-to-pyrene (F/P) have been used as a screening tool to assess the relative contributions of pyrogenic (combustion-related) versus petrogenic (uncombusted) sources of PAHs (Budzinski *et al.*, 1997). Higher levels of uncombusted PAHs would be more indicative of the presence of spilled fuels, such as gasoline or oil. P/A ratios less than 10 are more indicative of pyrogenic sources; F/P ratios greater than 1 are also thought to be associated with pyrogenic sources. Most of the sites in the STEER had P/A ratios of less than 10, and the F/P ratio close to or above 1, indicating the pyrogenic nature of the PAHs present in the sediments. At 1-3P in Mangrove Lagoon, which had the highest total PAH level in the samples analyzed from the STEER, the P/A ratio was 3.24, indicating pyrogenic sources, however the F/P ratio was 0.70, indicating that petrogenic sources may also have contributed to the mix of PAHs found in the sediments at this site.

Effects of Sediment Grain Size and TOC. As noted earlier, the adsorption of organic contaminants onto sediments is strongly influenced by grain size (Hassett *et al.*, 1980). The smaller grain sizes of the silts and clays have proportionally higher surface areas available for the adsorption of chemical contaminants and tend to have more TOC than coarse grained sediment. To assess this relationship for the samples collected in the STEER, a nonparametric correlation analysis (Spearman's) was run between percent fines

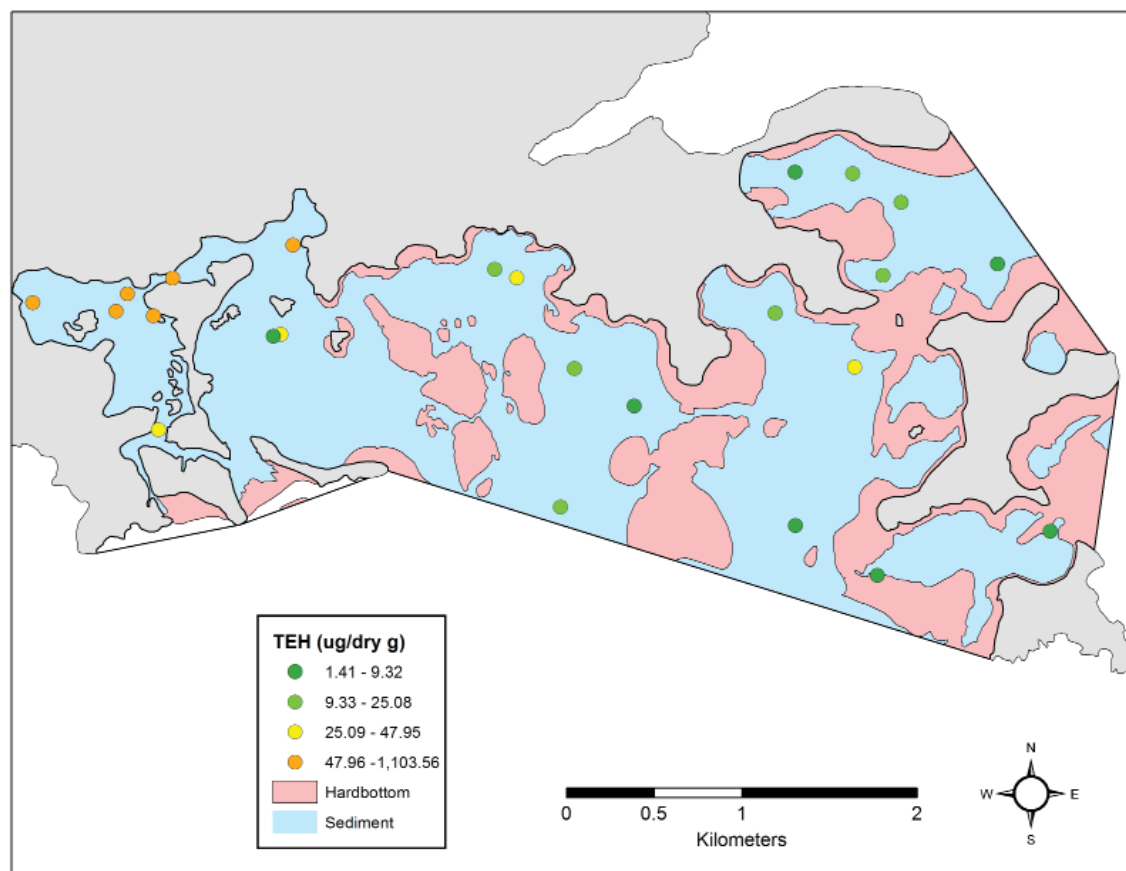


Figure 4.6. Total extractable hydrocarbons (TEH) detected in sediments from the St. Thomas East End Reserves.

(sum of percent silt and percent clay) and the concentration of total PAHs found in the sediment samples. There was a highly significant ($p < 0.0001$) and positive correlation between the percent fines and the concentration of total PAHs. There was also a significant negative correlation ($p < 0.0001$) between the concentration of PAHs and the sand fraction of the sediments, indicating that as the percent sand concentration increased in sediments, the amount of total PAHs present was correspondingly lower.

Typically, a positive relationship also exists between sediment total organic carbon (TOC) and chemical contaminants in freshwater, estuarine and coastal waters (Shine and Wallace, 2000; Hassett *et al.*, 1980). Because of this, organic contaminant concentrations are often normalized to the organic carbon content of sediments. However, a nonparametric correlation between TOC and total PAHs for the STEER samples was not significant (Spearman Rho = -0.1779, $p = 0.4057$).

Total Extractable Hydrocarbons

The results of the analysis of total extractable hydrocarbons, or TEH, is shown in Figure 4.6. TEH is not only composed of PAHs, such as those occurring from the combustion of fuels, but also of straight and branched aliphatics

(nonaromatic hydrocarbons) found in uncombusted fuels, in oil, and from natural sources (e.g., decaying vegetation). TEH provides a more complete assessment of the total mass of hydrocarbons present in a sediment sample.

It can be seen from Figure 4.6 that the relative concentration of TEH in the STEER mirrors total PAHs (Figure 4.5). It should be noted, however, that the units for total PAHs are in ng/g (ppb), while TEH is in $\mu\text{g/g}$ (ppm or parts per million). TEH in the sediments at a number of locations in the STEER were approximately an order of magnitude higher than total PAHs. The mean TEH concentration in the STEER was $167 \mu\text{g/g}$. The highest TEH concentration ($1,104 \mu\text{g/g}$) was found in Mangrove Lagoon at 1-3P.

Variation of TEH Across Strata. An ANOVA run on the log10 transformed data indicated that TEH varied by stratum ($p = 0.0018$), and a Tukey-Kramer HSD test indicated that Stratum 1 was significantly different (higher) than Strata 3, 4 and 5.

Sediment Quality Guidelines and TEH. There are no NOAA sediment quality guidelines for TEH. Boehm *et al.* (2008) carried out a regression analysis between total PAHs and TEH, and calculated a TEH “ERL” of $2,600 \mu\text{g/g}$, and a TEH “ERM” of $9,760 \mu\text{g/g}$. Using these values as rough TEH sediment quality guidelines, none of the TEH concentrations found in the STEER exceeded the lower threshold estimated by Boehm *et al.* (2008).

Effects of Sediment Grain Size and TOC. A nonparametric correlation run between TEH and percent fines indicated a significant positive correlation (Spearman Rho = 0.5852, $p = 0.0027$), however, there was no significant correlation between TEH and TOC (Spearman Rho = -0.0030, $p = 0.9887$).

Aliphatics. Straight chain and branched aliphatics, also a subset of TEH, are indicative of uncombusted fuels or oil, were analyzed in the sediment samples (Figure 4.7). There were a number of aliphatics in the C-19 (molecules containing 19 carbons), and the C-23 to C-31 range, with spikes at C-19 and C-27, particularly at Site 3-32P but also at Site

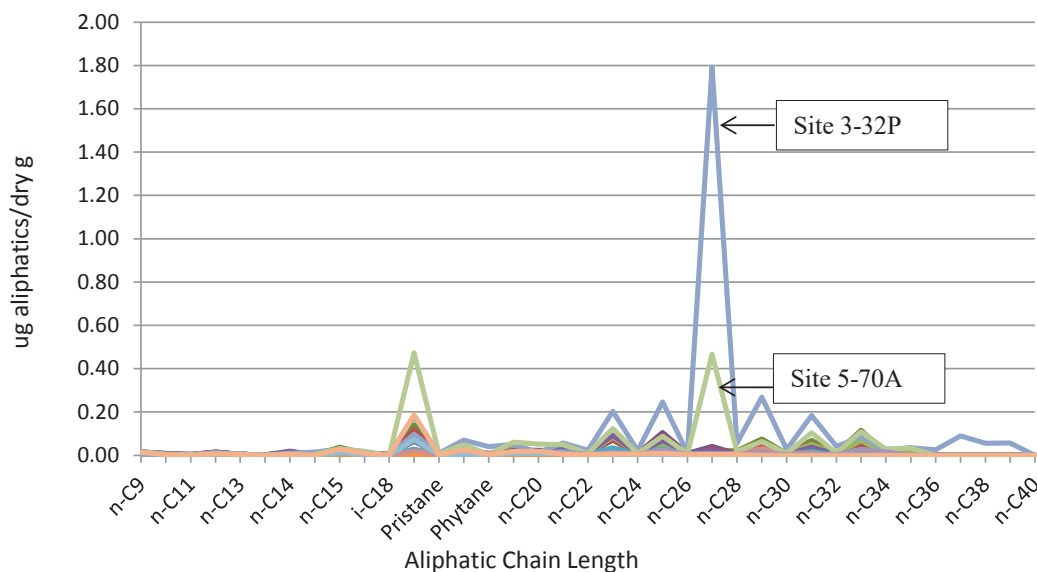


Figure 4.7. Straight chain and branched hydrocarbons (aliphatics) in sediments from the St. Thomas East End Reserves.

5-70A. The aliphatics present appear to be in the range of diesel fuel and lubricating oils (Libes, 1992), and could indicate some type of discharge in the past or perhaps low level, chronic (longer term) inputs at this site.

Polychlorinated Biphenyls

Total PCBs detected in the sediments are shown in Figure 4.8. Total PCBs as included in this report represents the sum of the 38 congeners analyzed for the project (Table 4.1).

The mean concentration of total PCBs found in the STEER was $1.00 \pm 0.32 \text{ ng/g}$; the median was 0.37 ng/g . The highest concentration of total PCBs from the stratified random sampling was 7.2 ng/g ; from the targeted 2010 sampling, the highest concentration was 65.9 ng/g at BB-2 (Figure 4.8). Eighteen, or 75%, of the sites sampled in the STEER, however, had a total PCBs concentration of less than 1 ng/g .

Variation of Total PCBs Across Strata. The variation of total PCBs by stratum was assessed using an ANOVA, run on the log10 normalized concentration values. The results indicated no significant variation ($p = 0.0573$) in total PCBs across the five strata established in the STEER.

Comparison with Other Data. EPA's EMAP sampled and analyzed sediments in the STEER from four locations in 2004 (Figure 4.1). A total of 21 congeners were analyzed in the sediment samples taken. No PCB congeners were detected in the four samples analyzed; the detection limit for each congener was 0.05 ng/g .

In a project conducted by NCCOS in Jobos Bay, the mean total PCBs concentration found in the sediments was 2.09 ± 0.50 ng/g (Pait *et al.*, 2012). In Vieques, that value was 2.86 ± 0.14 ng/g (Pait *et al.*, 2010), both of which are higher than the STEER. In southwest Puerto Rico, the mean total PCBs concentration in sediments was 12.1 ± 2.26 ng/g not including two very high total PCBs concentrations (1,022 ng/g and 2,710 ng/g) within Guanica Bay (Pait *et al.*, 2008). If the two sites in Guanica Bay are included, the mean for total PCBs in the southwest Puerto Rico study area was 74.7 ± 47.9 ng/g (Pait *et al.*, 2008).

Four of the five sites from the stratified random sampling (in Mangrove Lagoon and northern Benner Bay) had a total PCBs concentration above the NS&T median of 2.2 ng/g. All four of the 2010 targeted sample were above the NS&T median. BB-2 (65.9 ng/g) from the targeted sampling was above the NS&T 85th percentile (23.7 ng/g) for total PCBs.

NOAA Sediment Quality Guidelines for Total PCBs. The concentration of total PCBs at Site BB-2 (65.9 ng/g) from the 2010 targeted sampling was higher than the PCB ERL (Figure 4.8). At 21.8 ng/g, ML-10 was just below the ERL. Levels below the ERL indicate that effects on benthic infauna are not as likely. Concentrations above the ERL but below the ERM indicate that more sensitive species or life stages may be impacted.

Effects of Grain Size and TOC. A nonparametric analysis of the data revealed a significant positive correlation between total PCBs and the fines fractions of the sediment (Spearman Rho = 0.5452, $p = 0.0059$). No significant correlation existed between total PCBs and gravel, and there was a significant negative correlation (Spearman Rho = -0.4648, $p = 0.014$) between sand and total PCBs.

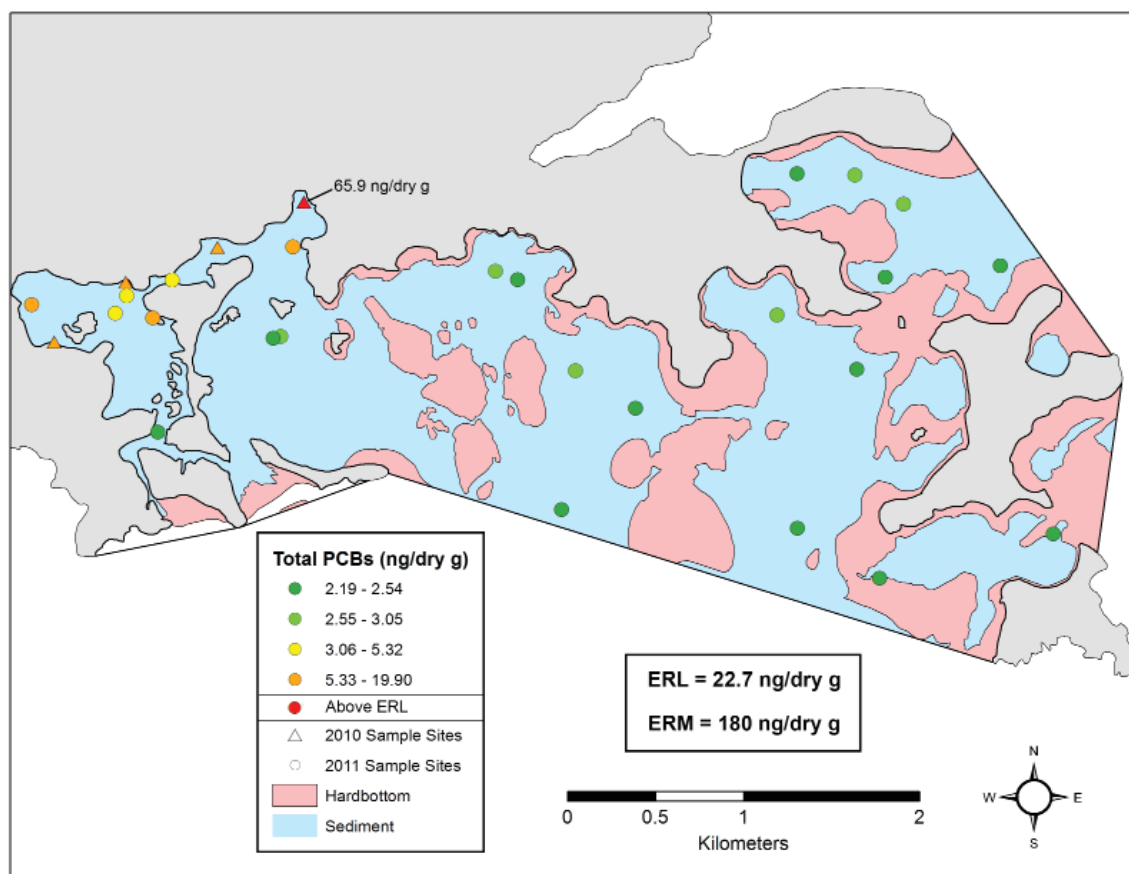


Figure 4.8 Total PCBs detected in sediments from the St. Thomas East End Reserves.

An analysis of total PCBs and TOC revealed no significant correlation (Spearman Rho = 0.0322, $p = 0.8813$), similar to what was observed for PAHs. For both PAHs and PCBs in the STEER, sediment concentrations appeared to be influenced more by sediment particle size than sediment TOC.

DDT and Other Organochlorine Pesticides

Figure 4.9 contains the results of the analyses of total DDT in sediment samples from the STEER. Total DDT as defined in this report is the sum of the parent isomers (4,4'-DDT and 2,4'-DDT), along with degradation products DDE, DDD and DDMU. The mean concentration of total DDT in the sediments in the STEER was 0.047 ± 0.025 ng/g; the median was 0.002 ng/g. As with a number of the other chemical contaminants, higher concentrations of total DDT were found in the Benner Bay and Mangrove Lagoon areas. The highest total DDT concentration from the stratified random samples collected in 2011 was 0.609 ng/g at 2-20P (Table 4.3). The highest concentration of total DDT from the 2010 targeted sampling was 3.61 ng/g at BB-2 (Figure 4.9) in northern Benner Bay. The second highest concentration was also in Benner Bay (1.31 ng/g) at BB-1. Currently, there does not appear to be much agricul-

ture on the island of St. Thomas, although in the past DDT could have been used on crops such as sugarcane, fruits and vegetables, along with use to control mosquitoes.

Variation of Total DDT Across Strata.

Although there were some elevated levels of DDT in Benner Bay and Mangrove Lagoon (Table 4.3), an ANOVA run on log10 normalized data indicated no significant variation ($p = 0.2575$) in total DDT concentration across the five strata in the STEER. In addition, the degradation products DDD and DDE appeared to make up much of the total DDT present, indicating that the parent compound had degraded over time.

Comparison with Other Data. EPA's EMAP analyzed DDT along with a number of DDT degradation products from four sites in the STEER. The only DDT-related compound detected was the degradation product 4,4'-DDD, at a concentration of 0.26 ng/g at one site. In southwest Puerto Rico, Pait *et al.* (2008) detected a mean total DDT concentration of 2.10 ± 1.26 ng/g, higher than in the STEER. In Jobos Bay, the mean total DDT concentration was 0.54 ± 0.10 ng/g. In Vieques, the mean concentration of total DDT in the sediments was substantially higher, 23.6 ± 16.5 ng/g, due in part to elevated concentrations (78 ng/g to 1,274 ng/g total DDT) at four sediment sampling sites (Pait *et al.*, 2010).

The concentration of total DDT in the STEER exceeded the NS&T median value of 0.395 ng/g at four sites,

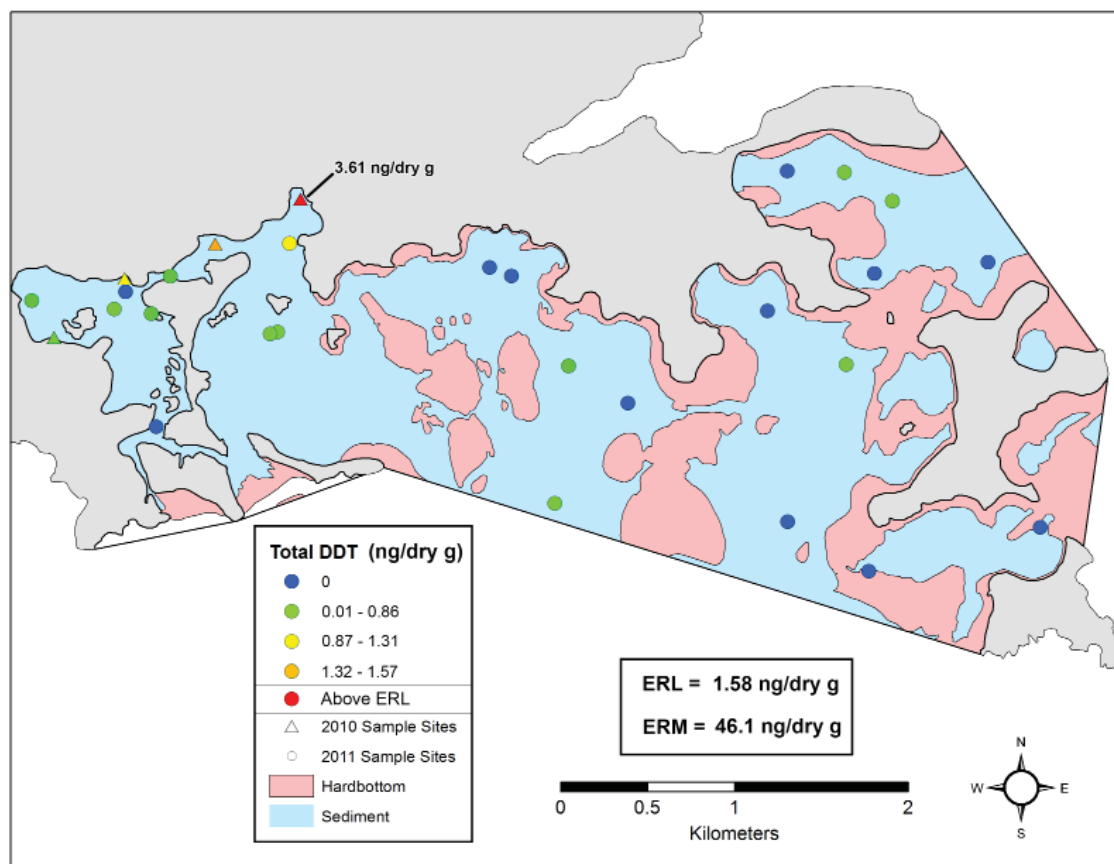


Figure 4.9. Total DDT detected in sediments from the St. Thomas East End Reserves.

three of those from the 2010 targeted sampling (Table 4.3). The concentration of total DDT at BB-2 was just above the NS&T 85th percentile of 3.49 ng/g.

NOAA Sediment Quality Guidelines for Total DDT. The concentration of total DDT at BB-2 (3.61 ng/g) from the 2010 targeted sampling was also above the ERL (1.58 ng/g). However, none of the sites sampled in the STEER had a concentration above the DDT ERM.

Effects of Sediment Grain Size and TOC. A nonparametric analysis of the data revealed no significant correlation between total DDT and percent fines (Spearman Rho = 0.2993, $p = 0.1554$), or total DDT and percent TOC (Spear-

Table 4.3. Comparison of higher concentrations (ng/g) of pesticides at STEER sites with NOAA NS&T data.

Compound	Sites						NS&T Statistics and Guidelines			
	1-5P	2-16P	2-20P	BB-1	BB-2	ML-10	Median	85th Percentile	ERL	ERM
Total DDT	0.10	0.082	0.609	1.31	3.61	0.86	0.395	3.49	1.58	46.1
Chlordane	0.07	0.18	0.02	0.22	0.2	0.85	0.195	0.975	0.5	6
Total Endosulfan	0.0	0.0	0.142	0.0	1.13	0.63	0.0	0.253	N/A	N/A
Chlorpyrifos	0.0	0.253	0.0	1.33	0.13	0.62	0.01	0.328	N/A	N/A

N/A, not available

man $Rho = -0.0060$, $p = 0.9776$), however, it should be noted that 13 of the 24 stratified random samples had no detectable level of total DDT.

Additional Pesticides. A number of other pesticides were analyzed in the sediments from the STEER as part of this project. Most of the results were below detection limits. Selected results are shown in Table 4.3. The highest concentration of chlordane (alpha and gamma isomers) detected in STEER sediments was 0.85 ng/g at site ML-10 at the mouth of Turpentine Gut, which empties into Mangrove Lagoon. This concentration is above the NS&T median but below the NS&T 85th percentile for chlordane (Table 4.3). NOAA has also established ERL and ERM values for chlordane. Chlordane was above the ERL only at ML-10. None of the sites had a chlordane concentration above the ERM.

The insecticides endosulfan and chlorpyrifos have had both agricultural and nonagricultural (e.g., homeowner) uses. Endosulfan, an organochlorine insecticide, is currently being phased out for all uses due to health risks to farmworkers and wildlife, and its persistence in the environment (EPA, 2010). Since there appears to be little commercial agriculture currently on St. Thomas, it is likely that endosulfan was used in the past, perhaps on vegetables and fruits in gardens, prior to being phased out for homeowner use. The highest concentration of total endosulfan (sum of endosulfan I and II and endosulfan sulfate) detected in the STEER (1.13 ng/g) was at BB-2 in Benner Bay, which is above the NS&T median and 85th percentile. There are no NOAA sediment quality guidelines for endosulfan.

Although the organophosphate insecticide chlorpyrifos has had widespread use both domestically and in agriculture, virtually all homeowner uses have been eliminated (EPA, 2002b). Chlorpyrifos can still be used on certain food crops, on golf courses, for wood treatment (nonstructural), and as an adult mosquitocide. The highest concentration of chlorpyrifos (1.33 ng/g) was in Benner Bay at BB-1. This concentration was above the NS&T median and also above the 85th percentile (Table 4.3). The concentration of chlorpyrifos at ML-10 was also above the NS&T median and 85th percentile. There are no NOAA sediment quality guidelines for chlorpyrifos.

Tributyltin (TBT)

The results of the analysis of butyltins in STEER sediments are shown in Figure 4.10. Mono-, di-, tri-, and tetrabutyltins were analyzed in the sediments. Tetrabutyltin is an intermediate in the manufacture of TBT compounds. As noted earlier, TBT has had extensive use in the past as an antifoulant on boat hulls. In the environment, TBT degrades to dibutyltin, then monobutyltin, and finally to inorganic tin. Experiments have shown that the half-life of TBT, the amount of time needed to convert half of the TBT

to dibutyltin in natural water samples, is on the order of days; degradation to monobutyltin takes approximately a month (Batley, 1996). Experiments with aerobic sediments have shown that the half-life of TBT is similar to that measured in solution. In deeper, anoxic sediments, however, the half life of TBT appears to be considerably longer, on the order of years (Batley, 1996) to decades (Mathiessen, 2013).



Sampling of sediments in the STEER.

The mean concentration of TBT in the sediments in the STEER was 1.85 ± 1.30 ng Sn/g; the median was zero. The highest concentration of TBT detected in STEER sediments collected using the stratified random sampling design was at site 2-20P in Benner Bay, with a concentration of 31 ng/g. However, of the samples analyzed from 2010, BB-1 and BB-2 had even higher TBT concentrations. The concentration of TBT at BB-1 was 77 ng/g; the TBT concentration at BB-2 was 248 ng/g.

Variation of TBT Across Strata. Because the targeted 2010 samples were collected nonrandomly, that data cannot be used to assess the variation of TBT in sediments across the STEER strata. An ANOVA run on the log10 transformed data for the 2011 samples collected using the stratified random design did not indicate any significant differences ($p = 0.4368$) in the concentration of TBT between strata.

Comparison with Other Data. The NS&T median concentration of TBT in sediments is 0.16 ng Sn/g. The NS&T 85th percentile for TBT is 1.38 ng Sn/g, lower than the mean concentration of TBT found in STEER sediments. The highest concentration of TBT detected in the STEER (248 ng Sn/g) represents the third highest detection ever in sediments in NOAA's NS&T Program (not just the 2006/2007 nationwide sediment sampling). The only two

higher TBT concentrations from NOAA's NS&T Program were from the Elizabeth River in the southern Chesapeake Bay area, and Elliot Bay in Puget Sound. Pait *et al.* (2008) detected a mean TBT concentration of 0.01 ± 0.01 ng Sn/g in southwest Puerto Rico. In Vieques, Puerto Rico, the mean concentration of TBT was 0.05 ± 0.02 ng Sn/g and in Jobos Bay, the mean concentration was 0.56 ± 0.28 ng Sn/g.

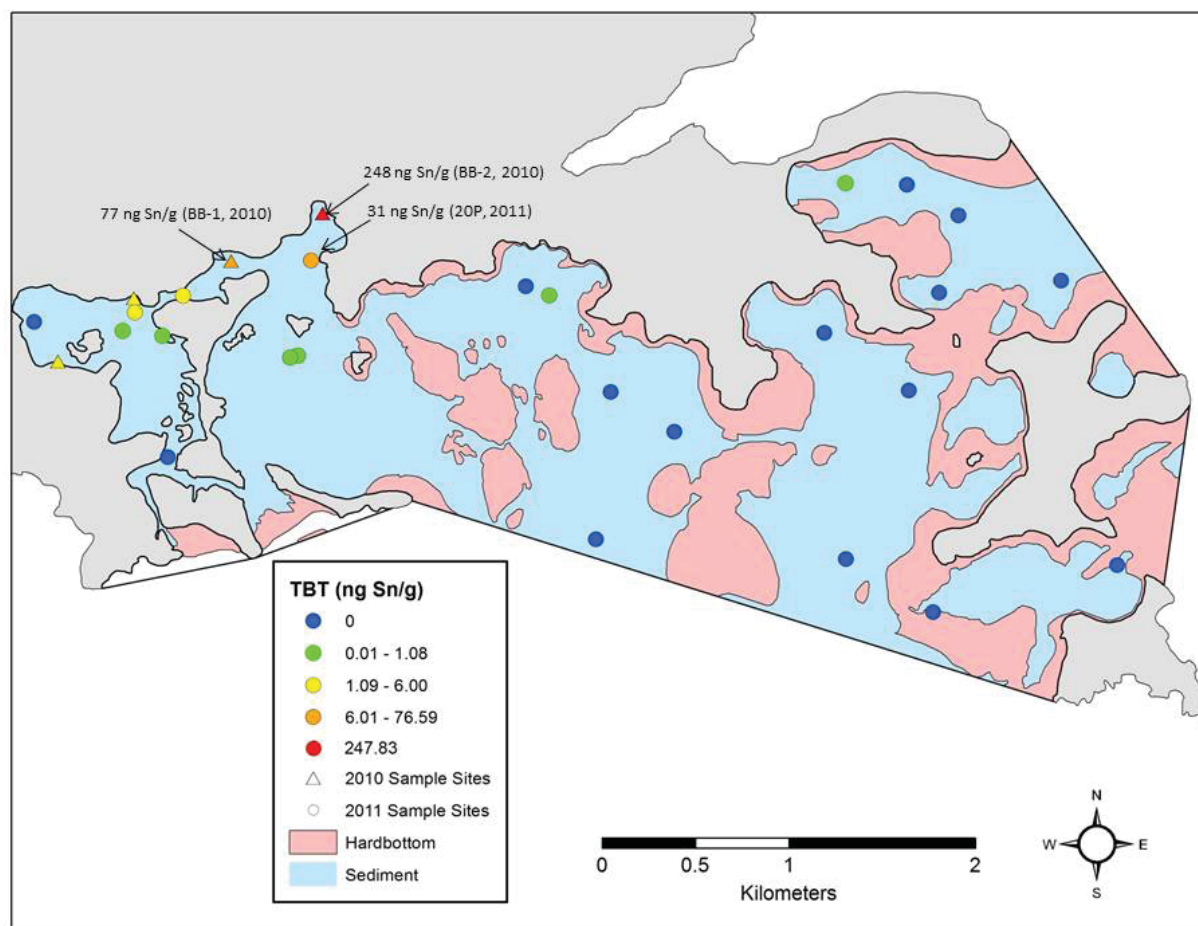


Figure 4.10. Tributyltin detected in sediments from the St. Thomas East End Reserves. BB-1 and BB-2 represent targeted, nonrandomized samples taken in 2010.

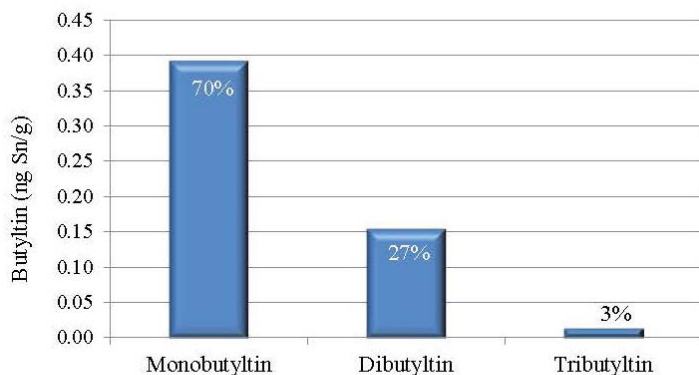
It should be noted, however, that 15 of the 24 sediment samples taken in the STEER had no detectable TBT at all. The higher concentrations of TBT tended to occur in the northern Benner Bay area. For the most part, this was also the case for the TBT degradation products dibutyltin and monobutyltin.

Sediment Quality Guidelines and TBT. There are no established sediment quality guidelines for TBT, due in part to the complex chemistry of this compound, including its bioavailability largely being governed by partitioning into porewater (water in between sediment particles), which is in turn is governed by a number of parameters of the sediment at a site.

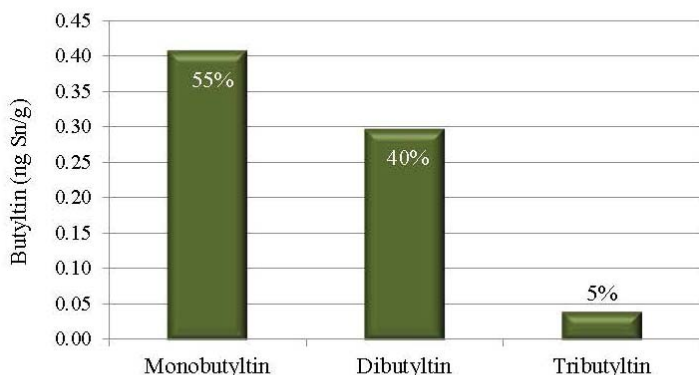
One of the few attempts at establishing guidelines for TBT was reported by Weston (1996). In that document, lower and upper screening values were established for TBT, based on a sediment organic carbon content of 2%. The intent was to establish thresholds for cleanup actions at EPA Superfund sites in Puget Sound. The lower and upper values are not analogous to NOAA ERL or ERM values, but rather

represent a range that was to be used to determine when additional testing (e.g., additional toxicity testing) at a site would be advisable. Using this approach, Weston (1996) recommended TBT values between 10 and 144 ng Sn/g as the lower and upper screening values, respectively, with the actual screening value selected being determined by an EPA site manager at a particular EPA Superfund site. If these two values are used as rough boundaries for determining when additional work (e.g., toxicity testing) would be recommended, it indicates there should be concern regarding the elevated TBT level at BB-2, especially as the concentration was above the higher screening value developed by Weston (1996). Sites BB-1 and 2-20P were above the lower screening value but below the upper screening value, but are also of concern. The results did not change even when the organic carbon content from each of these three sites was incorporated into the equations used in Weston (1996), including BB-2, which had an organic carbon content of roughly 2%. In the next chapter, results from follow-up sampling and analysis of surface sediments, and of sediment cores in northern Benner Bay are presented.

Mean butyltins in sediments from southwest Puerto Rico.



Mean butyltins in sediments from Vieques, Puerto Rico.



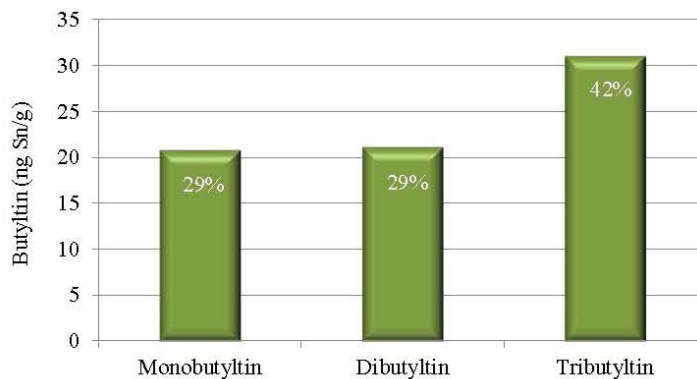
% indicates percent of butyltin total

Figure 4.11. Tributyltin detected in sediments from southwest Puerto Rico and Vieques, Puerto Rico. Values represent means.

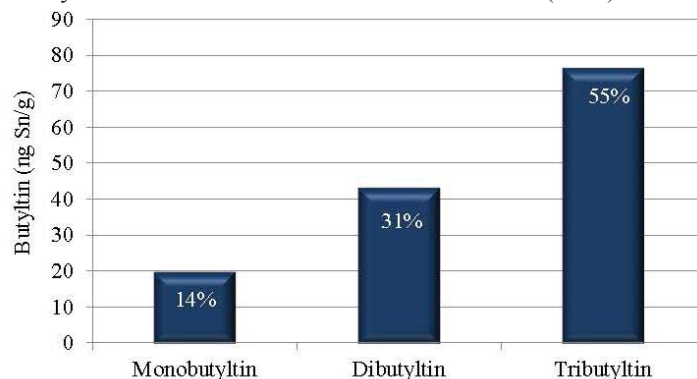
Butyltin Levels in the Sediments. A summary of the results for TBT and its metabolites in southwest Puerto Rico (Pait *et al.*, 2007) and Vieques, Puerto Rico (Pait *et al.*, 2010) is shown in Figure 4.11. From this figure, it can be seen that the majority of the butyltin present in the sediment samples taken from both southwest Puerto Rico and Vieques were primarily in the form of monobutyltin and dibutyltin, with only a small fraction of the total as the parent or undegraded TBT. This would be expected if TBT use had occurred primarily in the past.

Figure 4.12 shows the same type of information for the three sites in the STEER that had the highest TBT concentrations. Not only are the absolute concentrations much higher, the ratios of TBT to monobutyltin and dibutyltin at these three sites are different from those shown in Figure 4.11. The concentration of TBT at the three sites in the STEER was similar to, if not higher than, the concentration of monobutyltin and dibutyltin at these same sites. The higher concentrations of TBT relative to monobutyltin and dibutyltin, along with the overall higher concentration of

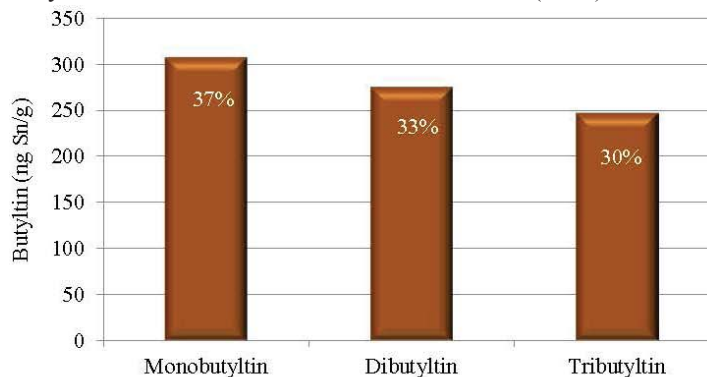
Butyltins in sediments from 20P in the STEER (2011).



Butyltins in sediments from BB-1 in the STEER (2010).



Butyltins in sediments from BB-2 in the STEER (2010).



% indicates percent of butyltin total

Figure 4.12. Tributyltin detected in sediments from three sites in the STEER.

TBT, may be indicative of recent TBT deposition. This may be a result of the mooring of boats and/or the cleaning and scraping of boat bottoms containing TBT-based paints, with subsequent input of these materials (e.g., paint chips or dust) through stormwater runoff into adjacent waters.

The higher levels of TBT found could also be related to past deposition. As noted, the sediment samples analyzed as part of this project were surficial or surface sediments. For this part of the project, the top three cm (centimeters)

of sediment were collected from the sediment grab. Given this, it is likely that the samples analyzed represent more recently deposited sediments. In order to confirm this, additional studies were used to assess the sedimentation rate in this part of Benner Bay. As part of the monthly monitoring in the STEER conducted by UVI, six sediment traps were installed in the STEER, including sites in Mangrove Lagoon, Benner Bay, Rotto Cay, Cowpet Bay, and adjacent to Great St. James and Little St. James. As will be seen in Chapter 9, terrestrial inputs into Benner Bay and Mangrove Lagoon were higher than the other sites in the STEER. Regardless of whether the TBT found in the Benner Bay area is from past inputs, more recent inputs, or both, the elevated levels suggests the need for additional work to quantify the distribution of this contaminant in the sediments along with effects (toxicity), particularly in mollusks. As noted earlier, Strand *et al.* (2009) found evidence of elevated levels of TBT and its degradation products in several gastropod species, as well as imposex at several locations, including Charlotte Amalie Bay in St. Thomas. As part of the project in the STEER, samples of queen conch (*Lobatus gigas*) and coral (*Porites astreoides*) were analyzed for chemical contaminants, including butyltins, which are reported in Chapter 7.

Follow-up sampling was conducted in 2013 in northern Benner Bay to assess TBT in sediment cores as well as additional surface samples, at the request of DPNR. The results of that work are reported in Chapter 5. This information is of particular importance if dredging operations (e.g., for deepening the navigation channels) in this part of Benner Bay were to occur in the future, as it could be used to help guide dredging operations to limit the transport and more importantly the impact of contaminated sediments.

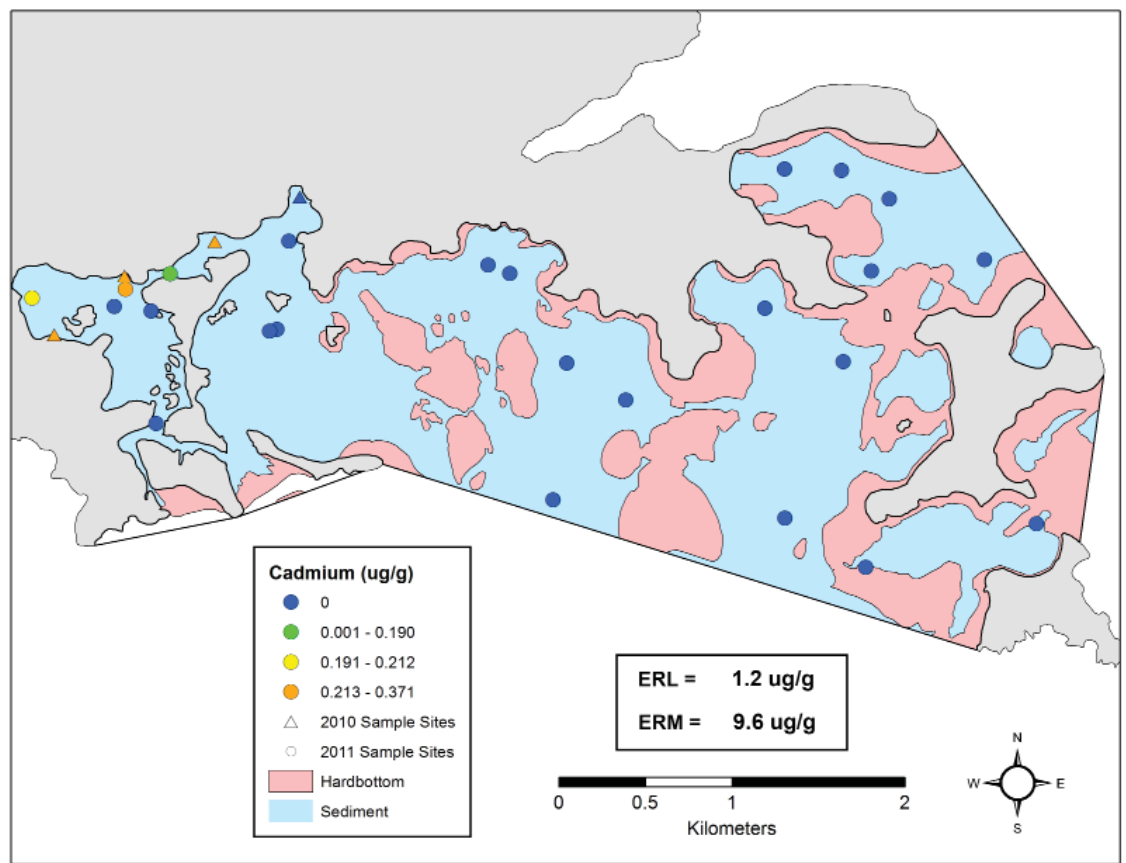


Figure 4.13. Cadmium detected in sediments from the St. Thomas East End Reserves.

Effects of Sediment Grain Size and TOC. The correlation of TBT concentrations to grain size and TOC was examined for sediments sampled in the STEER. A nonparametric analysis of the data revealed a significant correlation between TBT and percent fines (Spearman Rho = 0.5759, $p = 0.0032$). As noted earlier, smaller grain sizes (silt and clays) of sediments have more surface area available for the adsorption of contaminants. There was no correlation, however, found between TBT and percent TOC (Spearman Rho = 0.0430, $p = 0.8419$), similar to what was seen for a number of the other organic chemical contaminants analyzed for this project.

Cadmium

The range of cadmium concentrations in the sediments sampled in the STEER can be seen in Figure 4.13. The mean concentration in the STEER sediments was $0.03 \pm 0.02 \mu\text{g/g}$. Only three sediment samples from the stratified random sampling had a detectable level of cadmium, including 1-1P and 1-3P in Mangrove Lagoon, and 2-16P in Benner Bay. The highest concentration of cadmium in the samples was $0.264 \mu\text{g/g}$ at site 1-3P. Of the four samples collected in 2010, the highest concentration was $0.371 \mu\text{g/g}$ at ML-2 in Mangrove Lagoon. At BB-2, no cadmium was detected.

Variation of Cadmium Across Strata.

Because of the low number of detections of cadmium in the STEER, no analysis by stratum was carried out.

Comparison with Other Data.

EPA's EMAP analyzed cadmium in sediments from four locations in the STEER. The mean concentration of cadmium was 0.25 µg/g, higher than the mean detected in the STEER in the current project. In southwest Puerto Rico, the mean cadmium concentration in sediments was 0.01 µg/g, similar to that found in the STEER. In Vieques, the mean cadmium concentration in sediments was 0.13 µg/g (Pait *et al.*, 2010). In Jobos Bay, the mean cadmium concentration was 0.01 µg/g, similar to southwest Puerto Rico (Apeti *et al.*, 2012b). The NS&T median for cadmium in sediments is 0.19 µg/g, the 85th percentile is 0.44 µg/g. All three sites in the STEER where cadmium was detected from the stratified random sampling were at or above the NS&T median, but below the 85th percentile. Three of the four sites from the 2010 targeted sampling were also above the NS&T median, but all were below the 85th percentile.

NOAA Sediment Quality Guidelines for Cadmium. Figure 4.13 also contains the NOAA ERL and ERM. All cadmium levels detected in the STEER sediments, either from the stratified random sampling or from the 2010 targeted sampling, were below the ERL. Sites 1-1P and 1-3P are both in Mangrove Lagoon, which is adjacent to the Bovoni Landfill, and could be one source of the cadmium found in the sediments. At this time, it is not known whether the cadmium detected is associated with runoff or subsurface flow from the landfill, or perhaps input from septic systems or runoff from residential areas. At one time, a wastewater treatment plant emptied directly into Mangrove Lagoon (Grigg *et al.*, 1971). Additional work is needed to better

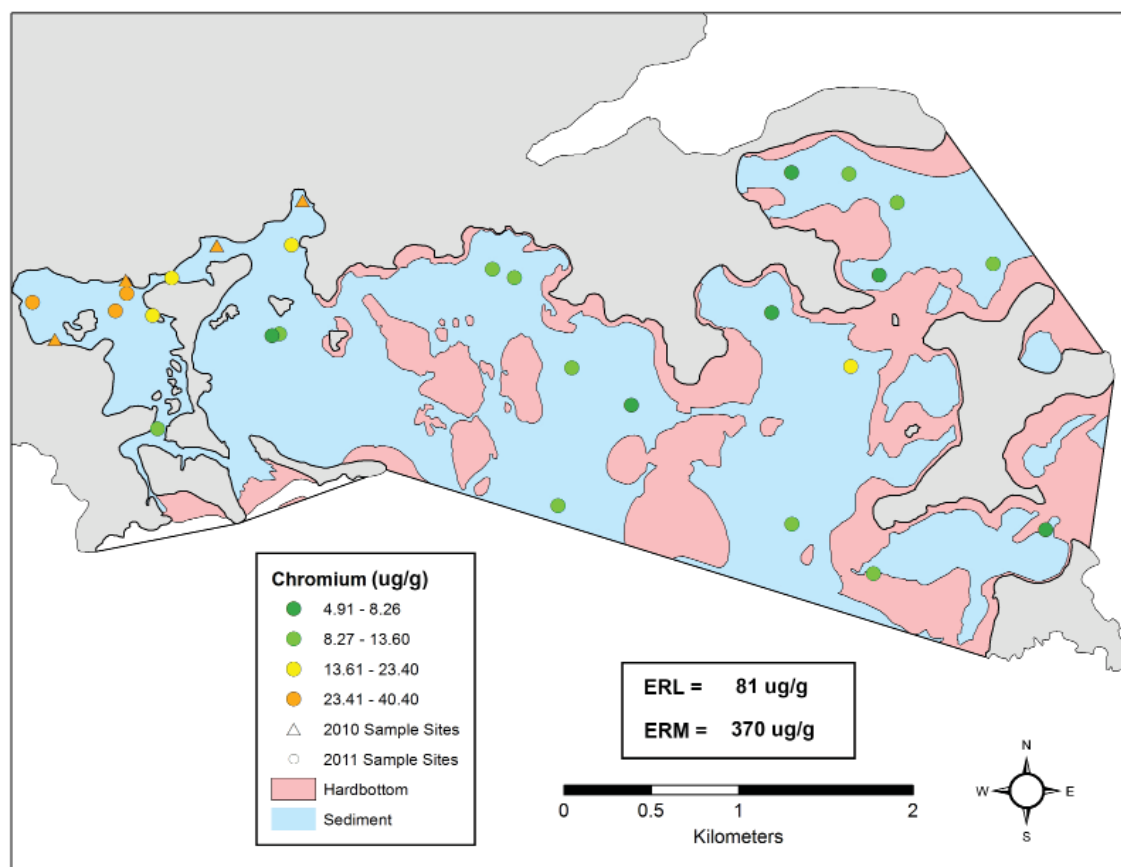


Figure 4.14. Chromium detected in sediments from the St. Thomas East End Reserves.

assess the possible sources of cadmium to the area of Mangrove Lagoon and Benner Bay.

Effects of Sediment Grain Size. Because of the low number of cadmium detections, an assessment of the effects of grain size and TOC was not carried out.

Chromium

The concentrations of chromium detected in the sediments collected from the STEER are shown in Figure 4.14. Higher concentrations were detected in the Mangrove Lagoon and Benner Bay areas, a pattern similar to a number of the organic compound classes (e.g., PAHs, PCBs) analyzed for this project.

The mean concentration of chromium in the STEER from the stratified random sampling was 14.1 ± 1.76 µg/g; the median was 11.4 µg/g. The highest concentration of chromium in the sediments analyzed from the stratified random sampling was 35.7 µg/g at site 1-1P in Mangrove Lagoon. The second highest concentration, 32.3 µg/g, was also in Mangrove Lagoon at site 1-3P. The lowest level of chromium detected from the stratified random sampling was at 2-24A (4.91 µg/g). Figure 4.14 also contains the results of the targeted sampling in 2010. The highest chromium

concentration detected was 40.4 µg/g at BB-2, similar to the concentration of chromium at the sites in Mangrove Lagoon.

Variation of Chromium Across Strata. An ANOVA carried out on the log10 transformed data indicated that chromium varied significantly ($p = 0.0090$) by stratum. A subsequent pair wise comparison (Tukey HSD) indicated that Stratum 1 was significantly different (higher) from Strata 3, 4 and 5, however, Stratum 2 (Benner Bay) was not significantly different from Stratum 1.

Comparison with Other Data. EPA's EMAP analyzed four samples from the STEER in 2004. The mean concentration from these four samples was 3.06 µg/g, lower than the mean found in the current study in the STEER. The highest chromium concentration found in the STEER by EMAP was 4.8 µg/g.

In southwest Puerto Rico, Pait *et al.* (2008) calculated a mean chromium concentration of 31.2 µg/g, higher than in the STEER. In Vieques, mean chromium levels in the sediment were somewhat higher as well, 22.5 µg/g (Pait *et al.*, 2010). In Jobos Bay, the mean chromium concentration was 18.3 µg/g (Apeti *et al.*, 2012b), similar to what was found in the STEER. The NS&T median for chromium in sediments is 66 µg/g, higher than any of the chromium levels found in sediments in the STEER in this study.

As with cadmium, it is not known if the chromium detected in the Mangrove Lagoon area is associated with inputs from the landfill, from septic systems, runoff from residential areas, or perhaps a combination of these sources. Keller *et al.* (2014) noted elevated aqueous concentrations of chromium in groundwater sampled from wells installed in the Bovoni Landfill in the mangrove area on the western border of Mangrove Lagoon. They also found evidence that the mangroves and associated clay-rich sediments in that area may be holding back higher levels of metals such as chromium from entering Mangrove Lagoon, likely another benefit that mangroves are providing in this area.

NOAA Sediment Quality Guidelines for Chromium. The ERL and ERM for chromium are also included in Figure

4.14. All STEER chromium values were below both of these NOAA guidelines.

Effects of Sediment Grain Size. Using the log10 normalized data, chromium was found to be correlated with the percent fines ($p = 0.0180$). A nonparametric analysis showed this metal had a slightly higher correlation with percent clay (Spearman Rho = 0.5441, $p = 0.0060$) than the percent silt content (Spearman Rho = 0.4767, $p = 0.0185$).

Copper

Results from the analysis of copper in the sediments from the STEER can be seen in Figure 4.15. The mean concentration of copper found in the sediments was 21 ± 7.46 µg/g; the median was 3.75 µg/g. The highest copper concentration from the stratified random sampling was 155 µg/g at site 2-20P, followed by 69.9 µg/g at site 1-1P in Mangrove Lagoon. From the targeted sampling in 2010, the lowest concentration was 60.6 µg/g at ML-10. At BB-1, the concentration of copper in the sediment was 145 µg/g. At BB-2, the copper concentration was quite high, 1,010 µg/g.



Sediment sample in the PONAR grab from Stratum 1 (Mangrove Lagoon).

Variation of Copper Across Strata. A Kruskal-Wallis test run on the ranked values from the stratified random sampling indicated a significant difference ($p = 0.0001$) in the concentration of copper across the strata. A Tukey-Kramer HSD analysis indicated that copper did not vary significantly between Strata 1 and 2 (i.e., Mangrove Lagoon and Benner Bay), but was different from Strata 3, 4, and 5.

Comparison with Other Data. EPA's EMAP collected and analyzed sediments from four sites within the STEER for copper in 2004. The mean concentration of copper in the sediments from the EMAP work was 3.9 µg/g, substantially less than the mean concentration found in the STEER in the current study. It is unclear why the means for the stratified random sampling done in both studies were quite different, although it may be related to the greater number of samples taken in the current study, particularly in the Mangrove Lagoon area. As noted earlier, Mangrove Lagoon was not included in the EMAP work.

In southwest Puerto Rico, Pait *et al.* (2008) detected a mean copper concentration of 5.21 µg/g, lower than the mean in

the STEER. In Vi-eques, the mean copper concentration was 25.9 $\mu\text{g/g}$, and in Jobos Bay, Apeti *et al.* (2012b) found a mean copper concentration of 34.1 $\mu\text{g/g}$, somewhat higher than the mean found in the STEER.

The NS&T median for copper in sediments is 16 $\mu\text{g/g}$. From the stratified random sampling in 2011, six sites (2-20P, 1-1P, 1-3P, 1-2P, 2-16P, 1-5P), all either in Mangrove Lagoon or northern Benner Bay, had copper levels above the NS&T median. The NS&T 85th percentile for copper is 38.3 $\mu\text{g/g}$. Five of these sites had copper levels above the NS&T 85th percentile level. All four of the targeted samples were above the NS&T median and 85th percentile. Finally, the concentration in the sediment at BB-2, is one of the 10 highest concentrations of copper that has been detected in NOAA's NS&T Program.

NOAA Sediment Quality Guidelines for Copper. The ERL for copper is 34 $\mu\text{g/g}$ (Figure 4.15); the ERM is 270 $\mu\text{g/g}$. From the stratified random sites, six sites (2-20P, 1-1P, 1-3P, 1-2P, 2-16P, 1-5P) exceeded the ERL, but none exceeded the ERM for copper, indicating that more sensitive life stages in these areas could, however, be experiencing effects related to the presence of copper.

All four of the 2010 targeted sites (i.e., BB-1, BB-2, ML-2 and ML-10) were above the NOAA ERL for copper. At Site BB-2 (1,010 $\mu\text{g/g}$), the level of copper substantially exceeded the copper ERM of 270 $\mu\text{g/g}$, indicating that effects from this trace element on biota inhabiting the sediments are likely.

Copper in the Sediments in Northern Benner Bay and Mangrove Lagoon. The pattern of copper seen in the sediments in Mangrove Lagoon and Benner Bay, with higher concentrations in the northern part of Benner Bay, and somewhat

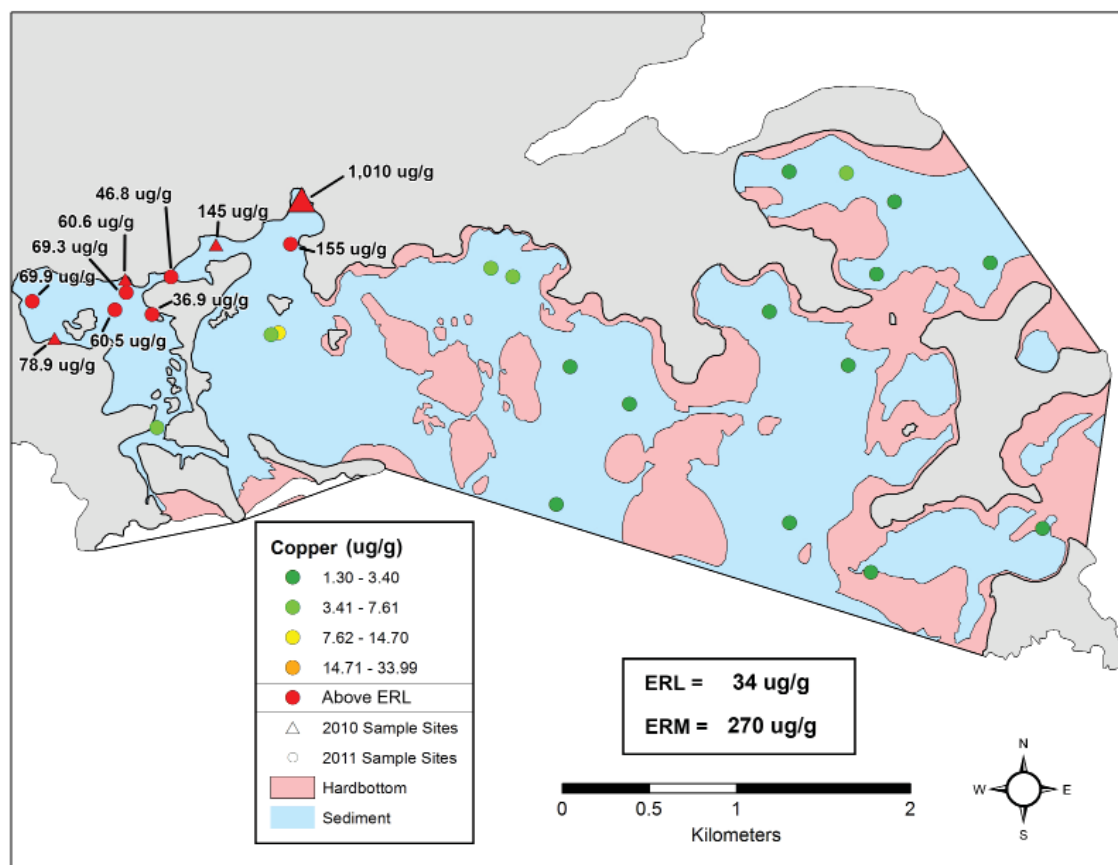


Figure 4.15. Copper detected in sediments from the St. Thomas East End Reserves.

lower concentrations in Mangrove Lagoon, suggests a possible gradient. The flow of water into and out of Mangrove Lagoon appears to have a number of driving forces, including a clockwise pattern through Cas Cay, a reversing tidal current, along with wind drift and runoff flow through both entrances (Tetra Tech, 2005). The three highest concentrations of copper were found at sites BB-1, 2-20P and BB-2, all in the northern part of Benner Bay. Given the variable direction of water currents in the area, a significant source of the higher copper concentrations in the sediments could be from the northern part of Benner Bay. However, additional work would be needed to assess this possibility.

Marinas and boat yards are a major commercial land use adjacent to the northern portion of Benner Bay. Copper has been used for years in antifoulant paint systems on boat hulls. Activities such as the mooring of vessels with hulls painted with copper-containing bottom paints, along with the cleaning, scraping and repainting of boat hulls, and subsequent runoff containing paint chips and dust, that might occur during a rainstorm, could lead to elevated concentrations of this metal in the environment. The elevated levels of TBT in the sediments in this same part of Benner Bay and lower levels of TBT in Mangrove Lagoon support the possibility that the copper found in the sediments in the northern part of Benner Bay are related to the use of

both copper and TBT over the years in boat-related activities. A nonparametric correlation analysis between copper and TBT in the STEER showed a very significant positive correlation (Spearman Rho = 0.7078, $p = 0.0001$). As noted earlier, TBT has had significant use as an antifoulant on boat hulls. For copper, however, inputs into Mangrove Lagoon from Turpentine Gut, runoff from residential areas, and inputs from the adjacent Bovoni Landfill could also be contributing sources.

In any case, the concentration of copper at BB-2 is indicative of levels that are likely impacting benthic organisms in this area. The next chapter contains the results of additional monitoring in northern Benner Bay, including an analysis of metals in sediment cores.

Effects of Sediment Grain Size. A nonparametric analysis revealed that copper was significantly correlated with the percent fines (Spearman Rho = 0.7852, $p < 0.0001$) fraction of the sediments.

Lead

The concentrations of the trace element lead detected in the STEER sediments are shown in Figure 4.16. The mean concentration was 5.87 ± 1.90 $\mu\text{g/g}$; the median was 0.82 $\mu\text{g/g}$. The highest concentration of lead in the sediments from the stratified random sampling was 31 $\mu\text{g/g}$ at 1-2P in Mangrove Lagoon. The lowest concentration detected was 0.371 $\mu\text{g/g}$ at 4-50P in Cowpet Bay. The highest concentration from the 2010 targeted sampling was 81.2 $\mu\text{g/g}$ at BB-2 in Benner Bay (Figure 4.16).

Variation of Lead Across Strata. A log10 transformation of the results failed to normalize the stratified-random sampling data. The data were subsequently ranked, and a Kruskal-Wallis test on the ranked data showed a significant

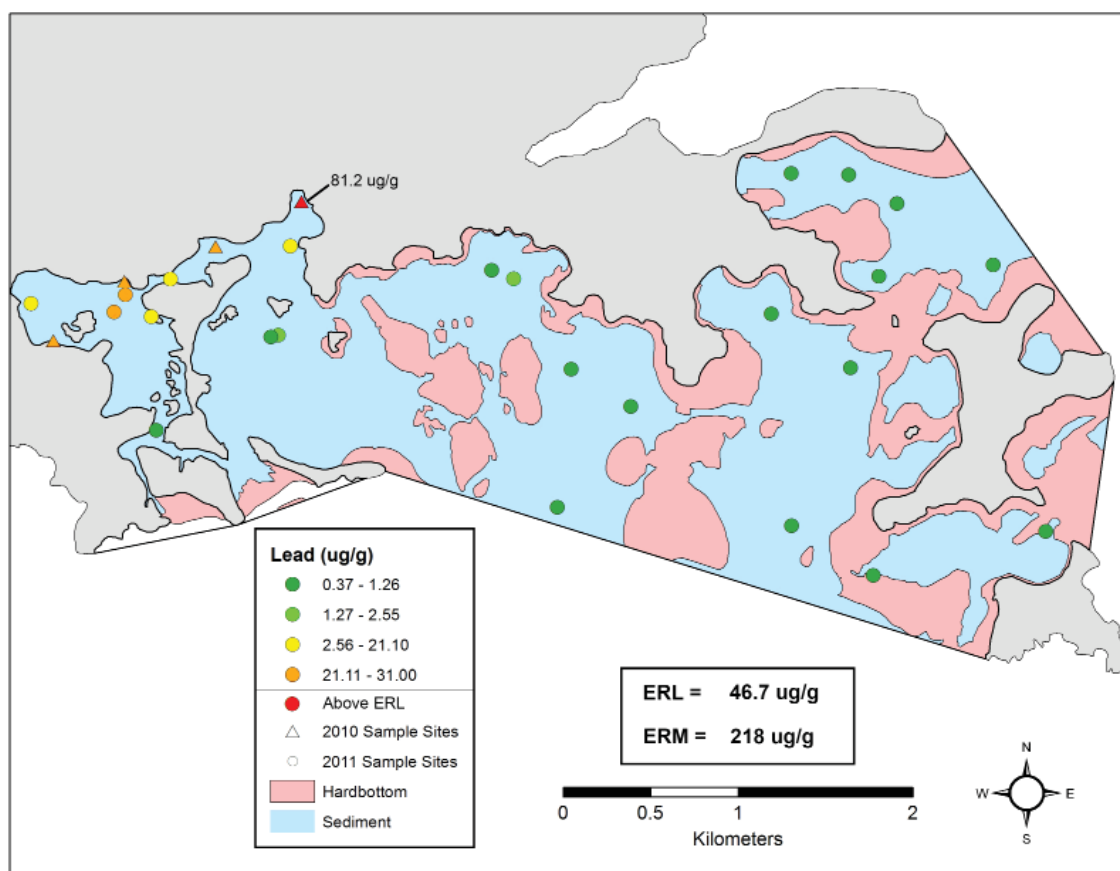


Figure 4.16. Lead detected in sediments from the St. Thomas East End Reserves.

($p < 0.0001$) difference in the concentration of lead across the STEER. A Tukey HSD test indicated that Stratum 1 was significantly higher than Strata 3, 4, and 5. The concentration of lead in Stratum 2 was not significantly different from Stratum 1 or Stratum 3, but was significantly higher than in Strata 4 and 5.

Comparison with Other Data. The mean concentration of lead in the sediments in the STEER analyzed by EMAP was 1.02 $\mu\text{g/g}$, lower than the mean concentration found in the STEER in the current study. In southwest Puerto Rico, Pait *et al.* (2008) detected a mean lead concentration of 1.93 $\mu\text{g/g}$, less than that found in the STEER. In Vieques, the mean lead concentration was 5.42 $\mu\text{g/g}$, close to that in the STEER. In Jobos Bay, the mean concentration of lead in the sediments sampled was 7 $\mu\text{g/g}$ (Apeti *et al.*, 2012b), also similar to the STEER.

The NS&T median sediment concentration for lead is 22.3 $\mu\text{g/g}$. Only two sites (1-2P and 1-3P) from the stratified random sampling, both in Mangrove Lagoon, had lead levels above the NS&T median. The NS&T 85th percentile for lead is 39.1 $\mu\text{g/g}$. None of the sites in the STEER from the stratified random sampling had a lead concentration in the sediments above this level. From the 2010 targeted

sampling, three of the four sites were above the NS&T median, with one site (BB-2) above the NS&T 85th percentile.

NOAA Sediment Quality Guidelines for Lead. The ERL for lead is 46.7 $\mu\text{g/g}$ (Figure 4.16); the ERM is 216 $\mu\text{g/g}$. None of the randomly chosen sites had a concentration above the ERL. However, BB-2 (81.2 $\mu\text{g/g}$) from the 2010 targeted sampling had a concentration above the ERL, but still well below the ERM of 218 $\mu\text{g/g}$.

Effects of Sediment Grain Size. A non-parametric correlation analysis between lead and percent fines in the sediment indicated a significant positive correlation (Spearman Rho = 0.7643, $p < 0.0001$).

Zinc

The results of the analysis of zinc in the sediments collected in the STEER are shown in Figure 4.17. The mean zinc concentration in the sediments from the stratified random sampling was $37.3 \pm 10.7 \mu\text{g/g}$; the median was 11.1 $\mu\text{g/g}$. The highest concentration of zinc from these collections was found at site 1-2P, at 159 $\mu\text{g/g}$. The second highest zinc concentration from the stratified random sampling was also in Mangrove Lagoon at site 1-3P, with a concentration of 154 $\mu\text{g/g}$.

From the 2010 targeted sampling, the highest zinc concentration detected was 392 $\mu\text{g/g}$ in Benner Bay (BB-2), followed by 192 $\mu\text{g/g}$ in BB-1, also in Benner Bay. As with a number of the other contaminants included in this report, higher concentrations were found in northern Benner Bay and in Mangrove Lagoon.

Variation of Zinc Across Strata. A Kruskal-Wallis test on the ranked values from the stratified random sampling indicated zinc varied significantly ($p = 0.0006$) by stratum,

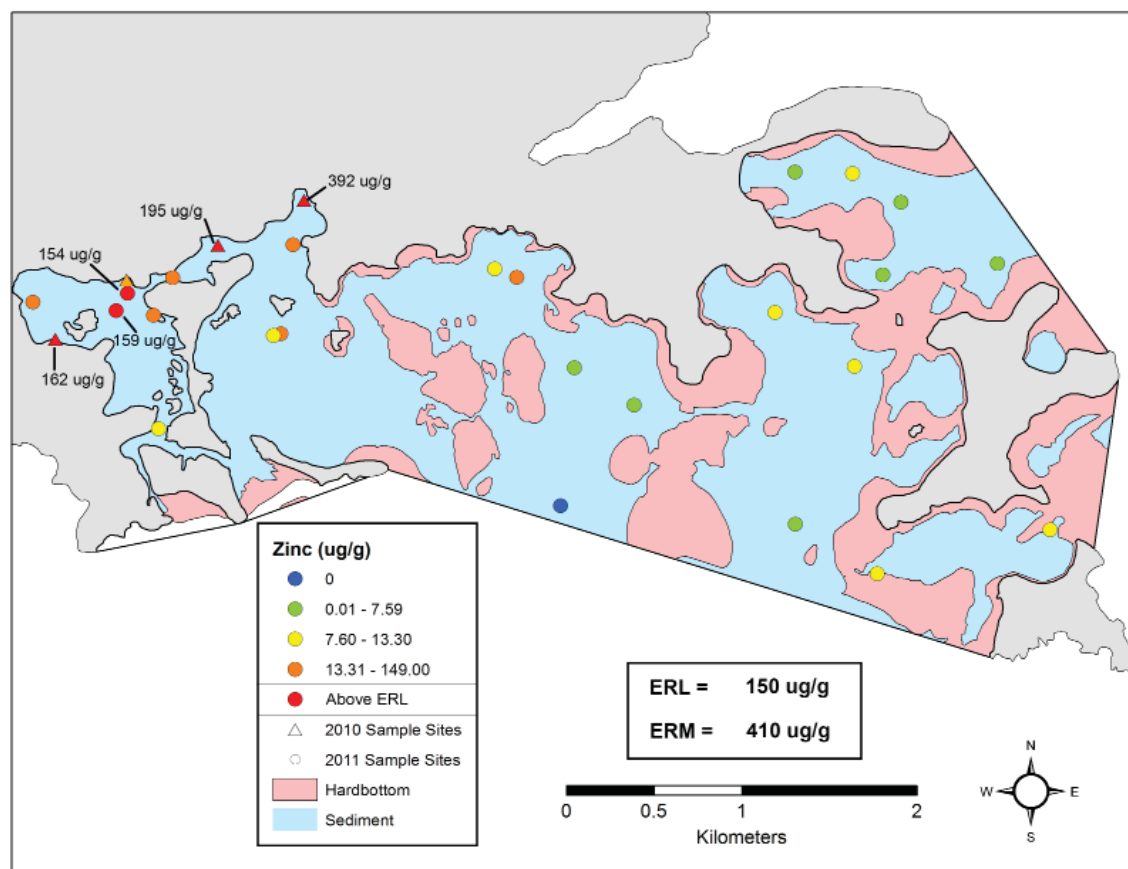


Figure 4.17. Zinc detected in sediments from the St. Thomas East End Reserves.

and the Tukey-Kramer HSD test indicated that Strata 1 and 2 were significantly different (higher) from Stratum 5.

Comparison with Other Data. The mean for the four samples that EPA's EMAP collected outside of Mangrove Lagoon in the STEER was 5.8 $\mu\text{g/g}$, substantially below the mean for the STEER in the current project. If the samples from Mangrove Lagoon are left out, the mean for the STEER is 18.4, still higher than was found by EMAP.

The concentration of zinc detected in the STEER can also be compared with work completed in southwest Puerto Rico. Pait *et al.* (2008) detected a mean zinc concentration of 8 $\mu\text{g/g}$, below the mean found in the STEER. In Viques, Pait *et al.* (2010) calculated a mean zinc concentration of 34.4 $\mu\text{g/g}$, similar to what was found in the STEER. In Jobos Bay, Apeti *et al.* (2012b) found a mean zinc concentration of 54.7 $\mu\text{g/g}$, somewhat higher than what was found in the STEER.

The NS&T median for zinc is 74 $\mu\text{g/g}$; the 85th percentile is 143 $\mu\text{g/g}$. From the stratified random sampling, there were six sites above the NS&T median, and two sites (1-2P and 1-3P) above the NS&T 85th percentile. From the 2010 targeted sampling, three (BB-2, BB-1, and ML-2) of

the four sites analyzed were above the NS&T 85th percentile.

NOAA Sediment Quality Guidelines for Zinc.

The ERL for zinc is 150 µg/g. There were two sites (1-2P and 1-3P) from the stratified random sampling that exceeded the ERL. From the 2010 targeted sampling, three of the four sites (BB-2, BB-1, and ML-2) analyzed exceeded the ERL. The results of this analysis indicate that some of the more sensitive species or life stages in both Benner Bay and Mangrove Lagoon could begin to experience effects related to the elevated levels of zinc. In addition, any additive effects that may be present as a result of multiple contaminants (e.g., TBT and copper) from this area are unknown. While none of the sites exceeded the ERM, BB-2 (392 µg/g) from the 2010 targeted sampling was fairly close to the zinc ERM of 410 µg/g.

Effects of Sediment Grain Size. The nonparametric Spearman's analysis between zinc and the sediment percent fines indicated a significant positive correlation (Spearman Rho = 0.5678, $p = 0.0038$) between zinc and the smaller grain sizes of the sediments sampled in the STEER. The correlation between zinc and percent clay (Spearman Rho = 0.7270, $p < 0.0001$) appeared somewhat higher than silt (Spearman Rho = 0.5235, $p = 0.0038$).

Clostridium perfringens

Although not a chemical contaminant, this bacterium is often included in the analyses done by NOAA's NS&T Program. This anaerobic, gram-positive staining rod-shaped bacterium frequently occurs in the intestines of humans, as well as in domestic and wild animals, and has been used as a human/animal waste indicator. The results of the analysis of sediments for *C. perfringens* are shown in Figure 4.18. Higher levels of *C. perfringens* were found primarily in

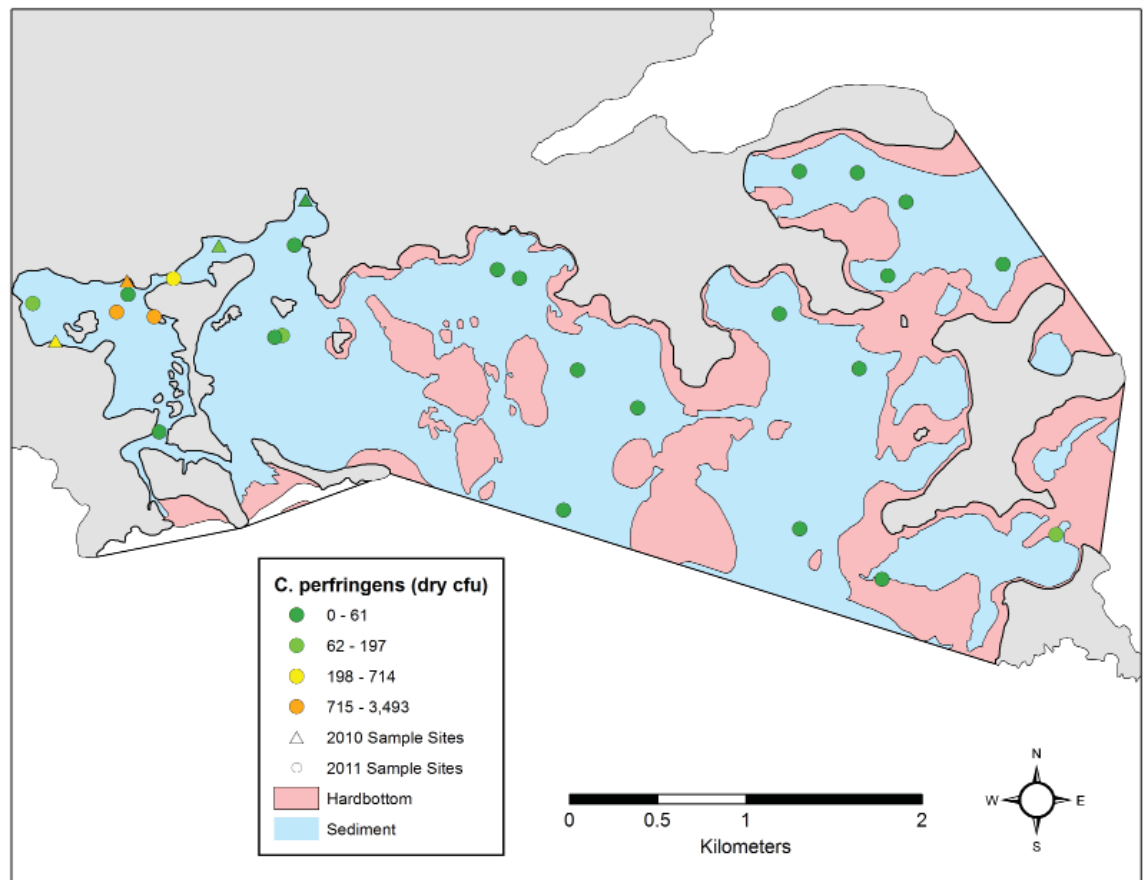


Figure 4.18. *Clostridium perfringens* detected in sediments from the St. Thomas East End Reserves.

Mangrove Lagoon, with lower concentrations in most other locations.

To assess the presence of viable *C. perfringens*, sediment extracts are plated on growth medium and the number of colonies that develop are counted. The mean *C. perfringens* concentration in the sediments was 291 ± 167 CFU/g (colony forming units per gram of dry sediment). The sediment sample from 1-2P contained 3,493 CFU. Site 1-3P had a *C. perfringens* count of 2,137 CFU/g. Both 1-2P and 1-3P are near the mouth of Turpentine Gut, as it enters Mangrove Lagoon, which may be indicative of input from septic systems, domestic animals such as dogs, and from a nearby horse race track. From the 2010 targeted sampling, the highest concentration of *C. perfringens* found was at ML-10, also at the mouth of Turpentine Gut (2,558 CFU/g).

Variation of *C. perfringens* Across Strata. A Kruskal-Wallis test run on the ranked *C. perfringens* data indicated a significant difference ($p < 0.0001$) between strata. A Tukey HSD test indicated that Strata 1 and 2 were significantly different (higher counts) from Strata 3, 4 and 5.

An analysis of *C. perfringens* and grain size was also carried out. A strong ($p = 0.0010$) correlation was found

between the log10 normalized *C. perfringens* data and percent fines. A nonparametric analysis indicated a significant negative correlation between *C. perfringens* and percent sand (Spearman Rho = -0.6342, $p = 0.0009$), but no correlation between *C. perfringens* and percent gravel (Spearman Rho = 0.1907, $p = 0.3722$).

Sediment Quality Guidelines and *C. perfringens*. No NOAA or other health guidelines exist for *C. perfringens* in sediments. *C. perfringens* is a common cause of foodborne illnesses. A more severe form of the disease can be fatal and results from ingesting large numbers of the active bacteria, typically from food. *C. perfringens* also has the capability of forming spores which can persist in soils and sediments.

The high levels of *C. perfringens* within Mangrove Lagoon indicate there is a need to reduce wastewater, stormwater and various sources of inputs for this pathogen, and other pathogenic microorganisms that may be present as well. Reducing the levels of bacterial contamination would not only benefit ecological health, but human health as well.

Sediment Toxicity

The use of sediment toxicity bioassays, along with the benthic infaunal community analysis, provides important information on the impacts of the chemical contaminants present in STEER sediments. While the NOAA ERMs and ERLs provide an indication of the likelihood of effects from one chemical contaminant or chemical contaminant class, the bioassays (and the benthic community analysis) integrate the effects of all contaminants present along with other environmental parameters. For this project, the sediment toxicity bioassays included amphipod (*Ampelisca abdita*) mortality, sea urchin (*Arbacia punctulata*) fertilization impairment, and cytochrome P450 Human Reporter Gene System (HRGS) response tests. The bioassay results for all tests are summarized in Table 4.4. All values are control corrected.

Table 4.4. Toxicity bioassay summary results from the STEER.

Site	Amphipod Mortality (%) Different from Control	Sig	Sea Urchin Fertilization Failure (%) Different from Control	Sig	P450 Response	BaP Eq. (ng/g)
1-1P	10.31		23.7	*	4.94	5.64
1-2P	14.43	*	14.5	*	13.79	12.98
1-3P	4.12		8.2		14.24	44.83
1-4P	7.22		6.3		9.86	0.12
1-5P	15.46		97.6	*	15.66	0.80
2-16P	12.37	*	40.4	*	11.12	8.33
2-19P	4.12		2.9		2.39	
2-20P	8.25		0.0		13.35	1.78
2-24A	13.40		5.1		2.88	
3-32P	10.31		15.5	*	15.64	8.58
3-33P	7.22		65.9	*	2.47	
3-37A	10.31		9.2		3.36	
3-38A	7.22		0.0		4.76	2.03
3-45A	47.42	*	0.0		2.49	
4-46P	29.90	*	30.5	*	2.20	
4-47P	21.65	*	13.8		2.75	
4-48P	12.37		13.3		1.23	
4-49P	10.31		18.2	*	2.73	
4-50P	37.11	*	6.8		1.49	
5-61P	9.28		0.0		1.45	
5-62P	22.68	*	3.6		1.90	
5-70A	53.61	*	0.2		5.57	
5-71A	4.12		5.8		3.14	0.97
5-75A	28.87	*	13.6		2.15	
* - statistically different from control						

Amphipod Toxicity

Significant amphipod mortality occurred throughout the STEER study area. The highest mortality values observed were in the eastern strata. This may be influenced by sediment grain size. The amphipod *A. abdita* normally live in silty sand habitats and may not thrive well in coarse sand (ASTM, 2008), however, this has not been tested rigorously. All the amphipod bioassays with mortality elevated above 20% were in sediments that were greater than 60% sand and gravel (Figure 4.19). Other parameters (e.g., higher TOC) may ameliorate this effect. Most of the highly toxic amphipod results were in coarse sand with very low TOC (Figure 4.20).

Sea Urchin Fertilization

Half of the significant sea urchin fertilization bioassays were in sediments from Mangrove Lagoon or the canal

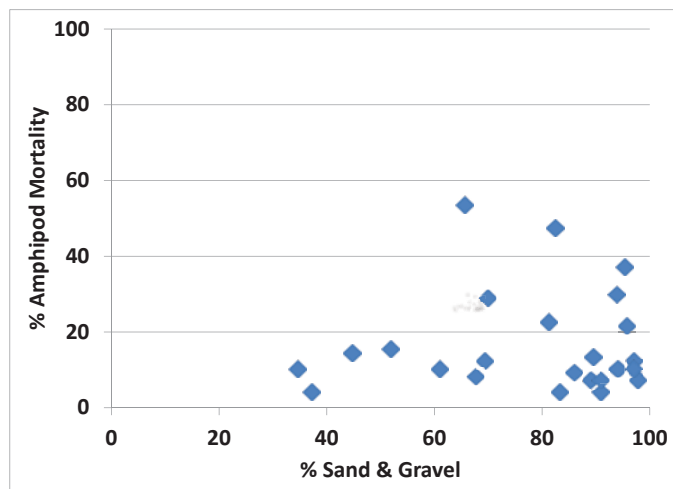


Figure 4.19. Amphipod mortality vs % sand and gravel in St. Thomas STEER sediments.

joining it to Benner Bay (Table 4.4). Jersey Bay and St. James Bay also showed significant results in two locations.

P450

Most of the significant P450 responses were in the western strata, including all of the Mangrove Lagoon sites. Further testing of those samples that exceeded 50% of the 10 nM TCDD (tetrachlorodibenzodioxin) standard are also presented in Table 4.4 in terms of B[a]P eq. The P450 response in terms of B[a]P eq is strongly correlated with PAH concentrations (Figure 4.21).

Results of the timed exposure (6 vs 24 hrs) to test for the relative contribution of labile versus persistent contaminants are shown in Table 4.5. In all cases, the 6 hr incubation showed a higher response than the 24 hr incubation (Figure 4.22). This indicates that the predominant chemical classes the cells were responding to were PAHs, as they are more easily degraded than the more recalcitrant PCBs. It is noteworthy that the level of response in Mangrove Lagoon and portions of Benner Bay and Cowpet Bay exhibit initial responses as high as the spiked sample and the positive control.

Taken together, the bioassay results indicate a gradient of effect from west to east within the STEER (Figure 4.23). Toxic responses occurred in all strata, but the western portion of the study area exhibited significant results from multiple bioassays. As noted above, several of the amphipod bioassay results may be an artifact of the coarse coral-line sediment present in many of the eastern sites (Strata 4 and 5).

With the exception of TBT and copper, extremely high concentrations of individual chemical pollutants were not seen.

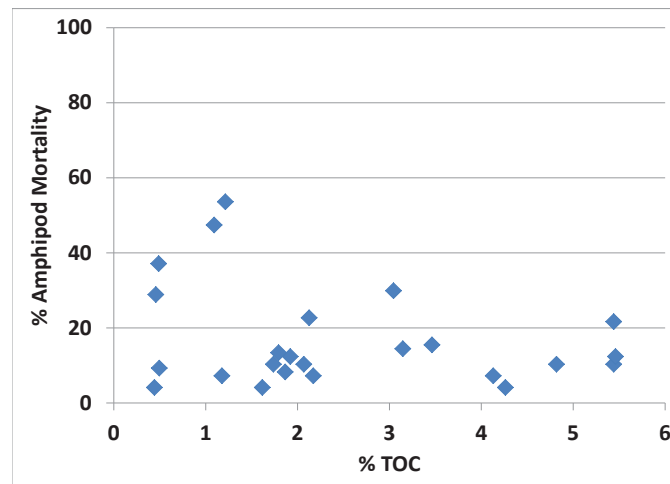


Figure 4.20. Amphipod mortality versus % total organic carbon (%TOC) in the STEER sediments.

However, the observed widespread toxicological responses indicate the interaction of a variety of factors, including multiple contaminants, physicochemical characteristics of the sediment, and likely chemical pollutants beyond the standard list of analytes that may vary from stratum to stratum.

Benthic Community Analysis

A total of 10,926 organisms were enumerated, comprised of 434 taxa (species or higher taxonomic level). Following elimination of epibenthic species (dwelling on hard surfaces, not within the sediment) and 'artificial' species (see page 84), there were 333 taxa and 10,605 individuals for analysis. There were 168 rare and unique taxa. One hundred fifty seven taxa were found at only one location. Annelids were the dominant taxa, accounting for 59.2% of all the organisms. Mollusks and arthropods accounted for 19.2 and 15.7%, respectively. Less than 1% of the animals were echinoderms. In the STEER, abundance was dominated by two dozen taxa that were found throughout most strata and a large number of taxa only represented by a few individuals (Figure 4.24).

Abundance was more uniform on average than diversity or number of species, but some locations had extremely low abundance and some sites were extremely high (Figure 4.25). Site 1-1P in Mangrove Lagoon only had 4 species and five organisms in total. Site 2-16P had over 1,000 organisms.

Gradients of diversity and species richness were seen from low in the west to higher in the eastern strata (Figure 4.26). With two exceptions (Sites 1-4P and 2-19P), diversity was higher in the eastern strata than in Mangrove Lagoon and Benner Bay. These are reflected in the calculated correla-

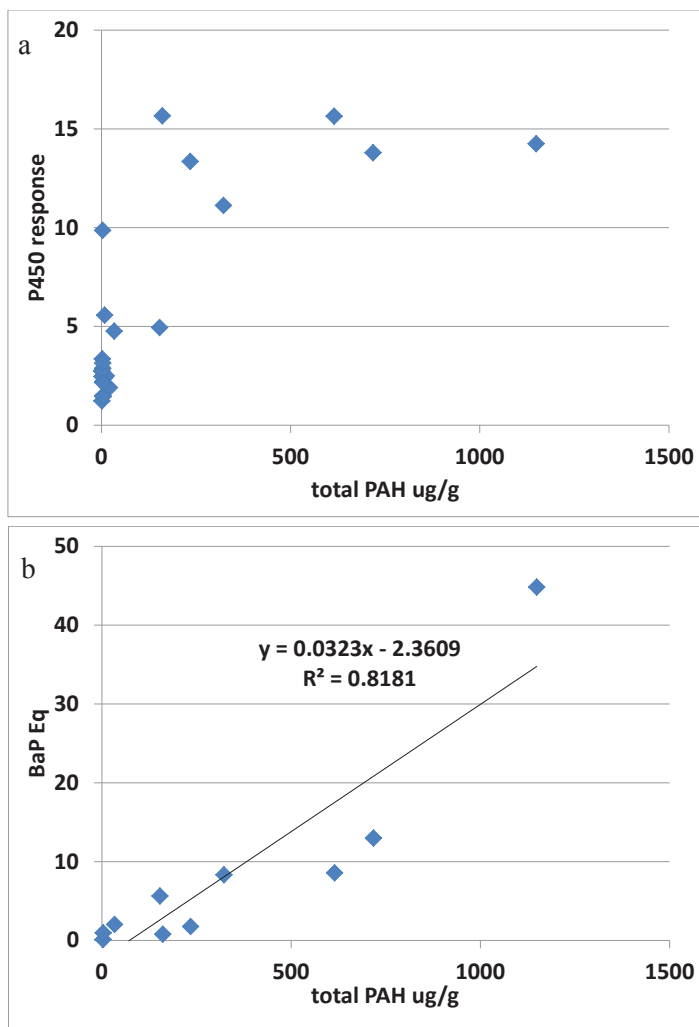


Figure 4.21. P450 response vs PAH concentrations (a), P450 response in terms of B[a]P eq for the subset of samples that exceeded the TCDD standard response (b).

tion coefficients. Number of species and diversity were negatively correlated with the ERMq, (Table 4.6). The ERMq is calculated by dividing the concentration of each contaminant analyzed in the sediment by its available ERM to produce a quotient. Number of species and diversity were positively correlated with stratum. There were no significant correlations with amphipod mortality or sea urchin fertilization failure. The number of species and diversity were significantly and negatively correlated with the PAH benzo[a]pyrene concentrations. A similar pattern was seen with respect to the percentage of fines in the sediments, and sand plus gravel showed the inverse.

Nodal Analysis

The nodal analysis revealed two site groups and three species groups. The site groups divided cleanly between Stratum 1 and 2 (Mangrove Lagoon and Benner Bay) vs 3, 4, and 5. There was almost no overlap in species makeup between the site groups. In Mangrove Lagoon and Benner

Table 4.5. Average (n = 3) P450 bioassay response following 6 and 24 hour incubation with STEER sediment extracts.

Site	6 Hour Response	24 Hour Response
1-1P	104.9	13.3
1-2P	113.3	24.7
1-3P	117.3	35.0
1-4P	78.5	10.4
1-5P	129.8	14.1
2-16P	89.7	22.6
2-19P	12.3	3.53
2-20P	113.4	29.0
2-24A	17.3	1.37
3-32P	134.4	41.0
3-33P	39.0	3.00
3-37A	21.6	9.24
3-38A	100.0	9.44
3-45A	39.8	5.22
4-46P	11.0	3.19
4-47P	16.1	3.28
4-48P	6.12	1.74
4-49P	15.9	1.86
4-50P	13.0	2.46
5-61P	11.9	0.30
5-62P	24.0	1.42
5-70A	42.0	3.84
5-71A	57.0	5.17
5-75A	30.6	3.26
clean	4.44	0.56
blank	-1.39	0.10
spike	125.5	46.5
contam	123.2	55.9

Values are expressed as % of the 10nM TCDD standard response. QA/QC samples: blank = solvent blank; clean = uncontaminated site in the Chesapeake Bay; contam = contaminated site in Chesapeake Bay; spike = spiked solvent solution.

Bay, there were 25 species, versus 109 species found in the other three strata. Species that were found in Mangrove Lagoon and Benner Bay were generally rarely found or were completely absent in the other three strata, and vice versa. The three species groups corresponded to the site groups. Mangrove Lagoon and Benner Bay have a different species assemblage than the other three strata, and were dominated by polychaete worm species that are neither widespread nor numerous in the rest of the STEER. Strata 3, 4 and 5 shared a common assemblage of species.

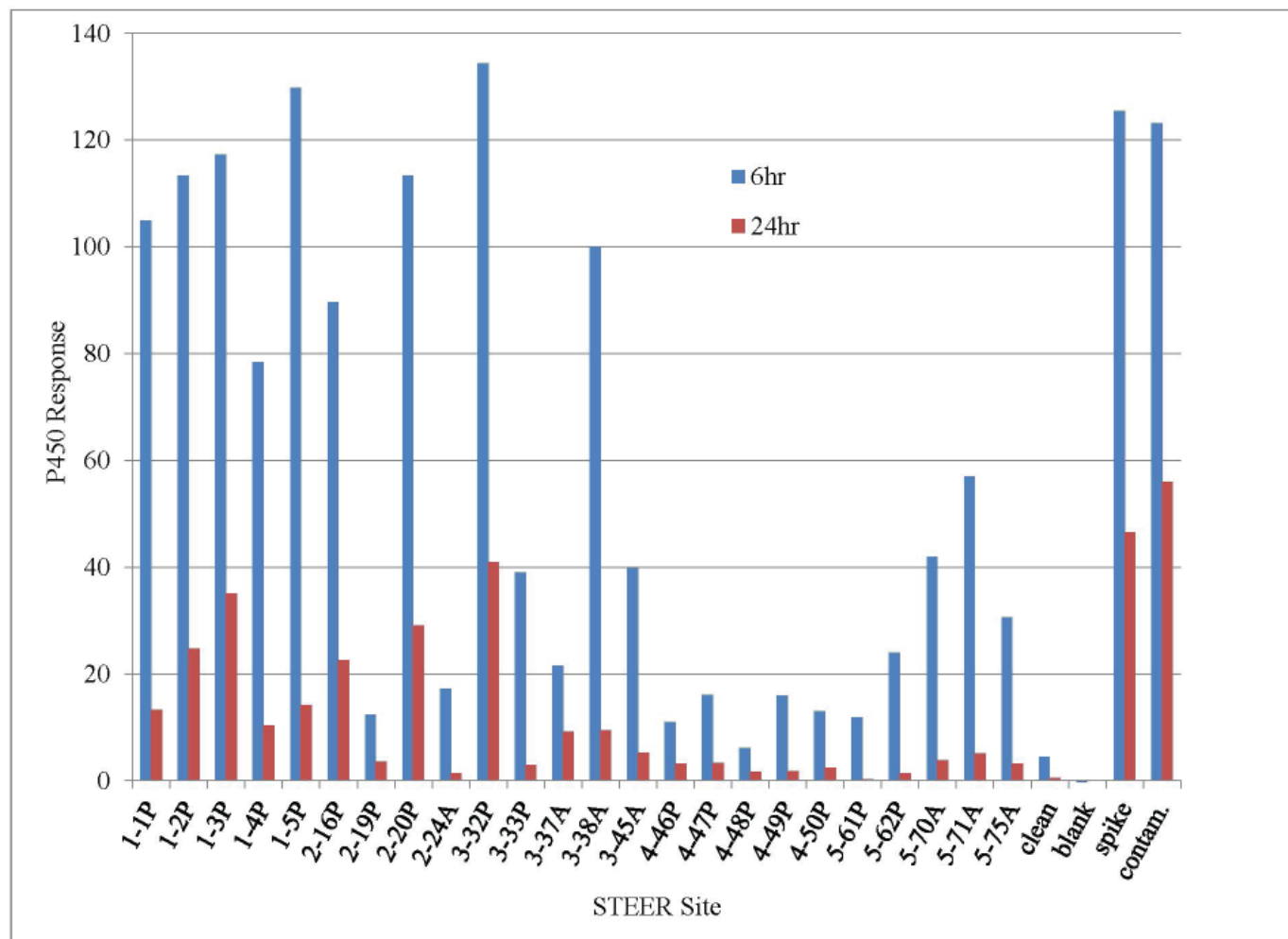


Figure 4.22. P450 response (normalized to standard 10nM TCDD) following 6 and 24 hour exposures to extracts from St. Thomas STEER sediment samples. (QA/QC samples; blank=solvent blank; clean=uncontaminated site in Chesapeake Bay; contam.=contaminated site in Chesapeake Bay; spiked solvent solution).

The third group of 26 species were found at sites 1-4P and 2-19P (hereinafter referred to as excluded sites). These species were rarely found elsewhere in either Mangrove Lagoon and Benner Bay, or the other three strata. These two sites had a unique species assemblage, different from the other areas. Site 1-4P in the lower part of Mangrove Lagoon away from the influence of Turpentine Gut and the landfill, and 2-19P in central Benner Bay have sediment that is predominantly sand, unlike most of the other sites in those strata. These two locations shared a number of species found within Strata 1 and 2, but also another set of species that were much more diverse and included polychaetes, crustaceans, bivalves and snails.

SIMPER Analysis

The difference in dominant species between strata is also illustrated in the results from the SIMPER analysis. Figure 4.27 shows the similarity index values for Strata 1 and 2 (Mangrove Lagoon and Benner Bay, respectively), and the

values for the excluded sites (excluded from the Stratum 1 and 2 lists), ranked from highest to lowest. Only a small number of species have high scores in Strata 1 and 2. That is, only a few are dominant. In contrast, none of the species in the two excluded sites have high scores. They are all equally important in defining the community makeup. The top 15 species in each group are listed in Table 4.7. Species highlighted in yellow were found in both Strata 1 and 2 (the colors are arbitrary). The strata have very similar dominant species makeup (there are many overlapping species that have high index values in both strata). Only two of the top species found in the excluded sites were present in both Strata 1 and 2. Thus, the species that are considered 'typical' in Strata 1 and 2 were generally not 'typical' in the two excluded sites (1-4P and 2-19P). The taxa highlighted in green were found in all five strata. Tubificids are a family of oligochaete worms. Nemerteans, also called proboscis worms, constitute an entire phylum. Both are difficult to identify to species without highly specialized expertise. Thus, these taxa actually represent potentially hundreds of

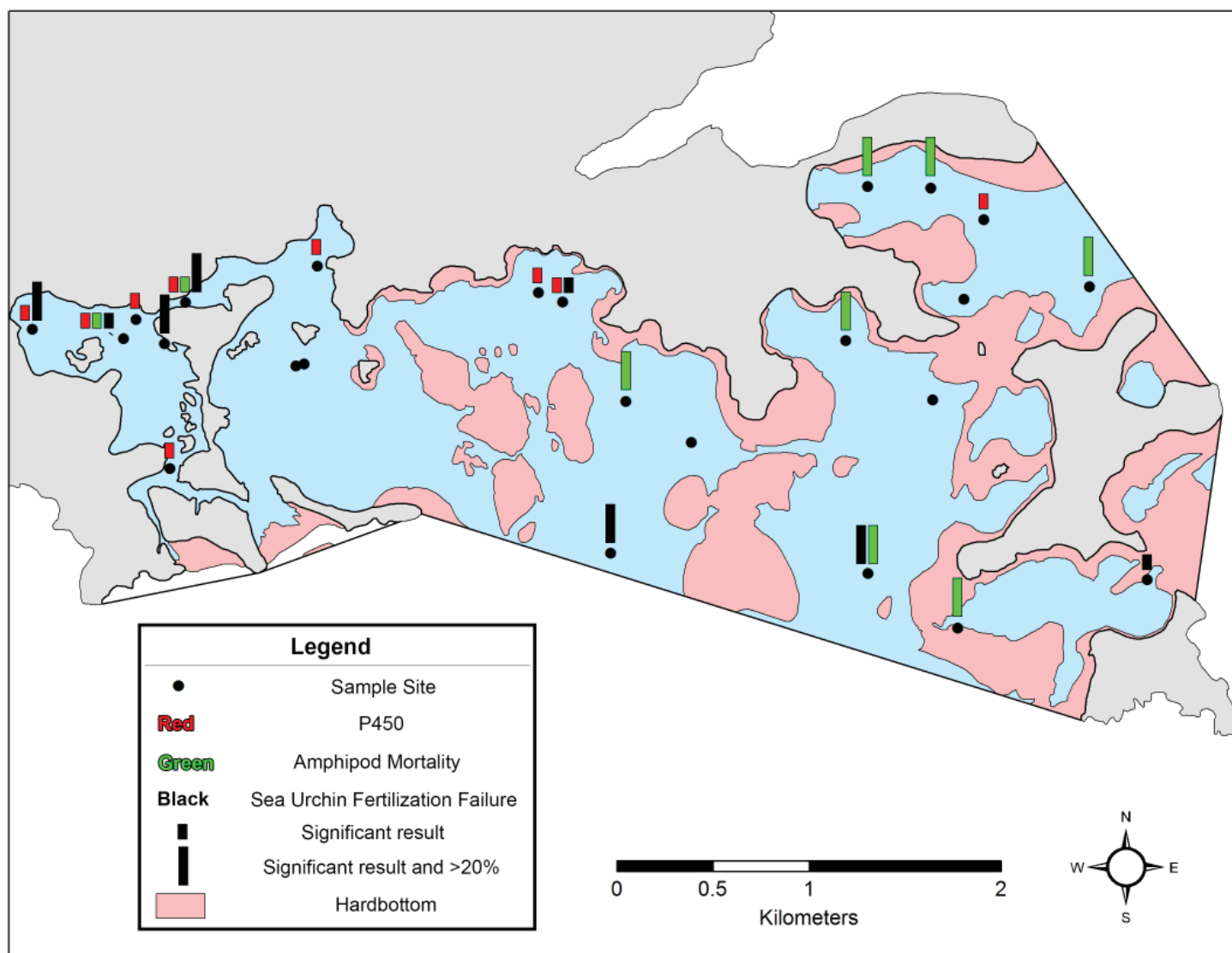


Figure 4.23. Distribution of bioassays showing responses that were significantly different than controls or greater than a standard threshold (P450) in St. Thomas STEER sediments.



Figure 4.24. Plot of total abundance of each taxa used in the analyses. Each triangle represents the total abundance of each individual taxa collected in the STEER.

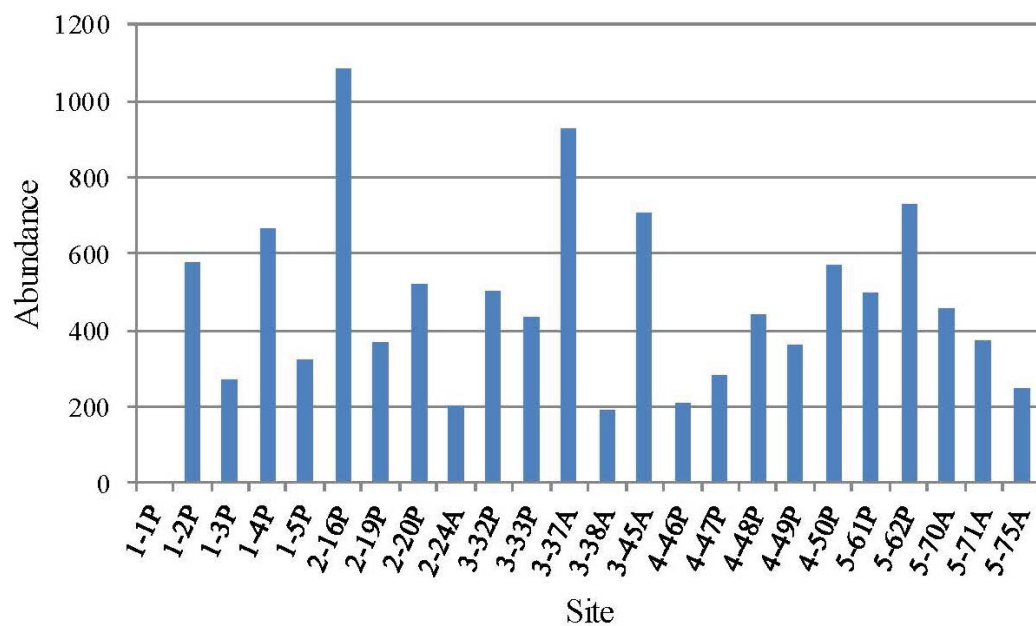


Figure 4.25. Total animal abundance at each station in the STEER.

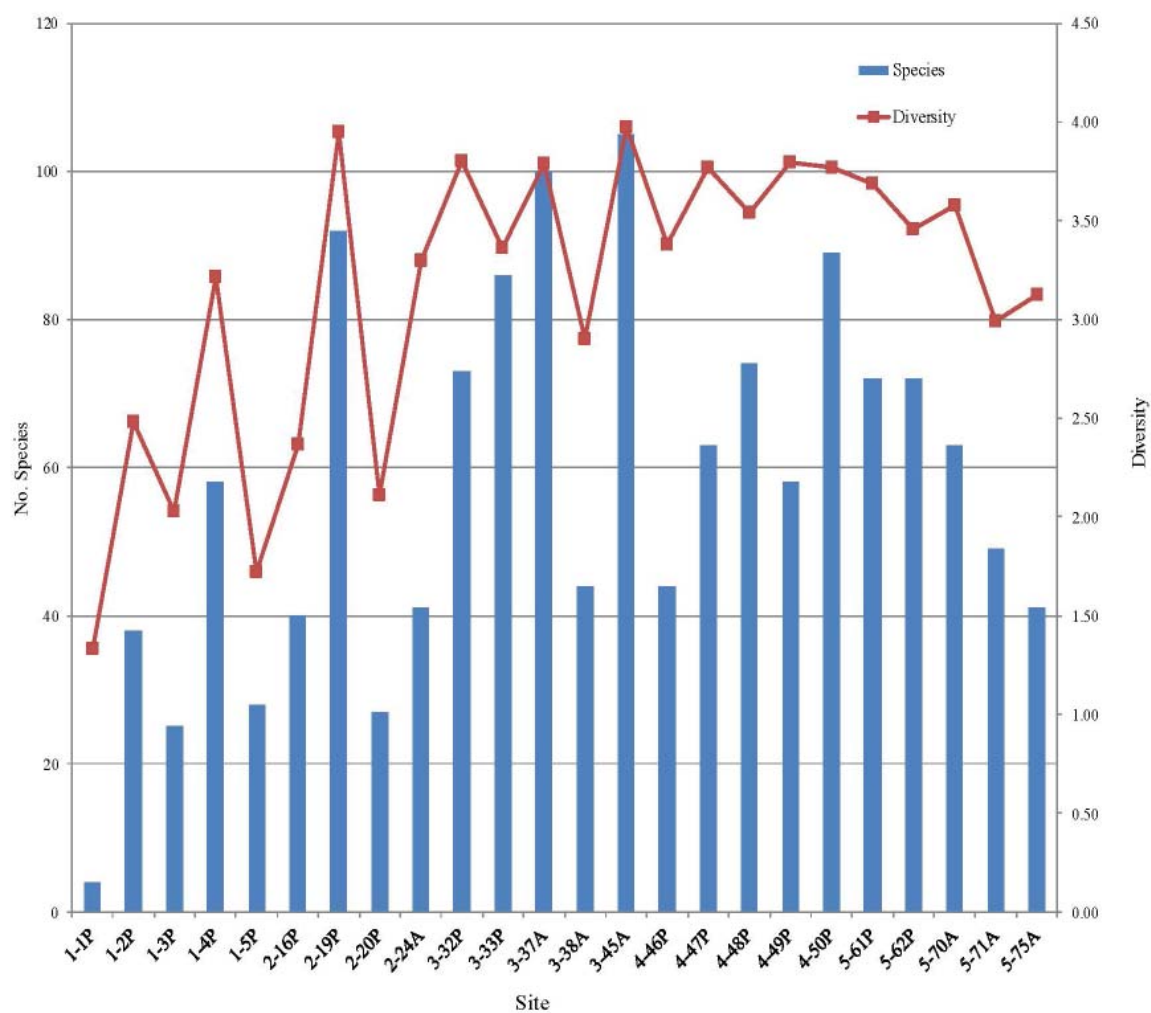


Figure 4.26. Total number of species (taxa) and calculated diversity at each station in the STEER.

Table 4.6. Spearman Rank correlation coefficients (bold) and significance level for community parameters and selected physical and chemical parameters, and toxicological results.

Parameter	Statistics	ERMq	Stratum	Amphipod Mortality	Fertilization Failure	B[a]P	%Silt and %Clay (Fines)	%Sand and %Gravel
Species	Spearman Rho	-0.65154	0.44292	0.12172	-0.25736	-0.64343	-0.62068	0.62068
	Significance	0.0006	0.0302	0.571	0.2247	0.0007	0.0012	0.0012
Abundance	Spearman Rho	0.16565	0.04081	0.13534	-0.16166	-0.01359	0.01913	-0.01913
	Significance	0.4392	0.8498	0.5283	0.4505	0.9497	0.9293	0.9293
Diversity	Spearman Rho	-0.56542	0.47125	0.22625	-0.24324	-0.65588	-0.55937	0.55937
	Significance	0.004	0.0201	0.2877	0.2521	0.0005	0.0045	0.0045

species and are not particularly informative. *Syllis cornuta* is a polychaete worm with a worldwide distribution. Thus, outside of these cosmopolitan taxa, the excluded sites had very little in common with the species makeup of Strata 1 and 2.

Figure 4.28 shows the indices for the species found in Strata 3-5. The numbers of species in these strata are much higher than in Strata 1 and 2. All the scores are relatively low because of the much higher diversity than in Strata 1 and 2, but the inflection of the curves shows that the top 15-20 species are the most important. Table 4.8 shows the top 15 species in each stratum, plus the excluded sites (1-4P and 2-19P) for comparison. Species names highlighted in blue were found in all three strata. Again, the taxa highlighted in green were cosmopolitan and are not informative. The species makeup of Strata 3, 4 and 5 is very similar. That is, the species highlighted in blue are typical in all three strata. The dominant species found in the excluded sites has almost no overlap with these strata. Thus there are three distinct species assemblages in the STEER. One in Mangrove Lagoon and Benner Bay, which are relatively depauperate. The two excluded sites that share some of the species found in Strata 1 or 2, but also another group of species not common anywhere else. The third assemblage is a much more diverse group, and occupies the bulk of the STEER area.

Overall Patterns in the STEER Benthic Community

The average diversity and species richness is lower in Mangrove Lagoon and Benner Bay than the other strata, but abundance varies throughout the study area. Diversity and species richness are better indicators of stress than abundance, however. Extremely depressed abundance is indicative of highly stressed habitats, but marginally stressed areas may have as high or higher an abundance of organ-

isms as healthy habitats, because those species that can thrive in stressed habitats may be released from competitive and predation pressure. Species in stressed environments also tend to have opportunistic and mobile life styles.

The composition of species present in various locations is also an indicator of stressed habitats. Many authors consider amphipods and echinoderms to be relatively sensitive to contaminant stress (Long *et al.*, 2001, Llanso *et al.*, 2002). Similarly, several types of polychaetes, such as Spionids and Capitellids, and oligochaete tubificids, are considered to be tolerant of contamination and/or other stressful conditions, such as hypoxia (Lenihan and Micheli. 2001; Llanso *et al.*, 2002).

The community makeup of specific taxonomic groups in the different strata is shown in Table 4.9. Average total abundance of the large taxonomic groups was not informative. However, pollution tolerant species of annelids make up a much larger proportion of the organisms in Mangrove Lagoon and Benner Bay than in the two excluded sites or any other strata. The number of amphipods show the opposite pattern.

Influence of Habitat Characteristics and Chemical Contamination on the Benthic Infaunal Community

Both species richness and diversity declined with ERMq (Figure 4.29, Table 4.6). Likewise, species richness and diversity declined with increasing percent fines (percent silt+clay) in the sediment (Figure 4.30). The relationship between contaminants and muddy sediment is clear (Figure 4.31), but which is the causative factor for reduced species community condition cannot be determined from the data. Resident communities found in muddy areas are inherently different from the areas with coarser grained sediments. However, it is clear that the occurrence of significant toxic-

Table 4.7. Results of SIMPER analysis for Strata 1 and 2. Taxa highlighted in green were cosmopolitan and were found in all strata. Taxa highlighted in yellow were found in strata 1 and 2.

Stratum 1		Stratum 2		Excluded Sites	
Index Value	Species	Index Value	Species	Index Value	Species
23.8	<i>Tubificidae (lpil)</i>	15.3	<i>Tubificidae (lpil)</i>	6.11	<i>Tubificidae (lpil)</i>
13.07	<i>Macoma brevifrons</i>	14.83	<i>Mediomastus (lpil)</i>	5.32	<i>Prionospio heterobranchia</i>
6.23	<i>Prionospio heterobranchia</i>	14.01	<i>Prionospio heterobranchia</i>	4.65	<i>Schistomeringos pectinata</i>
5.98	<i>Podarkeopsis levifuscina</i>	11.95	<i>Nemertea (lpil)</i>	4.65	<i>Branchiomma nigromaculata</i>
5.56	<i>Caecum pulchellum</i>	11.64	<i>Cirrophorus lyra</i>	4.65	<i>Maldanidae (lpil)</i>
4.71	<i>Exogone verugera</i>	7.19	<i>Podarkeopsis levifuscina</i>	4.47	<i>Podarkeopsis levifuscina</i>
4.01	<i>Nemertea (lpil)</i>	3.68	<i>Schistomeringos pectinata</i>	4.47	<i>Sabellidae (lpil)</i>
3.99	<i>Mediomastus (lpil)</i>	3.66	<i>Pseudopolydora (lpil)</i>	4.27	<i>Cumella (lpil)</i>
3.94	<i>Naineris setosa</i>	3.08	<i>Scoletoma verrilli</i>	4.27	<i>Leptochelia forresti</i>
3.89	<i>Pseudopolydora (lpil)</i>	2.87	<i>Syllis cornuta</i>	4.27	<i>Nemertea (lpil)</i>
3.83	<i>Caulleriella cf. alata</i>	2.6	<i>Aoridae (lpil)</i>	4.27	<i>Syllis cornuta</i>
3.67	<i>Podarke obscura</i>	2.59	<i>Capitella capitata</i>	3.76	<i>Chione cancellata</i>
3.58	<i>Sphaerosyllis piriferopsis</i>	2.41	<i>Exogone verugera</i>	3.76	<i>Mesanthura bivittata</i>
1.89	<i>Sabellidae (lpil)</i>	2.18	<i>Nereis acuminata</i>	3.76	<i>Cirratulidae (lpil)</i>
1.81	<i>Grandidierella bonnieroides</i>	2.03	<i>Schistomeringos rudolphi</i>	3.76	<i>Terebellidae (lpil)</i>

Table 4.8. Top 15 species in each stratum plus excluded sites. Taxa highlighted in green were cosmopolitan and were found in all strata. Taxa highlighted in blue were found in strata 3, 4, and 5.

Stratum 3		Stratum 4		Stratum 5		Excluded Sites	
Index Value	Species	Index Value	Species	Index Value	Species	Index Value	Species
5.01	<i>Nemertea (lpil)</i>	5.44	<i>Nemertea (lpil)</i>	6.83	<i>Nemertea (lpil)</i>	6.11	<i>Tubificidae (lpil)</i>
4.47	<i>Galathowenia oculata</i>	5.42	<i>Exogone lourei</i>	6.4	<i>Lucinidae (lpil)</i>	5.32	<i>Prionospio heterobranchia</i>
4.05	<i>Exogone lourei</i>	5.22	<i>Tubificidae (lpil)</i>	5.1	<i>Tubificidae (lpil)</i>	4.65	<i>Schistomeringos pectinata</i>
4.02	<i>Prionospio (lpil)</i>	4.82	<i>Lucinidae (lpil)</i>	4.97	<i>Prionospio steenstrupi</i>	4.65	<i>Branchiomma nigromaculata</i>
3.84	<i>Armandia maculata</i>	4.79	<i>Heteropodarke formalis</i>	4.94	<i>Galathowenia oculata</i>	4.65	<i>Maldanidae (lpil)</i>
3.69	<i>Lucinidae (lpil)</i>	4.52	<i>Armandia maculata</i>	4.82	<i>Exogone rolani</i>	4.47	<i>Podarkeopsis levifuscina</i>
3.59	<i>Maldanidae (lpil)</i>	3.21	<i>Pionosyllis gesae</i>	4.18	<i>Cumella (lpil)</i>	4.47	<i>Sabellidae (lpil)</i>
3.49	<i>Ischyroceridae (lpil)</i>	3.11	<i>Pseudoleptochelia (lpil)</i>	3.64	<i>Aricidea taylori</i>	4.27	<i>Cumella (lpil)</i>
3.4	<i>Sipuncula (lpil)</i>	2.96	<i>Terebellidae (lpil)</i>	3.62	<i>Codakia (lpil)</i>	4.27	<i>Leptochelia forresti</i>
2.48	<i>Aspidosiphon (lpil)</i>	2.89	<i>Fabricinuda trilobata</i>	3.47	<i>Leptochelia (lpil)</i>	4.27	<i>Nemertea (lpil)</i>
2.35	<i>Fabricinuda trilobata</i>	2.89	<i>Caulleriella cf. alata</i>	3.26	<i>Xenanthura brevitelson</i>	4.27	<i>Syllis cornuta</i>
2.34	<i>Sphaerosyllis piriferopsis</i>	2.87	<i>Saltipedis (lpil)</i>	3.13	<i>Exogone lourei</i>	3.76	<i>Chione cancellata</i>
2.33	<i>Exogone dispar</i>	2.85	<i>Tellinidae (lpil)</i>	3.1	<i>Sipuncula (lpil)</i>	3.76	<i>Mesanthura bivittata</i>
2.16	<i>Ophiuroidea (lpil)</i>	2.71	<i>Protodorrvillea kefersteini</i>	2.81	<i>Pitar simpsoni</i>	3.76	<i>Cirratulidae (lpil)</i>
2.13	<i>Ampharetidae (lpil)</i>	2.68	<i>Magelona sp. c</i>	2.6	<i>Armandia maculata</i>	3.76	<i>Terebellidae (lpil)</i>

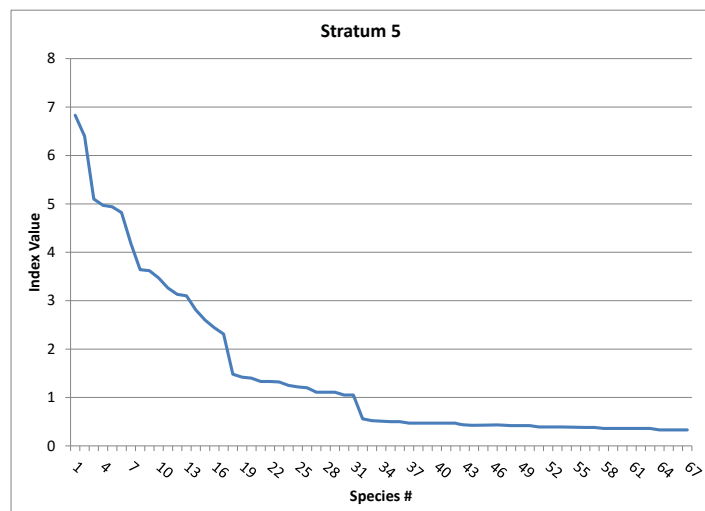
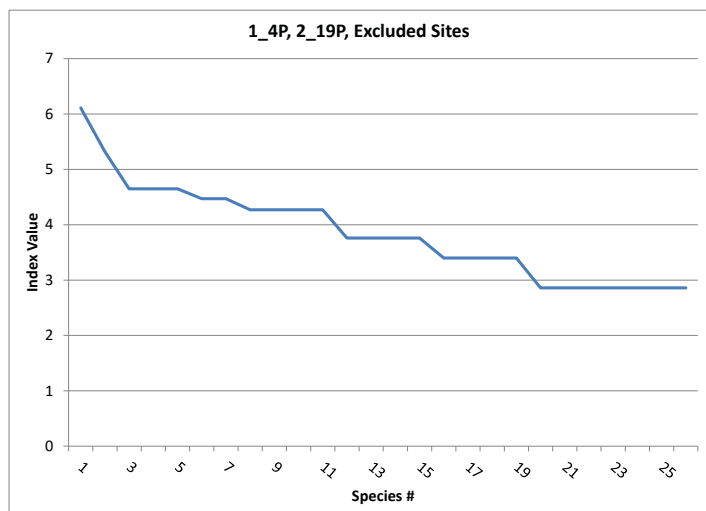
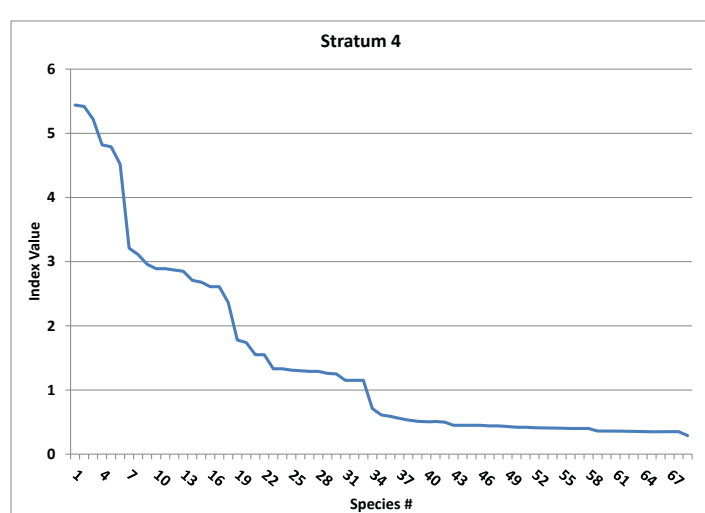
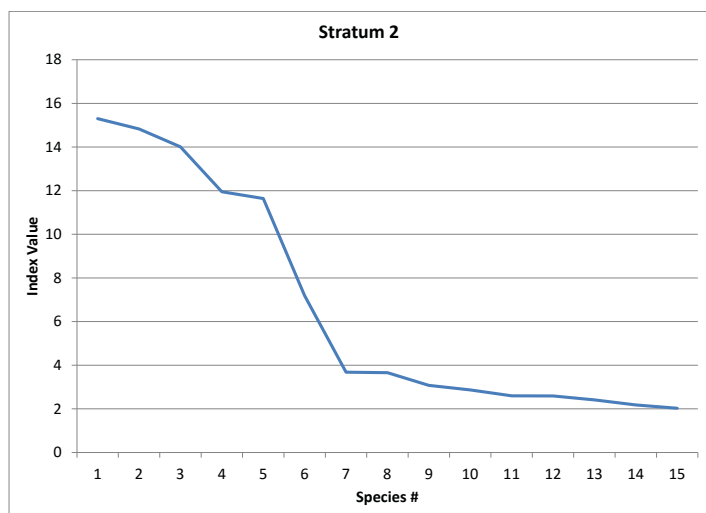
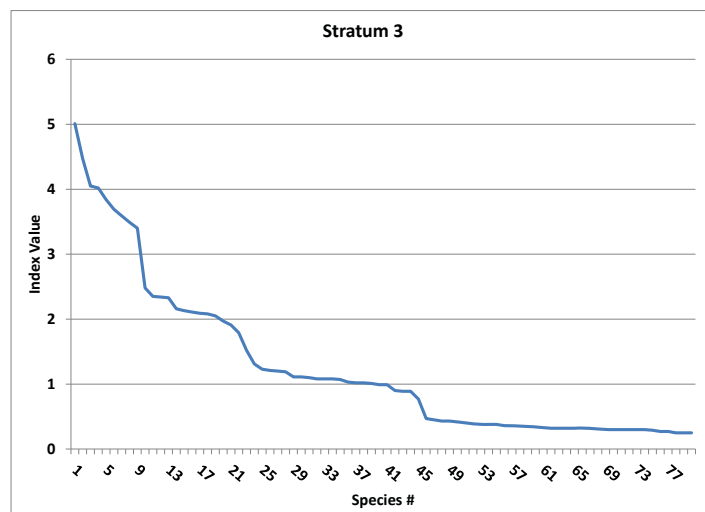
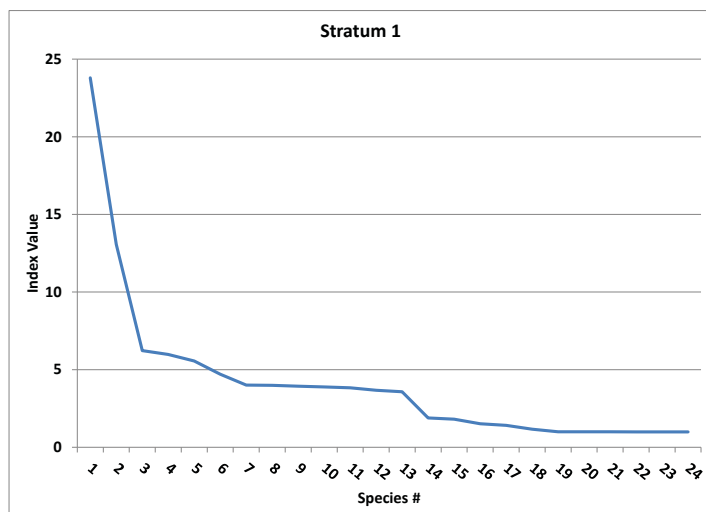


Figure 4.27. Results of the SIMPER analysis for Strata 1-2.

Figure 4.28. Results of the SIMPER analysis for Strata 3-5.

ity and the variety of significant endpoints is highest in Mangrove Lagoon (Figure 4.23). Many of the significant results from the amphipod bioassays are likely due to the coarse grain size of the sediments in the eastern strata. Consequently, amphipod mortality appears to be more widespread than the other toxicity endpoints. Abundance did not decline as sharply as species richness with increasing contamination, suggesting that pollution tolerant species are able to grow and reproduce in contaminated areas in the absence of competitors, predators, and/or indirect effects on the habitat. The weight of evidence between the toxicity, diversity, community makeup, and chemical contamination indicate pollution impacts in Benner Bay and, especially, in Mangrove Lagoon.

4.4 CONCLUSIONS

The degradation of coral reef ecosystems worldwide has led to intensive efforts to understand and mitigate the stressors responsible for the declines of these ecosystems. The role of pollution is often cited as a major factor, but the degree to which pollution, and more specifically, chemical contaminants, impact coral reefs and associated habitats is largely unknown. Because of this, coral reef managers may be missing an important, and in some locations, a critical piece of information required to effectively manage and, where needed, initiate restoration efforts.

For this part of the project in the STEER, a stratified random sampling design was utilized to characterize the distribution and concentrations of chemical contaminants, toxicity, and the benthic infaunal community on an areal basis. This allows for a quantitative analysis of habitat condition between strata. The Sediment Quality Triad, or SQT approach, which combines three types of analyses, was used to provide a means for more holistically assessing the presence and impacts of anthropogenic stressors. Sediment samples for the analysis of both organic contaminants (e.g., polycyclic aromatic hydrocarbons (PAHs), polychlorinated

Table 4.9. Average per-station abundance of selected taxonomic groups in Mangrove Lagoon and Benner Bay without stations 1-4P and 2-19P, stations 1-4P and 2-19P together, and the remaining strata in 3 (Nazareth Bay), 4 (Cowpet/St. James Bay), and 5 (Great Bay).

Organism	Taxa	Mangrove Lagoon and Benner Bay	Stations 1-4P and 2-19P	Stratum 3	Stratum 4	Stratum 5
Annelids		331.1	298.0	298.2	147.6	246.4
	<i>Tubificidae</i>	106.1	48.0	18.0	7.2	16.8
	<i>Capitellidae</i>	28.1	6.5	6.0	2.8	7.6
	<i>Spionidae</i>	77.4	28.0	27.2	8.4	38.4
Malacostraca		7.6	78.0	141.2	57.6	100.0
	<i>Amphipoda</i>	5.3	43.0	39.8	23.2	14.8
Echinoderms		0.0	17.0	8.8	1.2	1.6
Molluscs		73.4	84.0	77.0	41.8	172.0
	<i>Bivalva</i>	16.9	24.5	60.6	31.4	144.4
	<i>Gastropoda</i>	56.6	58.0	12.6	8.0	26.0
Miscellaneous		13.9	38.5	24.6	7.6	53.2
Total		426.0	515.5	549.8	255.8	573.2

biphenyls (PCBs) and pesticides), and inorganic contaminants (trace elements such as chromium, nickel and copper) were collected during a mission in June 2011. In 2010, a preliminary targeted sampling exercise resulted in the analysis of four of the sediment samples collected.

Elevated levels of chemical contaminants were primarily found in Mangrove Lagoon and in northern Benner Bay. There is a large landfill adjacent to Mangrove Lagoon, that likely contributes a variety of contaminants through runoff, groundwater seeps, and perhaps from atmospheric deposition (e.g., from tire/trash fires). Mangrove Lagoon also

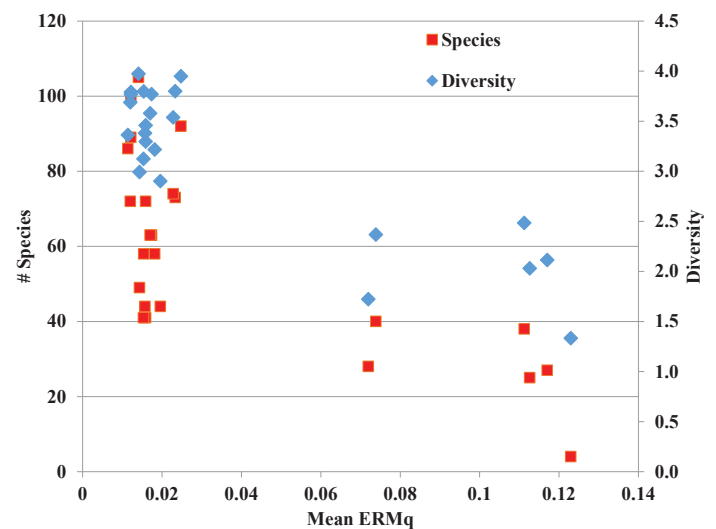


Figure 4.29. Relationship between number of species and species diversity and the ERMq.

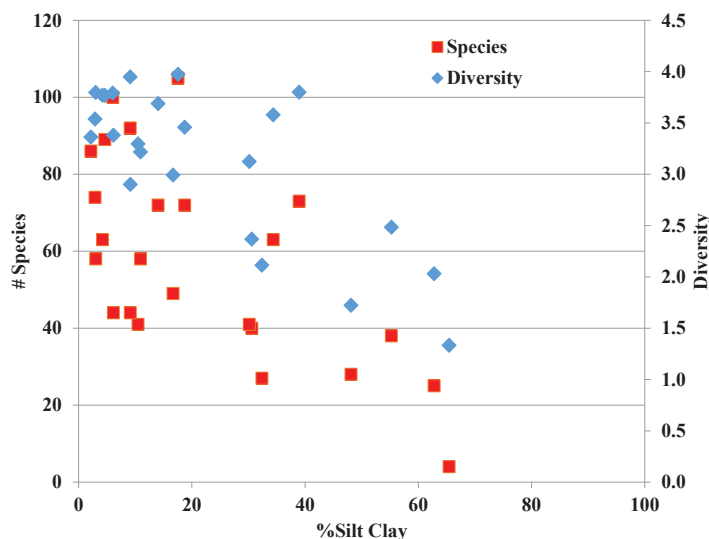


Figure 4.30. Relationship between number of species and species diversity and percent fines.

receives input from various commercial/industrial sources, as well as from adjacent residential/urbanized areas in the watershed via Turpentine Gut. Land use around northern Benner Bay appears dominated by marina-related activities, including the mooring, maintenance, and repair of boats, which is likely a source of chemical contaminants to the STEER.

Tributyltin, or TBT, was found at high levels at three sites in the northern Benner Bay area (two from the targeted 2010 sampling). The level of TBT detected at one site in Benner Bay was the third highest quantified in NOAA's NS&T Program. The presence of high concentrations of TBT likely represents the results of past application of TBT, mooring of vessels that contain TBT, along with the cleaning and scraping of hulls that may have had TBT applied at some point. Unfortunately, there are no established guidelines for TBT in sediment. A site specific numerical upper guideline established for an EPA Superfund site in the state of Washington was exceeded in northern Benner Bay. The purpose of the guideline developed was to inform EPA site managers when additional testing (e.g., toxicity testing) would be recommended.

Copper and zinc were elevated at several locations in Benner Bay and Mangrove Lagoon. The elevated copper level was above the NOAA ERM at one site from the 2010 targeted sampling, indicating that impacts to benthic organisms and within the broader food web are likely. This also appears to be associated with marina activities, including the mooring of vessels, along with the cleaning, and scraping of the hulls and subsequent transport (i.e., through rainfall and subsequent runoff) of these materials into northern Benner Bay. The locations where the three highest copper concentrations were found was also the location of the three highest TBT concentrations.

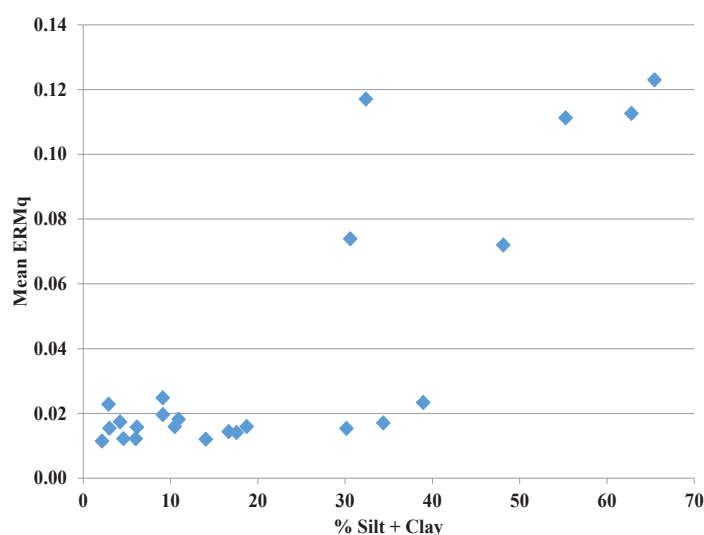


Figure 4.31. Relationship between mean ERMq and percent fines.

A number of other chemical contaminants analyzed for this project, including zinc, lead, copper, mercury, total PCBs, and total DDT, were above ERLs at one or more sites in the STEER, indicating that impacts may be occurring in some of the more sensitive species or life stages that may be present. Additive effects of these levels of contaminants on biota in the STEER are also possible. *Clostridium perfringens*, a pathogenic bacterium used as sewage indicator, was found at high levels in Mangrove Lagoon.

The elevated levels of chemical contaminants in the Mangrove Lagoon and northern Benner Bay areas were reflected in the results of a number of the toxicity tests. Overall, the bioassays indicated a significant gradient (high to low) of effects from west to east in the STEER. The widespread toxicological responses likely indicate the interaction of multiple chemicals, including those beyond the standard suite of NS&T analytes, along with other physicochemical characteristics which also vary between strata.

Half of the significant sea urchin fertilization failure bioassays in the STEER were in sediments from Mangrove Lagoon or the canal that joins it to Benner Bay. In addition, most of the significant P450 responses were in the western strata in the STEER, including all of the Mangrove Lagoon sites. The predominant class of contaminants to which the cells were responding to appeared to be the PAHs or polycyclic aromatic hydrocarbons, which were elevated in this part of the STEER, as opposed to PCBs.

The benthic infaunal analysis also correlated with the chemical contaminant and bioassay data, indicating gradients of diversity and species richness, with lower values in the western strata, especially in Mangrove Lagoon, and higher values towards the east. Similar to the results of the

P450 analysis, the number of species and diversity were significantly and negatively correlated with benzo[a]pyrene concentrations, indicating the likely impacts of this PAH and other chemicals present.

Finally, the nodal analysis showed that the community composition of animals in the sediments of Mangrove Lagoon and Benner Bay were distinct in terms of the species found from the other three strata in the STEER. Furthermore, the species found in Mangrove Lagoon and Benner Bay were for the most part absent from the other strata and vice versa, likely due in part to natural and anthropogenic stressors found in Mangrove Lagoon and Benner Bay.

The ecological health and condition of the interconnected mangroves, seagrass beds, and coral reefs within the STEER are a significant management concern for many, including the USVI DPNR and NOAA's CRCP. The proximity of the Reserves to the Bovoni Landfill, marinas, and other commercial and industrial activities, combined with likely inputs from residential sewer systems, has prompted concerns about negative impacts of chemical pollutants on natural resources of the STEER. Until this current study, very little was known about the types and concentrations of chemical contaminants present, or their spatial distribution patterns within these Reserves. The information generated from this assessment of chemical contaminants, along with the bioeffects, establishes a baseline of conditions so that managers can understand not only the status, but also the challenges that exist to improve the ecological functioning of the STEER.

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CHAPTER 5: AN ASSESSMENT OF TRIBUTYLTIN AND METALS IN SURFACE SEDIMENTS AND SEDIMENT CORES FROM BENNER BAY, ST. THOMAS EAST END RESERVES

S. Ian Hartwell¹, Dennis A. Apeti¹, Andrew L. Mason¹, and Anthony S. Pait¹

¹NOAA/NOS/National Centers for Coastal Ocean Science, Center for Coastal Monitoring and Assessment (CCMA), 1305 East/West Highway, Silver Spring, MD 20910

5.1 INTRODUCTION

This chapter summarizes the results of a follow-up study in northern Benner Bay to assess the distribution of tributyltin (TBT), metals and metalloids in surface sediments and in sediment cores. Results from the initial characterization of sediment contaminants (Chapter 4) in the area of Benner Bay indicated unusually elevated concentrations of TBT in sediments in the Benner Bay region of the STEER. After reviewing these results, the USVI DPNR requested that NCCOS conduct a follow-up study to better assess the extent of contamination in surface sediments and to determine contaminant concentrations in the deeper underlying sediments in the area. In 2013 we conducted the follow-up study to determine the distribution of TBT and other heavy metal residues in the area in more detail, and also to take sediment cores at a number of locations to assess the record of TBT concentrations and metals over time.

The antifouling properties of TBT compounds were discovered in the 1950s. For 40 years tributyltin (TBT) was used as a prime ingredient in antifouling paint applied to the hulls of boats and ocean going vessels. The function of the biocide in the antifouling paint is to prevent the settling of organisms on the hull and to poison the organisms that do. By the mid-1960s it became the most popular antifouling paint worldwide.

Although the paints were effective, the TBT slowly leaches out into the marine environment where it was highly toxic to a wide range of organisms.

The formulation of TBT paints changed over time. Initially it worked by contact leaching to release the TBT to water. However, the release rate proved inconsistent and unpredictable, and as a result self-polishing co-polymer paints were developed. These paints used a polymer base through which the biocide discharge rate is regulated by reacting with water, and resulted in the TBT being slowly released in water. Once a surface covering was worn away,

TBT release continued with the next layer. TBT-based paints were extremely effective and long lasting.

Bottom paint improves ship performance and durability by reducing biofouling on the ship's hull (Bray and Langston, 2006). This allowed increased ship speed and lower fuel consumption due to the lack of fouling organisms on the hull, and increased the time between scraping and repainting, all of which were economically advantageous. By the late 1970s, TBT paints were commonly used on commercial and recreational vessels.



NOAA scientists with sediment core from the STEER.

Negative aspects of TBT were suspected in the late 1960s when it was recognized that the release of organotin into aquatic environments was impacting non-target organisms. Toxic effects in some species occur at a concentration as low as one part per trillion (i.e., 1 nanogram per liter (ng/L)) of water (Bray and Langston, 2006). TBT is toxic to bacteria, algae, fungi, mollusks and crustaceans (Cruz *et al.*, 2015). There are implications of effects on cetaceans and bioaccumulation of TBT in the human food chain. A galvanizing event occurred in the late 1970s and early 1980s, when oyster crops in Arcachon Bay, France, failed. Subsequent research identified

that TBT had caused decreased spatfall, unnatural shell thickening and abnormal structure that weakened the shells (Bray and Langston, 2006). Similar observations were seen in United Kingdom oyster stocks. Away from hull cleaning operations, TBT sediment concentrations were higher in harbors with many small boats than in industrial harbors with commercial ships (Bryan and Gibbs 1991; Page *et al.*, 1996). In 1982, France banned TBT use on recreational vessels less than 25 meters long. Subsequent work showed that TBT was an endocrine disruptor in marine gastropods causing masculinization (imposex) in females and widespread population decline. By the early

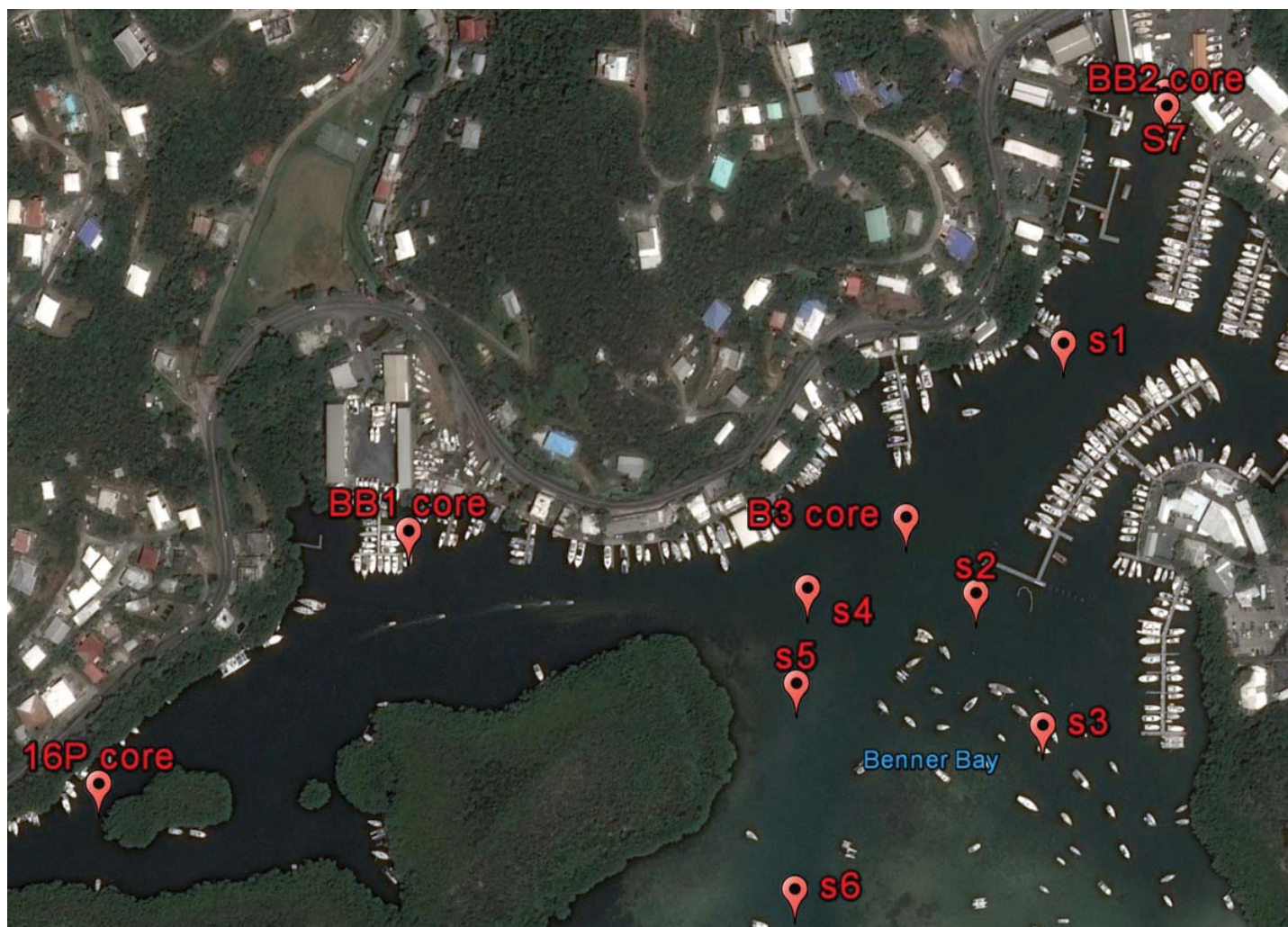


Figure 5.1 Sampling sites for surface and sediment core samples in Benner Bay within the STEER.

1990s, many nations had partial or complete bans on TBT. In 1999, the International Maritime Organization (IMO) a United Nations agency, came to agreement that TBT would be phased out between 2003 and 2008, with a total ban of organotin antifouling coatings by January 2008. The U.S. ratified the agreement in 2012. However, while the agreement requires compliance from the developed nations, much of the developing world are not signatories. Also, TBT is still used as a slimicide in power plant cooling towers and other industrial heat exchange equipment, as a wood preservative, and as a molluscicide. It was reportedly still available in bottom paint as recently as 2014 in the Caribbean and Central America through U.S. outlets (Turner and Glegg, 2014).

TBT is a persistent and bioaccumulative compound. The degradation pathways proceed from tributyltin to dibutyl-, to monobutyl-, and finally to elemental tin. The reported half-life in estuarine waters range from days to weeks (Omae, 2005). TBT is strongly sorbed to sediments particles, however, and the half-life of TBT in sediments is years, and in anaerobic sediments extends to decades

(Matthiessen, 2013). Also, paint chips from boat hulls, for example as might occur during the scraping and repainting process, can ultimately be flushed into a water body, and serve as a reservoir for release to the sediment. Thus high concentrations may persist in older buried sediments where uncontrolled releases have occurred in the past. This material may or may not become bioavailable depending on local sediment deposition rates and subsequent disturbance (e.g., dredging).

5.2 METHODS

Sampling

Surface Sediments

Surficial sediment samples were collected at six locations in Benner Bay leading away from the main marina facility on two transects, one out the approach channel (S4-6) and one through an offshore boat anchoring area (S1-3) (Figure 5.1). Sampling protocols were the same as those used in 2010 and 2011 (Chapter 4). Water quality measurements and sediments were collected using standard NOAA National Status and Trends (NS&T) protocols (Apeti *et al.*, 2012). A PONAR grab (see insert, next page) was

deployed to collect the samples, and retrieved by hand. As with the collection of sediments for the assessment of contaminants throughout the STEER (see Chapter 4), the top 3 cm of sediment were collected from the grab using a stainless steel sediment scoop. Surface sediment was used for chemical analyses to assess current depositional conditions.

Sediments were placed into certified clean (I-Chem®) 250 ml labeled jars, capped and then kept on ice in a cooler. Sediments for grain size analysis were placed in a WhirlPack® bag, sealed and then kept on ice in a cooler. At the end of each day, sediment samples for contaminant analysis were frozen. The WhirlPack® bags for the grain size analysis were placed in a refrigerator rather than frozen, to avoid altering the grain size structure.

Sediment Cores

The sediment core samples were collected at four locations starting from the approach channel leading from Mangrove Lagoon to the southwest, and up into the head of Benner Bay in the heart of the area where the marina facilities are located (Figure 5.1). The

BB2 and BB1 sites had been sampled for surface sediment in 2010. The B3 site was roughly equidistant between them, and site 16P was sampled in 2011 for surface sediment. The boat was anchored at the bow and stern to minimize drift so the core could be deployed vertically. The corer was designed to collect undisturbed cores of the sediment and mud-water interface. The corer drove a 7 cm diameter polycarbonate tube into the sediment with a hand-held

weight. A one-way check valve seated in the core head allowed water and sediment to move through the core barrel. During retrieval the check valve automatically seats, creating a partial vacuum which retains the sample in the core barrel. The core was returned to the dock where it was extruded in 2 cm sections by means of a plunger provided with the corer. Each section was placed into a certified clean (I-Chem®) 250 ml labeled jar and homogenized. A sub-sample was removed for grain size analysis. Sediments for grain size analysis were placed in a WhirlPack® bag, sealed and then kept refrigerated. Sediment samples for metals and TBT analyses were frozen. An additional

surface sediment sample was collected for TBT analysis with the PONAR grab at station BB2 as a check on potential gear bias.

Chemical Contaminants Analyzed

The sediments were analyzed for a suite of 16 major and trace elements, grain size analysis and for four butyltin compounds by TDI-Brooks International, using protocols established by the NS&T Program. The major and trace elements were analyzed using inductively coupled plasma mass spectrometry and atomic-fluorescence spectroscopy. Detailed descriptions of the NS&T protocols, including quality assurance/quality control (QA/QC) used in the analysis can be found in Kimbrough and Lauenstein (2006). The four butyltins were analyzed using gas chromatography/flame photometric detection after derivatization with hexyl-MgBr.

Radiochemical Dating

The core sections were also analyzed for two isotopes in order to estimate the age of the sediments down the core length. In depositional environments, sediments are continually laid down and compacted by overlying sediment. In a continuous sequence, the age of the sediment can be estimated and the history of chemical contamination can be recreated. Depending on the rate of sediment deposition, changes in contaminant inputs will be obvious. Bioturbation by burrowing organisms will tend to mix the surface layers as they accumulate and may blur the history. Also, powerful storms, like hurricanes, may scour out whole sections, or, conversely bury layers with

large deposits from runoff or resuspension. Also, human activities such as dredging and spoil disposal may cause breaks in the historical record. Activities on land that alter sediment delivery from runoff may increase or decrease the rate of sedimentation over time.

The age of the sediment sections were estimated by measuring an isotope of lead (^{210}Pb) and of cesium (^{137}Cs). Briefly, uranium-238 (^{238}U) decays through a series of isotopes to radium-236 (^{226}Ra). ^{226}Ra has a half-life of 16,000 yr, so the background supply is virtually constant. Radium eroded from rocks is deposited in sediments.



Sample of surface sediment being taken from PONAR grab by NOAA scientists.

Radium decays into radon-222 (^{222}Ra) which has a half-life of only 3.8 days. Finally, radon decays to ^{210}Pb with a moderate half-life of 22.3 yr., so a constant background level of ^{210}Pb is present in rock and eroded sediments. Because radon is a gas, some of it escapes from the earth into the atmosphere to yield atmospheric ^{210}Pb . This is washed out of the air by rain and is deposited into fresh sediments. Thus sediment deposits have two sources of ^{210}Pb , the background amount constantly derived from slow radium decay, and from atmospheric deposition. The older a layer of sediment is, the more of the atmospherically derived ^{210}Pb will have decayed until only background levels remain. Therefore, looking at ^{210}Pb levels down the length of a core you would expect to see concentrations decline in proportion to the sediment deposition rate, until only background levels are seen. ^{210}Pb is used to determine the age of sediments and accumulation rate of sediments in water bodies. In a typical application, the average accumulation rate over a period of 100 - 200 years is obtained. From the accumulation rate, the age of sediment from a particular depth in the sediment column can be estimated.

Cesium-137 (^{137}Cs) measurements are used as a check on calculated age profiles by providing date “markers” rather than concentration slopes. ^{137}Cs is derived from atomic bomb testing. The first appearance of ^{137}Cs in sediments marks the year 1954, which is the year when global concentrations generally achieved detectable levels. The other ^{137}Cs marker is the concentration maximum in the year 1963, after which atmospheric testing ceased.

Lead and cesium measurements were performed at the University of Maryland, Horn Point Environmental Laboratory. ^{210}Pb measurements were carried out via analysis of its short-term daughter product polonium-210 (^{210}Po), measured by alpha spectroscopy (Palinkas and Nittrouer, 2007). Briefly, 1-2 g of dry, ground sediment samples were spiked with a known volume of polonium-209 (^{209}Po), leaching first in 15.8 N HNO_3 , then in 6 N HCl . ^{210}Po and ^{209}Po were then electroplated onto silver planchets and counted for 24 hours in a Canberra Alpha Analyst alpha spectrometer (Nittrouer *et al.*, 1979). Age models were fit to the data, as appropriate, to determine sediment accumulation rates (Appleby and Oldfield, 1978; Carroll and Lerche, 2003; Hancock *et al.*, 2000). ^{137}Cs measurements were performed using gamma spectroscopy. Dry, ground sediment from each sampling interval (~3-5 g) was sealed in 60-mL plastic jars. The gamma emissions from each sample was counted for approximately 24 hr with a calibrated Canberra germanium detector, using the 661 KeV photopeak of the gamma



Figure 5.2. Photograph of core BB2 showing shell hash in the lower part of the core, grading to fine mud up the core in more recent sediments.

spectrum. Accumulation rates were calculated from both the depth of first appearance and maximum activity.

Core depth varied and was limited by the depth of dense and/or shell hash layers (Figure 5.2) which was the depth of refusal for the corer. Cores from 16P, BB1, and B3 were 16, 20 and 16 cm deep, respectively. Two cores were taken at BB2. The first core was 28 cm deep, but the core above 18 cm was disturbed by air bubbles during handling. A second core sample was taken which was only 14 cm deep so there is a break in the data. There was only enough material to do butyltins and the radiochemical analyses on the 18-20 cm section.

5.3 RESULTS AND DISCUSSION

Contaminants in Surface Sediments

Metals and butyltin concentrations in the surface samples and the top layer of the cores are shown in Table 5.1. The concentration of total butyltins (the sum of the butyltins analyzed) at BB2 was orders of magnitude above all other stations. There is a clear gradient of butyltins from BB2 out into Benner Bay and down the channel toward Mangrove

Tables 5.1. Concentrations of butyltins (ng Sn/g) and metals (µg/g) in surface sediments.

Site	LatDD	LongDD	Monobutyltin	Dibutyltin	Tributyltin	Total butyltin	%Tributyltin	Tetrabutyltin	Ag	As	Cd	Pb
16P 0-2	18.31690	-64.87339	6.61	10.43	66.4	83.5	79.6	0.31	0.00	11.0	0.16	15.7
B3 0-2	18.31849	-64.86831	13.3	11.2	12.4	36.9	33.6	0.20	0.00	10.0	0.09	17.4
BB1 0-2	18.31841	-64.87144	12.6	28.2	40.0	80.8	49.5	0.14	0.00	9.6	0.23	25.4
BB2 0-2	18.32103	-64.86668	940	700	1102	2,741	40.2	37.6	0.27	16.4	0.28	129
S1	18.31953	-64.86732	72.6	68.7	134	275.0	48.6	1.20	0.08	9.7	0.08	119
S2	18.31804	-64.86787	25.7	9.74	12.2	47.6	25.6	0.39	0.00	7.0	0.08	24.1
S3	18.31725	-64.86745	7.61	3.93	4.01	15.6	25.8	0.10	0.00	3.4	0.07	7.77
S4	18.31807	-64.86893	6.25	5.07	6.24	17.6	35.5	0.11	0.00	5.1	0.07	10.2
S5	18.31750	-64.86900	11.2	8.29	5.57	25.0	22.2	0.11	0.00	4.0	0.00	15.9
S6	18.31627	-64.86901	8.33	6.21	217	231.3	93.7	1.82	0.00	4.5	0.07	7.51
S7	18.32103	-64.86668	692	592	993	2,277	43.6	12.8				
ERM									3.7	70	9.6	218
ERL									1	8.2	1.2	46.7

Site	Sb	Sn	Al	Cr	Cu	Fe	Mn	Ni	Zn	Si	Se	Hg
16P 0-2	0.46	1.99	38,600	16.8	92.9	23,200	176	5.28	118	140,000	0.746	0.061
B3 0-2	0.22	1.76	21,400	10.3	97.4	11,700	94.1	3.12	92.9	57,000	0.34	0.050
BB1 0-2	1.00	3.32	48,900	19.7	88.7	27,000	200	5.09	145	222,000	0.605	0.096
BB2 0-2	0.73	22.2	64,700	61.5	1,520	36,200	225	8.86	574	141,000	0.521	0.410
S1	1.49	5.25	29,500	18.9	373.0	17,100	115	4.4	206	62,600	0.24	0.126
S2	0.19	3.06	26,500	15.9	115.0	15,000	124	3.32	104	76,400	0.238	0.083
S3	0.10	0.82	7,660	2.5	51.7	4,760	38.9	2.59	47.5	22,700	0	0.030
S4	0.13	1.21	13,300	6.13	50.6	7,810	61.7	3.01	52.7	32,600	0.237	0.032
S5	0.12	1.2	12,400	5.13	54.8	6,840	55.3	2.57	49.4	29,800	0.151	0.033
S6	0.09	0.901	9,450	4.43	41.6	6,210	49.3	2.37	45.4	26,200	0.092	0.033
S7												
ERM				370	270			51.6	410			0.71
ERL				81	34			20.9	150			0.15

Abbreviations: LatDD, latitude in decimal degrees; LongDD, longitude in decimal degrees; ERL, effects range low; ERM, effects range median

Note - Sediment from site S7 was analyzed for butyltins, to check on potential gear bias (PONAR versus sediment corer) with BB-2. No trace or major element analysis performed at S7.

Lagoon. There was good agreement between the top core section at BB2 and sample S7, indicating no gear bias. The concentration of Cu was also orders of magnitude higher at station BB2 than all other stations. Copper concentrations exceeded the ERM by 5X at BB2. Zinc also exceeded the ERM at BB2. Impacts to benthic organisms in this area are likely as a result of these concentrations.

given errors and assumptions inherent in both methods, and the profiles are very clear. The accumulation rates using ^{137}Cs are slightly higher than from the ^{210}Pb data, indicating somewhat younger sediment at depth. This becomes more obvious when the TBT data is plotted vs YBP (years before present).

Isotope Analysis of Cores

Plots of ^{210}Pb and ^{137}Cs activities with depth are shown for each core in Figures 5.3 - 5.6. Core 16P had the best profile of all the cores, showing the characteristic shape of logarithmic ^{210}Pb decay with depth until the background activity (0.1 dpm/g) is reached (Figure 5.3). The accumulation rate calculated from the ^{210}Pb data is 0.073 cm/yr. For ^{137}Cs , the depth of first appearance (1954) is 6-8 cm; the depth of maximum activity (1963) is 4-6 cm. Calculated accumulation rates based on the ^{137}Cs data are 0.10-0.14 cm/y and 0.08-0.12 cm/y, respectively. All of these rates are in rough agreement,

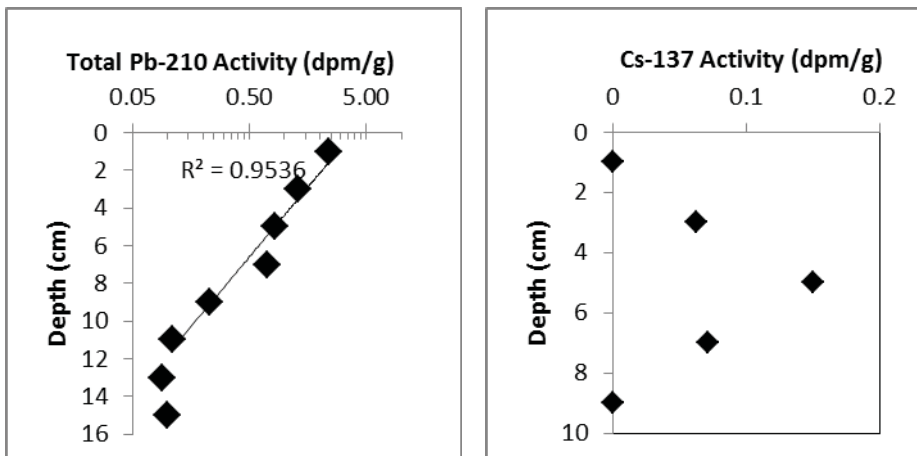


Figure 5.3. Plots of ^{210}Pb and ^{137}Cs activities with depth for core 16P.

Figure 5.4 shows the profile for core BB1. The accumulation rate calculated from the ^{210}Pb data is 0.06 cm/y. The profile is straightforward – ^{210}Pb decays logarithmically with depth to a background level of 0.1 dpm/g. For ^{137}Cs , the depth of first appearance (1954) is 6-8 cm, yielding an accumulation rate of 0.10-0.14 cm/y. The depth of maximum ^{137}Cs activity (1963) is 2-4 cm, yielding an accumulation rate of 0.04-0.08 cm/y. There is good agreement between the ^{137}Cs and ^{210}Pb data. There is some evidence for increased accumulation rates over time, but increased depth resolution would be needed for further evaluation.

Figure 5.5 shows the profile for core B3. The ^{210}Pb data for this core are subject to interpretation. It does not appear that the core was long enough to reach the background ^{210}Pb level, so the background activity was assumed to be equal to that used for 16P and BB1 (0.1 dpm/g). The background level within a small region should be the same. If a higher background activity is used, the accumulation rate will decrease – for example, using 0.5 dpm/g as the background level yields a rate of 0.085 cm/y. Also, there is some question as to whether all data points reflect sedimentation, and thus should be used in the rate calculation, or whether the upper 4 data points reflect mixing and should be neglected in the rate calculation. The accumulation rates are 0.32 cm/y (all data) or 0.16 cm/y (only the lower 4 data points).

For ^{137}Cs data, the depth of first appearance (1954) is 8-10 cm, yielding an accumulation rate of 0.14-0.17 cm/y. This is in agreement with the ^{210}Pb rate from the lower four points. There are two depths, above and below the 6-8 cm interval, with identically high activities that could be

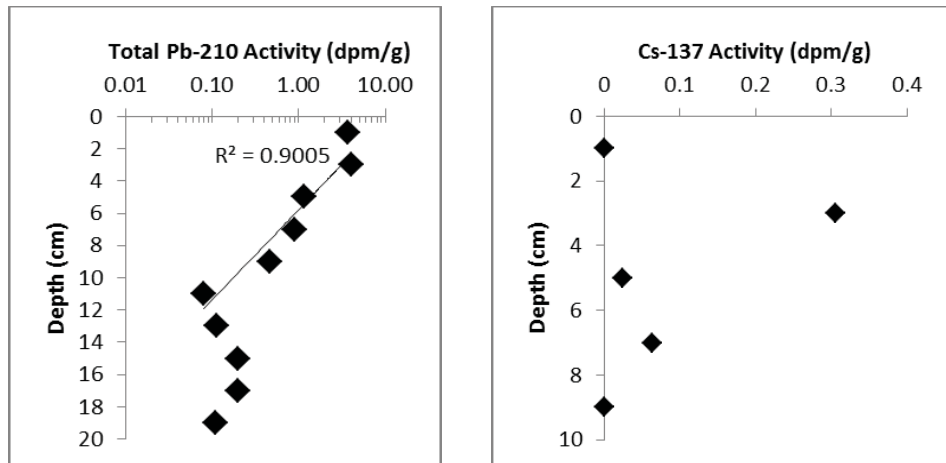


Figure 5.4. Plots of ^{210}Pb and ^{137}Cs activities with depth for core BB1.

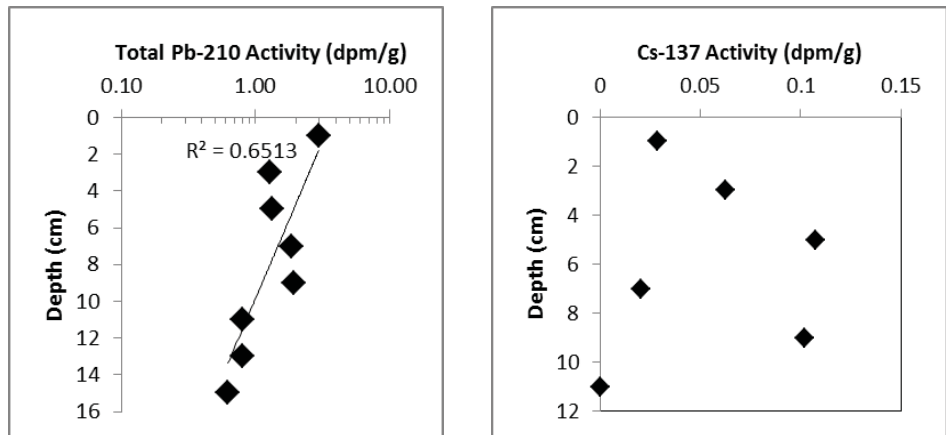


Figure 5.5. Plots of ^{210}Pb and ^{137}Cs activities with depth for core B3.

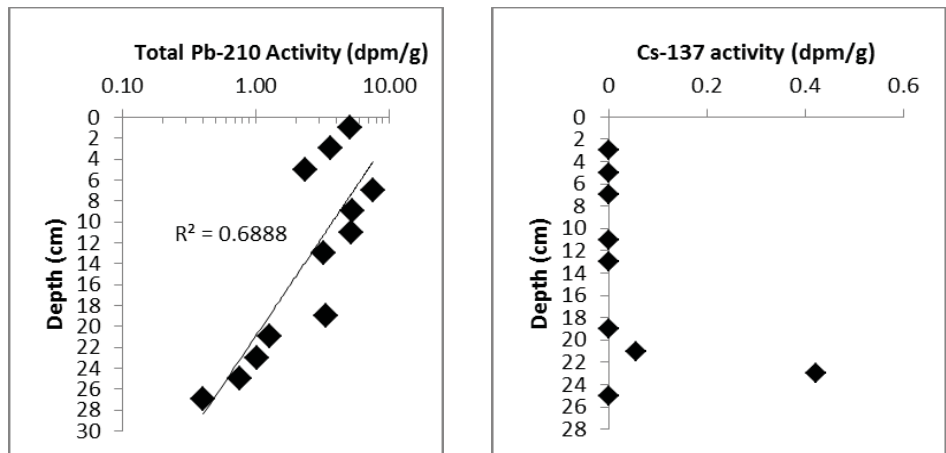


Figure 5.6. Plots of ^{210}Pb and ^{137}Cs activities with depth for core BB2.

considered the depth of maximum ^{137}Cs activity (1963). The first, at the top of the higher-activity layer, is 4-6 cm and yields an accumulation rate of 0.08-0.12 cm/y. One could also assume a typical profile shape and assume that the sediment in the 6-8 cm interval has anomalously low activities. If 6-8 cm is used as the depth of maximum activity, the accumulation rate would be 0.12-0.16 cm/y. However, the anomaly in the ^{210}Pb data occurs at the 6-8

cm depth interval, so it may be an event-related disturbance of the sediment layer is responsible.

Figure 5.6 shows the profile for core BB2. The ^{210}Pb profile for this core is also subject to interpretation because of the break in the data set.

Accumulation rates were

calculated both using all points and the lower points. Accumulation rates are 0.35 cm/y for all data points and 0.12 cm/y for only the lower points. A supported value of 0.1 dpm/g was used, consistent with cores 16P and BB1, since the core does not appear to be long enough for ^{210}Pb activities to have reached background activities. As noted previously, a higher background value would yield a lower accumulation rate. There is an anomaly at the 6–8 cm depth interval, which may be an event-related disturbance of the sediment layer similar to core B3. Station BB2 was in the heart of the marina facilities, adjacent to the boat ramp and travel lift.

^{137}Cs samples down to 12 cm were originally analyzed, with no detectable ^{137}Cs . The reason for this is unclear; one potential explanation is that sediment properties differ between this core and the others. The remaining samples were counted with detections for the 20–22 cm and 22–24 cm intervals. However, these detections occurred at the 659 keV photopeak (part of gamma spectrum); the photopeak that represents Cs-137 is 662 keV. In practice, a deviation of ± 2 keV is accepted when evaluating gamma spectra, using information about sample and site to guide decisions.

Thus, the 20–22 and 22–24 sections are considered borderline; they would likely be recorded as detectable ^{137}Cs , unless other evidence suggested otherwise. It is difficult to determine whether these detections represent the depth of first appearance or maximum activity. Using the point as a first occurrence (1954) yields an accumulation rate of 0.42 cm/yr. Using the point as a maximum (1963) occurrence yields an accumulation rate of 0.50 cm/yr.

Table 5.2. Estimated ages of sections within sediment cores from the STEER.

Mean Core Depth cm	16P		BB1		B3		BB2		
	^{210}Pb age	^{137}Cs age	^{210}Pb age	^{137}Cs age	^{210}Pb age	^{137}Cs age	^{210}Pb age	^{137}Cs age	^{137}Cs age
							1954	1963	
1	13.7	9.1	16.7	11.1	3.1	6.7	2.9	2.4	2.0
3	41.1	27.3	50.0	33.3	9.4	20.0	8.7	7.1	6.0
5	68.5	45.5	83.3	55.6	15.6	33.3	14.5	11.8	10.0
7	95.9	63.6	116.7	77.8	21.9	46.7	20.3	16.5	14.0
9	123.3	81.8	150.0	100.0	28.1	60.0	26.1	21.3	18.0
11	150.7	100.0	183.3	122.2	34.4	73.3	31.9	26.0	22.0
13	178.1	118.2	216.7	144.4	40.6	86.7	37.7	30.7	26.0
15	205.5	136.4	250.0	166.7	46.9	100.0			
17			283.3	188.9					
19			316.7	211.1			55.1	44.9	38.0
21							60.9	49.6	42.0
23							66.7	54.4	46.0
25							72.5	59.1	50.0
27							78.3	63.8	54.0
Rate cm/yr	0.07	0.11	0.06	0.09	0.32	0.15	0.35	0.42	0.50

The age of each layer of the cores as calculated by the ^{210}Pb and ^{137}Cs methods are shown in Table 5.2. The sections representing 1963 (upper) and 1954 (lower) are highlighted (shaded). These represent 50 and 59 years before 2013 respectively. The ages determined by the ^{137}Cs method appear to more closely reflect years before present (YBP). Years at 16P, BB1 and BB2 are overestimated while B3 is underestimated by the ^{210}Pb method. It is unclear which ^{137}Cs accumulation rate to use at BB2 so the most recent marker (1963) was used.

Grain Size in the Sediment Cores

The characteristics of the sediment in the area have changed dramatically over the years. Figures 5.7 and 5.8 show the proportion of fine grained (silt + clay) and gravel-sized particles in the cores over time and down the length of the cores. All the cores show a much higher percentage of gravel-sized material at the bottom of the cores than the top. These particles, however, were not gravel but were in fact shell hash (Figure 5.2). The shift toward fine-grained material is most dramatic at BB2. Station BB2 is surrounded by bulkheads and sits beneath constant boat activity. It is next to the main marina ramp. While BB2 was the deepest core, it covers the shortest time span due to the much higher accumulation rate (Table 5.2). Recall that there were anomalies in the cores from BB2 and B3 and that the calculated accumulation rates in the lower half of the cores were much lower than in the top half. A much lower accumulation rate in the deeper, older, layers at BB2 is logical as human activity has clearly and drastically altered the bottom sediment characteristics. Note also that

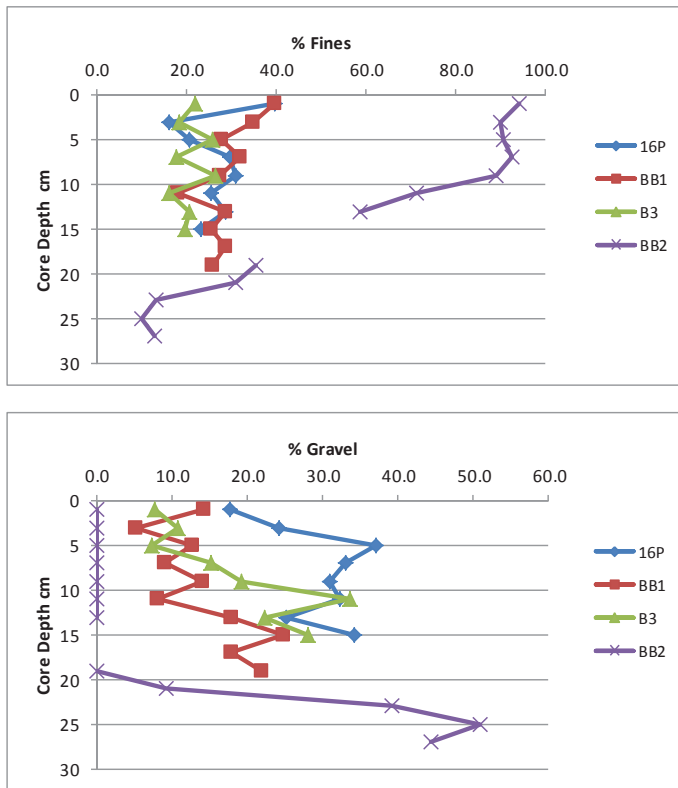


Figure 5.7. Percentage of fine grained sediment and gravel sized material (shell hash) down the length of the cores.

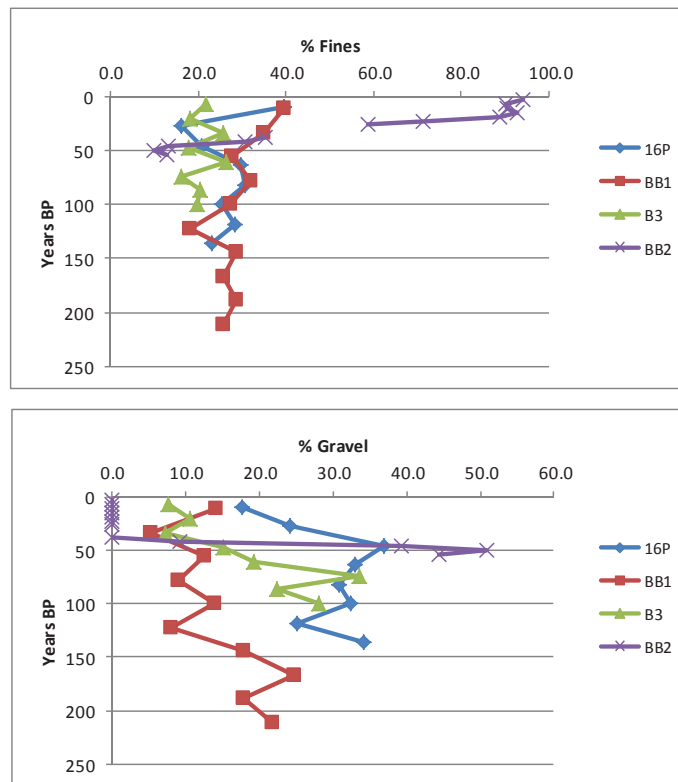


Figure 5.8. Percentage of fine grained sediment and gravel sized material (shell hash) as a function of time. Years BP, years before present.

the most dramatic shifts have occurred in the last 50-75 years.

Reviews done in the 1980s to evaluate the potential outcomes of declaring the STEER a NOAA National Marine Sanctuary noted that Mangrove Lagoon and Benner Bay were dominated by turtle grass beds in the 1960s, but in the 1970s the bottom was becoming mud, and was converting to a calcareous macroalgae-dominated benthic community (NOAA, 1981). Extensive shoreline development, increasing boating activity, pollution from terrestrial runoff and sewage discharges were all impacting Benner Bay water quality by the early 1970s (Grigg *et al.*, 1971). The watersheds immediately adjacent to Benner Bay that drain into it are the Nadir and Compass Pt. subwatersheds. More recent assessments describe the Nadir shoreline and watershed as being “a highly dense chain of marinas and commercial properties that transitions to single family residential area moving uphill”, and Compass Pt. includes “a residential area that extends from the top of the subwatershed down to Benner Bay; the main gut flows behind single family homes on the hillside, then under or on the road through the marina complex” (Horsley Witten, 2013).

Butyltin Concentrations in the Sediment Cores

The concentrations of total butyltins are shown in Figure 5.9. The peak concentrations occur at less than 50 years ago, which is consistent with historical use patterns. There are low concentrations of butyltins in sections 2-4 cm below 50 years ago which may reflect the initial buildup of TBT contamination and/or an indication of how deep sediments are churned by storms and bioturbation. Butyltin concentrations are vastly higher at BB2 which also shows a peak in the past. The break in the data unfortunately occurs where the use of TBT was initiated so it is impossible to conclude if concentrations were even higher in the past, or if TBT use continued into more recent years.

Concentrations have declined in recent years. Nevertheless, the observed concentrations at depth are vastly higher than anywhere else in Benner Bay or the rest of the STEER. Concentrations of this magnitude have only been observed in a few places (Page *et al.*, 1996, 24-12,400 ng/g - Maine, USA; Shim *et al.* 2002, 33-19,780 ng/g - S. Korea; Diaz *et al.* 2002, 123-6,692 and 574-1,970 - NE and SE Spain; EVS, 1999, 8-6,200 - Seattle, USA). All of these studies were sampling in marinas and/or ship yards for the purpose of locating hot spots. Also, our data is reported as ng/g of tin (ng Sn/g), as opposed to TBT which has a 60% higher molecular weight than elemental tin. Of 1,506 data points in the NS&T data base with TBT sediment analyses, the

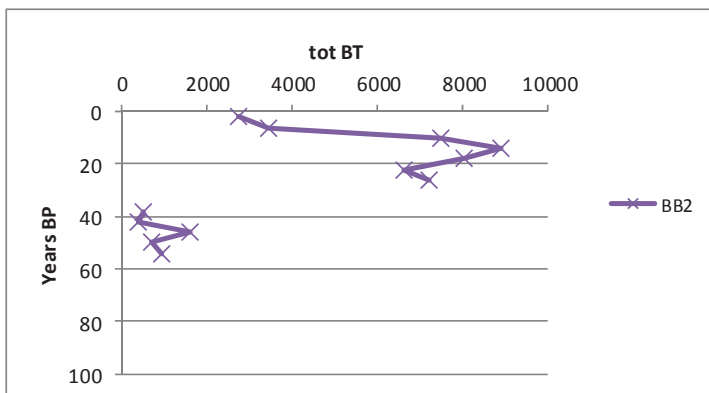
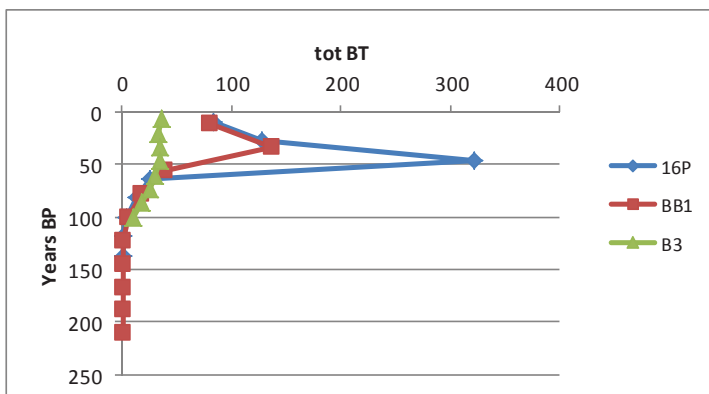


Figure 5.9. Concentration (ng/g) of total butyltins as a function of time.

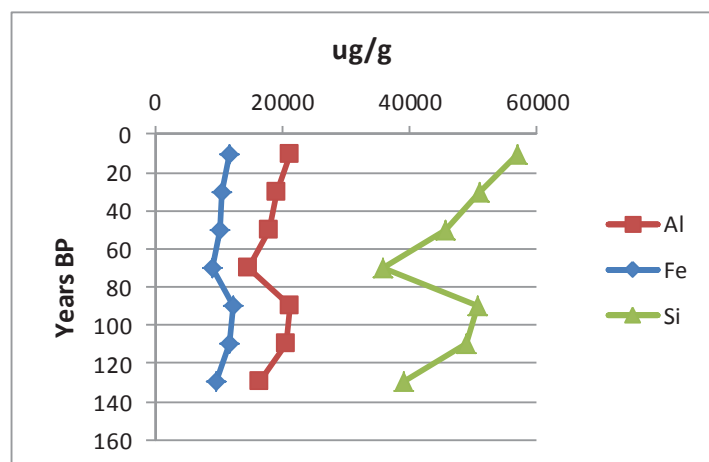


Figure 5.10. Changes in major element concentrations over time at station B3.

median total butyltin value is 0.95 ng/g, the average is 8.9 ng/g. The highest value is 990 ng/g from Elliott Bay, a Superfund site in Puget Sound sampled in 1989.

There was relatively little butyltin in the B3 core. Neither is there evidence of historical changes over the years. Relative to the ^{137}Cs date markers, the ^{210}Pb analyses severely underestimated the age of the B3 sediments unlike the other cores (Table 5.2). There are obvious discontinuities in both the ^{137}Cs and ^{210}Pb records (Figure 5.4). Its location is in the middle of the channel, perhaps the natural channel,

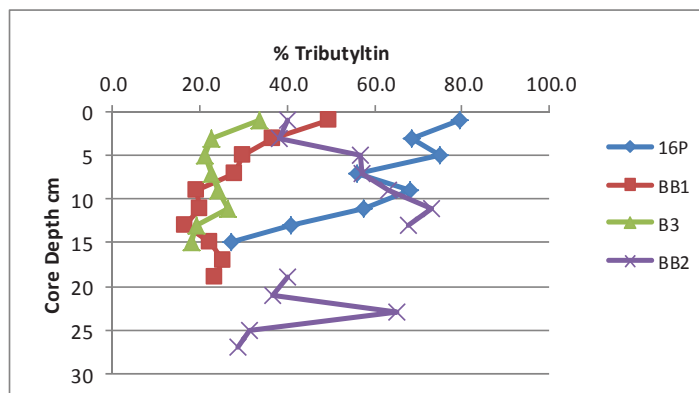
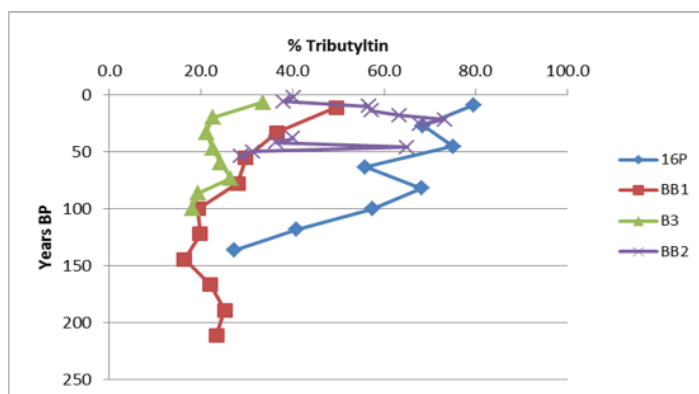


Figure 5.11. Percent tributyltin as a proportion of the total butyltins as a function of time and core depth.

leading into inner Benner Bay. What events or processes (e.g., storms, dredging, etc.) may have contributed to the history of deposition at this location is unknown. The concentrations of the major soil elements of aluminum, iron and silicon all show a major shift at B3 in the mid-1940s (Figure 5.10), which coincides with the discontinuity in the ^{210}Pb and ^{137}Cs anomalies (Figure 5.4). Interestingly, the same is true for all the trace metals as well. Clearly, some event or change in circulation or terrestrial input occurred then.

Another puzzling aspect is the percentage of tributyltin as a proportion of total butyltins in the sediment. While the concentration of butyltins is decreasing over time, the proportion of tributyltin is increasing at 16P, BB1 and B3 up to the present, indicating continuing fresh inputs (Figure 5.11). The peak concentrations at BB2 in the marina are only 4-10 cm deep. The proportion of tributyltin at those depths is 50-70%. Prop wash from boats and storm-driven tidal currents may be the source of fresh tributyltin at the outer stations. Unlike BB2, the proportions below 10 cm depth in the cores at 16P, BB1 and B3 are meaningless as the concentrations are essentially zero and they date back to before TBT existed. Station 16P was located behind a mangrove island in the middle of the waterway between Benner Bay and Mangrove Lagoon (Figure 5. 1). There

are moored boats, but no marina facilities in the immediate vicinity, so the TBT load there must be drifting in from other areas. The currents in the waterway are highly variable and dependent on tidal flux and wind driven currents. The last tropical storm to hit St. Thomas was Irene in 2011, which passed just south of the island with sustained gale force winds and heavy rain.

Gradients in Chemical Contamination

Chemical results from the top sections in the cores and the surface sediment grabs illustrate consistent gradients away from inner Benner Bay for almost all constituents. With three exceptions (Se, Sb, Si) the highest metals concentrations are all found at BB2. Metals concentrations were higher in general at the stations located closest to shore and declined further out into Benner Bay. Mercury was an order of magnitude higher at BB2 than all other stations except S1. Zinc exceeded the ERM. As noted above, the copper concentration was more than five times the ERM. Arsenic, Pb, Hg, and Zn exceeded the ERLs at multiple stations. Aluminum, silicon, and iron are the most common elements in the earth's crust. Decreasing gradients of these elements leading away from Benner Bay all indicate a greater contribution of land-based sediment material close to the shoreline, as opposed to marine sources. The pattern of calcium decreasing from offshore toward Benner Bay and Mangrove Lagoon locations in the 2011 data are consistent with this interpretation (Figure 5.12). All of these observations, plus the historical patterns revealed in

the cores indicate a disturbed habitat that is heavily and increasingly impacted by land-based sediment input and chemical contamination from boating related activities.

The gradient of butyltins in the surface from BB2 out into Benner Bay and down the channel toward Mangrove Lagoon clearly illustrate the impact of boating related activities (Table 5.1). Surface concentrations in the marina complex are one to two orders of magnitude higher than anywhere else. Butyltins at S1 leading out of the harbor were higher than at core B3 and those stations further out. Stations 2-5 were laid out on two transects leading away from core B3 on different routes. Concentrations drop off rapidly further out. The one exception was station S6 that

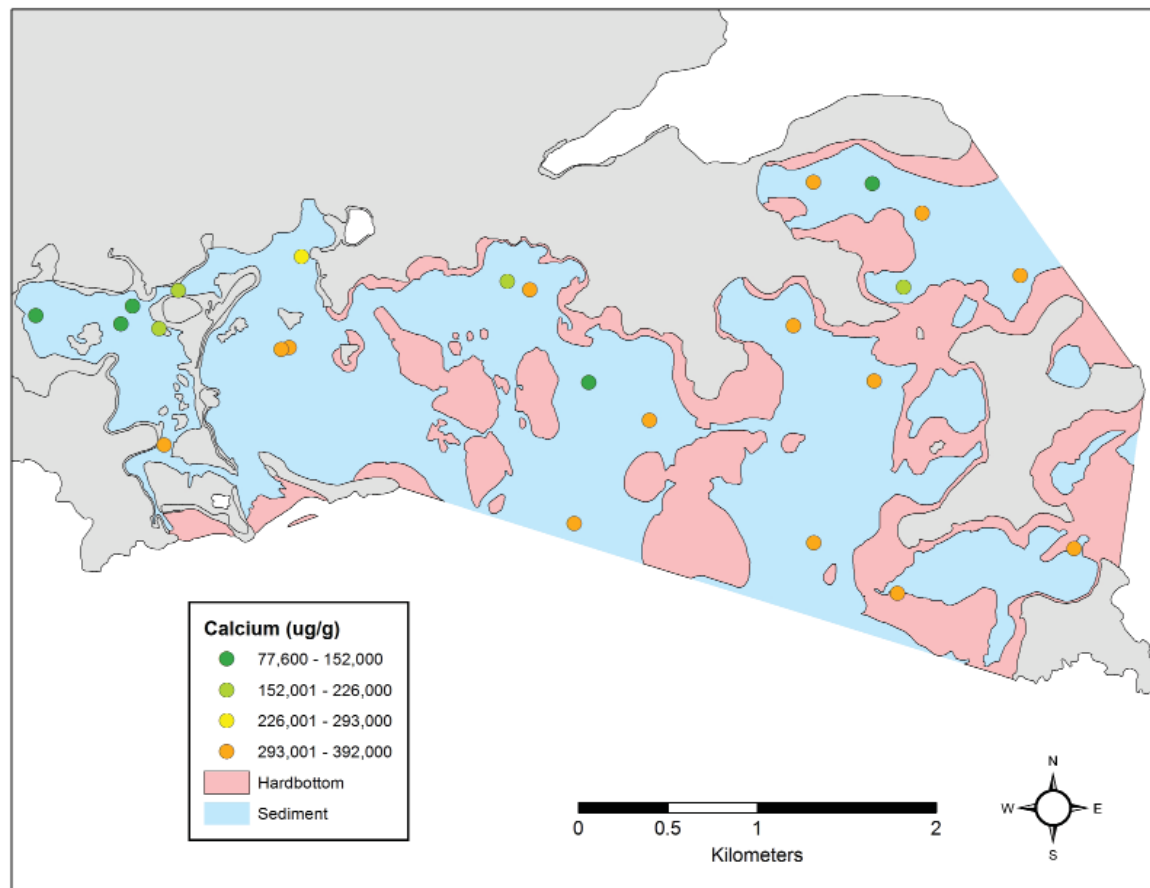


Figure 5.12. Calcium detected in sediments collected in the STEER.

had a total butyltins concentration of 231 $\mu\text{g/g}$. Station S6 was taken on the edge of the channel leading out to open waters. The spike in concentration at that point, was almost as high as seen at S1. Notably, the percentage of tributyltin (relative to total butyltins) at that site was 93.7%, indicating fresh contamination. This strongly suggests a recent spill or perhaps grounding of a freshly painted vessel. It is unclear why butyltins are elevated at 16P and BB1, unless there are

other sources in the area as well.

There is also a strong gradient of copper leading away from the marina complex (Figure 5.13, Table 5.1). Copper-based bottom paints were used before the advent of TBT, and copper-based paints have replaced the TBT paints. Note also, that copper at Station S6 is not elevated, in contrast to TBT. The concentration of copper in the cores show a clear increase in concentrations over time at all locations (Figure 5.14), including at B3. Copper concentrations exceeded the ERL at all surface stations.

5.4 CONCLUSIONS

The inner reaches of northern Benner Bay are degraded by marina operations, shoreline development and watershed changes. Sediment dynamics and sediment quality are, and have been for decades, heavily impacted. The benthic community was once a thriving ecosystem as evidenced by the remnants of shelled species, that, at the bottom of some cores exceeded the volume of sediment present. That community has vanished and was smothered by very fine sediment that accumulates at rates an order of magnitude above rates seen at other core locations in the STEER. The sediment is highly contaminated with butyltin paint residues, copper, and other toxic metals. The sediment is contaminated below the surface as well. Dredging new or deeper channels will spread these contaminants over a wide area. Dredging for remediation purposes is advisable, but will be expensive to do with methods that will properly prevent dredge spoil release to the water column. Otherwise, the benthic community will likely never recover, and environmental damage may expand if contaminants are diffused over a larger area. Matthiessen (2013) reported that benthic communities do not recover

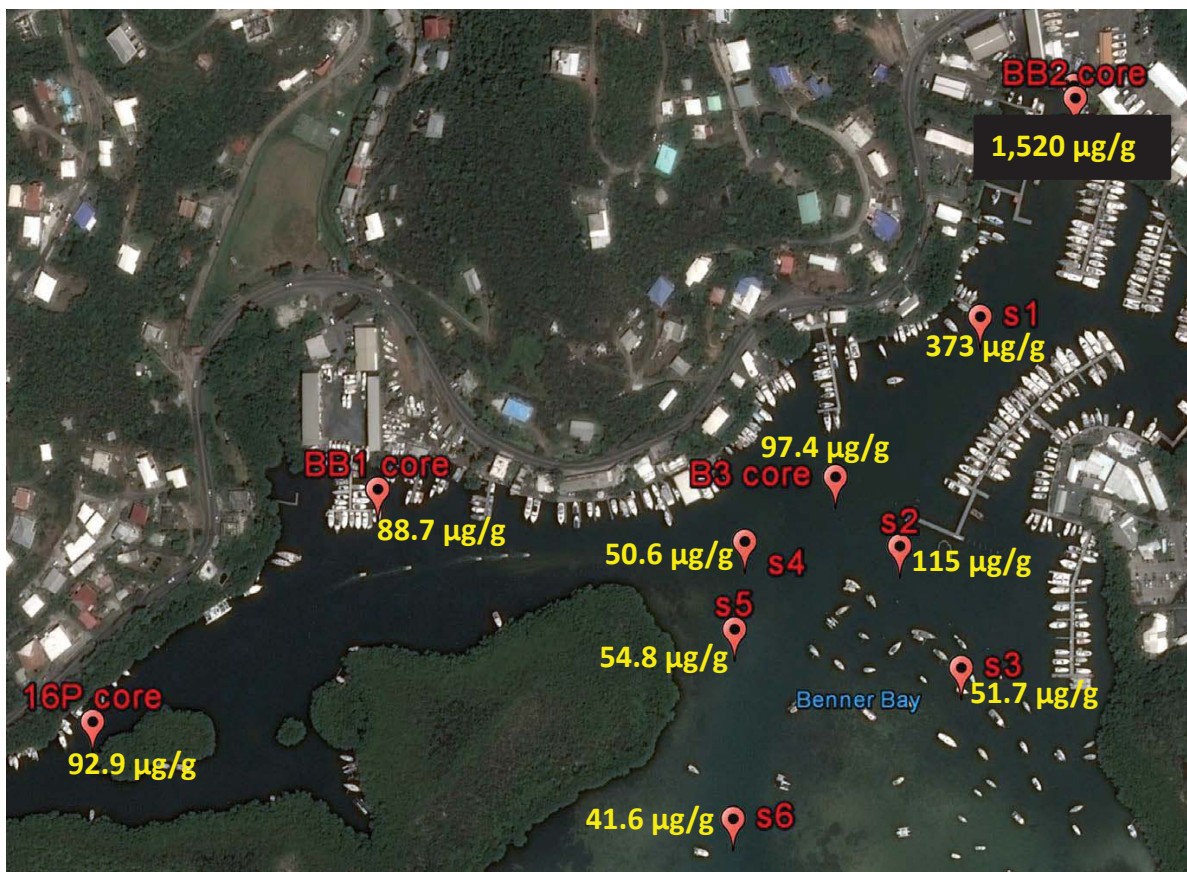


Figure 5.13. Concentration of copper in surface sediments in northern Benner Bay.

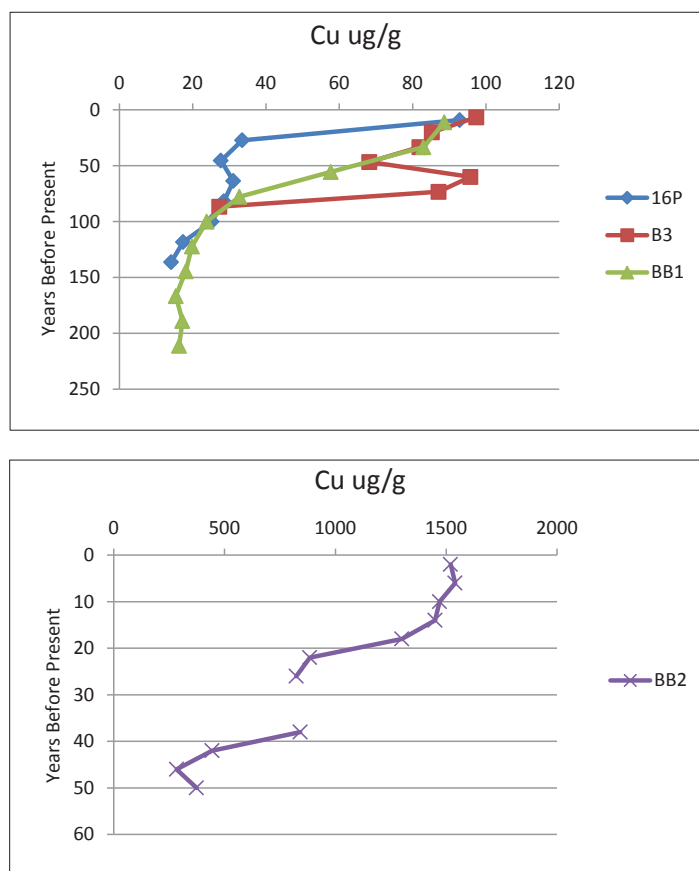


Figure 5.14. Copper concentrations as a function of time.

from TBT contamination until the concentration of butyltins is reduced to 10-40 ng/g. The concentration of butyltins at a depth of 6-8 cm was 8,871 ng/g at Station BB2. The system may be receiving fresh inputs of TBT as evidenced by the percentage of tributyltin residues at station S6. The system is dynamic. Between sampling in 2011 and 2013, the concentration of TBT had more than doubled at station BB2. Copper had increased by 50%. What event(s) may have caused such large changes in such a short time are unknown. Boat groundings, prop wash, and pile driving are all likely activities present at a marina that would stir up the bottom, and bring up contaminants from deeper in the sediment column.

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CHAPTER 6: AN ASSESSMENT OF CHEMICAL CONTAMINANTS DETECTED IN PASSIVE WATER SAMPLERS DEPLOYED IN THE ST. THOMAS EAST END RESERVES (STEER)

Anthony S. Pait¹, S. Ian Hartwell¹, Andrew L. Mason¹, Francis R. Galdo Jr.², Robert A. Warner¹, Christopher F. G. Jeffrey^{1,3}, Anne M. Hoffman⁴, Dennis A. Apeti¹, and Simon J. Pittman^{1,5}

¹NOAA/NOS/National Centers for Coastal Ocean Science, Center for Coastal Monitoring and Assessment (CCMA), 1305 East/West Highway, Silver Spring, MD 20910

²The University of the Virgin Islands, St. Thomas, USVI

³CSS-Dynamac Consolidated Safety Services, Fairfax, VA 22030, under CSS-Dynamac Contract #EA-133C-14-NC-1384

⁴The Nature Conservancy, St. Thomas, USVI

⁵The Marine Institute, Plymouth University, United Kingdom

6.1. INTRODUCTION

Polar Organic Chemical Integrative Samplers or POCIS were developed by the U.S. Geological Survey (USGS) as a tool to detect water soluble contaminants in the environment. They were deployed in the STEER at five locations in February 2012.

The goal of using POCIS in the STEER was to assess the presence of and when possible, estimate ambient concentrations of water soluble contaminants. While the concentration and effects of chemical contaminants in sediments in the STEER were reported in earlier chapters, there are many chemical contaminants that don't readily accumulate in sediments, but are toxic to biota (e.g., some current use pesticides). Determining the presence and estimating the concentration of water soluble chemicals, specifically stormwater contaminants, provides additional information on stressors present in the STEER. As noted earlier, the STEER watershed contains hotels/resorts, an active landfill, an EPA Superfund site, residential areas with individual sewage treatment systems, all of which can be sources of a variety of stormwater contaminants to the STEER.

The POCIS (Figure 6.1) is a disk consisting of a sorbent matrix (Waters Oasis[®] HLB) sandwiched between two membrane sheets (Alvarez *et al.*, 2008). The porous membrane sheets allow water to flow through the sorbent, enabling the sorbent material to sequester (accumulate) the contaminants of interest. For deployment, the POCIS disks are assembled onto a frame and then placed in a deployment canister (Figure 6.2).

Many stormwater chemical contaminants, if present in the aquatic environment, are at very low concentrations, typically in the low parts per billion (ppb or µg/L) or even parts per trillion (ppt or ng/L) ranges. A classic approach for detecting compounds at very low concentrations in the aquatic environment has been to collect, filter and extract

large volumes of water, to enable their detection by analytical instrumentation. The POCIS disk, however, can potentially sample tens to hundreds of liters of water during deployment, providing time-weighted concentration estimates of contaminants present in the water column (Alvarez *et al.*, 2008). This technology also has an advantage over discrete water samples, as POCIS can sequester chemical contaminants that may be present in the water only episodically, such as during storm events or during a small discharge or spill (Alvarez *et al.*, 2008). Typical deployments of the POCIS last approximately 30 days.



Figure 6.1. Example of a POCIS disk. Image courtesy of the USGS.

The USGS (Alvarez *et al.* (2004) has also developed methods to estimate the ambient water concentration for a number of stormwater contaminants sequestered by the POCIS, using individual sampling rate values (R_s). The R_s values are experimentally determined for each chemical, and incorporated into the following equation:

$$C_w = \frac{N}{R_s t}$$

where

C_w is the ambient water concentration of the chemical
 N = the amount (e.g., ng) of chemical accumulated by the POCIS

R_s is the sampling rate (L/d), i.e., liters of water cleared of analyte per day, and
 t is the number of days of deployment for the POCIS.

The number of days the POCIS is deployed is used in calculating estimated concentrations of the chemicals of interest, providing some flexibility in the length of time the POCIS is deployed. Additional information on this technology can be found in Alvarez *et al.* (2004, 2008).

6.2. METHODS

The POCIS deployment canisters containing the POCIS disks were shipped on ice to the University of the Virgin Islands in sealed solvent-rinsed metal cans. On arrival at UVI, the cans with the canisters were placed in a freezer until deployment.

The POCIS deployment canisters were placed in the STEER on 16 February 2012 at five locations (Figure 6.3), by scientists from UVI, and DPNR's Division of Fish and Wildlife. The five sites where the POCIS were de-

ployed were the same targeted (not randomly selected) sites used for the monthly sampling in the STEER for nutrients, TSS and sedimentation (see Chapter 9). At each of these sites, sediment traps were attached to rods secured in the sediments (Figure 6.4). The POCIS deployment canisters were tethered to these same rods, by UVI and DPNR SCUBA divers.

A summary of conditions at each site at the time of POCIS canister

deployment is provided in Table 6.1. The depth of the POCIS deployment canisters ranged from 1 meter at the site

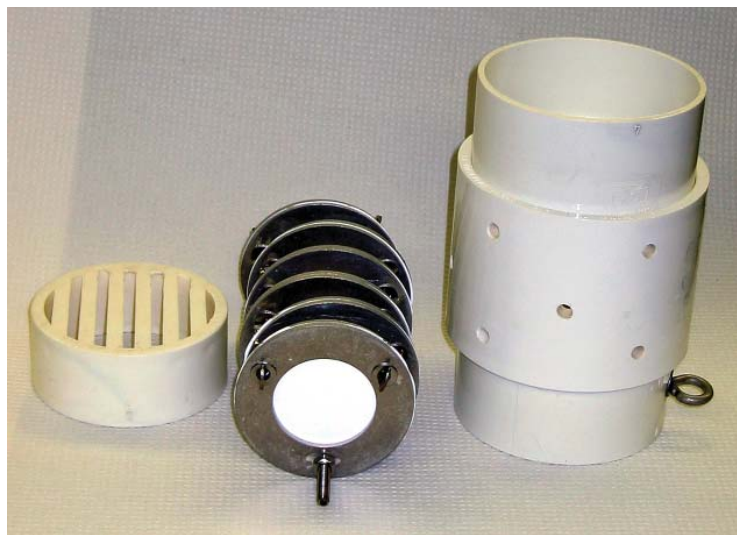


Figure 6.2. A disassembled POCIS deployment canister with the POCIS disks. Image courtesy of the USGS.

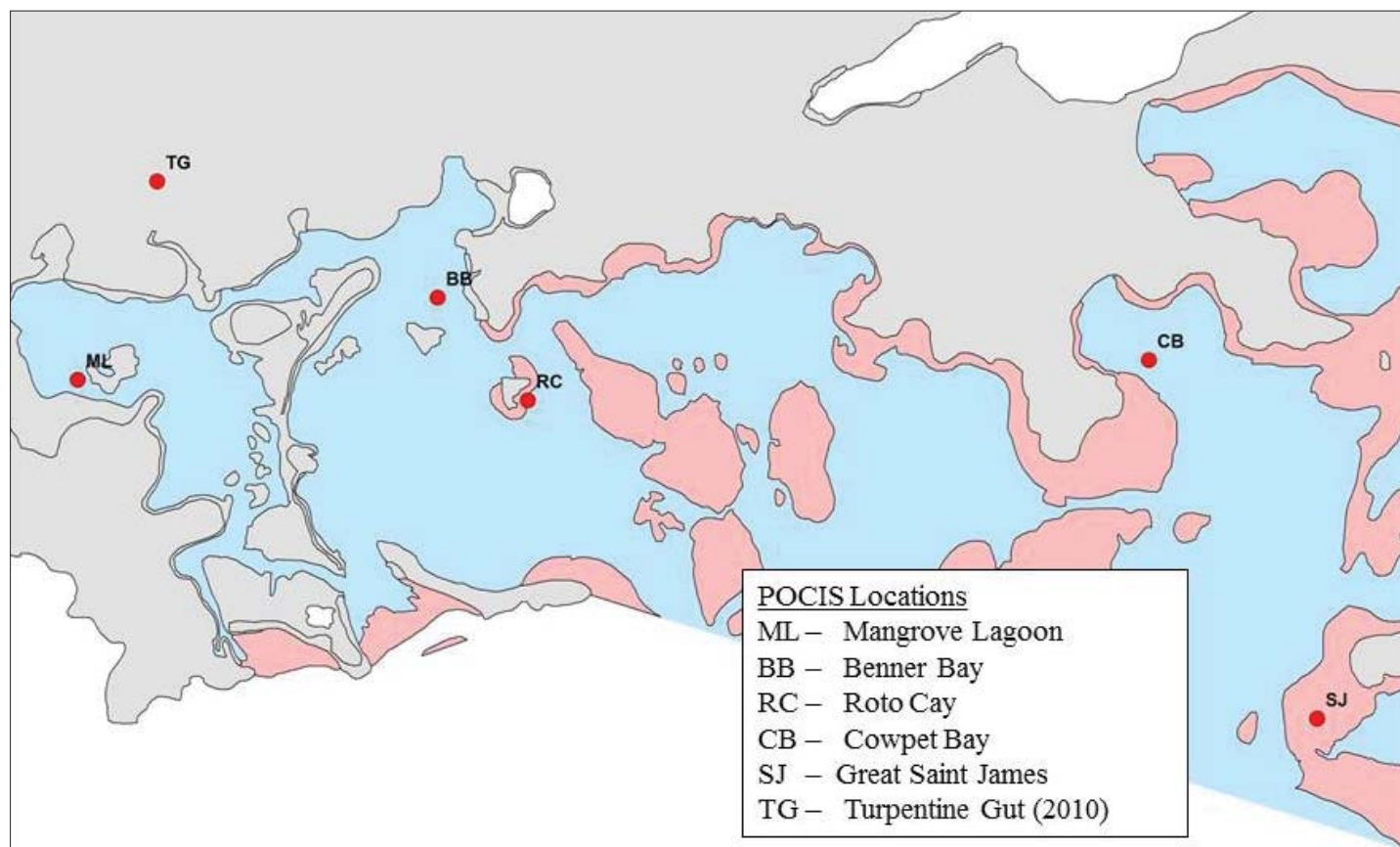


Figure 6.3. Locations of passive water samplers (POCIS) in the STEER.

in Benner Bay, to 12.2 meters at the site adjacent to Great St. James Island. Each POCIS deployment canister was set between 20 and 50 cm from the bottom.

As part of the preliminary work for the project in 2010, a POCIS deployment canister was placed in Turpentine Gut, the only perennial stream in St. Thomas (Nemeth and Platenberg, 2007). Although Turpentine Gut (site TG) is not within the boundaries of the STEER, it drains directly to Mangrove Lagoon (Figure 6.3), and is not only a significant source of freshwater to the STEER, but also of sediments (STEER, 2011), and likely other types of nonpoint source pollution including stormwater contaminants.

The POCIS deployment canister in Turpentine Gut was suspended in the water column at a depth of approximately 1.5 meters (Table 6.1). As with the 2012 deployment, two POCIS stormwater disks were included in the deployment canister. NOAA and DPNR Division of Fish and Wildlife personnel placed the POCIS deployment canister approximately 20 meters south of the unfinished bridge adjacent to Bovoni Road, on the east bank of Turpentine Gut.

Field blank canisters were used in both the 2012 and 2010 deployments. The field blank, a POCIS disk and frame, was exposed to the air during the time the POCIS canisters

were being deployed, in order to assess the presence of airborne contaminants.

During the 2012 deployment in the STEER, the field blank was exposed to the air at each of the five sites, and served as a cumulative field blank. For the 2010 deployment

in Turpentine Gut, one field blank was used. After a POCIS deployment canister entered the water, the POCIS field blank was placed back in its protective canister (solvent-rinsed metal container also used for shipping), sealed and then returned to UVI and placed in a freezer.

The five POCIS deployment canisters from the STEER were retrieved by UVI and DPNR Division of Fish and

Wildlife scientists using SCUBA on 14 March 2012. During the time the POCIS deployment canisters were out of the water, the field blank was again opened and exposed to the air. Once retrieved, the POCIS deployment canisters were returned to their protective canisters, and then placed in the freezer at UVI until they were shipped on ice along with the field blank in its own protective canister to the USGS for analysis. Additional information on POCIS deployment techniques can be found in Alvarez (2010).



Figure 6.4. POCIS deployment canister (arrow) among the sediment traps at a site in the STEER.

Table 6.1. Location and conditions during deployment of the POCIS canisters in February 2012 and May 2010.

Site	Date	Latitude	Longitude	Deployment Depth (m)	Water Conditions
Great St. James (SJ)	2/16/2012	18.30302	-64.83671	12.2	Clear, calm
Cowpet Bay (CB)	2/16/2012	18.31487	-64.84267	5.5	Clear, calm
Rotto Cay (RC)	2/16/2012	18.31331	-64.86423	5.5	Clear, calm
Benner Bay (BB)	2/16/2012	18.3167	-64.8674	1	High turbidity
Mangrove Lagoon (ML)	2/16/2012	18.31385	-64.87988	1.3	High turbidity
Turpentine Gut (2010) (TG)	5/5/2010	18.32046	-64.87719	1.5	Moderate flow, high turbidity

Note - depth of POCIS canister in meters (m)

At the USGS laboratory, the POCIS disks were removed from the deployment canisters, and dichloromethane and methyl-tert-butyl ether were used to extract the stormwater contaminants accumulated by the POCIS, for analysis. The analysis of the extracts from the POCIS was carried out using gas chromatography/mass spectrometry in the selected ion monitoring mode, and results were reported as a quantity of each chemical per POCIS extract (i.e., ng/ampoule). The sampling rate values (R_s) mentioned earlier were then used to estimate ambient water concentrations of those compounds found to be above the Quantitation Level. The Quantitation Level is the minimum concentration of the stormwater contaminant in the POCIS extract needed to ensure confidence in the reported values. Additional information on the analysis of the POCIS disks can be found in Alvarez *et al.* (2008).

The 69 stormwater contaminants analyzed from the POCIS are listed in Table 6.2, and include a number of pesticides, detergent metabolites, animal and plant sterols, flame retardants, fecal indicators and ingredients in personal care products. They represent a suite of water soluble contaminants analyzed by USGS, and are primarily associated with urban and domestic human use.

Pesticides include the herbicides atrazine, metolachlor and bromacil, and the insecticides chlorpyrifos, carbaryl, and diazinon. The metabolites of the surfactants/detergents included 4-octylphenol and para-nonylphenol, along with nonyl- and octylphenol polyethoxylates such as NPEO1 (nonylphenol monoethoxylate) and OPEO2 (octylphenol diethoxylate). The environmentally ubiquitous phthalate ester plasticizers diethyl phthalate (DEP) and diethylhexyl phthalate or DEHP, are also sequestered by POCIS.

A number of animal and plant-related compounds accumulated by the POCIS include the animal/plant sterol cholesterol, and the plant sterols beta-sitosterol and stigmasterol. Fecal indicators 3-beta-coprostanol and skatol are also sequestered by POCIS.

6.3. RESULTS AND DISCUSSION

A total of 26 stormwater contaminants were detected at least once in the STEER, although the majority (65%) of detections for these 26 were below the Quantitation Level, the minimum concentration sequestered in the POCIS disk needed to estimate concentrations. In Turpentine Gut, 31 stormwater contaminants were detected, a little over half (55%) of these 31 were below the Quantitation Level.

Table 6.2 presents the estimated ambient water concentrations of the stormwater contaminants accumulated by the

POCIS disks in the STEER and in Turpentine Gut. Additional information on the results, including the raw data can be found in Pait *et al.* (2013). The shading in Table 6.2 indicates those stormwater contaminants in the POCIS that were high enough to estimate ambient water concentrations. The Reporting Limit represents the lower limit for estimating the ambient water concentration of a stormwater contaminant.

Estimated Concentrations of Stormwater Contaminants in the STEER in 2012

From the February 2012 deployment at the five sites in the STEER, ambient water concentrations could be estimated for nine stormwater contaminants which are highlighted in Table 6.2. The highest estimated concentration for any of the compounds detected in the STEER was the animal/plant sterol cholesterol at the Benner Bay site (1,100 ng/L), followed by Cowpet Bay (800 ng/L) and Rotto Cay (550 ng/L). Cholesterol is an essential and natural component of cell membranes in animals, as well as an ingredient in some personal care products. Cholesterol is found to a lesser degree in some plant and fungal species, and has sometimes been used as a fecal indicator (Francy *et al.*, 2003).

Bargar *et al.* (2013) conducted a study on nearby St. John, in the Virgin Islands National Park and the Virgin Islands Coral Reef National Monument. Cholesterol concentrations detected by Bargar *et al.* (2013) ranged from 240 ng/L at Round Bay, to 440 ng/L at Whistling Cay in the POCIS deployed, similar to the concentrations found in the STEER. Along the South Florida coast (Miami to Looe Key), concentrations ranged from below the detection limit to 2,896 ng/L (Singh *et al.*, (2010).

Another class of stormwater contaminant detected in the STEER by POCIS were two phthalate ester plasticizers, diethylhexyl phthalate (DEHP), and diethyl phthalate (DEP). The highest estimated concentration of DEHP detected in the STEER was 300 ng/L at Great St. James. Due to health concerns regarding DEHP, including evidence that DEHP is an endocrine disruptor (Gray *et al.*, 2000) and a possible carcinogen (USDHHS, 2011), the use of DEHP has been reduced or eliminated for some applications.

The highest estimated concentration of DEHP detected in St. John, USVI by Bargar *et al.* (2013) was 820 ng/L at Whistling Cay, somewhat higher than what was estimated at Great St. James. As noted earlier, the deployment of the POCIS in the STEER was made in February 2012, during the dry season. Lower rainfall levels would likely lead to less runoff, which in turn could result in lower concentra-

Table 6.2. Estimated water column concentrations of stormwater contaminant residues sampled by POCIS in the STEER and in Turpentine Gut.

Compound	Use	February 2012 POCIS Deployment						May 2010 POCIS Deployment	
		Mangrove Lagoon	Benner Bay	Rotto Cay	Cowpet Bay	Great St. James	Reporting Limit	Turpentine Gut	Reporting Limit
		ng/L	ng/L	ng/L	ng/L	ng/L	ng/L	ng/L	ng/L
1,4-Dichlorobenzene	Moth repellent, fumigant, deodorizer	<RL	<RL	<RL	<RL	<RL	130	<RL	100
3,4-Dichlorophenyl isocyanate (m)	Degradation product of diuron or linuron	<RL	<RL	<RL	<RL	<RL	240	<RL	190
3-Beta-coprostanol (m)	Fecal indicator	<RL	<RL	<RL	<RL	<RL	240	<RL	190
4-Cumylphenol (m)	Detergent metabolite	<RL	<RL	<RL	<RL	<RL	12	<RL	10
4-n-Octylphenol (m)	Detergent metabolite	<RL	<RL	<RL	<RL	<RL	5.2	<RL	4.2
4-Tert-octylphenol	Detergent metabolite	<RL	<RL	<RL	<RL	<RL	5.2	17	4.2
5-Methyl-1H-benzotriazole	Anticorrosive used in antifreeze	<RL	<RL	<RL	<RL	<RL	110	140	86
Acetophenone	Fragrance used in detergent and tobacco	<RL	<RL	<RL	<RL	<RL	29	41	24
Anthraquinone (m)	Used in the manufacture of dyes/textiles	<RL	<RL	<RL	<RL	<RL	18	<RL	15
Atrazine	Herbicide	<RL	<RL	<RL	<RL	<RL	20	<RL	16
Benzophenone	Component of perfumes and soaps	<RL	<RL	<RL	<RL	<RL	36	<RL	30
Beta-sitosterol (m)	Plant sterol	<RL	<RL	620	<RL	<RL	480	<RL	380
BHA (m)	Food additive, preservative	<RL	<RL	<RL	<RL	<RL	73	<RL	61
Bisphenol A (m)	Manufacturing resin	<RL	<RL	<RL	<RL	<RL	1.9	6.2	1.5
Bromacil	Herbicide	<RL	<RL	<RL	<RL	<RL	72	<RL	60
Bromoform	Byproduct of water chlorination	230	84	230	160	160	49	<RL	41
Caffeine	Stimulant	<RL	<RL	<RL	<RL	<RL	19	<RL	16
Camphor	Fragrance in personal-care products	<RL	<RL	<RL	<RL	<RL	35	45	29
Carbaryl (m)	Insecticide	<RL	<RL	<RL	<RL	<RL	35	<RL	29
Carbazole	Used in the manufacture of dyes and lubricants	<RL	<RL	<RL	<RL	<RL	8.1	<RL	6.7
Chlorpyrifos	Insecticide	<RL	<RL	<RL	<RL	<RL	180	<RL	150
Cholesterol (m)	Plant/animal sterol	<RL	1100	550	800	380	240	610	190
Cotinine (m)	Nicotine metabolite	<RL	<RL	<RL	<RL	<RL	70	<RL	58
Cumene	Paint thinner	<RL	<RL	<RL	<RL	<RL	16	<RL	13
Diazinon	Insecticide	<RL	<RL	<RL	<RL	<RL	22	<RL	18
Dichlorvos	Insecticide (e.g., used in pet collars)	<RL	<RL	<RL	<RL	<RL	110	<RL	94
Diethyl phthalate	Plasticizer	97	92	72	100	120	20	120	16
Diethylhexyl phthalate	Plasticizer	77	66	65	83	300	29	1100	24
d-Limonene	Antimicrobial, fungicide, fragrance	<RL	<RL	<RL	<RL	<RL	70	<RL	58
Ethanol,2-butoxy-,phosphate	Flame retardant	<RL	<RL	<RL	<RL	<RL	170	220	140
Ethyl citrate	Plasticizer	<RL	<RL	<RL	<RL	<RL	4.5	<RL	3.7
Galaxolide (HHCb)	Synthetic fragrance	<RL	<RL	<RL	<RL	<RL	5.4	18	4.4

Contaminant residues sampled by POCIS in the STEER; concentrations above the Reporting Limit for a compound are indicated by shading. POCIS, polar organic chemical integrative sampler; ng/L, estimated water concentration reported; (m), indicates a highly variable compound; <RL, less than the reporting limit.

Table 6.2. Estimated water column concentrations of stormwater contaminant residues sampled by POCIS in the STEER and in Turpentine Gut (continued).

Compound	Use	February 2012 POCIS Deployment						May 2010 POCIS Deployment	
		Mangrove Lagoon	Benner Bay	Rotto Cay	Cowpet Bay	Great St. James	Reporting Limit	Turpentine Gut	Reporting Limit
		ng/L	ng/L	ng/L	ng/L	ng/L	ng/L	ng/L	ng/L
Indole	Component of fragrances	<RL	17	<RL	<RL	<RL	14	110	11
Isoborneol	Natural insect repellent	<RL	<RL	<RL	<RL	<RL	35	<RL	29
Isophorone	Solvent	<RL	<RL	<RL	<RL	<RL	4.5	<RL	3.6
Isoquinoline	Flavor and fragrance ingredient	<RL	<RL	<RL	<RL	<RL	14	<RL	12
Menthol	Decongestant	<RL	200	<RL	<RL	<RL	150	<RL	120
Metaxyl	Fungicide	<RL	<RL	<RL	<RL	<RL	90	<RL	74
Methyl salicylate	Liniment, flavoring agent	<RL	<RL	<RL	<RL	<RL	30	<RL	25
Metolachlor	Herbicide	<RL	<RL	<RL	<RL	<RL	2.7	<RL	2.2
n-n-Diethyltoluamide (DEET)	Mosquito repellent	7.6	<RL	<RL	<RL	<RL	4.9	95	4.0
NPEO1-total (m)	Detergent metabolite	<RL	<RL	<RL	<RL	<RL	100	<RL	83
NPEO2-total (m)	Detergent metabolite	<RL	<RL	<RL	<RL	<RL	170	<RL	140
OPEO1 (m)	Detergent metabolite	<RL	<RL	<RL	<RL	<RL	17	<RL	13
OPEO2 (m)	Detergent metabolite	<RL	<RL	<RL	<RL	<RL	17	21	14
Para-cresol	Wood preservative	<RL	<RL	<RL	<RL	<RL	27	520	23
Para-nonylphenol-total (m)	Detergent metabolite	<RL	<RL	<RL	<RL	<RL	76	<RL	63
Phenol	Disinfectant, manufacturing intermediate	150	250	<RL	29	<RL	25	35	20
Prometon	Herbicide	<RL	<RL	<RL	<RL	<RL	10	<RL	9
Skatol	Present in feces and coal tar	<RL	<RL	<RL	<RL	<RL	21	20	17
Stigmastanol (m)	Plant sterol	<RL	<RL	<RL	<RL	<RL	240	<RL	190
Tetrachloroethylene (m)	Dry cleaning	<RL	<RL	<RL	<RL	<RL	54	<RL	45
Tonalide (AHTN)	Synthetic fragrance	<RL	<RL	<RL	<RL	<RL	5.2	<RL	4.3
Tri(2-chloroethyl)phosphate	Plasticizer, flame retardant	<RL	<RL	<RL	<RL	<RL	15	<RL	13
Tri(dichlorisopropyl)phosphate	Flame retardant	<RL	<RL	<RL	<RL	<RL	190	<RL	150
Tributylphosphate	Anti-foaming agent, fire retardant	<RL	<RL	<RL	<RL	<RL	7.9	7.3	6.5
Triclosan	Disinfectant, antimicrobial	<RL	<RL	<RL	<RL	<RL	4.9	<RL	4.0
Triphenyl phosphate	Plasticizer, flame retardant	<RL	<RL	<RL	<RL	<RL	47	<RL	39

Contaminant residues sampled by POCIS in the STEER; concentrations above the Reporting Limit for a compound are indicated by shading. POCIS, polar organic chemical integrative sampler; ng/L, estimated water concentration reported; (m), indicates a highly variable compound; <RL, less than the reporting limit.

tions of contaminants in the water column. The deployment of POCIS in St. John occurred in May, which Bargar *et al.* (2013) identified as the beginning of the rainy season. Alvarez *et al.* (2014) quantified contaminants of emerging concern at 11 sites along the California coast including the Southern California Bight and San Francisco Bay. The mean estimated concentration of DEHP using POCIS was 400 ng/L, and ranged from not detected to 1,100 ng/L. In a study to assess organic micropollutants in coastal waters from the northwestern Mediterranean Sea (Catalonia to Valencia, Spain), Sánchez-Avila *et al.* (2012) sampled coastal seawater from 22 sites. DEHP was detected in over 90 percent of the water samples collected. The mean concentration was 145 ng/L, the maximum detected in coastal seawaters was 617 ng/L (Sánchez-Avila *et al.*, 2012).

The highest estimated ambient water concentration of DEP from the STEER was 120 ng/L, at the Great St. James site. The mean concentration of DEP estimated by Alvarez *et al.* (2014) in California from the POCIS was 150 ng/L, similar to the highest estimated concentration in the STEER; the highest concentration estimated by Alvarez *et al.* (2014) was 600 ng/L. In the northwestern Mediterranean, the mean concentration of DEP detected in coastal seawaters was 253 ng/L with a maximum of 483 ng/L. DEP has also been identified as a possible endocrine disruptor (Cólon *et al.*, 2000).

Phenol had an estimated ambient water concentration of 250 ng/L at the Benner Bay site, and 150 ng/L in Mangrove Lagoon. Phenol is a common intermediate used in the production of many types of products including detergents and plastics, and is also used in oral antiseptics and as a disinfectant. Phenols can be released into the environment through wastewater treatment systems, from either the direct use of phenol or degradation of phenolic compounds, and also as leachate from landfills, as materials containing phenols degrade (Masoner and Mashburn, 2004). Phenol was also a low level contaminant in the POCIS fabrication and field blanks. Phenol was not detected in the POCIS deployed by Bargar *et al.* (2013) in St. John.

In the marine environment, the presence of bromoform is often the result of algal metabolism (Palmer and Reason, 2009). In freshwater, bromoform can be an indicator of the

effects of chlorination or ozonation of drinking water, or from the chlorination of treated wastewater. The highest estimated bromoform concentration from the POCIS deployed in the STEER was 230 ng/L in Mangrove Lagoon and Rotto Cay, followed by 160 ng/L in Cowpet Bay and Great St. James Bay. In St. John, Bargar *et al.* (2013) detected concentrations of bromoform ranging from 73 ng/L to 170 ng/L, similar to what was found in the STEER. In their work along the California coast, Alvarez *et al.* (2014), detected concentrations of bromoform ranging from 5 ng/L to 77 ng/L; bromoform was detected in all POCIS samples.



The Bovoni Landfill in the western end of the STEER receives solid waste from both St. Thomas and St. John.

A number of fragrance-related compounds were detected in the POCIS in the STEER as well. The concentration of indole at the Benner Bay site was estimated at 17 ng/L, just above the Reporting Limit of 14 ng/L (Table 6.2). Indole was detected at two of the four sites in St. John, by Bargar *et al.* (2013) and the highest estimated concentration was 17 ng/L, the same as the Benner Bay site in the STEER. A number of other fragrances, including acetophenone, benzophenone, d-limonene, and

galaxolide (HHCB) were detected at most of the STEER sites, but the concentrations were below the Reporting Limit (Table 6.2). Acetophenone and galaxolide were also below the Reporting Limit at the sites sampled by POCIS in St. John (Bargar *et al.*, 2013).

Estimates of the concentration of two other personal care product ingredients, including menthol and n,n-diethyl-toluamide (DEET) could be made for the STEER. There was only one detection of menthol, at an estimated ambient water concentration of 200 ng/L from the POCIS placed in Benner Bay. The menthol could be related to its use in cigarettes, however cotinine, a degradation product of nicotine, could not be quantified in any of the POCIS deployed in the STEER or in Turpentine Gut.

The highest estimated concentration of DEET in the STEER was 7.6 ng/L from the POCIS deployed in Mangrove Lagoon. DEET is an insect repellent and its use would not be surprising, given the likely need for protection against mosquitos and other insect pests in the area. In St. John, DEET was detected but was below the method detection limit (Bargar *et al.*, 2013). On the California coast,

DEET was detected in 60 percent of the POCIS deployed, with an average concentration of 10 ng/L and a maximum concentration of 69 ng/L, lower than in the STEER (Alvarez *et al.*, 2014). Along the South Florida coast, DEET ranged from not detected to 68 ng/L (Singh *et al.*, 2010).

The presence of beta-sitosterol typically results from the decay of plant materials from natural sources, and from the use and decay of paper products. The only occurrence for which an estimate of the ambient water concentration could be made in the STEER was at Rotto Cay with an estimated concentration of 620 ng/L, although beta-sitosterol was detected (but below the Reporting Limit) at all the other sites.

Although Mangrove Lagoon receives inputs from the Bovi Landfill through runoff, and inputs from surrounding residential/commercial areas and from Turpentine Gut, this did not seem to result in a greater number of stormwater contaminants in Mangrove Lagoon compared to the other POCIS sites in the STEER, as was expected. The sites in the STEER in Figure 6.5 are ordered in a west to east fashion. There did not appear to be an obvious pattern or gradient in the number of compounds in the STEER, moving west to east away from Turpentine Gut and Mangrove Lagoon. There were four compounds with estimated ambient water concentrations in Mangrove Lagoon, however, there were five compounds each at Benner Bay, Rotto Cay and Cowpet Bay. Additional POCIS, perhaps offshore of these areas would provide more information, including the possibility of gradients.

Estimated Concentrations of Stormwater Contaminants in Turpentine Gut (2010)

The deployment of the POCIS in Turpentine Gut was almost two years earlier than the POCIS deployed in the STEER. In addition, the deployment in Turpentine Gut and the STEER were done at somewhat different times of the year; mid-spring for Turpentine Gut, and mid to late winter for the POCIS deployment in the STEER. Nevertheless, the earlier POCIS deployment in Turpentine Gut provides a useful snapshot of stormwater contaminants present in St. Thomas' only perennial stream during the deployment of the POCIS there.

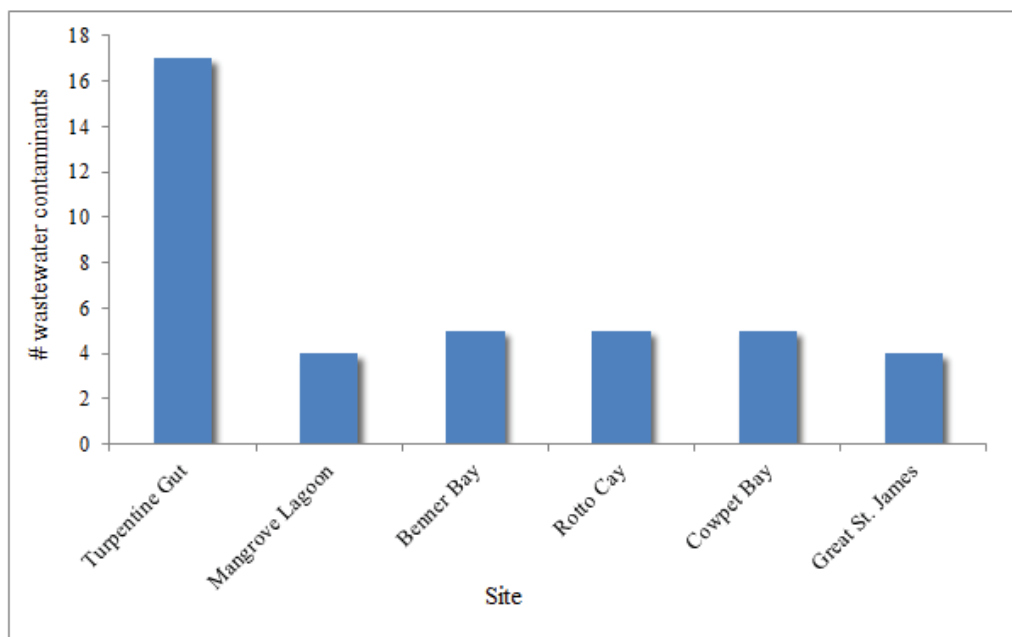


Figure 6.5. Number of stormwater contaminants with estimated water column concentrations in the STEER and in Turpentine Gut.

As was noted earlier, ambient water concentrations could be estimated for a greater number of compounds in Turpentine Gut than in the STEER (Figure 6.5). Disregarding the temporal aspects of the deployment, these findings are not surprising, as Turpentine Gut runs through the heart of the Jersey Bay watershed before it empties into Mangrove Lagoon, and likely receives input from a variety of sources. Land use in the watershed surrounding Jersey Bay is roughly 30% low- and mid-density urban development, with the remaining land use primarily forest and shrub (62%) (Cadmus, 2011).

Two compounds, the plasticizer DEHP (1,100 ng/L) and the insect repellent DEET (95 ng/L) could be quantified in Turpentine Gut. Both DEHP and DEET are common stormwater/wastewater contaminants, and their appearance in Turpentine Gut is not surprising. The DEHP concentration in Turpentine Gut was similar to the maximum concentration found by Bargar *et al.* (2013) in St. John (820 ng/L), and was the same as the maximum found by Alvarez *et al.* (2014), along the California coast (1,100 ng/L). The estimated concentration of DEET found in Turpentine Gut was higher than the highest concentrations estimated by Alvarez *et al.* (2014) (69 ng/L) along the California coast, and by Singh *et al.* (2010) (68 ng/L) in South Florida.

There were 10 compounds in the POCIS for which ambient water concentration estimates could be made from Turpentine Gut, that were either not found or were below Reporting Limits at the sites in the STEER. Two of these were the

detergent metabolites 4-tert-octylphenol and OPEO2. The estimated concentration of 4-tert-octylphenol in Turpentine Gut was 17 ng/L (Table 6.2). Bargar *et al.* (2013) did not detect 4-tert-octylphenol in the POCIS deployed in St. John.

The estimated ambient water concentration of bisphenol-A or BPA in Turpentine Gut was 6.2 ng/L. Bisphenol-A was detected by Singh *et al.* (2010) at concentrations up to 190 ng/L in South Florida. In coastal seawater, Sánchez-Avila *et al.* (2012) detected an average bisphenol-A concentration of 18 ng/L, ranging from 2.3 ng/L to 102 ng/L. Bisphenol-A has also been identified as an endocrine disruptor in rainbow trout (Lindholm *et al.*, 2000).

There were three personal care product ingredients detected above the Reporting Limit in Turpentine Gut. Camphor (45 ng/L) and galaxolide (18 ng/L) are fragrance ingredients. Acetophenone was also identified in Turpentine Gut at an estimated ambient concentration of 41 ng/L. As noted earlier, acetophenone was detected in the STEER at all sites, but below the Reporting Limit. Along the California coast, galaxolide was present in 80 percent of the POCIS samples, with a mean concentration of 150 ng/L, and acetophenone was also present in 80 percent of the samples from POCIS, with a mean concentration of 11 ng/L (Alvarez *et al.*, 2014).

The corrosion inhibitor, 5-methyl-1H-benzotriazole was detected at an estimated concentration of 140 ng/L in Turpentine Gut, and may be related to its use in antifreeze formulations for automobiles. Two flame retardants, ethanol,2-butoxy-,phosphate (220 ng/L), and tributylphosphate (7.3 ng/L) were only found in the Turpentine Gut POCIS. In addition to being a flame retardant, tributylphosphate has a number of other uses including as a defoamer in detergent solutions and in paints and adhesives. Alvarez *et al.* (2014), estimated a mean tributylphosphate concentration of 6.6 ng/L along the California coast, similar to the concentration estimated in Turpentine Gut.

Para-cresol, used as a wood preservative, was detected in Turpentine Gut with an estimated water concentration of 520 ng/L. As with a number of the other stormwater contaminants, para-cresol was detected in the POCIS from all sites in the STEER, but was below the Reporting Limit.

The fecal marker 3-methyl-1(H)-indole, or skatol, was detected at an estimated ambient water concentration in Turpentine Gut of 20 ng/L, just above the Reporting Limit of 17 ng/L. A second fecal indicator, 3-beta-coprostanol was also detected in Turpentine Gut, however, at a level

Table 6.3. Comparison of estimated concentrations of stormwater contaminants with available water quality criteria.

Wastewater Contaminant	Water Quality Criteria			
	Marine (µg/L)		Freshwater (µg/L)	
	Acute	Chronic	Acute	Chronic
Bromoform	—	—	2,300	320
DEP	2,944	3.4	1,800	110
DEHP	400	360	400	0.3-16
Para-cresol	—	—	<230	<13
Phenol	5,800	400	10,200	180-320
Tetrachloroethylene	10,200	450	830	45

DEP, diethyl phthalate; DEHP, diethylhexyl phthalate.

Marine and freshwater criteria represent water guidelines developed by the USEPA. Water Quality Criteria values taken from NOAA Screening Quick Reference Tables (SQuiRTs; Buchman, 2008).

too low to estimate a water concentration. The presence of both these fecal indicators from Turpentine Gut would seem indicative of inputs from septic systems and from animals (e.g., dogs and horses).

No cotinine or caffeine was detected in the POCIS deployed in Turpentine Gut or in the STEER, which was somewhat surprising given the density of the population in this part of St. Thomas, and likely presence of these compounds in tobacco products or in caffeinated beverages. Neither of these compounds were detected in POCIS deployed in St. John (Bargar *et al.*, 2013).

Comparison with Water Quality Criteria

Table 6.3 lists a number of water quality criteria developed by the U.S. Environmental Protection Agency (EPA) and others. Almost all the estimated water concentrations of the contaminants from both the STEER and Turpentine Gut (Table 6.2) were below, in most cases orders of magnitude below, the criteria listed in Table 6.3. The only exception was the plasticizer DEHP from Turpentine Gut. The estimated water concentration in Turpentine Gut (1.1 µg/L or 1,100 ng/L) is above a lower chronic (longer-term exposure) criteria (0.3 µg/L) (Table 6.3).

Finally, the compound tetrachloroethylene is also included in Table 6.3. Within the watershed that drains to Turpentine Gut and then to the STEER is the Tutu Wellfield Superfund Site. This EPA Superfund site was established due to contamination of groundwater and wells in the area by chlorinated volatile organic compounds (CVOC), including tetrachloroethylene. This compound was used extensively for dry cleaning by a textile plant located in the watershed,

that operated from 1969 to 1982 (EPA, 2011). Contamination of the groundwater and commercial and private wells in the Tutu area resulted in EPA establishing the Superfund site and installing groundwater treatment systems to remove CVOCs, including tetrachloroethylene (EPA, 2011).

In Table 2, it can be seen that there were no estimated water concentrations for tetrachloroethylene in either Turpentine Gut or in Mangrove Lagoon. There was one detection of tetrachloroethylene from the POCIS deployed adjacent to Great St. James Island, however, the level was below the Quantitation Level, that would have allowed an ambient water concentration estimate to be made. The location of this POCIS adjacent to Great St. James (Figure 6.3), would make a connection to the Tutu Wellfield Superfund Site appear unlikely, due to the distance from the Superfund site, along with the lack of detections in the POCIS deployed in Turpentine Gut and Mangrove Lagoon, both of which are closer to the Superfund site.

6.4. CONCLUSIONS

The deployment of five POCIS in the STEER in February 2012, and one in Turpentine Gut in May 2010 provides information on the presence of stormwater contaminants that might be missed if sediments alone were sampled. Although Turpentine Gut is not within the boundaries of the STEER, this perennial stream drains most of the Jersey Bay watershed and empties into the STEER through Mangrove Lagoon. The stormwater contaminants detected appear fairly representative of the low- and mid-density urban development in the watershed.

The deployment and analysis of POCIS from the STEER and Turpentine Gut represents a snapshot of stormwater contaminants in these systems. The analysis resulted in the identification of a number of stormwater contaminants, however, the majority at concentrations were below reporting limits established by the USGS for the compounds analyzed. Stormwater contaminants that could be quantified at least once included phthalate ester plasticizers, wood preservatives, personal care product/fragrance ingredients, plant sterols and a fecal indicator.

Significantly, none of the nine pesticides, primarily agricultural use herbicides and insecticides, were detected in the POCIS. This would appear to reflect the low level of agriculture in the watersheds surrounding the STEER and in St. Thomas in general.

The most common contaminants identified in the POCIS were the phthalate ester plasticizers DEP and DEHP. Both were present in the POCIS at all sites, and their concentra-

tions were high enough to estimate ambient water concentrations. Both of these phthalates are ubiquitous environment contaminants. The estimated water concentration of DEHP in Turpentine Gut (1,100 ng/L) was above a chronic freshwater quality criteria, and was the only stormwater contaminant found to be above an available water quality criteria.

The POCIS passive water samplers represent a fairly new technology used by NOAA's National Centers for Coastal Ocean Science along with project partners. The use of POCIS provides the opportunity to assess the presence of water soluble chemical contaminants in coral reef environments. It would be useful to do additional deployments of POCIS in the STEER and perhaps in Turpentine Gut during different times of the year, to assess how the concentration of the stormwater contaminants might vary during the wet and dry seasons in St. Thomas. It would also be useful to target additional POCIS in Mangrove Lagoon and in northern Benner Bay, the areas that appear to be most impacted by LBSP.

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CHAPTER 7: AN ASSESSMENT OF CONTAMINANT BODY BURDENS IN CORAL (PORITES ASTREOIDES), CONCH (LOBATUS GIGAS) AND FISH (HOLOCENTRUS RUFUS AND LUTJANUS APODUS) FROM THE ST. THOMAS EAST END RESERVES (STEER)

Dennis A. Apeti¹, Andrew L. Mason¹, S. Ian Hartwell¹, Anthony S. Pait¹, Laurie J. Bauer^{1,2}, Christopher F. G. Jeffrey^{1,2}, Anne M. Hoffman³, Francis R. Galdo, Jr.⁴, and Simon J. Pittman^{1,5}

¹NOAA/NOS/National Centers for Coastal Ocean Science, Center for Coastal Monitoring and Assessment (CCMA), 1305 East/West Highway, Silver Spring, MD 20910

²CSS-Dynamac Consolidated Safety Services, Fairfax, VA 22030, under CSS-Dynamac Contract #EA-133C-14-NC-1384

³The Nature Conservancy, St. Thomas, USVI

⁴The University of the Virgin Islands, St. Thomas, USVI

⁵The Marine Institute, Plymouth University, United Kingdom

7.1 INTRODUCTION

Contaminant body burdens in coral, conch and fish were assessed for this part of the project in the STEER. Samples were collected from each of the five previously identified strata and analyzed for more than 150 chemical contaminants, including organic contaminants (e.g. polycyclic aromatic hydrocarbons, polychlorinated biphenyls and pesticides), along with trace (e.g. cadmium, copper, and mercury), and major (aluminum, iron and manganese) elements. Although the number of samples that could be collected for this project was low due to harvest limitations and funding, the information generated will benefit management by providing additional information on contaminant body burdens for informed coastal resource management decisions, as well as providing data for the Reserves-wide assessment of contaminant issues.

7.2. METHODS

Sampling Methods

Samples of coral were collected under the Department of Planning and Natural Resources (DPNR) permit STT-0023-12. Samples of coral, conch and fish were collected under DPNR permit STT-022-12. Collection of biota for this study followed the standard protocols established by the NOAA National Status and Trends (NS&T) Program (Apeti *et al.*, 2012). Sampling was conducted in collaboration with USVI partners who also provided logistical and boat support for the fieldwork.

Coral

Mustard hill coral, *Porites astreoides*, was chosen as it is abundant and has been used in a number of other NOAA NCCOS projects for body burden analyses. Coral tissue sampling locations for this study, with the exception of the coral collected in Mangrove Lagoon (Stratum 1), were chosen from a subset of pre-selected hard bottom (HB) locations where NCCOS Biogeography divers had determined that *P. astreoides* were present (Chapter 3). The hard bottom habitats were grouped into four separate strata (stratum 2 – 5), which in addition to the Mangrove Lagoon (ML) stratum, represented five strata for sampling. A coral sample site was defined as a single dive area with a 50 meter radius where enough *P. astreoides* colonies (“heads”) were available for sampling multiple heads. While in stratum 1, only one *P. astreoides* colony was found at site MLC01; within each of the HB strata two sample sites were selected (Figure 7.1). The coral sample



Image of the coral Porites astreoides in the southern portion of Mangrove Lagoon.

collected at HBI24p, was considered part of Stratum 2 because of the proximity of the site to the stratum boundary.

The coral samples were collected by SCUBA divers using a hammer and a titanium coring punch. At each site,

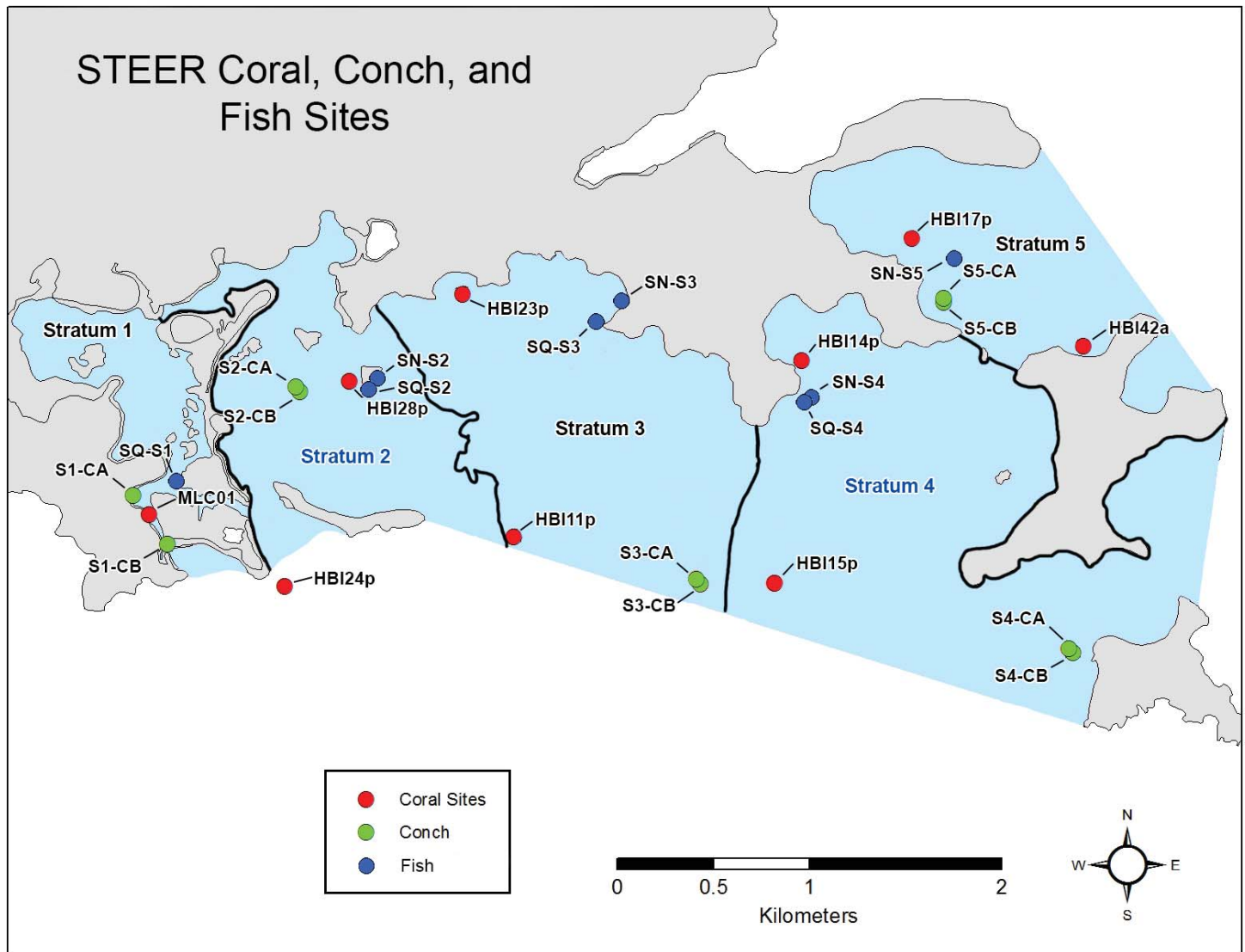


Figure 7.1. Coral, conch and fish sites collected in 2012 in the St. Thomas East End Reserves (STEER).

coral cores were collected from 5 different coral heads to constitute a site-composite. The titanium coring punch was driven into the coral colonies with a hammer, to extract cores of coral tissue approximately 1.5 cm in diameter and 1-1.5 cm in depth. Coral cores were dislodged from the corer with a Teflon stir stick. At each sampling site two sets of coral tissues were collected, one for contaminant analyses and the other for histopathology assessments. Care was taken to avoid removal of large amounts of unnecessary skeletal material, however, whatever skeletal material was removed along with the tissue was analyzed for this study. The cores of coral tissue were placed inside pre-labeled 250 ml IChem® jars and then capped underwater. The jars were brought to the surface, drained of water and placed on ice. The coral samples were then frozen at -15 °C until shipped overnight on ice to the analytical laboratory. At the laboratory, coral skeletal material and tissue were cryogrinded using liquid nitrogen.

Conch

Queen conch (*Lobatus gigas*) were chosen for this study because they are herbivorous gastropods that live in sand, seagrass beds and coral reefs, feeding on seagrass and various species of algae (Davis, 2005). It has been determined that through their feeding process, conch ingest considerable amounts of sediment particles (Brownell and Stevely, 1981), along with any contaminants that may be associated with the sediment.

Conch specimens were collected at locations within the previously defined five strata (Figure 7.1). Due to limited conch resources, only two specimens were collected per stratum. Collections of conch were made using SCUBA or snorkeling. Thus, from 10 separate locations (2 per stratum) (Figure 7.1) a total of 10 conch were collected by hand and placed in labeled 2 gallon Ziplock™ bags.

The bags containing the specimens were placed in a cooler with ice, and at the end of the day, frozen at the University of the Virgin Islands. At the end of the field mission, conch specimens were partially thawed, removed from their shells, weighed, and placed into labeled 1 liter Teflon jars and refrozen. Once completely frozen, the samples were shipped on ice to TDI-Brooks, International. There, the soft tissues were homogenized prior to contaminant analysis. This resulted in the production of only one data point per stratum, and consequently limited statistical comparisons of contaminant concentrations in conch between strata. However, lateral (east-west) and nearshore versus off shore contrasts could be made.

Fish

Fish were collected using nets and with hook and line. Spearfishing was avoided to reduce the possibility of biasing sample results via loss of fluids during and after collection, and possible introduction of additional metals into the tissues. Two species of fish were targeted for this study, the longspine squirrelfish (SQ) (*Holocentrus rufus*) and the schoolmaster snapper (SN) (*Lutjanus apodus*). Due to the difficulty in collecting the targeted species, a total of only eight fish, representing four of each species, were collected from across the STEER (Figure 7.1). Squirrelfish were taken in strata 1-4. Snappers were taken in strata 2-5. Because of the limited spatial nature of the data, statistical comparisons of contaminant concentrations between the strata identified during phase 1 of the study cannot be made, but lateral (east-west) contrasts can be made. All fish samples were stored at -15° C prior to shipment to the laboratory for analysis. At the laboratory the whole fish was homogenized, including both edible and inedible portions.

Water Quality Measurements

A series of water quality parameters (dissolved oxygen, temperature, and salinity) were also measured at each site using a YSI® salinity/conductivity/temperature meter. The instrument probe was submerged to a depth of approximately one meter.

Analytical Methods

The list of chemical contaminants analyzed in the coral and conch tissues is shown in Table 7.1. This contaminant list constitutes the suite of compounds regularly quantified nationwide as part of NOAA's NS&T Program.

For over 20 years, the Mussel Watch and NS&T Programs have monitored the Nation's estuarine and coastal waters for chemical contaminants in bivalve mollusk tissues and in sediments. Work to characterize chemical contaminants

as part of the NCCOS Center for Coastal Monitoring and Assessment's (CCMA) ecological characterizations in tropical waters, represents a fairly recent expansion of NS&T activities. The compounds analyzed included 59 polycyclic aromatic hydrocarbons (PAHs), 31 organochlorine pesticides, 38 polychlorinated biphenyls (PCBs), four butyltins, and 15 trace and major elements.

Organic Contaminants

Coral, conch and fish tissues were subjected to the same procedures for the determination of organic contaminant concentrations. Whole tissues were analyzed for both fish and conch, while coral tissues were analyzed along with attached skeletal material. Aliquots of tissue samples were chemically dried using Hydromatix®. Tissue/Hydromatix mixtures were then extracted with 100% dichloromethane using accelerated solvent extraction (ASE). Detailed analytical protocols are provided in Kimbrough and Lauenstein (2006) for organic compounds.

PAHs and their alkylated homologues (Table 7.1) were quantified using gas chromatography mass spectrometry (GC/MS) in the selected ion monitoring mode (SIM). Concentrations of PCBs and chlorinated pesticides were determined by capillary gas chromatography with an electron capture detector (ECD). All results are reported in ng/g dry weight (dw). Analysis for butyltins was based on high resolution, capillary gas chromatography using flame photometric detection (GC/FPD), which quantitatively determined tributyltin (TBT), dibutyltin (DBT), and monobutyltin (MBT). The concentration of butyltins was expressed as the concentration of tin (ng Sn/g).

Major and Trace Elements

The major and trace elements measured are presented in Table 7.1. Most of these elements are metals, however, antimony, arsenic and silicon are metalloids, and selenium is a nonmetal. Coral, conch, and fish were subjected to the same digestion and analytical methods (Kimbrough and Lauenstein, 2006). The whole animal was first homogenized, freeze-dried to a constant weight, and acid digested in Teflon bombs. For all metals except mercury, the tissue samples were prepared for inductively coupled plasma mass spectrometric (ICP-MS) analysis. For mercury quantification, tissue homogenates were acid digested based on a modified version of the Environmental Protection Agency (EPA) method 245.5 and measured using cold vapor atomic absorption spectroscopy.

Metals can exist in the environment in several forms (e.g., organic versus inorganic arsenic), but the analytical methods used by the NS&T does not distinguish between

Table 7.1. Chemical contaminants analyzed in STEER coral, conch and fish samples.

PAHs - Low MW	PAHs - High MW	Organochlorine Pesticides	PCBs	PCBs (continued)	Butyltins
Naphthalene	Fluoranthene	Aldrin	PCB8/5	PCB170/190	Monobutyltin
1-Methylnaphthalene	Pyrene	Dieldrin	PCB18	PCB174	Dibutyltin
2-Methylnaphthalene	C1-Fluoranthenes/Pyrenes	Endrin	PCB28	PCB180	Tributyltin
2,6-Dimethylnaphthalene	C2-Fluoranthenes/Pyrenes	Heptachlor	PCB29	PCB183	Tetrabutyltin
1,6,7-Trimethylnaphthalene	C3-Fluoranthenes/Pyrenes	Heptachlor-Epoxide	PCB31	PCB187	
C1-Naphthalenes	Naphthobenzothiophene	Oxychlorane	PCB44	PCB194	Major and Trace Elements
C2-Naphthalenes	C1-Naphthobenzothiophenes	Alpha-Chlordane	PCB45	PCB195/208	
C3-Naphthalenes	C2-Naphthobenzothiophenes	Gamma-Chlordane	PCB49	PCB201/157/173	Aluminum (Al)
C4-Naphthalenes	C3-Naphthobenzothiophenes	Trans-Nonachlor	PCB52	PCB206	Antimony (Sb)
Benzothiophene	Benz[a]anthracene	Cis-Nonachlor	PCB56/60	PCB209	Arsenic (As)
C1-Benzothiophenes	Chrysene	Alpha-HCH	PCB66		Cadmium (Cd)
C2-Benzothiophenes	C1-Chrysenes	Beta-HCH	PCB70		Chromium (Cr)
C3-Benzothiophenes	C2-Chrysenes	Delta-HCH	PCB74/61		Copper (Cu)
Biphenyl	C3-Chrysenes	Gamma-HCH	PCB87/115		Iron (Fe)
Acenaphthylene	C4-Chrysenes	DDMU	PCB95		Lead (Pb)
Acenaphthene	Benzo[b]fluoranthene	2,4'-DDD	PCB99		Manganese (Mn)
Dibenzofuran	Benzo[k]fluoranthene	4,4'-DDD	PCB101/90		Mercury (Hg)
Fluorene	Benzo[e]pyrene	2,4'-DDE	PCB105		Nickel (Ni)
C1-Fluorenes	Benzo[a]pyrene	4,4'-DDE	PCB110/77		Selenium (Se)
C2-Fluorenes	Perylene	2,4'-DDT	PCB118		Silicon (Si)
C3-Fluorenes	Indeno[1,2,3-c,d]pyrene	4,4'-DDT	PCB128		Silver (Ag)
Carbazole	Dibenzo[a,h]anthracene	1,2,3,4-Tetrachlorobenzene	PCB138/160		Tin (Sn)
Anthracene	C1-Dibenzo[a,h]anthracenes	1,2,4,5-Tetrachlorobenzene	PCB146		Zinc (Zn)
Phenanthrene	C2-Dibenzo[a,h]anthracenes	Hexachlorobenzene	PCB149/123		
1-Methylphenanthrene	C3-Dibenzo[a,h]anthracenes	Pentachloroanisole	PCB151		
C1-Phenanthrene/Anthracenes	Benzo[g,h,i]perylene	Pentachlorobenzene	PCB153/132		
C2-Phenanthrene/Anthracenes		Endosulfan II	PCB156/171/202		
C3-Phenanthrene/Anthracenes		Endosulfan I			
C4-Phenanthrene/Anthracenes		Endosulfan Sulfate	PCB158		
Dibenzothiophene		Mirex			
C1-Dibenzothiophenes		Chlorpyrifos			
C2-Dibenzothiophenes					
C3-Dibenzothiophenes					

Abbreviations: MW, molecular weight; PAH, polycyclic aromatic hydrocarbons; HCH, hexachlorocyclohexane; DDMU, 1-chloro-2,2- (p-chlorophenyl)ethylene, DDT, dichlorodiphenyltrichloroethane; DDD, dichlorodiphenyldichloroethane; DDE, dichlorodiphenyldichloroethylene; PCB, polychlorinated biphenyl

these various forms. Instead, analytical results are reported as total metal concentration (aggregation of all species of a metal) in microgram per gram ($\mu\text{g/g}$) for dry tissue weight (dw).

Statistical Analyses

Concentration values for individual compounds that were lower than the method detection limits (MDL) were qualified as undetected and assigned a value of zero. Concentration values of organic contaminants were presented as “total” concentration (e.g., PAHs), were derived as the arithmetic sum of all the individual congeners or homologues of the same group of compounds as listed in Table 7.1.

Primary statistical analyses were conducted using the JMP-5.1[®] system statistical package. The non-parametric Wilcoxon test was used for data comparisons, while relationships between variables (e.g. inter-metal correlations) were assessed using the Pearson correlation coefficient. A three-group classification scheme based on ArcGIS Jenks grouping method was used to assess the spatial distribution of the contaminants. Significance of statistical tests was reported at a probability level of 0.05.

A non-parametric Wilcoxon test showed that with the exception of percent tributyltin in total butyltins, there were no statistical differences between longspine squirrelfish and schoolmaster snapper for any contaminant for the limited number of samples collected in this study. As such we treat all fish as one population for calculating contaminant means and spatial analysis.

To put the concentrations found in coral, conch and fish from the STEER into context, results were compared to previously published studies conducted in the Caribbean. Additionally, contaminant body burdens of toxic metals and organic compounds in conch and fish were compared to available FDA and EPA guidelines for chronic consumption limits. FDA reports concentrations on a wet weight basis. The average measured percent moisture content of the conch from the STEER was 76%. A factor of four was therefore used to convert wet weight concentrations to dry weight in order to compare to results from other non-NS&T studies. Since acceptable concentrations of metals in fish tissue are determined on a case-by-case (including factors like region, and subsistence vs. recreational fishing) basis by the FDA, and because this study was not designed to measure the impacts of metals in fish tissues on human health, we used EPA’s Guidance for Assessing Chemical Contaminant Data for Use in Fish Advisories (EPA, 2000) as a guideline, where possible.

7.3. RESULTS AND DISCUSSION

Comparisons between strata were conducted using nonparametric Kruskal-Wallis tests ($\alpha = 0.05$). None of the strata were significantly different from any other for any contaminant in any matrix. To further explore spatial patterns, the strata were pooled into two groups, Western (strata 1 and 2) and Eastern (strata 3, 4, and 5). With the exception of mercury concentrations in fish tissues, there were no significant differences between the pooled groups of Eastern and Western for any contaminant for fish, coral, or conch.

Field Data

Sampling for the coral and conch in the STEER occurred 18-22 June 2012. The mean water depth at the coral sites was 5.1 ± 1.67 m; the mean surface water temperature was 29.1 ± 0.34 °C. The average surface salinity was 35.9 ± 0.07 ppt. The average surface water dissolved oxygen for coral sites was 3.96 ± 0.18 mg/L.

The mean water depth for the sites where conch were sampled was 5.97 ± 1.19 m; the mean surface water temperature was 29 ± 0.21 °C. The average surface salinity was 36 ± 0.01 ppt. The average surface water dissolved oxygen for conch sites was 4.17 ± 0.21 mg/L. Sampling for fish in the STEER occurred between July and November of 2013. No water quality measurements were taken during fish collections.

Organic Contaminants

Polycyclic Aromatic Hydrocarbons

Also referred to as PAHs, polycyclic aromatic hydrocarbons are associated with the use and combustion of fossil fuels (e.g., oil and gas) and other organic materials (e.g., wood). Natural sources of PAHs include forest fires and decaying plant material.

A number of PAHs can bioaccumulate to toxic concentrations in aquatic biota, and some PAHs including benzo[a]pyrene, benz[a]anthracene, chrysene, benzo[b]fluoranthene, benzo[k]fluoranthene, dibenzo[a,h]anthracene, and indeno[1,2,3-c,d]pyrene have been linked to carcinogenicity in vertebrates (USDHHS 1995). Significant relationships have also been established between PAHs in sediment and prevalence of liver lesions in English sole in Puget Sound, Washington (Malins *et al.* 1984). Very little research has been carried out to address the effects of PAHs on coral or conch. Vertebrates can metabolize PAHs, often into more harmful substances (e.g. carcinogens); mollusks do not. Solbakken *et al.* (1984) showed that both phenanthrene and naphthalene accumulate to detectable levels in brain coral *Diploria*

strigosa and green cactus coral *Madracis decatis*. While the simple accumulation of a PAH is not an impact in and of itself, the accumulation of exogenous chemicals in living tissue increases the likelihood of adverse effects. The PAHs fluoranthene and pyrene at $\mu\text{g/L}$ level concentrations have been shown to be toxic to coral, particularly in the presence of increased ultraviolet radiation, termed phototoxicity (Peachey and Crosby 1996; Guzman-Martinez *et al.*, 2007). Peters *et al.* (1981) found that exposure to No. 2 fuel oil at concentrations as low as $70 \mu\text{g/L}$ in the rose coral (*Manicina areolata*) over a 12 week period resulted in substantial histologic changes in both the somatic and reproductive tissues, along with the loss of the zooxanthellae.

PAHs in Corals. The concentrations of total PAHs found in coral tissues are presented in Figure 7.2. The mean concentration of total PAHs in the tissues of *P. astreoides* ($22.0 \pm 8.6 \text{ ng/g}$) (Table 7.2) was lower than those found in sediments ($142 \pm 59 \text{ ng/g}$) (Pait *et al.*, 2013) indicating that coral tissue integrate less PAHs than sediment in the surrounding area. The mean of $22.0 \pm 8.6 \text{ ng/g}$ in STEER *P. astreoides* tissues was comparable, but more variable, than the mean concentration of total PAHs in the tissues of *P. astreoides* ($15.0 \pm 0.6 \text{ ng/g}$) found in Vieques, Puerto Rico (Pait *et al.*, 2010) (Table 7.3), the closest geographical location where we have

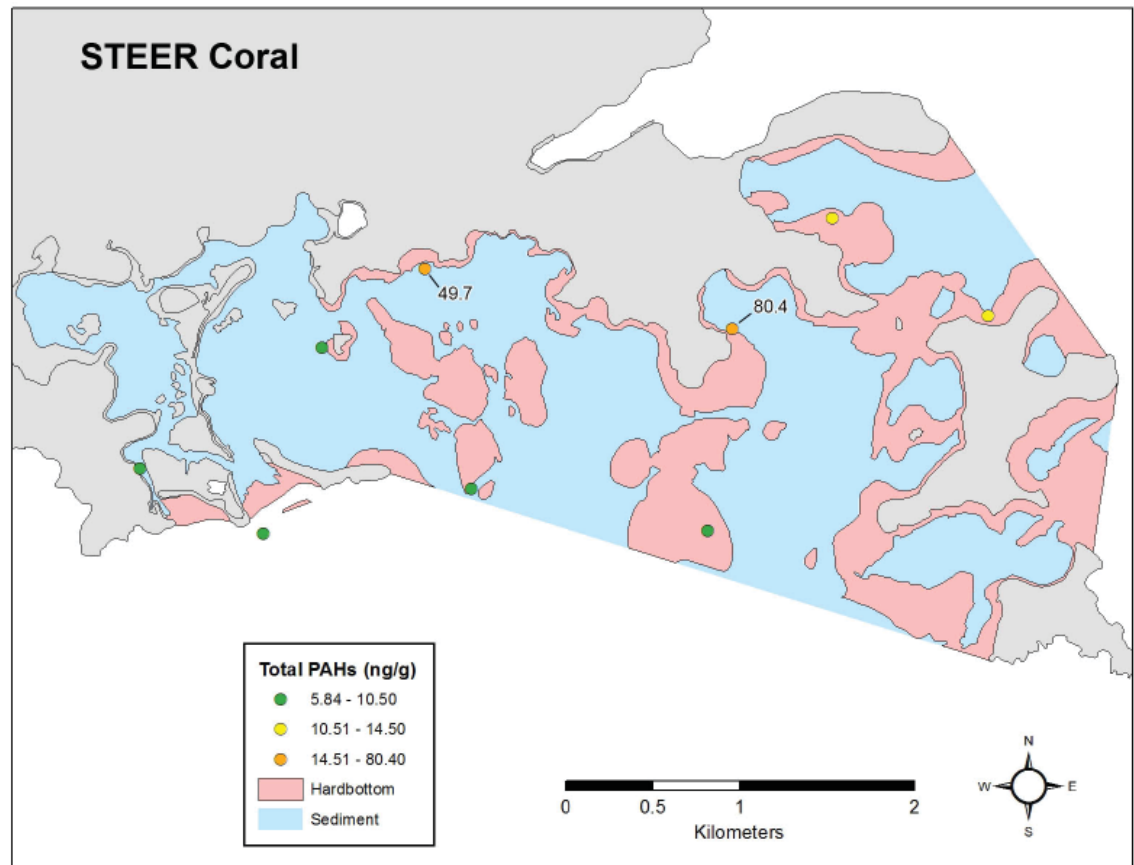


Figure 7.2 Total PAHs detected in the coral *Porites astreoides*.

comparable data. The highest total PAH concentration in coral tissues, 80.4 ng/g was found at the HBI23P site in stratum 3 (Figure 7. 1). Pait *et al.* (2013) calculated a mean total PAH concentration of $46.9 \pm 18.5 \text{ ng/g}$ in *P. astreoides* from southwest Puerto Rico, comparable to corals from both STEER and Vieques, Puerto Rico. Looking at the means of total PAHs across four NCCOS Caribbean studies, the STEER falls above the means of Guanica Bay and Jobos Bay.

Table 7.2. Summary of means, standard error, and maximum values for organic chemical contaminants analyzed in STEER coral, conch and fish.

Contaminant/Class	Coral (ng/g)		Conch (ng/g)		Fish (ng/g)	
	Mean \pm SE	Maximum	Mean \pm SE	Maximum	Mean \pm SE	Maximum
Total PAHs	22.0 ± 8.6	80.4	32.7 ± 9.07	113	15.0 ± 0.97	18.9
Monobutyltin	0.13 ± 0.04	0.29	5.11 ± 2.77	23.2	2.60 ± 0.63	5.39
Dibutyltin	0.03 ± 0.02	0.16	1.07 ± 0.45	4.27	5.47 ± 1.95	14.2
Tributyltin	0.08 ± 0.03	0.29	0.12 ± 0.03	0.38	9.37 ± 5.73	48.7
Total BTs	0.24 ± 0.08	0.74	6.30 ± 3.24	27.9	17.4 ± 7.66	68.3
Total DDT	0.01 ± 0.01	0.08	0	0	2.32 ± 2.02	16.5
Mirex	0	0.04	0	0	0.42 ± 0.11	0.64
Total Chlordane	0.01 ± 0.01	0.05	0	0	1.41 ± 0.44	3.92
Total PCBs	0	0	0	0	65.9 ± 50.1	416

BTs, butyltins; SE. standard error

Table 7.3. PAHs, TBT and total butyltins in coral tissues from NOAA Caribbean studies.

Location	PAHs			TBT			Total Butyltins		
	Mean	SE	Maximum	Mean	SE	Maximum	Mean	SE	Maximum
Guanica Bay, Puerto Rico ^a	4.96	± 0.48	10.1	ND	N/A	ND	ND	N/A	ND
Jobos Bay, Puerto Rico ^b	5.61	± 0.31	8	ND	N/A	ND	ND	N/A	ND
Southwest Puerto Rico ^c	46.9	± 18.5	158.9	ND	N/A	ND	2.62	± 0.25	3.53
Vieques, Puerto Rico ^d	15.0	± 0.64	24.9	0.08	± 0.07	2.36	4.65	± 0.45	9.37
STEER, USVI	22.04	± 8.57	80.4	0.08	± 0.03	0.29	0.24	± 0.08	0.74

^aWhitall *et al.*, 2013; ^bWhitall *et al.*, 2011; ^cPait *et al.*, 2007; ^dPait *et al.*, 2010. TBT, tributyltin; SE, standard error.

Units for PAHs are ng/g for TBT, and total butyltins ng Sn/g.

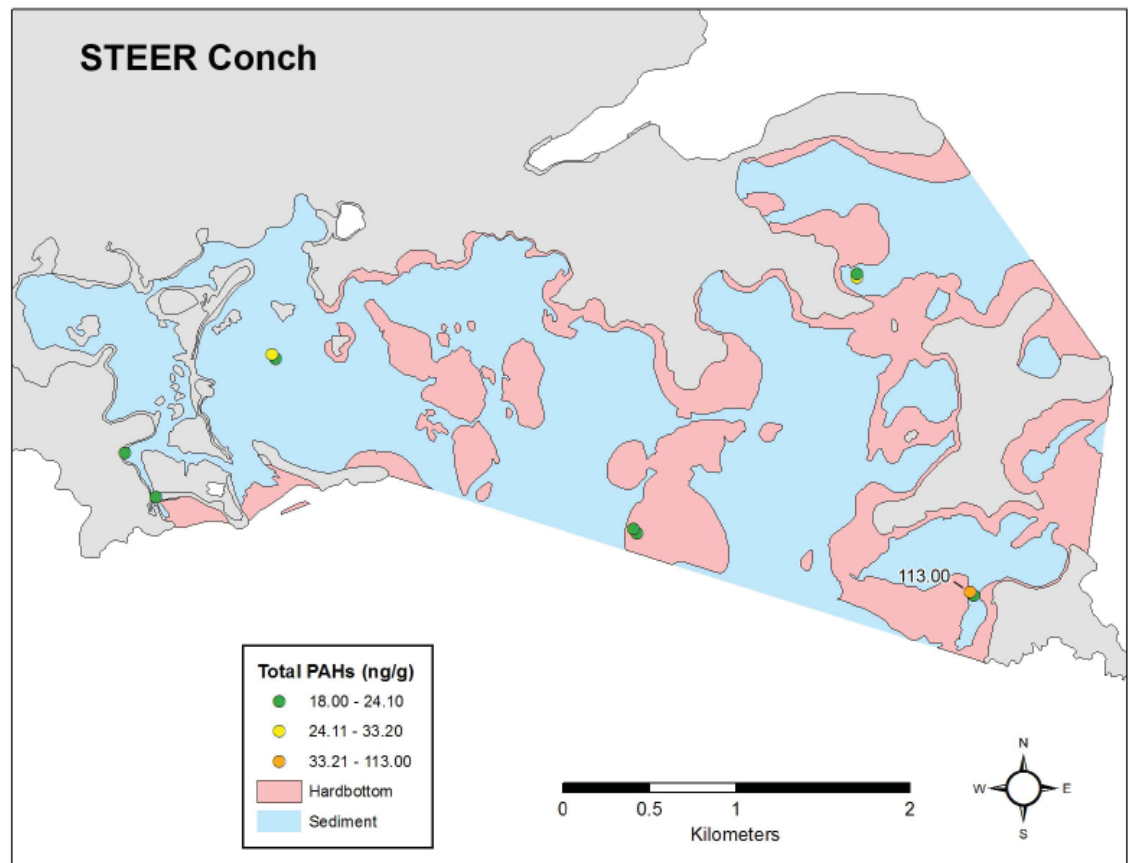
PAHs in Conch. The concentrations of total PAHs found in conch tissues are presented in Figure 7.3. The mean concentration of total PAHs in the tissues of the queen conch (32.7 ± 9.07 ng/g) were similar to those found in corals (22.0 ± 8.6 ng/g) (Table 7.2). In their global survey of mollusk tissues, Vorkamp *et al.* (2010) found that total PAH ranged between 177 and 5,966 ng/g, calculated from 30 PAHs (versus the 59 used in this study). In the STEER, the sum of our 59 PAHs in general fell well below this range, with the highest concentration of total PAHs in *L. gigas* tissues being 113 ng/g at site S4-CB (Table 7.2 and Figure 7.3).

(SVs) for PAHs in fish tissues for recreational fishers (EPA, 2000).

Polychlorinated Biphenyls (PCBs)

PCBs or polychlorinated biphenyls are a group of synthetic compounds that have been used in numerous applications ranging from electrical transformers and capacitors, to hydraulic and heat transfer fluids, to pesticides and in paints. Although no longer manufactured in the U.S., ecosystem contamination by PCBs is widespread due to their environmental persistence and tendency to bioaccumulate.

PAHs in Fish. The mean concentration of total PAHs (Table 7.2) in the tissues of all fish (15.0 ± 0.97 ng/g) were not significantly different (nonparametric Kruskal-Wallis) than those found in sediments (142 ± 59 ng/g), corals (22.04 ± 8.57 ng/g), or conch (32.7 ± 9.07 ng/g). The mean concentration of total PAHs in the tissues of schoolmaster snapper was 16.5 ± 1.16 ng/g while those for longspine squirrelfish were 13.5 ± 1.20 ng/g. None of the values exceeded EPA recommended screening values

Figure 7.3. Total PAHs detected in the conch *Lobatus gigas*.

PCBs in Coral.

No detectable levels of PCBs were found in any of the coral samples from the STEER.

PCBs in Conch.

No detectable levels of PCBs were found in any of the conch samples from the STEER.

PCBs in Fish.

PCBs were detected in all the fish analyzed; the mean concentration was 65.9 ± 50.1 ng/g (Table 7.2).

The highest concentration of total PCBs

in fish tissues in the STEER was found in the fish caught at site SQ-S2 (Figure 7.4). The concentration in fish SQ-S2 (longspine squirrelfish) was 419.2 ng/g (150.9 ng/g wet weight) which is in the top 10th percentile of all National Status and Trends (NS&T) data for PCBs. This sample also exceeded the EPA screening value (SV) for PCBs in fish tissues for recreational fishers for noncarcinogens of 80 ng/g wet weight (EPA, 2000). Exposure to PCBs in fish has been linked to reduced growth, reproductive impairment and vertebral abnormalities (EPA 1997).

Organochlorine Pesticides

A series of man-made chlorine-containing hydrocarbon pesticides (insecticides and herbicides) were developed and used from the 1940s through the 1970s. Organochlorine pesticides are toxic to varying degrees to aquatic life including crayfish, shrimp and fish. One of the best known organochlorine pesticides was the insecticide DDT (dichlorodiphenyltrichloroethane). The use of many of the organochlorine pesticides, including DDT, was banned in the U.S. due to their environmental persistence and toxicity to nontarget species (ATSDR, 2002). Because

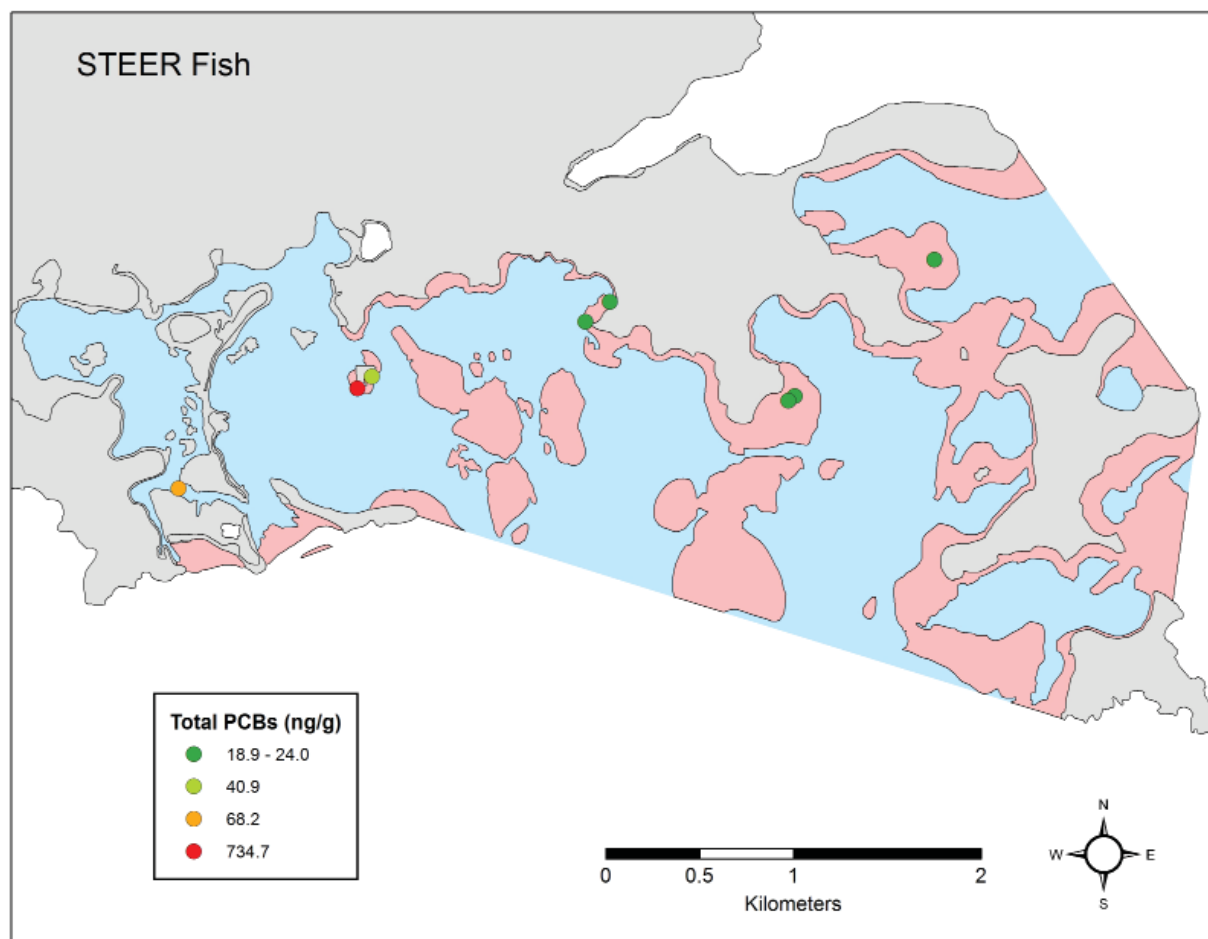


Figure 7.4. Total PCBs detected in fish in the STEER.

of their persistence and heavy use in the past, residues of many organochlorine pesticides can still be found in the environment (Butler, 1973).

DDT in Coral. Only the coral sample from the HBI14P site in stratum 4 (Figure 7.1), had a detectable concentration of both DDT and its metabolites, and only then at a total concentration of 0.08 ng/g. In comparison, the mean concentration of total DDT in coral tissues found in Vieques, Puerto Rico was 0.13 ± 0.07 ng/g. The mean concentration of total DDT in the sediments in the STEER was 0.047 ± 0.025 ng/g (Pait *et al.*, 2013).

DDT in Conch. None of the conch analyzed from the STEER contained detectable levels of total DDT.

DDT in Fish. Only the squirrelfish sample collected at the site SQ-S2 in stratum 2 had a DDT level above the detection limit. The total DDTs concentration in the fish was 16.5 ng/g, which was comprised solely of DDT breakdown products (56.7% 2,4 – DDD, 6.1% 4,4 – DDD, and 37.2% 4,4 – DDE). The absence of the parent DDT compounds (2,4'-DDT or 4,4'-DDT) likely indicates an

older source of the contamination as DDT was phased out for use in the U.S. beginning in 1972.

Other Pesticides. A number of additional chlorinated pesticides were analyzed, however with only one exception (chlordane), there were no detectable concentrations in coral and conch. Total chlordane was detected at HBI14P at a concentration of 0.05 ng/g in coral. In comparison, the mean concentration of total chlordane detected in coral tissues in Vieques, Puerto Rico was 0.12 ± 0.03 ng/g (Pait *et al.*, 2010).

Chlordane was detected in fish tissues in the STEER. The mean total chlordane concentration in fish was 1.41 ± 0.44 ng/g. The mean total chlordane concentration in the tissues of longspine squirrelfish was 1.32 ± 0.87 ng/g, while the mean tissue concentration in schoolmaster snapper was 1.51 ± 0.35 ng/g.

Endosulfan, mirex, and dieldrin were also detected in fish tissues collected from the STEER. Mirex was detected in two squirrelfish samples at sites SQ-S2 (0.64 ng/g) and SQ-S3 (1.11 ng/g). Dieldrin was detected in a squirrelfish from site SQ-S4 (0.53 ng/g) and in a snapper from site SN-S4 (3.58 ng/g). Endosulfan was detected in only one squirrelfish from SQ-S2 (0.69 ng/g).

Butyltins

A class of organometallic compounds, butyltins have had a variety of uses ranging from biocides in antifoulant paints to catalysts and glass coatings (Birchenough *et al.* 2002; Bennett, 1996). In the environment, tributyltin or TBT degrades to dibutyltin (DBT), then monobutyltin (MBT), and finally to inorganic tin. Tetrabutyltin is an intermediate in the manufacture of TBT. Experiments have shown that the half-life of TBT is on the order of days; degradation to monobutyltin takes approximately a month, however in deeper anoxic sediments, the half-life of TBT appears to be on the order of 2-4 years or longer (Batley, 1996).

The presence of TBT has been linked to endocrine disruption, specifically an imposex (females developing male characteristics) condition in marine gastropods, while in other mollusks (e.g., oysters), abnormal shell development, and poor weight gain have been seen (Batley, 1996; Strand *et al.* 2009). TBT has also been shown to have effects on larval growth of fish at concentrations below 100 ng/L in water (Newman and McIntosh, 1991). Beginning in 1989, the use of TBT as an antifouling agent was banned in the U.S. on non-aluminum vessels smaller than 25 meters in length (Gibbs and Bryan, 1996). Because of its widespread use in the past, TBT and its metabolites

continue to be detected in many components of the environment. Recent work by Titley-O'Neil *et al.* (2011) showed that high concentrations of antifouling paint-based butyltin compounds were linked to imposex in conch.

Negri *et al.* (2002) investigated the effects of TBT in sediments on the coral *Acropora microphthalma*. They found that the effective concentration of TBT which caused 50 percent inhibition (EC50) of fertilization after four hours was 200 µg/L, and the concentration needed to inhibit 50 percent larval metamorphosis was only 2 µg/L.

Total Butyltins in Coral. The sum of the three butyltins (total butyltins), was calculated to better visualize total butyltin exposure to corals in the STEER (Figure 7.5). The mean concentration in coral was 0.24 ng Sn/g. The highest concentration of total butyltins (0.74 ng Sn/g) was found in stratum 2 just outside of Benner Bay at site HBI28P. The mean concentrations of total butyltins in coral tissues in the STEER appeared somewhat lower than that detected in southwest and Vieques, Puerto Rico in studies conducted by NOAA (Table 7.3).

Total Butyltins in Conch. The mean concentration of butyltins in conch tissues increased from TBT (0.12 ng Sn/g), to DBT (1.07 ng Sn/g), and the highest being MBT (5.11 ng Sn/g) (Table 7.2). This appears to follow the natural degradation pattern of TBT in the environment (Batley, 1996). The two highest concentrations of total butyltins of 27.9 ng Sn/g and 23.3 ng Sn/g were both found in stratum 2 in Benner Bay (Figure 7.6). This appears to correlate with the elevated concentrations of butyltins found in the sediments of STEER in northern Benner Bay (Pait *et al.*, 2013; Chapters 4 and 5), which has a number of marinas and boatyards.

The two highest total butyltin concentrations are at least seven times higher than the next highest concentration (3.25 ng Sn/g) found in stratum 5. Strand *et al.* (2009) looked at total butyltins in different non-*L. gigas* conch species in the U.S. Virgin Islands. They found a mean concentration of total butyltins in tissues collected from St. Thomas of 104.7 ± 44.8 ng Sn/g, however, none of the samples collected by Strand *et al.* (2009) were from the STEER. In contrast, the mean concentration of total butyltins in *L. gigas* tissues from the STEER in the current study was much lower, 6.30 ng Sn/g ± 3.24 .

Total Butyltins in Fish. The mean concentration of total butyltins in all fish was 17.4 ng Sn/g (Table 7.2). For schoolmaster snapper, the mean concentration of total butyltins was 24.9 ng Sn/g while the mean concentration

of total butyltins for longspine squirrelfish was 10.0 ng Sn/g. The highest concentration of total butyltins (68.3 ng Sn/g) was at site SN-S4.

TBT in Coral. The highest concentration of TBT in *P. astreoides* tissues collected from the STEER was 0.29 ng Sn/g at site HBI28P just outside Benner Bay (Figure 7.7). The mean concentration of TBT in STEER corals was 0.08 ng Sn/g \pm 0.03 (Table 7.2). The average concentration of TBT in the sediments in the STEER found by Pait *et al.* (2013) was 1.85 \pm 1.30 ng Sn/g. The highest concentration of TBT detected in STEER sediments from the same study using a stratified-random sampling design was in Benner Bay, at 31 ng Sn/g. The mean concentration of TBT in coral tissues in the STEER was the same as that found in Vieques, Puerto Rico (Table 7.3). From the NOAA NCCOS studies, only Vieques, Puerto Rico and STEER had detectable TBT in *P. astreoides*. The higher detection of TBT in coral in Benner Bay, may be associated with the elevated concentrations found in the sediments of STEER in this same area (Pait *et al.*, 2013). There was a significant relationship between TBT in coral

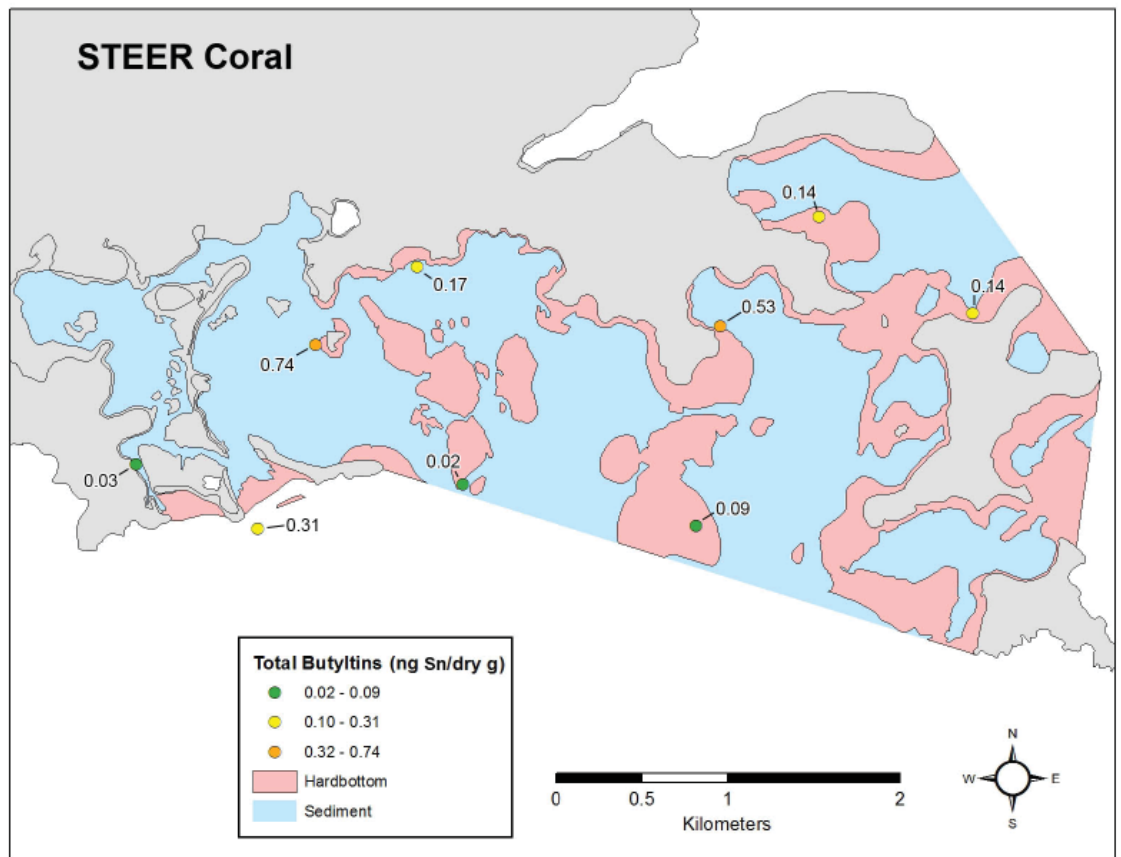


Figure 7.5. Total butyltins detected in the coral *Porites astreoides*.

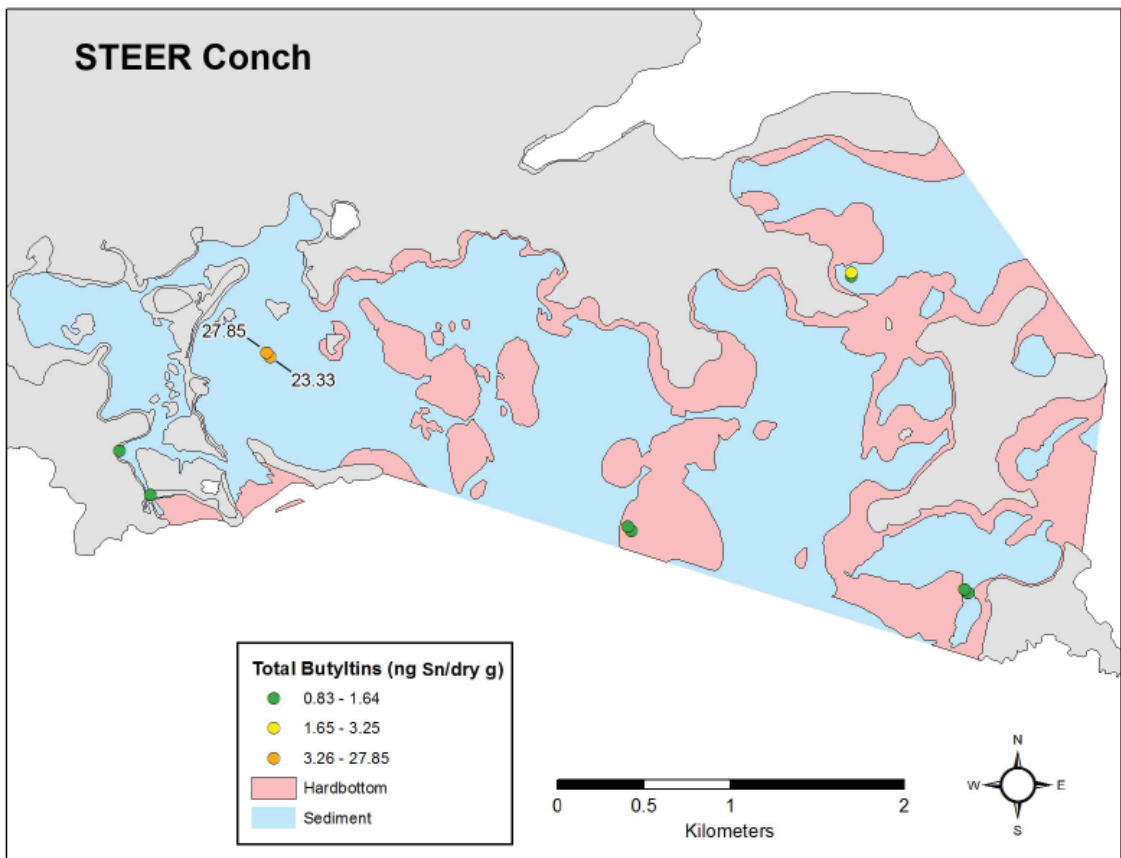


Figure 7.6 Total butyltins detected in the conch *Lobatus gigas*.

tissues in the STEER and a basic inshore vs. off shore designation, with the inshore being significantly higher than the offshore using a nonparametric Wilcoxon's rank sums test ($p = 0.0304$).

TBT in Conch. The highest concentration of TBT in *L. gigas* tissues in the STEER was 0.38 ng Sn/g in stratum 2 at site Conch 2B (Figure 7.8). There was also a significant relationship between TBT in conch tissues in the STEER and a basic inshore vs. offshore designation, using nonparametric Wilcoxon's rank sums ($p = 0.0304$).

TBT in Fish. The highest concentration of TBT was 48.7 ng Sn/g at site SN-S4 (Figure 7.9). The mean concentration of butyltins in all fish tissues decreased from TBT (9.37 ng Sn/g), to DBT (5.47 ng Sn/g), to MBT (2.60 ng Sn/g). This relationship is inverse of that found in conch and may be caused by fish being exposed more recently to TBT. For schoolmaster snapper, the mean concentration of butyltins followed the same pattern, decreasing from TBT (16.95 ng Sn/g), to DBT (5.18 ng Sn/g), to MBT (2.75 ng Sn/g). However, in longspine

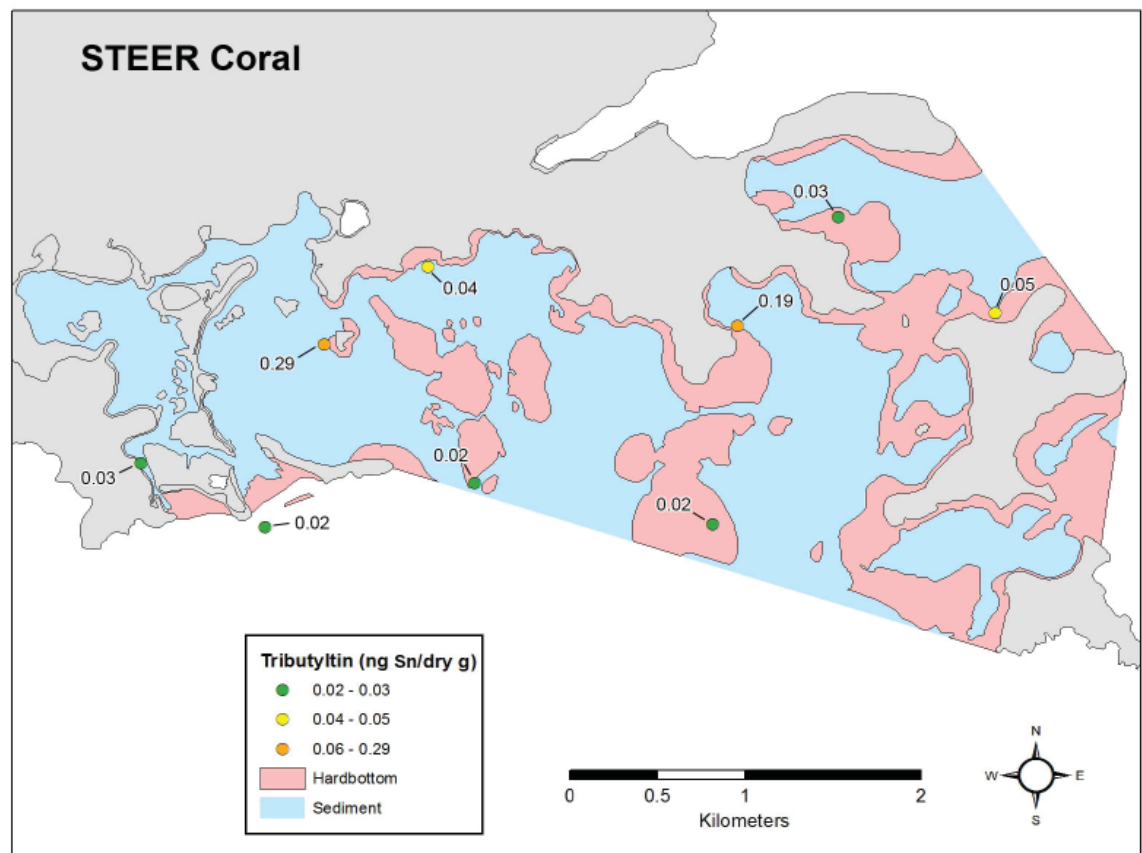


Figure 7.7. Tributyltin detected in the coral *Porites astreoides*.

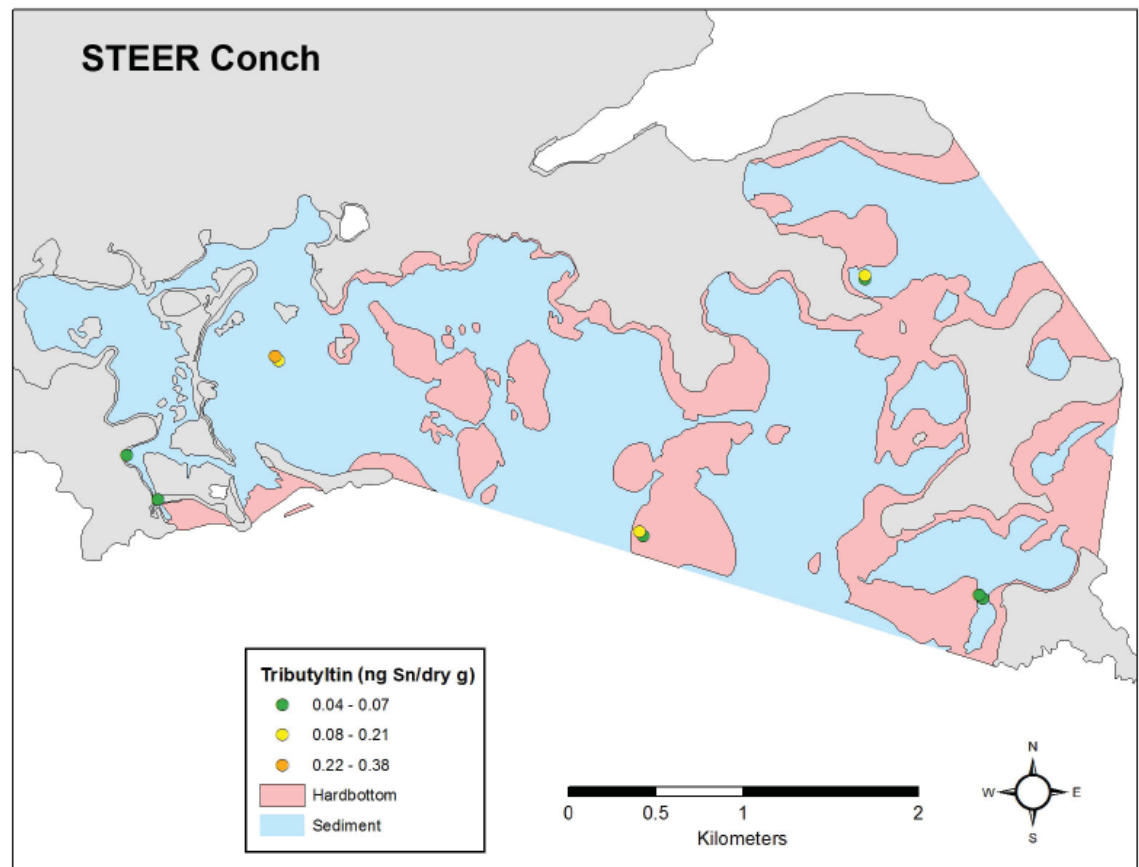


Figure 7.8 Tributyltin detected in the conch *Lobatus gigas*.

squirrelfish the mean for DBT (5.77 ng Sn/g) was higher than MBT (2.46 ng Sn/g), or TBT (1.79 ng Sn/g). TBT is broken down in fish tissues usually on the scale of days to weeks (Newmann and McIntosh, 1991) suggesting the presence of TBT in fish tissues was from a more recent exposure. Figure 7.9 shows the distribution of TBT, DBT, and MBT in fish tissues collected from the STEER.

While there was no statistically significant differences in the concentrations of TBT or any of its metabolites between the two fish species, there was a statistically significant difference (non-parametric Wilcoxon) between the percentage of TBT as compared to total butyltins, with schoolmaster snapper having a significantly higher percentage of TBT in their tissues than longspine squirrelfish. It is not clear why the distribution of TBT, DBT and MBT differed between the two fish species. One possibility is that the longspine squirrelfish are more efficient at metabolizing TBT than the schoolmaster snapper.

Trace and Major Elements

All 14 of the trace and major elements (Table 7.1) analyzed in the samples were detected in coral, conch and fish. Coral tissue body burdens for each element varied broadly from one collection site to another within the STEER. With the exception of arsenic, copper, zinc, lead and the major elements (Al, Fe, Mn), the trace metal concentrations in coral are illustrated in Tables 7.4a and 7.4b. A summary of average concentrations of individual trace and major elements are presented in Figures 7.10 - 7.13, to show the relative abundance of each metal.

Silver

Silver in Corals. Body burdens of silver in *P. astreoides* were relatively low; the maximum value of 0.02 µg/g (Table 7.4a, Figure 7.10) was found in coral from HBI42A site in stratum 5. In coral from various bays in Puerto Rico Pait *et al.*, (2009 and 2010) and Whitall *et al.* (2011), have documented comparable ranges of silver in *P. astreoides* tissues. Because silver was not detected in sediment from the STEER (Pait *et al.*, 2013), the results suggest naturally low silver concentration related to the geological formation of the island.

Silver in Conch. Silver body burdens in conch samples from the STEER varied from 0.16 µg/g to 3.75 µg/g, with a mean value of 0.88 µg/g (Table 7.6, Figure 7.11). Mean

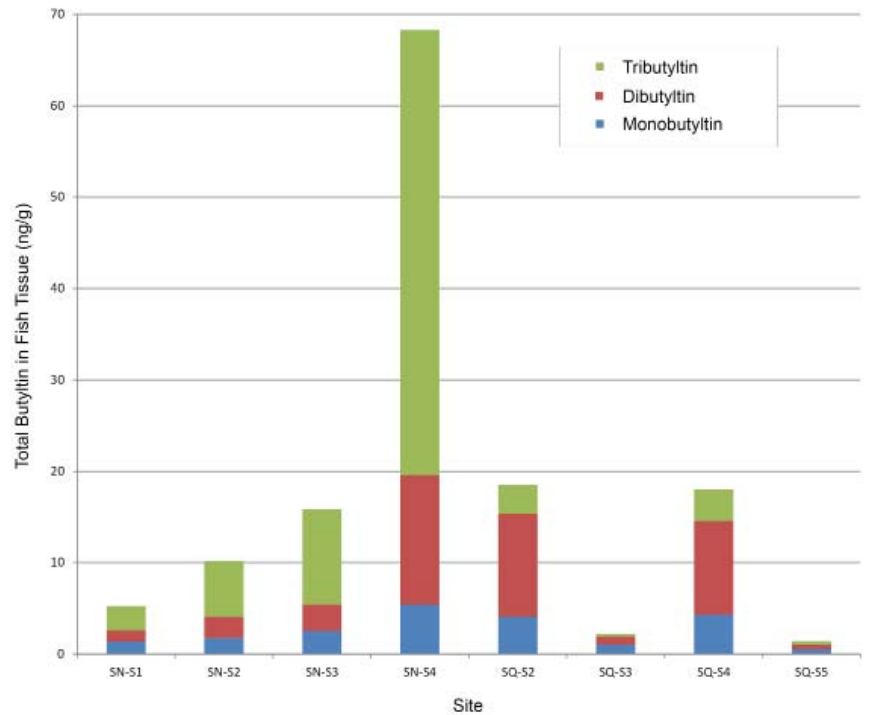


Figure 7.9 Distribution of tributyltin (TBT), dibutyltin (DBT), and monobutyltin (MBT) from fish sampled in the STEER.

values of silver were similar in conch across all strata in the STEER with the exception of stratum 5 where silver was found to be nearly an order of magnitude higher (Table 7.7). Glazer *et al.* (2008) investigated heavy metal concentrations in queen conch from south Florida, and found a mean body burden for silver of 1.03 µg/g in conch from the offshore habitats, and 2.54 µg/g in conch from nearshore habitats. Although causative effects were not established, these authors hypothesized that reduced reproductive fitness of conch in the nearshore habitats in south Florida was due to elevated metal concentrations including silver.

Silver in Fish. Silver was detected at only three of the eight fish sites (Table 7.8). The maximum concentration of silver in any fish tissue was only 0.04 µg/g observed in snappers from SN-S1 site (Table 7.8). The mean concentration of silver for all fish was 0.01 ± 0.005 µg/g (0.01 ± 0.009 µg/g in schoolmaster snapper, 0.01 ± 0.005 µg/g in longspine squirrelfish). The mean concentration of silver in fish tissues from the STEER were similar to the mean concentration of silver in fish tissues from Vieques, Puerto Rico (Table 7.9).

Aluminum

Aluminum in Corals. In the STEER, *P. astreoides* had aluminum body burdens ranging from zero to a maximum

Table 7.4a. Summary statistics for contaminants (µg/g) in coral tissue, including comparison with other studies.

Species	Location	Value	Ag	Al	As	Cd	Cr	Cu	Fe	Reference
<i>Porites astreoides</i>	STEER	Maximum	0.0202	201	1.76	0.0467	1.52	4.07	275	This study
<i>Porites astreoides</i>	STEER	Mean	0.01293	22.33333	1.255	0.011867	0.389111	2.692222	72.31111	This study
<i>Porites astreoides</i>	STEER	Minimum	0	0	0.611	0	0	1.96	27.9	This study
<i>Porites astreoides</i>	Southwest Puerto Rico	Mean	0	37.8	0	0	0	2.06	90.8	Pait et al., 2009
<i>Porites astreoides</i>	Vieques, Puerto Rico	Mean	0.013	30.75	0.241	0.194	0.183	0.757	51.2	Pait et al., 2010
<i>Porites astreoides</i>	Jobos Bay, Puerto Rico	Range	0	100-333	0.94-2.44	0.21-0.31	0	2.37-97.2	110-480	Whitall et al., 2011
<i>Porites astreoides</i>	Punta brava, Venezuela	Range							1.32-369	Bastidas and Garcia, 1999
	Bajo Caiman, Venezuela	Range							nd-88.7	
<i>Porites sp.</i>	Misima Island, Papua NG									Fallon et al., 2002
<i>Porites lobata</i>	Ulan Reef, Philippines	Mean						3.1		David, 2003
<i>Porites sp.</i>	Dafangji Island, China									Peng et al., 2006
<i>Porites sp.</i>	Daya Bay, China	Range							41.4-226.4	Chen et al., 2010

Table 7.4b. Summary statistics for contaminants (µg/g) in coral tissue, including comparison with other studies.

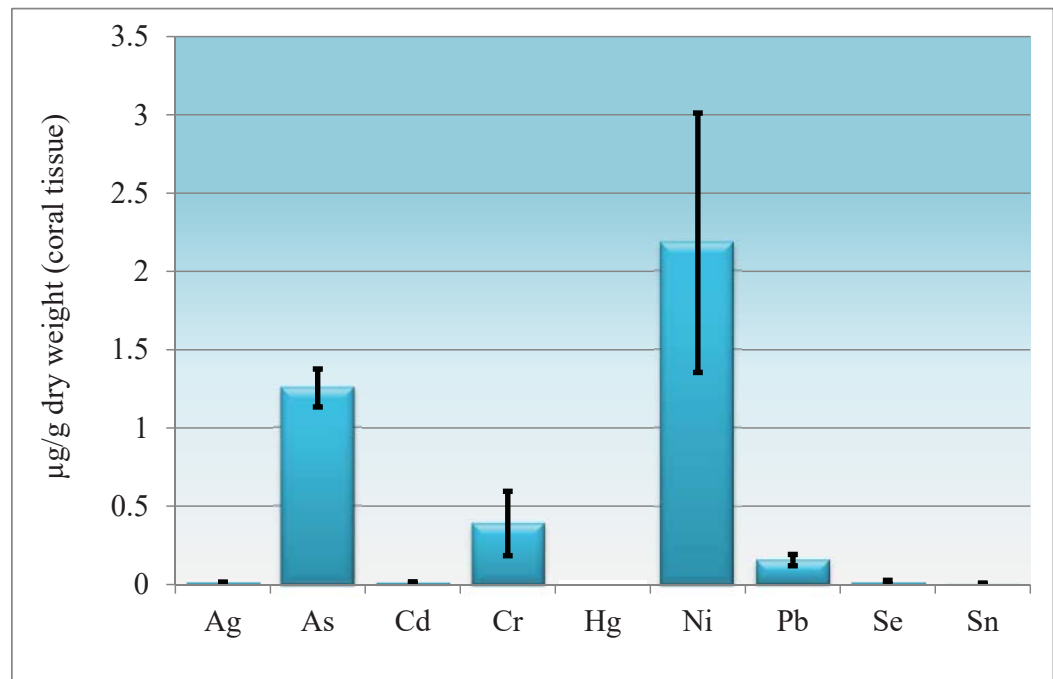
Species	Location	Value	Hg	Mn	Ni	Pb	Se	Sn	Zn	Reference
<i>Porites astreoides</i>	STEER	Maximum	0.003	19.6	8.33	0.42	0.116	0.0393	14.8	This study
<i>Porites astreoides</i>	STEER	Mean	0.001	10.1	2.18	0.16	0.012889	0.004367	5.983333	This study
<i>Porites astreoides</i>	STEER	Minimum	0	7.25	0	0.07	0	0	1.87	This study
<i>Porites astreoides</i>	Southwest Puerto Rico	Mean	0	3.01	1.32	0	0.05	0.02	6.09	Pait et al., 2009
<i>Porites astreoides</i>	Vieques, Puerto Rico	Mean		2.66	0.9	0.07	0.096	0.246	3.43	Pait et al., 2010
<i>Porites astreoides</i>	Jobos Bay, Puerto Rico	Range	0.001-0.004	8.33-24.6	0.8-6.84	0.08-12.5	0.13-0.26	nd-0.10	2.56-16.9	Whitall et al., 2011
<i>Porites astreoides</i>	Punta brava, Venezuela	Range							3.59-42.5	Bastidas and Garcia, 1999
	Bajo Caiman, Venezuela	Range							0.83-23.1	
<i>Porites sp.</i>	Misima Island, Papua NG			0.19-1.6					0.68-36.5	Fallon et al., 2002
<i>Porites lobata</i>	Ulan Reef, Philippines	Mean		1					1.8	David, 2003
<i>Porites sp.</i>	Dafangji Island, China			2.76-6.85					4.2-55.1	Peng et al., 2006
<i>Porites sp.</i>	Daya Bay, China	Range		0.79-5.38					0.02-22.3	Chen et al., 2010

value of 201 µg/g (Table 7.4a). Figure 7.12 shows the overall mean of aluminum in coral tissue, however, the results indicated that aluminum concentrations were below the detection limit at most of the sampling locations in the STEER except at HBI28P where the maximum value was detected. Pait *et al.*

(2010) and Whitall *et al.* (2011) have respectively reported maximum values of 37 µg/g in coral from Vieques, Puerto Rico and 333 µg/g in Jobos Bay, Puerto Rico indicating that concentrations found in the STEER were within the range of aluminum concentration in coral from the Caribbean.

Aluminum in Conch. In *L. gigas*, aluminum ranged from 38.5 to 828 µg/g (Table 7.6). The highest concentration of aluminum occurred in one of the conch collected from stratum 4 (Table 7.7).

Aluminum in Fish. Aluminum concentrations in the tissue of all fish in the STEER ranged from 1.67 µg/g to 14.4 µg/g (Table 7.8) with a mean of 6.32 ± 2.03 µg/g. The highest concentration of aluminum was found in snappers at site SN-S3. Similar ranges of aluminum concentrations were

Figure 7.10. Concentrations (mean ± SE) of metals detected in the coral *Porites astreoides*.

found in fish tissues from Vieques, Puerto Rico (Table 7.9).

Arsenic

Arsenic in Corals. Arsenic body burdens in *P. astreoides* varied from 0.61 µg/g to 1.76 µg/g. Table 7.4a indicates that the overall range of arsenic concentrations were similar to those reported elsewhere in southwest Puerto Rico and Jobos Bay, Puerto Rico (Pait *et al.*, 2009 and Whittall *et al.*, 2011). Sublethal thresholds for arsenic in coral have not been established, but Pichler *et al.* (1999) found that coral in Ambide Island, Papua New Guinea, exposed to elevated concentrations of arsenic in seawater from hydrothermal vents did not show any obvious toxic effects.

Arsenic in Conch. Arsenic was detected in the tissue of all *L. gigas* collected in the STEER. Body burdens varied from 17.6 µg/g to 67.6 µg/g (Table 7.6). The highest concentrations in conch were found in stratum 4 (Table 7.7). With a mean value of 32.7 µg/g, arsenic had the second highest concentration of the trace metals measured in conch after copper (Figure 7.11). Said *et al.* (2013) assessed elemental concentrations including arsenic in the conch *S. canarium* in the western region of Johor Straits, Malaysia, and reported an arsenic value of 0.125 µg/g wet weight (Table 7.6). Using the average 76% moisture content measured in the conch from the STEER, we derived an equivalence maximum concentration of 16.9 µg/g wet weight (ww) and minimum of 8.2 µg/g ww of arsenic in *L. gigas*. The sublethal threshold toxicity of arsenic for conch is unknown at this time. For human protection however, the U.S. FDA (FDA, 2009) has set the maximum permissible action level of 86 µg/g arsenic wet weight (ww) in shellfish. The derived wet weight equivalence of the maximum arsenic concentration value found

Table 7.5. Mean metal body burden (µg/dry g) in coral by stratum.

Element	Stratum 1	Stratum 2	Stratum 3	Stratum 4	Stratum 5
Ag	0.0138	0.0134	0.0131	0.0169	0.0134
Al	0	201	0	0	0
As	0.611	1.76	1.57	0.984	1.11
Cd	0	0	0	0.0177	0.0205
Cr	0	0	0	0	1.52
Cu	2.37	4.07	2.06	2.44	2.77
Fe	54.7	275	82.6	32.3	42
Hg	0	0.00227	0	0	0
Mn	8	19.6	11.8	8.07	7.25
Ni	1.58	2.3	0.954	1.78	2.28
Pb	0.111	0.415	0.217	0.0693	0.0741
Se	0	0	0.116	0	0
Sn	0	0.0393	0	0	0
Zn	2.04	14.8	10.2	1.87	2.38

n = 2 for each stratum except for stratum 1 (only one site located).

in conch from the STEER (16.9 µg/g ww) was low relative to the FDA criterion.

Arsenic in Fish. Arsenic was also detected in all tissue samples of both fish species with concentrations ranging from 4.7 µg/g to 27.9 µg/g (Table 7.8). The highest concentration was found at site SQ-S2 just outside of Benner Bay. The mean concentrations of arsenic for both

Table 7.6. Summary statistics for contaminants (µg/g) in conch tissue (n=10), including comparison with mean concentration values derived from study of conch contamination in South Florida (Glazer *et al.*, 2008) and Johor Straits, Malaysia (Said *et al.*, 2013).

Metal	STEER			Glazer, 2008 (ww)		Said et al., 2013 (ww) Johor Straits, Malaysia
	Mean	Min. Conc.	Max. Conc.	PS (offshore)	TI (nearshore)	
Ag	0.88	0.16	3.75	1.03 (1.4)	2.54 (3.4)	0.125 (0.17) 0.01 (0.01)
As	32.7	17.6	67.6			
Cd	1.96	0.89	3.75	2.62 (3.5)	24.14 (31.9)	
Hg	0.24	0.05	0.88	0.01 (0.01)	0.00 (0)	
Al	229	38.5	828			
Cr	3.41	1.45	8.57			1.36 (1.8)
Fe	785	284	1720			
Mn	113	40	355			
Ni	5.79	3.03	13.6	16.28 (21.53)	9.59 (12.7)	
Zn	484	170	1320	30.53 (40.39)	660.32 (873.6)	
Cu	84.7	36	122	14.06 (18.6)	84.34 (111.6)	
Pb	0.61	0.21	1.32			
Se	1.19	0.68	2.38			
Sn	5	0.03	12.7	0.00 (0)	0.00 (0)	

Min. Conc., minimum concentration; Max. Conc., maximum concentration; TI, Tinger Island; PS = Pelican Shoal. Values in parentheses are dry weight equivalence of the wet weight concentrations; ww, wet weight.

fish species was $11.8 \pm 2.8 \mu\text{g/g}$ ($9.7 \pm 2.7 \mu\text{g/g}$ for schoolmaster snapper, $13.8 \pm 5.1 \mu\text{g/g}$ for longspine squirrelfish). The mean concentrations of arsenic in fish tissues from the STEER were similar to the mean concentration of arsenic found in fish tissues from Vieques, Puerto Rico (Table 7.9).

Using the laboratory recorded percent dry and percent moisture for each sample, a wet weight concentration of arsenic for each sample was calculated for comparison to

EPA's recreational fishers recommended screening values (SVs) (EPA, 2000). Based on the SV of $1.2 \mu\text{g/g}$ inorganic arsenic, all of the STEER samples exceeded the threshold by at least $0.15 \mu\text{g/g}$. However, it is important to note that our numbers represent total arsenic, including both organic and inorganic fractions. According to the EPA (2012), inorganic arsenic makes up only 2% of total arsenic in marine organisms.

Given that 2%, our inorganic arsenic concentrations in wet weight would range from $0.027 \mu\text{g/g}$ to $0.185 \mu\text{g/g}$ with a mean concentration of $0.079 \pm 0.051 \mu\text{g/g}$ wet weight (schoolmaster snapper $0.054 \pm 0.028 \mu\text{g/g}$, longspine squirrelfish $0.104 \pm 0.061 \mu\text{g/g}$). As such all of the fish samples from the STEER would fall below the EPA SV of $1.2 \mu\text{g/g}$ wet weight.

Cadmium

Cadmium in Corals. Cadmium concentrations in coral tissues ranged from zero to a maximum of $0.047 \mu\text{g/g}$ (Table 7.4a), in stratum 5, at HBI42A. As with the other trace metals, the coral body burdens of cadmium were low (Figure 7.10). Cadmium body burdens in coral from

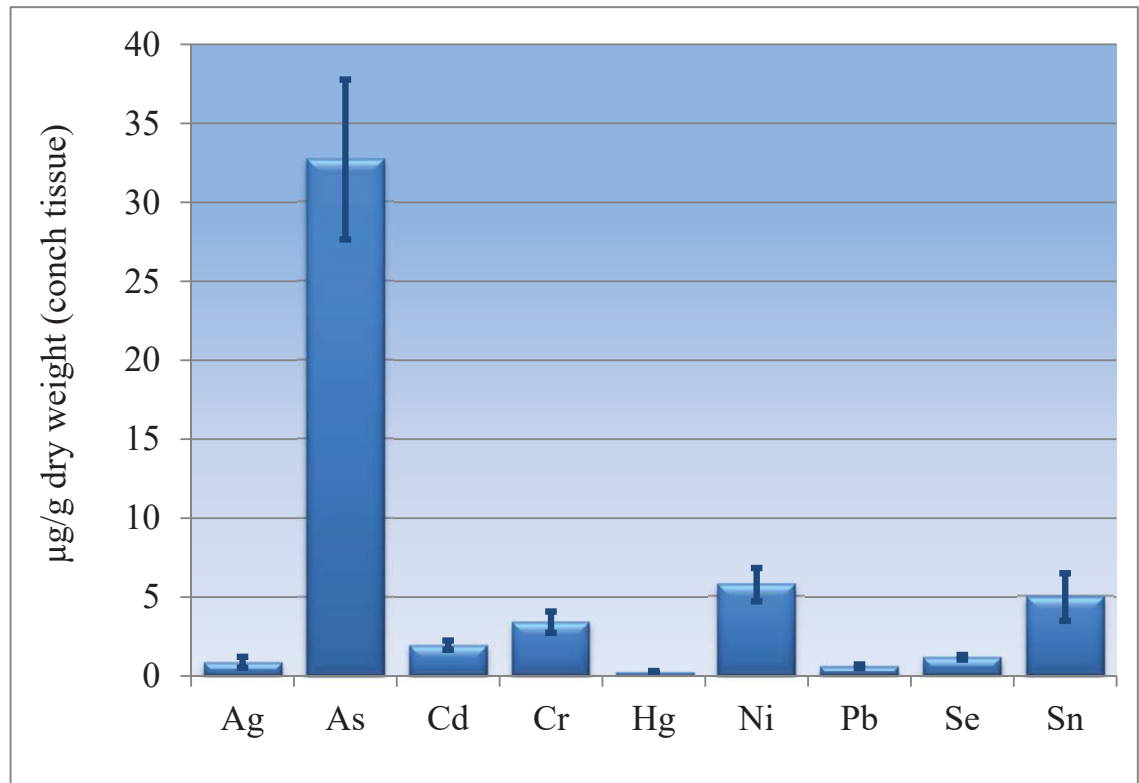


Figure 7.11. Concentrations (mean \pm SE) of metals detected in the conch *Lobatus gigas*.

the STEER were virtually an order of magnitude lower than concentrations reported by Whitall *et al.* (2011) in Jobos Bay, Puerto Rico (Table 7.4a). These authors linked the high cadmium concentrations observed in coral from Jobos Bay to elevated concentrations observed in bed sediments in the vicinity of the reef. Laboratory studies

Table 7.7. Mean metal body burden in conch collected from the five strata. in the STEER ($\mu\text{g/g}$).

Element	Stratum 1	Stratum 2	Stratum 3	Stratum 4	Stratum 5
Ag	0.238	0.172	0.8755	0.985	2.116
Al	113	141.5	38.65	767.5	77.1
As	25.3	19.9	32.25	51.4	38.1
Cd	1.45	1.044	2.66	2.725	1.745
Cr	3.03	1.81	3.085	6.33	3.16
Cu	48.8	113	62.2	114.5	85.5
Fe	783	776	389	1625	380
Hg	0.054	0.097	0.298	0.196	0.549
Mn	55.8	258	86.2	103.9	69.55
Ni	5.18	6.06	4.08	11.045	3.545
Pb	0.424	1.26	0.29	0.7415	0.3305
Se	0.758	0.962	1.22	1.19	1.845
Sn	1.74	4.75	12.25	4.011	0.03765
Zn	307	920.5	670	235.5	288.5

n = 2 from each stratum.

have shown that cadmium can affect coral metabolic processes by inhibiting photosynthetic electron transport in symbiotic zooxanthellae (Kuzminov *et al.*, 2013). Additionally, cadmium has been shown to impair development and reproduction in several invertebrate species including coral (Eisler, 1985; Mitchelmore *et al.*, 2007).

Cadmium in Conch.

Cadmium body burdens in conch tissues ranged from 0.89 µg/g to a maximum of 3.75 µg/g (Table 7.6) which was found in stratum 3. The results showed little variation between strata for cadmium body burden in conch relative to the STEER-wide mean value of 1.96 µg/g (Table 7.7; Figure 7.11). Cadmium body burdens found in the STEER were similar to concentrations reported by Glazer *et al.* (2008) in *L. gigas* collected from the offshore environment in south Florida. However, the same authors reported concentrations of 31.9 µg/g in conch collected from nearshore environments in Florida (Table 7.6). Cadmium's toxicity to aquatic organisms is well documented (Lin and Dunson, 1993; Omer *et al.*, 2012). However, threshold guidelines for cadmium sublethal effects in conch are unknown at this time. The FDA action level for cadmium in molluscan shellfish is 4 µg/g wet weight. Using the measured 76% moisture content in conch, we derived an equivalence value of 0.94 µg/g (ww) for cadmium in STEER conch. The concentrations of cadmium in conch from the STEER were below the FDA threshold.

Cadmium in Fish. Cadmium was detected in all fish tissues collected in the STEER and ranged from 0.017 µg/g up to 0.047 µg/g. The highest concentration of cadmium was detected at site SQ-S3. The mean concentration of cadmium for all fish in the STEER was 0.028 ± 0.004 µg/g (0.023 ± 0.003 µg/g in schoolmaster snapper, 0.034 ± 0.008 µg/g in longspine squirrelfish) (Table 7.8). Using the laboratory recorded percent dry and percent moisture for each sample, a wet weight concentration of cadmium for each sample was calculated for comparison to EPA's recreational fishers recommended screening value (SV) (EPA, 2000). Based on the SV of 4.0 µg/g, all detections of

Table 7.8. Trace and major element concentrations (µg/g) in fish from the STEER.

Element	All Fish	Schoolmaster Snapper			Longspine Squirrelfish		
	Mean	Mean	Minimum	Maximum	Mean	Minimum	Maximum
Ag	0.0084	0.0087	0	0.0349	0.0080	0	0.0174
As	11.76	9.72	4.70	17.00	13.80	6.34	27.90
Cd	0.0283	0.0226	0.0178	0.0318	0.0339	0.0173	0.0472
Hg	0.2548	0.2953	0.1970	0.4660	0.2144	0.0496	0.5020
Al	6.32	5.40	1.67	14.40	7.24	1.64	13.50
Cr	20.1	24.6	11.1	35.2	15.5	11.5	18.1
Fe	177.1	203.5	119	276	150.8	131	192
Mn	2.52	2.73	2.16	3.48	2.32	1.94	2.77
Ni	2.575	3.758	0.911	10.100	1.392	0.966	1.880
Zn	65.8	40.8	35.8	43.8	90.8	25.3	217.0
Cu	3.07	2.32	1.79	2.96	3.83	1.86	6.87
Pb	0.0865	0.0482	0	0.0653	0.1247	0.0590	0.2600
Se	2.69	1.71	1.53	2.11	3.67	1.18	9.84
Sn	0.0436	0.0511	0.0231	0.0930	0.0360	0	0.1040

Note - all values in dry weight; whole fish analysis.

cadmium were two to three orders of magnitude below the SV.

Cadmium concentrations in fish tissues in the STEER were an order of magnitude lower than those found in the sediments. Cadmium concentrations in fish and corals were two orders of magnitude lower than those observed in conch in the STEER. The mean concentration of cadmium in fish tissues in the STEER were similar to the mean

Table 7.9. Comparison of average elemental composition in fish tissues (µg/g) in STEER and Vieques.

Element	STEER* (NOAA)	Vieques (ATSDR)
Aluminum (Al)	2.15	7.66
Arsenic (As)	3.95	3.95
Cadmium (Cd)	0.01	0.08
Chromium (Cr)	6.62	0.16
Copper (Cu)	1.07	0.56
Iron (Fe)	58.8	6.81
Lead (Pb)	0.04	0.27
Manganese (Mn)	0.85	0.33
Mercury (Hg)	0.08	0.05
Nickel (Ni)	0.81	0.81
Selenium (Se)	0.91	0.98
Silver (Ag)	0.01	0.08
Zinc (Zn)	22.7	3.13

*Note - STEER means converted to approximate wet weight concentrations.

concentration of cadmium found in fish tissues in Vieques, Puerto Rico (Table 7.9).

Chromium

Chromium in Corals. The average concentration of chromium in *P. astreoides* was 0.39 µg/g (Table 7.4a), and ranged from zero to a maximum of 1.52 µg/g, at sampling location HBI7P in stratum 5. Mean chromium body burdens in coral tissue from the STEER were similar to the mean value reported in coral from Puerto Rico (Table 7.4a), although the maximum level found at HBI7P was nearly an order of magnitude higher than concentrations found in Vieques, Puerto Rico.

Chromium in Conch. Summary statistics for the concentrations of chromium in *L. gigas* from the STEER are presented in Table 7.6. Mean chromium concentrations in conch varied from 1.81 µg/g to a maximum value of 6.33 µg/g. Relative to the STEER-wide mean concentration of 3.41 µg/g, chromium appeared to be fairly well distributed across the strata in the STEER except in Stratum 4 where the maximum values were measured (Table 7.7).

Chromium effects in conch are unknown. To limit human exposure to chromium through seafood consumption, the U.S. FDA (FDA, 2009) has set a chromium action level in molluscan shellfish at 2.14 µg/g wet wt. Using the measured 76% moisture content in conch, we derived an FDA equivalence value of 17.2 µg/g chromium (dry weight) in mollusks. Levels of chromium found in conch tissue from the STEER (maximum of 8.57 µg/g) are below the FDA action levels. Chromium has been shown to reduce survival and fecundity in the cladoceran *Daphnia magna*, and result in reduced growth in fingerling chinook salmon (*Oncorhynchus tshawytscha*) (Eisler, 1986).

Chromium in Fish. Chromium was detected in all fish samples from the STEER and ranged from 11.5 µg/g to 35.2 µg/g with a mean concentration of 21.4 ± 3.04 µg/g (schoolmaster snapper 24.6 ± 5.92 µg/g, longspine squirrelfish 15.5 ± 1.49 µg/g) (Table 7.8). The highest concentration of chromium was detected at site SN-S3.

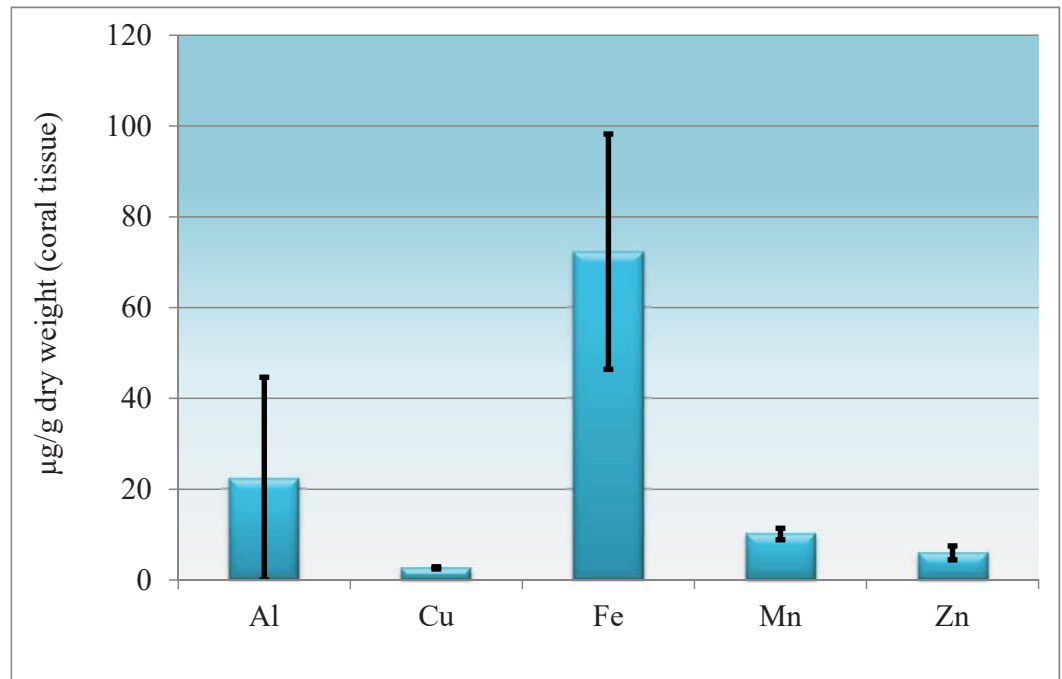


Figure 7.12. Concentrations (mean \pm SE) of metals detected in the coral *Porites astreoides*.

The mean concentration of chromium in fish tissues in the STEER were an order of magnitude higher than the mean concentration of chromium found in fish tissues in Vieques, Puerto Rico (Table 7.9). Compared to sediment concentrations in the STEER chromium in fish tissues are similar in range.

Copper

Copper in Corals. Copper body burdens in *P. astreoides* ranged from 1.96 µg/g to 4.07 µg/g, with a mean of 2.69 µg/g (Table 7.4a and Figure 7.12). Like a number of the other metals measured, the highest copper concentration was found at HBI28P within stratum 2 in Benner Bay (Figure 7.1). A number of metals (e.g. Al, Fe, Mg, As, Cu, and Zn) were more elevated in the coral at this site than coral from other sites, and this may be related to inputs from point and nonpoint sources in the area, including runoff from terrestrial areas (e.g., roads and boatyard activities), and the resuspension of sediments as a result of boat traffic.

Copper found in *P. astreoides* from the STEER were in the range of concentrations found in southwest and in Vieques, Puerto Rico (Table 7.4a). However, in *P. astreoides* from Jobos Bay, Puerto Rico, Whitall *et al.* (2011) reported a higher median value, 69 µg/g. These authors suggested there have been copper contamination problems in Jobos Bay.

The toxicity of copper to corals is well demonstrated (Downs *et al.*, 2005; Reichelt-Brushett and Harrison, 1999,

Goh and Chou, 1997; Reichelt-Brushett and Michalek-Wagner, 2005). Downs *et al.* (2005) showed that copper as cuprous oxide affected cell vitality and mitochondrial function. Reichelt-Brushett and Michalek-Wagner (2005) investigated the effects of copper on the soft coral *Lobophytum compactum*. A significant reduction in fertilization success was found at a copper concentration of 117 µg/L. Also, in corals, Reichelt-Brushett and Harrison (2004) found that a copper concentration of 20 µg/L significantly reduced fertilization success in brain coral *Goniastrea aspera*. Goh and Chou (1997) found that a copper concentration of 40 µg/L in the zooxanthellae *Symbiodinium microadriaticum*, isolated from the rice coral *Montipora verrucosa* resulted in growth inhibition in the symbiotic dinoflagellate. Goh and Chou (1997) noted a synergistic effect when the zooxanthellae were exposed to both copper and zinc.

Copper in Conch. Copper body burdens in *L. gigas* from the STEER ranged from 36 µg/g to 122 µg/g with a mean of 84.7 µg/g (Table 7.6 and Figure 7.13). Mean concentration of copper found in conch from the STEER were similar to concentrations found in conch from nearshore environments in south Florida (Table 7.6). However, the mean copper value measured in conch from the STEER was elevated relative to published values by Glazer *et al.* (2008) and Said *et al.* (2013), in conch from offshore environments of south Florida and Johor Straits, Malaysia (Table 7.6), respectively.

While an essential element especially for mollusks, which use copper as the oxygen carrier in their blood, elevated concentrations of copper can impact aquatic organisms, including the functioning of gills, along with reproduction and development in fish and mollusks (Eisler, 1998a; Spade *et al.*, 2010). Spade *et al.* (2010) found copper concentrations of 34.8 µg/g ww (46 µg/g dw) and 84.0 µg/g (111 µg/g dw) respectively in the testis and digestive gland of conch from south Florida, and speculated that copper may be contributing to testis regression, and hence to reproductive failure of the conch in the nearshore environment of south Florida. Note, our data is whole body burden, and not tissue specific.

Copper in Fish. Copper was detected in all fish tissues from the STEER with body burdens ranging from 1.79 µg/g to 6.87 µg/g and a mean of 3.07 ± 1.66 µg/g (schoolmaster snapper 2.32 ± 0.27 µg/g, longspine squirrelfish 3.83 ± 1.08 µg/g) (Table 7.8). The highest concentration of copper was found in the squirrelfish caught at site SQ-S2. The mean concentration of copper in fish tissues from the STEER was

an order of magnitude higher than the mean concentration of copper in fish tissues from Vieques, Puerto Rico (Table 7.9). However, in the STEER, the mean concentration of copper in conch tissues was elevated relative to mean copper concentration found in fish tissues.

As mentioned above, copper can impact the functioning of gills in fish and mollusks (Eisler, 1998a; Spade *et al.*, 2010). Reduced hatching rates in seabass *Dicentrarchus labrax* has also been seen with concentrations of copper as low as 5 µg/L in seawater (Newman and McIntosh, 1991). Overall in the STEER, less copper was found in fish and coral tissues relative to copper concentrations found in conch tissue (non-parametric Kruskal-Wallis test, p-value = <0.0001).

Iron

Iron in Coral. The major element iron ranged from a minimum of 27.9 µg/g to a maximum of 275 µg/g in *P. astreoides* (Table 7.4a). Similar to copper, the maximum iron body burden in coral was found at the HBI28P in stratum 2. The STEER-wide mean of iron of 72.3 µg/g indicates that iron is the most abundant metal in coral from the STEER (Figure 7.12).

Similar coral body burden ranges of iron were reported in southwest Puerto Rico, Vieques, Puerto Rico and in Daya Bay, China (Table 7.4a). Like aluminum and manganese, iron is regarded as a marker of metals from terrestrial sources (Chen *et al.* 2010). However, there was no significant correlation between iron and other trace metals to infer any reasonable source identification aside from that of natural sources.

Iron in Conch. Iron detected in conch ranged from 284 to 1,720 µg/g, with a mean of 785 µg/g (Table 7.6). As with coral, iron had the highest mean concentration of any element measured in conch in this study.

Iron in Fish. The range of iron detected in fish tissues in the STEER ranged from 119 µg/g to 276 µg/g with a mean of 177 ± 21 µg/g (schoolmaster snapper 204 ± 36 µg/g, longspine squirrelfish 151 ± 14 µg/g) (Table 7.8). The highest concentration of iron measured in fish in the STEER was found in the snapper caught at site SN-S1. As with coral and conch, iron was the most abundant metal measured in fish tissues. The mean iron concentration in fish tissues from the STEER was an order of magnitude higher than the mean concentration of iron from fish tissues in Vieques, Puerto Rico (Table 7.9).

Mercury

Mercury in Coral.

Mercury was detected at low concentrations in *P. astreoides* (Figure 7.10) in the STEER with concentrations ranging from zero to 0.003 $\mu\text{g/g}$ (Table 7.4b). Similar concentration ranges were reported for mercury in coral from Puerto Rico (Table 7.4b). The ecotoxicity of mercury includes neurological effects in vertebrates, which are well documented (Murphy *et al.* 2008). Bastidas and Garcia (2004) found reduced zooxanthellae density in *Porites astreoides* exposed to mercury as well reduced levels of symbiotic algae either through death or expulsion.

Mercury in Conch. Mercury body burdens in conch were elevated in the STEER (ranging from 0.05 $\mu\text{g/g}$ to 0.88 $\mu\text{g/g}$) relatively to other studies (Table 7.6). Mercury was detected in all conch specimens collected. With a STEER-wide mean of 0.24 $\mu\text{g/g}$, conch in the STEER appeared to have a slightly higher mercury tissue content than conch from south Florida, which averaged 0.01 $\mu\text{g/g}$ in a study published by Glazer *et al.* (2008).

Mercury in Fish. Mercury was detected in all fish tissues in the STEER with body burdens ranging from 0.050 $\mu\text{g/g}$ to 0.502 $\mu\text{g/g}$ and a mean concentration of 0.255 ± 0.056 $\mu\text{g/g}$ (schoolmaster snapper 0.295 ± 0.061 $\mu\text{g/g}$, longspine squirrelfish 0.214 ± 0.100 $\mu\text{g/g}$) (Table 7.8). The highest concentration of mercury in fish tissues in the STEER was found in the squirrelfish caught at site SQ-S2 (0.502 $\mu\text{g/g}$) followed by the snappers caught at site SN-S1 (0.466 $\mu\text{g/g}$). The mean concentration of mercury was elevated relative to the mean concentration of mercury in fish from Vieques, Puerto Rico (Table 7.9). Relative to the coral and conch tissues that were measured, there was more mercury in fish tissue in the STEER.

Using the laboratory recorded percent dry and percent moisture for each sample, a wet weight concentration of

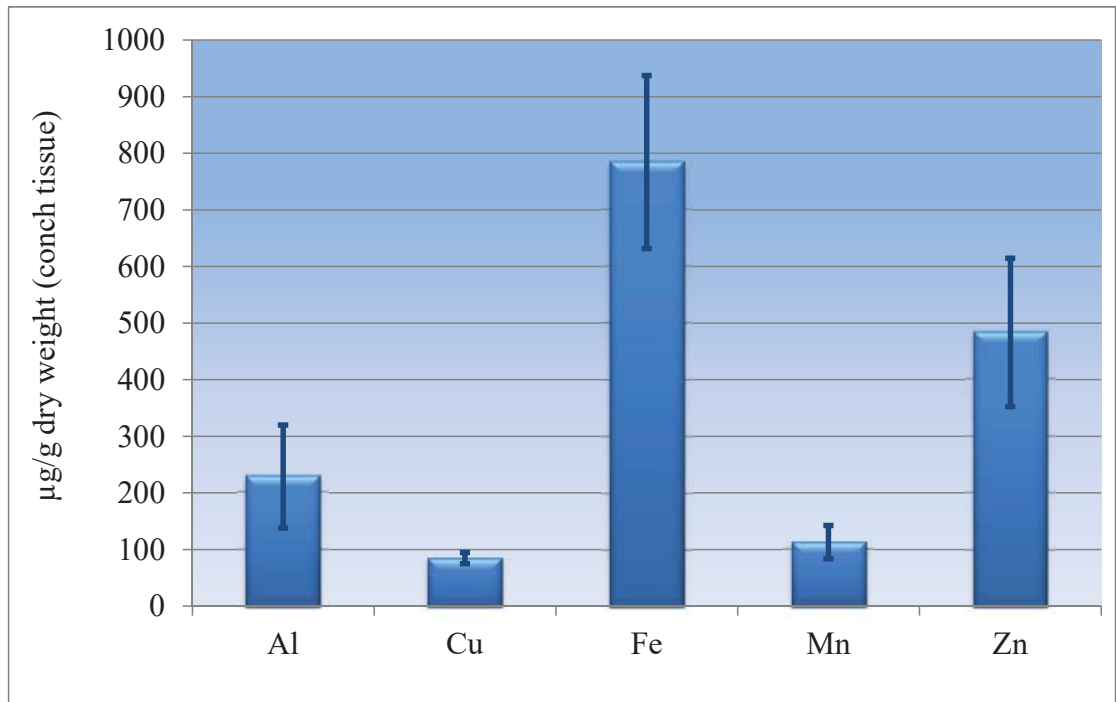


Figure 7.13. Concentrations (mean \pm SE) of metals detected in the conch *Lobatus gigas*.

mercury for each sample was calculated for comparison to EPA's recreational fishers recommended screening values (SVs) (EPA, 2000). Based on the SV of 0.4 $\mu\text{g/g}$ methyl mercury, none of our total mercury (methyl mercury plus inorganic mercury) concentrations exceeded the threshold. According to Kannan (1998), methyl mercury makes up 83% of total mercury in marine organisms.

Due to the low number of fish collected from the STEER, comparison between individual strata was not possible. However, by pooling the strata into two groups, Western (strata 1 and 2) and Eastern (strata 3, 4, and 5), a comparison was possible. Using a non-parametric Wilcoxon test, concentrations of mercury in pooled fish tissues were significantly higher in fish from the Western region than the Eastern region (p -value = 0.0253). While no guidelines for mercury were exceeded in sediments from the STEER, the same spatial pattern of the Western region being significantly higher than the Eastern region exists in sediments as it does for fish (nonparametric Wilcoxon, p -value = 0.0012). For the coring portion of this study (see Chapter 5), surficial sediments in Benner Bay (BB-2) were an order of magnitude higher than those nearby suggesting that, while below all guidelines and thresholds, Benner Bay may be a source of mercury for the STEER.

Mercury has no known biological function, and is hazardous to exposed organisms. Accumulation of mercury at high concentrations in aquatic systems can pose serious

environmental threats to wildlife (EPA, 1997; Murphy *et al.*, 2008). Signs of neurological effects including abnormal behavior, convulsions, reduced fitness and death, have been observed in wildlife exposed to mercury (EPA, 1997; Murphy *et al.*, 2008).

Manganese

Manganese in Coral. Manganese body burdens in *P. astreoides* varied from a minimum value of 7.25 µg/g to a maximum value of 19.6 µg/g (Table 7.4b). In the STEER, manganese was detected in all the coral samples, with the maximum concentration measured in stratum 2. Manganese concentrations were similar in range to reported values in *P. astreoides* from Puerto Rico (Table 7.4b). However, these values were nearly an order of magnitude higher than concentrations reported in Misima Island, Papua New Guinea, Ulan Reef, Philippines, and Daya Bay, China (Table 7.4b). With a mean body burden value of 10.1 µg/g, manganese had the third highest elemental concentration in coral after iron and aluminum.

Accumulation of metals in coral tissue reflects the environmental condition, and because manganese is also considered a marker of terrigenous metal inputs in the aquatic environment (Chen *et al.*, 2010), manganese-to-metal ratio in coral tissue could indicate possible metal enrichment. A non-parametric Spearman correlation test indicated that, in the STEER, with the exception of lead, all other metals were correlated with manganese (Figure 7.14). However, because there were no such positive correlations with aluminum and iron, which are also terrigenous marker elements, the indication is that rather than enrichment, lead along with the other metals are of natural origin. More research is needed to understand uptake processes of metals in coral species.

Manganese in Conch. In *L. gigas*, manganese was also detected in all samples. The highest concentration detected was 355 µg/g, in conch from stratum 2 (Table 7.7). There does not currently appear to be any research available on the effects of manganese in conch.

Manganese in Fish. Manganese was detected in all fish sampled in the STEER with body burden concentrations ranging from 1.94 µg/g to 3.48 µg/g and a mean concentration of 2.52 ± 0.49 µg/g (schoolmaster snapper 2.73 ± 0.28 µg/g, longspine squirrelfish 2.32 ± 0.18 µg/g) (Table 7.8). The highest concentration of manganese found in fish tissues in the STEER was in the fish caught at site SN-S3. The mean manganese concentration was elevated relative to the mean concentration of manganese found in fish tissues from Vieques, Puerto Rico (Table 7.9). On a

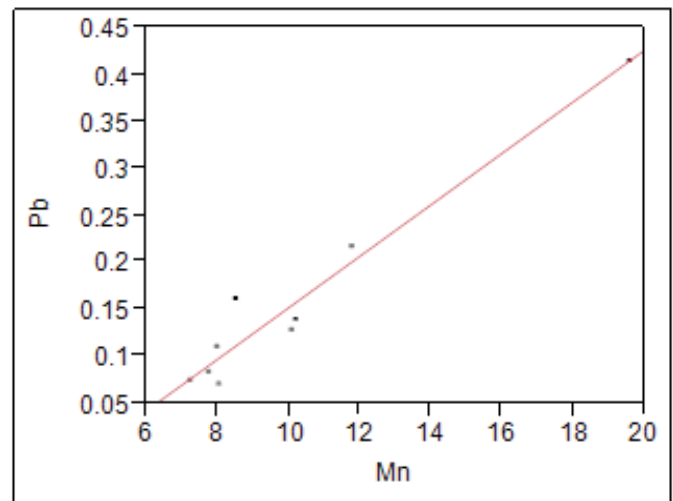


Figure 7.14. Scatter plot showing relationship between manganese and lead in coral.

STEER-wide basis, there was less manganese in fish tissue than in both coral and conch.

Manganese can be toxic to aquatic organisms (Reimer, 1999). The most manganese sensitive aquatic organism that Reimer (1999) studied had a 48 hour LC50 at 0.8 mg/L manganese while the majority of other aquatic fishes had LC50s between 2.1 mg/L up to 94 mg/L.

Nickel

Nickel in Coral. Body burdens of nickel in coral tissues ranged from zero to a maximum value of 8.33 µg/g, found at the HBI42A in stratum 5. Nickel was detected in all coral samples with concentrations that were elevated relative to other trace metals measured (Figure 7.10). Whittall *et al.* (2011) reported a maximum body burden of 6.84 µg/g in *P. astreoides* for nickel, indicating that concentrations found in the STEER were within the regional range.

Reichert-Brushett and Harrison (2005) exposed the gametes of *Goniastrea aspera* to nickel concentrations ranging from 5 to 2,000 µg/L. Concentrations of nickel at or above 100 µg/L resulted in decreased fertilization success. In another study, Goh (1991) exposed larvae of the coral *Pocillopora damicornis* to nickel. The larvae were exposed to concentrations ranging from 1 to 25 mg/L (ppm) for durations of 12 to 96 hours. After the exposure, the larvae were placed in natural filtered seawater, and settlement success was followed over a period of days. Goh (1991) found significantly lower settlement success of the larvae nine days after the recovery period in all concentrations, regardless of the duration of the treatment. In experimental settings, nickel was found to cause serious toxicity to sea anemones *Condylactis gigantea* and *Stichodactyla*

helianthus (Shimek, 2008). The author reported that nickel ambient concentrations as low as 4 ppb could induce sublethal effects such as reduced carbonic anhydrase activity. Sea anemones are sometimes used as a lab model for coral (Tollete *et al.* 2013).

Nickel in Conch. Body burdens of nickel in conch tissues ranged from 3.03 µg/g to 13.6 µg/g (Table 7.6). Nickel was detected in conch from all strata. The mean concentrations of nickel in *L. gigas* tissue by stratum are shown in Table 7.7. While in all other strata, nickel body burdens in conch were fairly similar, relative to the STEER-wide mean, high nickel concentrations (~14 µg/g) were observed in stratum 4. Glazer *et al.* (2008) reported concentrations of 9.59 µg/g ww in conch from the nearshore coastal environment in Florida. Using the measured 76% moisture content in conch, we derived a maximum wet weight value of 3.4 µg/g, indicating that nickel concentrations in conch from the STEER were low relative to values reported by Glazer *et al.* (2008). For nickel, the FDA action level in shellfish of 80 µg/g (ww), is much higher than the wet weight nickel concentration of 3.4 µg/g derived for conch from the STEER.

Nickel in Fish. Nickel was detected in all fish sampled in the STEER with body burdens ranging from 0.911 µg/g to 10.1 µg/g and a mean concentration of 2.57 ± 1.09 µg/g (schoolmaster snapper 3.76 ± 2.13 µg/g, longspine squirrelfish 1.39 ± 0.20 µg/g) (Table 7.8). The highest concentration of nickel found in fish tissues in the STEER was in the fish caught at site SN-S3. The mean nickel concentration was the same as that in Vieques, Puerto Rico (Table 7.9), and was higher than the mean nickel concentration in coral but lower than that of conch.

Nickel toxicity in aquatic organisms varies by species with the concentration necessary to cause negative effects decreasing as exposure time increases (Eisler, 1998b; Svecevicus, 2010). Eisler (1998b) found that for most non-human mammals, concentrations of nickel in the liver above 3.0 µg/g and 10.0 µg/g in the kidneys was evidence of significant nickel exposure. Our highest concentration of 10.1 µg/g falls above those ranges, but represents whole body burden nickel and is not tissue specific. Nickel concentrations above 40 µg/L, have no effect on larval or embryonic survival in fishes, but have been shown to delay hatching time (Eisler, 1998b).

Lead

Lead in Coral. Lead body burdens in *P. astreoides* from the STEER ranged from 0.07 µg/g to 0.42 µg/g (Table 7.4b). With a mean value 0.16 µg/g, lead was detected

in coral from all five strata (Table 7.5). The highest lead concentration in *P. astreoides* from the STEER was found at HBI28P (0.415 µg/g). A lead body burden as high as 12.50 µg/g was reported in *P. astreoides* from Jobos Bay, Puerto Rico (Whitall *et al.*, 2011). However, other studies in southwest and Vieques, Puerto Rico have reported lead mean values of below detection to 0.07 in *P. astreoides* (Pait *et al.*, 2009 and 2010). In a laboratory experiment, Reichelt-Brushett and Harrison (2004) demonstrated that a seawater concentration of lead of 2,900 µg/L seriously impacted coral larvae survival. When lead concentrations in water exceeded 500 ppb, enzymes needed for photosynthesis in the algae were inhibited (Taub, 2004).

Lead in Conch. The lead body burden in *L. gigas* from the STEER ranged from 0.21 µg/g to 1.32 µg/g (Table 7.6). With a mean value of 0.61 µg/g, lead was detected in conch from all five strata (Table 7.7). The highest lead body burden in conch from the STEER was found in stratum 2 at 1.32 µg/g. In a 48h laboratory study, toxicity of lead to the shellfish *Meretrix meretrix* embryogenesis was observed with an EC₅₀ of 297 µg/L in water (Wang *et al.* 2009). The FDA action level for lead in molluscan shellfish is 1.7 µg/g (ww). Using the measured 76% moisture content in conch, we derived an equivalence maximum value of 0.33 µg/g lead ww, indicating that lead concentrations in conch from the STEER were lower than the FDA action level for shellfish.

Lead in Fish. Lead was detected in all but one fish sampled in the STEER at concentrations ranging from 0.086 µg/g to 0.027 µg/g and a mean concentration of 0.0865 ± 0.0268 µg/g (schoolmaster snapper 0.0483 ± 0.0161 µg/g, longspine squirrelfish 0.1247 ± 0.0459 µg/g) (Table 7.8). The highest concentration of lead found in fish tissues in the STEER was at site SQ-S4. Lead tissue concentrations in the STEER appear to be on the low end of the normal range of coastal marine fishes in the U.S. (Eisler, 1988). Mean lead concentrations in the STEER were an order of magnitude lower than those in Vieques, Puerto Rico (Table 7.9) and were also lower than mean concentrations found in coral and conch in the STEER. Rainbow trout (*Salmo gairdneri*) had an LC50 at 72 hours after exposure to 3.5 µg Pb/L of water (Eisler, 1988). Other deleterious effects of high concentrations of lead on fish include anemia, reduced stamina, vertebral deformities, caudal fin erosion, and inhibition of hatching (Eisler, 1988).

Selenium

Selenium in Coral. The body burden of selenium in *P. astreoides* varied from zero to 0.12 µg/g. Selenium was only detected in coral samples from stratum 3 (Table 7.5).

Similar concentration values were observed elsewhere in *P. astreoides* from the Caribbean (Pait *et al.*, 2009 and 2010; Whitall *et al.*, 2011).

Selenium in Conch. Selenium body burdens in *L. gigas* varied from 0.68 µg/g to 2.38 µg/g in the STEER (Table 7.6). The highest selenium concentration was found in stratum 5 (Table 7.7). There is no FDA action level for selenium in shellfish tissue.

Selenium in Fish. Selenium was detected in all fish tissues in the STEER and ranged from 1.18 µg/g to 9.84 µg/g with a mean concentration of 2.69 ± 1.03 µg/g (schoolmaster snapper 1.71 ± 0.14 µg/g, longspine squirrelfish 3.67 ± 2.06 µg/g) (Table 7.8). The highest concentration of selenium found in fish tissues in the STEER was in snappers caught at site SQ-S2 and exceeded the EPA selenium criterion (EPA, 2015). It is important to note that the EPA criterion is for freshwater fish, but there currently is no EPA selenium criterion for saltwater fish. All other measured selenium detections in fish were below the EPA freshwater selenium criterion of 8.0 µg/g dry weight. Mean selenium concentrations in the STEER were similar to those found in fish tissues in Vieques, Puerto Rico (Table 7.9). However, elevated concentrations were found in coral and conch tissues in the STEER. High concentrations of selenium (> 50 µg/L) in water can cause adverse effects to aquatic organisms (Taylor *et al.*, 1992), and exposure to fish larvae can cause skeletal anomalies, decreased growth, and mortality (Newman and McIntosh, 1991). Studies in both marine and fresh water show mortality and other adverse effects at or above 50 µg/L selenium (Hamilton *et al.*, 1986; Phillips, 1988). Some data indicate effects at lower concentrations, for example hematological effects in freshwater trout (15-53 µg/L) (Hodson *et al.*, 1980).

Zinc

Zinc in Coral. In the STEER, *P. astreoides* body burdens ranged from 1.87 µg/g to a maximum value of 14.8 µg/g (Table 7.4b), with an overall average of 5.98 µg/g. Zinc was detected in all five strata (Table 7.5). Zinc body burdens in *P. astreoides* from the STEER were similar to reported values (Table 7.4b). Mean zinc body burdens of 6.09 µg/g and 8.59 µg/g were reported by Pait *et al.* (2009) and Whitall *et al.* (2011) in *P. astreoides* from southwest Puerto Rico and Jobos Bay, Puerto Rico, respectively. These published data indicate that zinc concentrations found in the coral from the STEER are within the concentration ranges seen in the region. However, higher zinc body burdens (Table 7.4b) have been reported elsewhere in Punta Brava, Venezuela (Bastidas and Garcia, 1999), Misima Island, Papua New Guinea

(Fallon *et al.*, 2002), and Dafangji Island, China (Peng *et al.* 2006). Zinc is an essential element, however at elevated concentrations, it can be toxic to coral (Chen *et al.*, 2010). Several studies have linked excess zinc to harmful effects in zooxanthellae (Goh and Chou, 1997) and fertilization impairments in coral (Reichelt-Brushett and Harrison, 2005).

Zinc in Conch. In the STEER, *L. gigas* body burdens ranged from 170 µg/g to a maximum value of 1,320 µg/g (Table 7.6). With an overall average of 484 µg/g, zinc was detected in all five strata (Table 7.7). Concentration ranges of 40.4 µg/g to 874 µg/g have been reported in *L. gigas* from south Florida (Glazer *et al.* 2008), indicating that zinc concentrations found in conch from the STEER are within the concentration range seen in Florida. Zinc has been associated with reproductive inhibition in mollusks. Spade *et al.* (2010) reported that zinc concentrations of 84 µg/g in testis have been linked to testis regression in gastropods such as *L. gigas* from the Florida Keys.

Zinc in Fish. Zinc was detected in all fish tissues in the STEER and ranged from 25.3 µg/g to 217 µg/g with a mean concentration of 65.79 ± 22.0 µg/g (schoolmaster snapper 40.75 ± 1.73 µg/g, longspine squirrelfish 90.83 ± 40.87 µg/g) (Table 7.8). The highest concentration of zinc found in fish tissues in the STEER was in the snappers caught at site SQ-S2. Mean zinc concentrations in the STEER were an order of magnitude higher than those found in fish tissues in Vieques, Puerto Rico (Table 7.9), and in coral tissues in the STEER. Mean zinc concentrations in conch were higher than those found in fish however.

7.4. CONCLUSIONS

In general, organic contaminant concentrations in the tissues of coral, conch, and fish in the STEER appeared to be relatively low, and similar to results seen in other studies from the region. A significant correlation between higher TBT concentrations closer to shore (inshore vs. offshore) existed for both coral and conch. The correlation was not significant for fish tissues. Total butyltins in fish tissues were significantly higher than those found in coral (non-parametric Kruskal-Wallis, p-value = <0.0001) but not conch, yet did not seem to follow the spatial patterns observed in sediments. This may be a factor of the number of fish we were able to collect or the temporal difference between when the sediments and fish tissues were collected, among other confounding factors. The percentage of TBT as compared to its breakdown products DBT and MBT in fish tissues was significantly higher in schoolmaster snapper than in longspine squirrelfish, possibly pointing to long-

spine squirrelfish being more efficient at breaking down TBT into its metabolites than schoolmaster snapper.

With the exception of the fish caught at SQ-S2, total PCBs concentrations in the tissues of fish caught in the STEER were generally an order of magnitude lower than those observed by NS&T in the round goby caught in the Great Lakes. The fish at site SQ-S2 had a sum total PCB value of 150.9 ng/g wet weight (419.2 ng/g dry weight) which exceeded EPA's screening value for noncarcinogenic PCBs of 80 ng/g wet weight (EPA, 2000).

Trace and major elements are incorporated into corals, conch, and fish tissue by a variety of pathways. In coral, metal accumulation can occur by direct replacement of calcium by dissolved metals in the aragonite lattice, inclusion of detritus materials into skeletal pore spaces, uptake of organic materials, incorporation of metals into coral skeletons, or coral feeding (Howard and Brown, 1984). Bioaccumulation of metals in conch can occur via exposure to dissolved metals in the gills, through feeding, and by direct contact through the skin. Conch have been shown to ingest considerable amounts of sediment particles (Brownell and Stevely, 1981). It has been observed that corals (David, 2003; Chen *et al.*, 2010) and conch (Glazer *et al.*, 2008 and Said *et al.*, 2013) from polluted areas show much higher concentrations of trace metals in their tissues than corals from unpolluted areas. In the STEER, the most elevated metal concentrations in corals (skeleton and tissue) were found in strata 1 and 2, which are the strata where sediments had the most elevated concentration of metals (see Chapter 4). This pattern was not observed in conch. The most elevated metal concentrations seen in conch were from stratum 4 instead of strata 1 and 2, where elevated metal concentrations were seen in sediment. It is unclear why there is a difference in the relative metals concentrations in conch tissues as compared to coral and fish. Based on the concentrations of metal body burdens found, the coral and conch tissues do not appear to be very contaminated.

Since there are currently no ecotoxicity thresholds, and the fact that trace and major element concentrations were similar to published data from many other coastal areas in the Caribbean, we conclude that concentrations of the trace and major elements in the coral and conch tissue were background concentrations. More research is needed to understand ecotoxic processes of metals in coral species.

Trace and major elemental contamination in the tissues of fish were not able to be statistically analyzed by individual stratum due to the low number of samples collected. For mercury there appears to be significantly higher mean con-

centrations in the western region (pooling strata 1 and 2) than in the eastern region (pooling strata 3, 4, and 5). Mean metals concentrations in the tissues of all fish in the STEER were comparable to those found in Vieques, Puerto Rico by ATSDR (2003) with the exception of chromium, copper, iron, lead, and zinc which had means an order of magnitude higher than those found in Vieques. No metal concentrations in any fish tissue exceeded any health guidelines or thresholds.

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CHAPTER 8: HISTOLOGICAL EXAMINATION OF CORAL TISSUE SAMPLES FROM ST. THOMAS EAST END RESERVES (STEER), USVI

Esther C. Peters, Ph.D.

Principal, Pathobiology Consulting Services, 4107 Parkedge Lane, Annandale, VA 22003

8.1. INTRODUCTION

Samples of the coral *Porites astreoides* (Figure 8.1) were obtained from the same sites and colonies from which the coral tissue samples were taken for chemical contaminant analyses (see Chapter 7). Sampling sites were located on a transect from most suspected contaminant exposure near the Bovoni Landfill (Stratum 1), nearshore shallow Bener Bay (Stratum 2) and Nazareth Bay (Stratum 3), to least exposure in deeper water Jersey Bay (Stratum 4) or shallow Great Bay (Stratum 5).

This encrusting to massive species is widespread and considered to be relatively hardy (Szmant 1986; Fauth *et al.*, 2011); it was found in varying abundances at the STEER stations. This species has also been used in recent studies on biomarkers of contaminant or stressor exposure (by measuring physiological or biochemical changes) indicating functional or molecular changes (Downs *et al.*, 2011, Fauth *et al.*, 2011, Kenkel *et al.*, 2011).

Changes in an organism's health can also be evaluated using histopathological examination, a biomarker of effect, based on microscopic anatomy that is related to changes in

molecular composition and function of the cells and tissues (Yevich and Yevich 1994; Jagoe 1996; Peters *et al.*, 2005). Some histological studies of *P. astreoides* have been conducted to describe its structure, reproduction, and suspected diseases (Peters, 1984a, 1984b; Szmant, 1986; Chornesky, and Peters, 1987). Toxicant exposures are anticipated to affect this coral's health, which might be reflected in its microscopic anatomy (structure = function), since other coral species have been adversely affected by field and laboratory exposures to oil hydrocarbons, heavy metals, and pesticides, as well as sedimentation (e.g., Peters *et al.*, 1981, Peters and Pilson 1985; Peters *et al.*, 1997; Downs, 2005; Downs *et al.*, 2005, 2006; Rotchell and Ostrander, 2011).

Downs *et al.* (2011) identified site-specific changes in cellular-stress marker proteins in *P. astreoides* sampled from St. John, U.S. Virgin Islands, indicating DNA lesions, which were associated with polyaromatic hydrocarbons (PAHs), biocides, and semi-volatile organochlorines, as well as fecal contamination. These were not detected in their control population, furthest removed from anthropogenic activities. Fauth *et al.* (2011) collected samples of *P. astreoides*

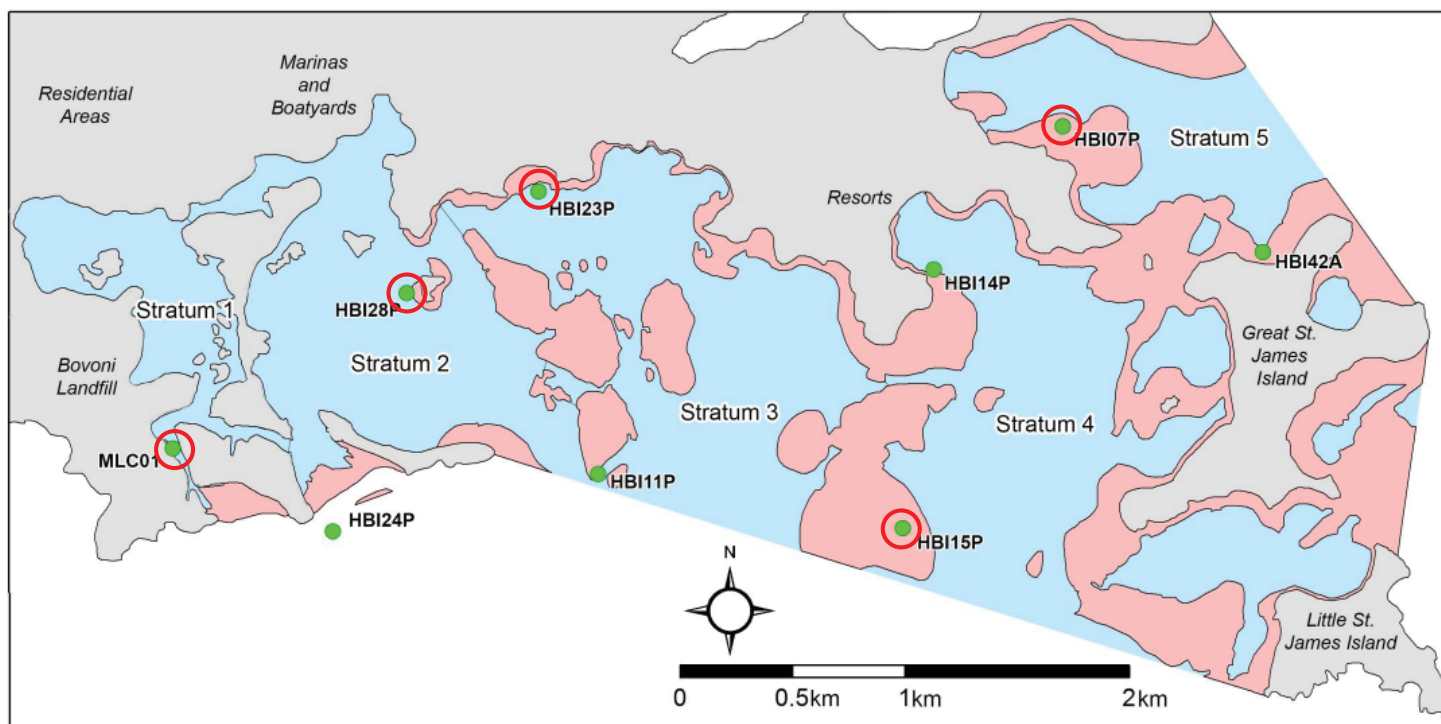


Figure 8.1. Map of the St. Thomas East End Reserves showing site locations (in red circles) where the samples for histopathological examination were collected in June 2012. Coral samples for tissue chemistry analyses were also collected at all sample sites.

from sites near an ocean outfall, inlets, and reefs off Broward County, FL, and also observed increased frequencies of DNA damage and higher concentrations of the antioxidant enzymes catalase and copper/zinc superoxide dismutase at mid-water stations near Port Everglades and Hillsborough Inlets, and an overall indication of intermittent, chronic stress from land-based sources of pollution. However, no histopathological examinations were included in those reports.

8.2. METHODS

Samples of *P. astreoides* were collected by NOAA divers from five sites (Figure 8.1) in the STEER on June 18 and June 22, 2012, according to “Standard Operating Procedure: Collecting

Coral Samples with a Coring Tube for Histology”, provided by the author of this chapter. One or two cores were obtained from each coral colony (“head”) by driving a 2.2-cm diameter stainless steel coring tube into the colony with a sledge hammer and placing the tube and sample into a clean labeled 50-mL plastic centrifuge tube with ambient seawater. In some cases, portions of a colony were removed by chisel and sledge hammer and placed in a tube. On return to the surface, the seawater was replaced with fixative solution, 1 part Z-Fix Concentrate (Anatech, Ltd.) mixed with 4 parts 35 ppt seawater prepared from marine salts and deionized water (about 4% formaldehyde).

Sample Preparation

The 44 samples from 23 coral colonies (specimens) were left in the fixative, shipped to Dr. Peters at George Mason University (GMU), and received on June 26, 2012. Samples were photographed in the laboratory (Figure 8.2) and then decalcified using Immunocal™, a 5% formic acid decalcifying solution, to remove the aragonite exoskeleton from the polyps. Some samples needed only 24 hours to completely decalcify, but most required 48–72 hours.

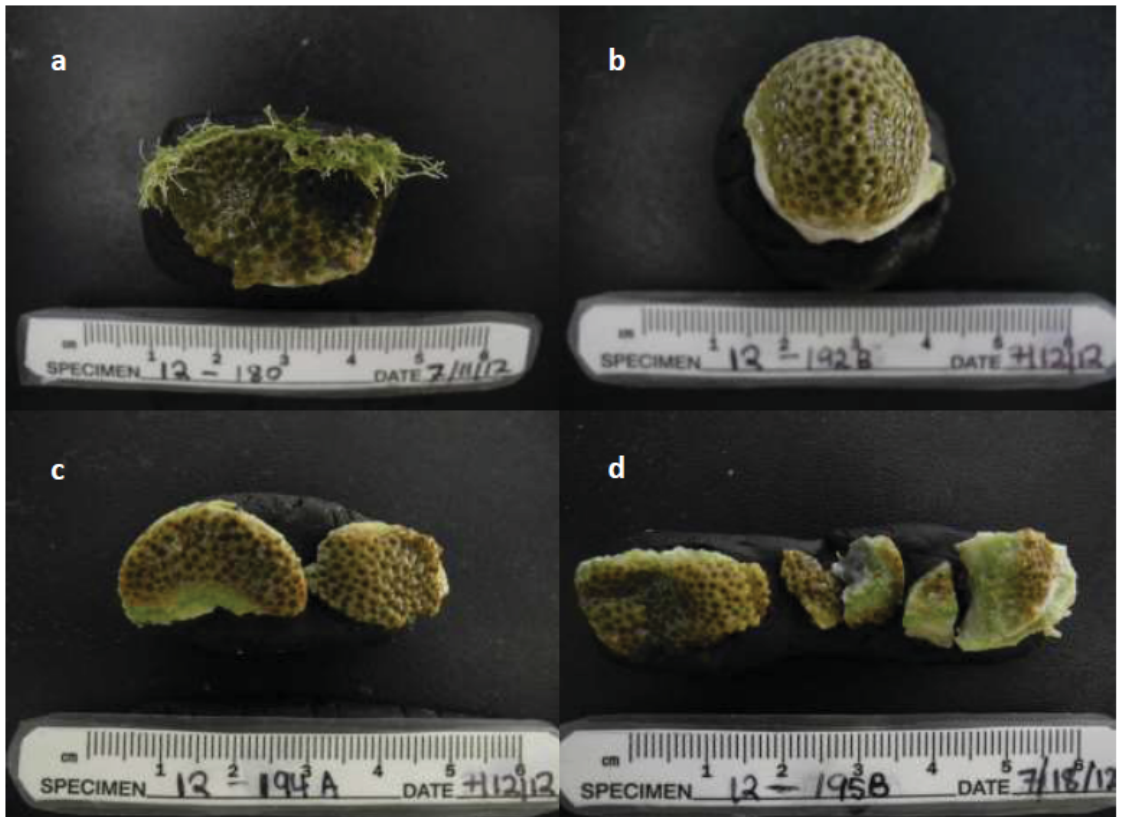


Figure 8.2. Examples of fixed coral samples as photographed in the GMU Histology Laboratory. (a) MLC 01 Head 1, small portion from edge of colony, note abundant algae underneath the sample. (b) HBI 15P Head 5 B, intact core sample. (c) HBI 23P Head 2A, two pieces received, note surface mucus and sediment on larger piece, and green endolithic community (d) HBI 23P Head 3B, three smallest shattered pieces were discarded, two largest pieces were decalcified and trimmed for embedding, slides read but data not used in final evaluation, because the other core from the colony was intact.

Endolithic algae and fungal filaments and sponge tissue remaining with the sample were removed only when it was easy to do so (most samples had visible endolithic communities). When decalcified, samples were put in a running tap water rinse bath for 30 minutes or more to remove the solution and placed in 70% undenatured ethanol. Each sample was trimmed into three or more pieces to fit into one or two tissue processing cassettes.

The approximate locations where the tissue was sectioned were marked on the gross undecalcified images which had been printed out to document the trimming. For those samples that had broken into pieces when collected in the field, the largest piece(s) were retained; smaller ones were discarded as they were probably damaged, and grossly damaged margins were trimmed away when feasible. In general, one or more 2–3 mm wide strips were cut from the middle of the sample core for examining the polyps sagittally, and the ends of the core were placed in the cassette oral and aboral sides up to prepare cross (transverse) sections of the polyps.

Tissue samples were processed through a graded series of diluted undenatured ethanol (two changes of 70%, one each of 80% and 95%), three changes of 100% reagent alcohol, three changes of SafeClear II (xylene substitute), and three changes of molten Paraplast® Plus. They were then embedded in Paraplast® Xtra in stainless steel molds and cooled to solidify into blocks. Blocks were sectioned on a rotary microtome set at 5- μ m thickness. Sections were floated on a warm-deionized water bath and one or two sections were mounted on clean microscope slides. Two slides were prepared from each block. The first slide for each block was stained with Harris's hematoxylin and alcoholic eosin procedure (H&E) and coverslipped with Permount™ mounting medium. The second slide was stored unstained, if needed for a special staining procedure later.

The H&E-stained histoslides were examined with a Zeiss microscope and photomicrographs were taken using an attached Nikon COOLPIX 5000 camera. Histoslides of *P. astreoides* collected from reference sites (Jamaica, Belize, 1980s) were used to develop the “within normal limits” criteria for coral tissue condition and zooxanthellae condition/abundance scores, specific parameters thought to indicate sampling damage or exposure to environmental stressors, and suspect parasites or pathogenic microorganisms (protozoans from the phylum Apicomplexa and bacterial aggregates previously reported in this species, Peters, 1984; Rickettsiales-like bacteria, Casas *et al.*, 2004, C. S. Friedman, pers. comm; and three “unknowns”). Sections were first scanned at 100x magnification to obtain data on larger features (e.g., general condition, epidermal ruptures, gonads, planulae, endolithic organisms) and then scanned at 250x to obtain data on smaller features (e.g., zooxanthellae condition, epithelial necrosis, suspect bacteria, protozoan parasites).

Sample Analysis

Semi-quantitative data were collected from each sample based on relative condition (0=Excellent, 1=Very Good, 2=Good, 3=Fair, 4=Poor, 5=Very Poor) or the severity or intensity of tissue changes from normal (0=Within Normal Limits, 1=Minimal, 2=Mild, 3=Moderate, 4=Marked, 5=Severe). The extent of the lesions was also recorded (1=Focal, 2=Multifocal, 3=diffuse or systemic). Only presence was recorded for the three “unknowns” (unusual cells or metazoans whose identity is unknown and were rarely found).

The developmental stages of gonads were noted and number of planulae in the sections were counted, if present. The criteria for these data are presented in Table 8.1. Data for each colony sample were entered into a spreadsheet,

grouped by Stratum. The approach for analyzing the data was simply exploratory, because no one has done this sort of histopathological examination of a population of *P. astreoides* to determine effects of environmental stressors on the coral. A mean score for each parameter for each colony was calculated from the observations made on the two cores (A and B) or from multiple histoslides of tissue sections, if available, but damaged (broken) core scores were not used.

For gonad development and “unknowns,” percentage of colonies containing gonads, planulae, or “unknowns” in each stratum were determined. For changes related to suspect sampling damage, epidermis, gastrodermis, calicodermis, skeleton, and suspect apicomplexa, the severity was multiplied by the extent to obtain a final score in the range of 0–15 (= not present to the most severe and widespread) for each parameter. These final scores were summed for each sample to provide a Specific Condition Scores Sum as an indicator of the colony's health status. Means and standard deviations were calculated for the scores for each parameter from the samples for each Stratum. The frequency distributions of data for all the colonies sampled from the STEER were plotted to determine whether the scores were normally distributed.

Quality Assurance and Quality Control

Preparation of the histoslides included frequent checks that samples had been carried through all steps of the procedures, including gross photography (images were retaken if they were not in focus), decalcification and trimming (damaged fragments were noted and some discarded, marked on the trimming sheets), processing and embedding (clearing and infiltration with paraffin occurred, tissues placed in bottom center of molds as they had been trimmed into the cassettes), microtomy (sectioned at 5 microns, one section mounted on each of two microscope slides, dried), and staining with H&E (procedure followed and staining quality of each rack checked microscopically).

Histoslides were briefly reviewed for staining quality and to detect any lesions that should be included in the spreadsheet for analysis. Then each sample was examined in more depth to record lesion and reproduction data. Sections were viewed at 100x to note general condition and zooxanthellae distribution, then at 250x to detect lesions, and 400x to confirm observations. After recording the data for all histoslides, the spreadsheet columns were adjusted to separate lesions that were probably associated with sample collection and handling from those that might be indicative of site-specific stressors. Randomly selected histoslides were reexamined to confirm the data or make corrections

Table 8.1. Characteristics and scoring of changes noted in cells and tissues using light microscopic examination of *P. astreoides* samples collected during this study.

Parameter Viewed at 100x or 250+x, Description of "Normal"	Numerical Score Intensity or Severity Score				
0 (No Change)	1	2	3	4	5
Low Magnification	Very Good	Good	Fair	Poor	Very Poor
General Condition 0 = Excellent, similar to reference samples, thick and intact epithelia and mesoglea, highly cellular, polyp and coenenchyme architecture well developed	Similar to reference samples, but epithelia and mesoglea not as thick, epidermal mucocytes slightly hypertrophied	Hypertrophy of epidermal mucocytes, intact epithelia and mesoglea not attenuated, mesentery and filament architecture still normal	Hypertrophy of epidermal mucocytes, minimal to mild attenuation of epithelia and mesoglea noted, damage to epithelia noted (lysing cells)	Loss of mucocytes, moderate attenuation of epithelia and mesoglea, mesentery and filament architecture degenerating, vacuolation of basal body wall epithelia	Severe attenuation of epithelia and mesoglea, vacuolation of mesogleal pleats, necrosis and dissociation of mesenterial filaments, necrosis and lysing of epithelial cells
Zooxanthellae 0 = Gastrodermal cells packed with well-stained algal symbionts in surface body wall, tentacles; scattered algal symbionts deeper in gastrovascular canals	Similar to reference samples, slightly fewer well-stained algal symbionts in gastrodermis of surface body wall, tentacles, and scattered cells in gastrovascular canals	Layer of well-stained algal symbionts, but less abundant than in reference samples	Algal symbionts fewer in gastrodermis which is mildly attenuated, most still stain appropriately, some may be degenerating	Even fewer algal symbionts in surface body wall gastrodermis and in tentacle gastrodermis, some have lost acidophilic staining as proteins no longer produced or nucleus/cytoplasm lysed, vacuole enlarged compared to algal cell	No zooxanthellae present in gastrodermis of colony (bleached)
Low or High Magnification	Minimal	Mild	Moderate	Marked	Severe
Melanin-like Granular Amoebocytes, Surface Body Wall 0 = None present	One to a few noted in some areas of surface body wall (epidermis or gastrodermis)	About one-quarter of epithelial area contains these cells	About half of epithelial area contains these cells	About three-quarters of epithelial area contains these cells	Both epidermis and gastrodermis are heavily infiltrated by these cells
Melanin-like Granular Amoebocytes, Mesenteries and Mesenterial Filaments 0 = None present	One to a few noted in some areas of mesenterial lobes or cnidoglandular bands of mesenterial filaments	About one-quarter of the area of mesenterial lobes or cnidoglandular bands contains these cells	About half of the area of mesenterial lobes or cnidoglandular bands contains these cells	About three-quarters of the area of mesenterial lobes or cnidoglandular bands contains these cells	Area of both lobes and mesenterial filaments heavily infiltrated by these cells
High Magnification	Minimal	Mild	Moderate	Marked	Severe
Suspect Sampling Damage, Surface Body Wall Ruptures 0 = Epithelia and mesoglea intact	One or a few breaks in epithelia noted on sagittal sections	About one-quarter of area of epithelia has gaps with missing epithelia and mesoglea, mucus discharge	About one-half of area of epithelia has gaps with missing epithelia and mesoglea, mucus discharge	About three-quarters of area of epithelia has gaps with missing epithelia and mesoglea, mucus discharge	Entire surface body wall dissociated, extensive release of mucus
Suspect Sampling Damage, Lysing Gastrodermal Cells 0 = Gastrodermis of surface body wall and lining basal body wall of oral coenenchyme gastrovascular canals intact	One or a few areas of gastrodermal cells have lysed, mucus filling gastrovascular canals or gastrovascular cavity, granular gland cells, holotrichous isorhiza nematocysts, zooxanthellae also released	About one-quarter of the areas of gastrodermal cells have lysed, mucus filling gastrovascular canals or gastrovascular cavity, granular gland cells, holotrichous isorhiza nematocysts, zooxanthellae also released	About one-half of the areas of gastrodermal cells lysed, mucus filling gastrovascular canals and gastrovascular cavities, granular gland cells, holotrichous isorhiza nematocysts, zooxanthellae also released	About three-quarters of the area of gastrodermal cells lysed, mucus filling gastrovascular canals and gastrovascular cavities, granular gland cells, holotrichous isorhiza nematocysts, zooxanthellae also released	All gastrodermal cells have lysed, mucus filling gastrovascular canals and gastrovascular cavities, granular gland cells, holotrichous isorhiza nematocysts, zooxanthellae also released

Table 8.1. Characteristics and scoring of changes noted in cells and tissues using light microscopic examination of *P. astreoides* samples collected during this study (continued).

Parameter Viewed at 100x or 250x, Description of "Normal"	Numerical Score Intensity or Severity Score				
	1	2	3	4	5
0 (No Change)					
Epidermis, Epidermal Mucocytes Hypertrophy 0 = In reference sample, short columnar cells, uniform distribution and not taller than ciliated supporting cells, pale mucus	Slightly hypertrophied, numerous, pale- staining frothy mucus	Many cells hypertrophied, abundant release of pale- staining mucus	Uneven appearance of mucocytes, some hypertrophied but some reduced in size and secretion, darker staining mucus	Some epidermal foci lack mucocytes entirely, attenuation of epidermis evident, darker staining and stringy mucus	Loss of many mucocytes, epidermis is attenuated to at least half of normal thickness or more, if mucus present stains dark, thick
Epidermis, Necrosis or Apoptosis? 0 = None present	Single cells rarely seen having pycnotic or blebbed nuclei in the epidermis	Single cell necrosis or apoptosis present in about one- quarter of area of epidermis	Single cell necrosis or apoptosis present in about one-half of the area of epidermis	About three-quarters of the area of epidermis contains necrotic or apoptotic cells	Complete necrosis of epidermis
Gastrodermis, Black Particles in Released Mucus of Gastrovascular Canals 0 = None present	One or a few black spherules present in mucus within gastrovascular canals of oral region	Cluster of black spherules present in mucus within gastrovascular canals of oral region	Two clusters of black spherules present in mucus within gastrovascular canals of oral region	Three clusters of black spherules present in mucus within gastrovascular canals of oral region	Multiple clusters of black spherules scattered in mucus with gastrovascular canals of oral region
Gastrodermis, Necrosis or Apoptosis? 0 = None present	Single cells rarely seen having pycnotic or blebbed nuclei in the gastrodermis (surface body wall or elsewhere)	Single cell necrosis or apoptosis present in about one- quarter of area of gastrodermis	Single cell necrosis or apoptosis present in about one-half of the area of gastrodermis	About three-quarters of the area of gastrodermis contains necrotic or apoptotic cells	Complete necrosis of epidermis
Calicodermis, Attenuation, Lack of Acidophilic Granules 0 = squamous to low columnar calicodermis, fine acidophilic granules of organic matrix proteins present	Slight thinning of calicodermis or fewer acidophilic granules	More variable in thinning of calicodermis, fewer acidophilic granules, more areas affected	Calicodermis squamous, fewer acidophilic granules, lysing in some areas	Squamous calicodermis, necrotic or lysing, no acidophilic granules	Loss of calicoblasts along mesoglea, necrotic or lysing
Calicodermis, Suspect Bacteria on Apical Surface or Necrosis? 0 = None present	Fine basophilic granules rarely seen along apical surface of calicodermis	About one- quarter of calicoblasts attenuated, fewer acidophilic proteins in cytoplasm, increase in basophilic granules on surface	About half of calicoblasts attenuated, fewer in number, thin layer of basophilic granules on apical surface on about half of calicodermis, some nuclei may be pycnotic	Most calicoblasts attenuated, fewer in number, calicoblasts cytoplasm basophilic, fine darker purple granules present, nuclei swollen and pale or pycnotic	Calicoblasts attenuated or vacuolated, cytoplasm basophilic, fine darker purple granules present and being released from cells, forming thick layer, nuclei swollen and pale or absent
Skeleton, Endolithic Algae and Fungi 0 = None present	A few filaments or suspect hyphae (special stain must be done to confirm fungi) present in subcalicodermal spaces where skeleton was removed	About one- quarter of subcalicodermal space where skeleton was removed contain filaments or suspect hyphae	About half of subcalicodermal space where skeleton was removed contain filaments or suspect hyphae	About three-quarters of subcalicodermal space where skeleton was removed contain filaments or suspect hyphae	Thick tangle of filaments or suspect hyphae present in subcalicodermal spaces where skeleton was removed

Table 8.1. Characteristics and scoring of changes noted in cells and tissues using light microscopic examination of *P. astreoides* samples collected during this study (continued).

Parameter Viewed at 100x or 250x, Description of "Normal"	Numerical Score Intensity or Severity Score				
0 (No Change)	1	2	3	4	5
Apicomplexa, Coccidia? Oocysts in gastrodermal cells in lobes of mesenterial filaments; Sporozoites (stage hatching from oocysts) and Sporozoa (four-celled organism, one cell resembles sporozoite nucleus) in gastrodermis of basal body wall lining gastrovascular canals and polyps	One to a few oocysts, sporozoites, or sporozoa rarely seen in specific cells, deep aboral region of tissue	About one- quarter of specific cells in deep aboral region contain oocysts, sporozoites, or sporozoa, scattered	About half of specific cells in deep aboral region contain oocysts, sporozoites, or sporozoa, scattered	About three-quarters of specific cells in deep aboral region contain oocysts, sporozoites, or sporozoa, scattered	Specific cells contain one or more oocysts, sporozoites, or sporozoa throughout entire deep aboral region of tissue
Bacterial Aggregates in Tentacles (Peters 1984) 0 = Not present	One bacterial aggregate in mesoglea/ gastrodermis of tentacle section of sample (rare)	One bacterial aggregate in tentacle tissue sections of more than one polyp on slide (occasional)	Density increases, one bacterial aggregate in tentacle tissue of all polyps on slide (common)	Density increases, multiple bacterial aggregates in tentacle tissue of polyps on slide (frequent)	Numerous bacterial aggregates present in tentacle tissue, multiple adjacent aggregates (abundant)
Other Suspect Bacteria (RLOs, CGB) 0 = Not present	One infected cell in cnidoglandular bands of mesenterial filaments of sample (rare)	One infected cell in a few cnidoglandular bands of mesenterial filaments of sample (occasional)	One or more infected cells in about half of cnidoglandular band sections of sample (common)	Multiple infected cells in about three- quarters of cnidoglandular band sections of sample (frequent)	Infected cells in all cnidoglandular band sections of sample (abundant)
Unknowns - Presence only noted of these organisms – one to a few seen in some samples					
Unknown Cells?	Spherical cells containing two or three cells packed within, each has large dark-staining nucleus, pale pink cytoplasm, multiple paler vacuoles				
Calicodermis/skeleton Unknown Metazoan	Ovoid multicellular organism, pink cuticle, present in a space adjacent to calicodermis (next to skeleton?), one end appears to be ciliated, seen in <i>P. astreoides</i> occasionally from other areas (e.g., Belize, Florida Keys, Peters unpubl. observ.), unidentified				
Skeletal Space Containing "Blobs"	In spaces where skeleton should be at tips of septa, small (about twice the size of a zooxanthella or about 20 µm), ovoid to irregularly shaped cells with pink cytoplasm, single nucleus does not seem to be present, but fine dark purple granules scattered in cytoplasm, have been seen in other species (Renegar et al. 2008, Peters unpubl. observ.), unidentified				
Gonad Staging 0 = None present	1	2	3	4	5
Oocytes	Single germ cell surrounded by mesoglea in mesentery	Early oocyte, nucleus with distinct nucleolus but little development of lipid and protein in cytoplasm	Mid-development, uniform distribution of lipid droplets and protein granules, nucleus and cytoplasm enlarge	Mature, development of cortical granules and vitelline membrane, beginning to separate from mesoglea	Spawned, hole present where ovum released to gastrovascular cavity
Spermaries	Germ cells aggregate in mesoglea, forming one or a few clusters	Early spermaries, multiplication of germ cells, mitotic figures present	More spermaries present, spermatocytes undergo meiosis, spermatids fill lumen	Mature spermatozoa fill lumen, may still have earlier stages surrounding these, but eventually all change to spermatozoa	Spawned, remnants of spermatozoa endocytosed by absorptive gastro- dermal cells on mesentery

to the spreadsheet. Photomicrographs of selected structures and conditions were taken; a couple of them were sent to experts to obtain additional diagnosis insights (the project name, species, and sampling locations were not provided to the experts). A complete review of the histopathology data by another comparative histopathologist (the most common QC approach in anatomic pathology) has not been conducted.

Samples that had broken or were grossly visibly damaged during collection were removed from the calculations. Condition scores between the A and B subsamples were usually close in value, if not the same. Calculations were rechecked and reviewed.

Statistical Analysis

A single-factor analysis of variance was calculated to determine whether the mean parameter scores and condition sums of samples collected for each stratum were equal. Because of the small sample sizes and nonnormality of some of the data, a Kruskal-Wallis test was also run on the Specific Condition Scores Sums for each stratum.

8.3. RESULTS AND DISCUSSION

A total of 23 colonies of *P. astreoides* were sampled from the STEER stations, with duplicate cores or fragments obtained from most specimens. The colonies were widely scattered at the station nearest to shore, and only three colonies were sampled at this station (MLC 0). Five colonies were sampled at each of the other four stations (HBI7P, 15P, 23P, 28P). On examination of the gross specimens, most appeared to be apparently healthy—having a covering of uniformly brown pigmented tissue and polyps, with normal morphology for the species and no visible lesions—and in good condition, except for obvious breakage during sampling. Many, however, had mild to moderate mucus and sediment particles visible on the tissue surface.

Samples taken from the growing edge of colonies often had green filamentous or calcified algae attached to the skeleton (e.g., Figures 8.2a and 8.2c). After decalcification, endolithic fungi, algae, and suspect boring sponge tissue were found in most samples, deep within the gastrovascular canal region of coenosteum between the polyps. This mat community was removed if it could be done easily without tearing the coral tissue, because this material often harbors siliceous sponge spicules, which can cause knicks in the microtome blades and tears in tissue sections. None of the sampled colonies were judged to be in excellent condition microscopically under low magnification, even disregarding what was probably the result of sampling damage: surface body wall ruptures and lysing of gastrodermal cells

in the oral region of the polyp and coenenchyme tissue (Figure 8.3a-d).

It appeared that crushing of the sample margins by the stainless steel corers released mucus into the gastrovascular cavity and canals throughout each sample. On closer examination, the impact of hitting the corer with the sledge hammer caused lysing of gastrodermal cells, largely mucocytes, which line the basal body wall and gastrovascular canals below the surface body wall. Only the primitive connective tissue, the mesoglea, and calicodermis remained to mark where the epithelium should have been. The epidermis of the surface body wall was usually thin, resting on a thinner layer of mesoglea and having a thicker gastrodermal epithelium underneath it filled with zooxanthellae, and this surface body wall often remained intact.

Most samples, however, had multifocal areas of rupture of the epidermis and release of mucus, nematocysts, and zooxanthellae over the surface from the hammering. No changes in zooxanthellae condition were noted at any station (intact, not misshapen, no vacuolation, no necrosis, no enlargement of the gastrodermal cell vacuoles containing the algae) and all samples contained these symbiotic algae; some colonies might have fewer than others, but this could not be reliably detected in the histopathological examinations. The relative amount of melanin-like granular amoebocytes (formerly known as chromophore cells, Peters, 1984a) was noted for each sample (Figure 8.4), with differences found in their distribution. These cells support the coral's immune system (Palmer *et al.*, 2008). Some of the samples had high levels of these cells in the epidermis/gastrodermis and tentacles of the polyps, some had fewer cells there but more in the mesenteries and mesenterial filaments, and in others they were distributed among the basal body wall epithelial cells.

Toward the base of the tissue layer (polyps with associated gastrovascular canals) in this perforate coral, these amoebocytes become rounded and dark, indicating that the cells have died, and are present within gastrodermal cells of the basal body wall as residual bodies, remnants of pigment that are only slowly broken down. Although it appears that the number and distribution of these cells changes with exposure to stressors, such as fungi (Domart-Coulon *et al.*, 2006), it could also be related to genetics as individual expression of their innate immune system.

The distribution and size of epidermal mucocytes also varied greatly, and some samples had markedly hypertrophied mucocytes. This might be related to sedimentation (Peters and Pilson, 1985) or contaminant exposure as a

response to a toxic substance, but other studies will be needed to determine this. Probably the most unusual finding was minute particles of basophilic cell debris in the basal portion of the epidermal cells or in gastrodermal cells of the surface body wall, which was otherwise intact and had normal-appearing nuclei in the columnar cells and mucocytes (Figure 8.5). The condition of the calicodermis was often poor, degenerating, or appeared to have basophilic debris or suspect bacteria on the surface of calicoblasts. Occasionally such particles appeared to be sequestered within phagocytic amoebocytes.

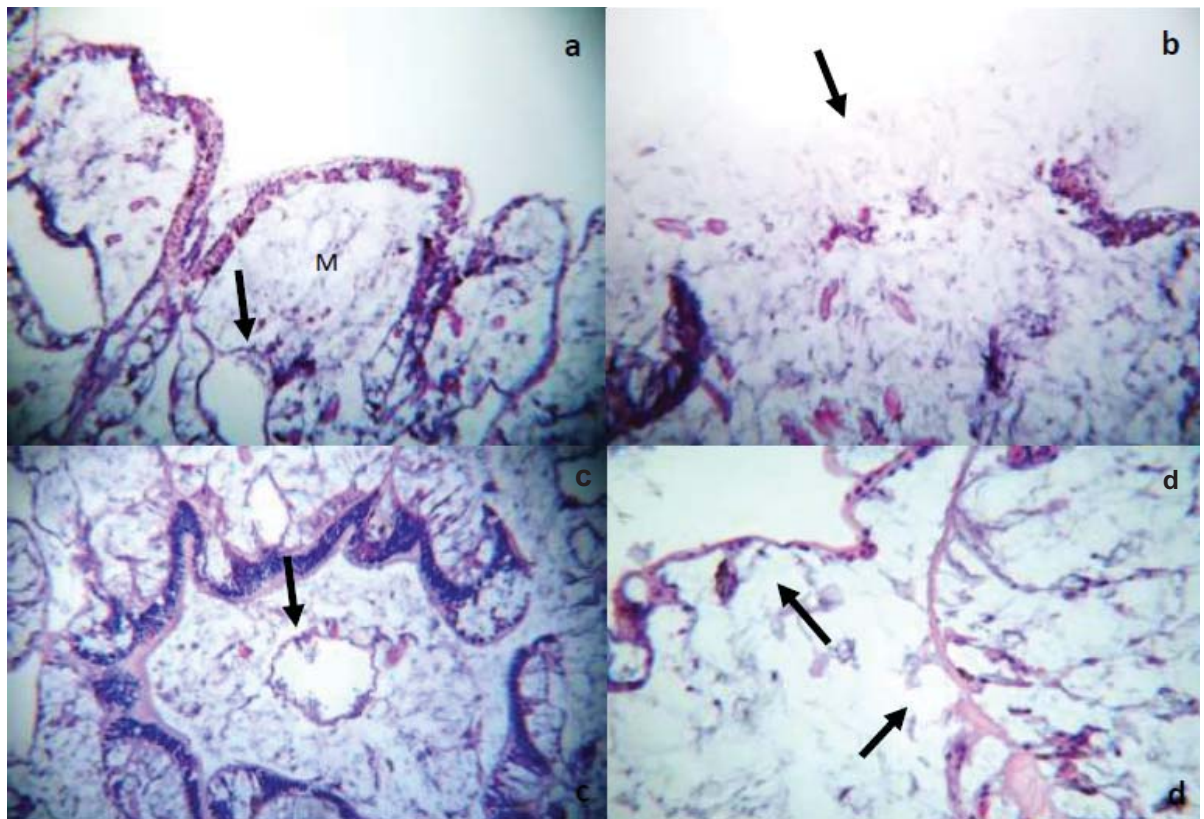


Figure 8.3. Examples of surface body wall and gastrodermis ruptures and lysing. (a) Intact surface body wall, but gastrovascular canal is filled with discharged mucus and lysed cells (M) from basal body wall gastrodermis (arrow), 10x. (b) Sagittal view of surface body wall rupture, discharge of mucus containing holotrichous isorhiza nematocysts, zooxanthellae, and cell debris, 10x. (c) Cross-sectional view of gastrovascular cavity and canals filled with mucus, lysed gastrodermis from basal body wall surrounding an area where skeleton was removed, 10x. (d) Lysed gastrodermal cells along mesentery, mesogleal pleats in lower right-hand corner, 40x.

More often, the particles were present in that region and did not appear to have been phagocytosed. This could mean that mild necrosis of cells was occurring chronically. It did not seem to be associated with the sampling damage,

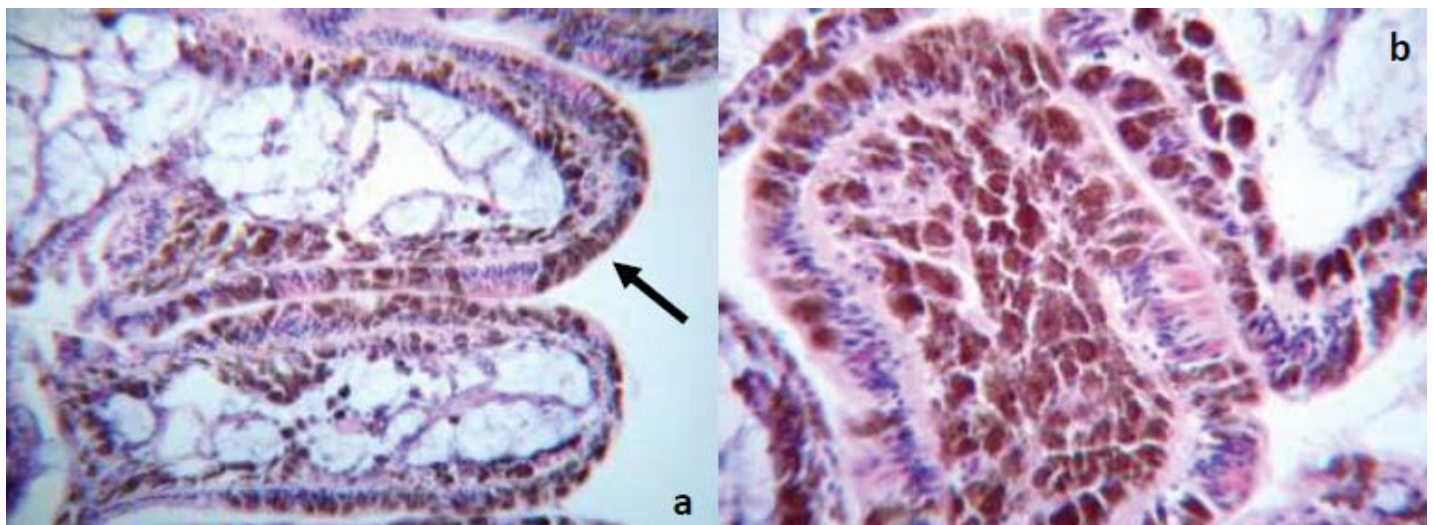


Figure 8.4. Melanin-like granular amoebocytes (brown cells) infiltrating between cells of the epidermis and gastrodermis of tentacles (arrow), rated severe (= 5). (a) Sagittal section, 10x. (b) Cross section, 40x.

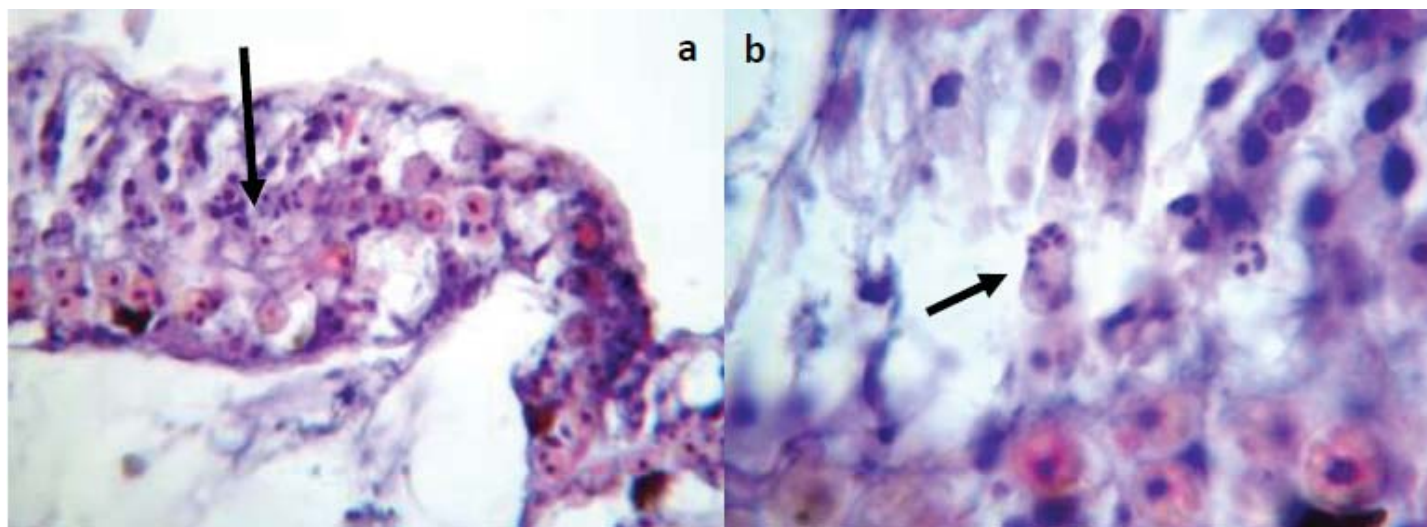


Figure 8.5. Examples of apoptosis in the epidermis (at arrows). (a) Surface body wall, 40x. (b) High magnification of apoptotic cells, 100x.

although a delay in putting the cores into the fixative might begin this process; however, no delay was reported.

Dr. Taylor Reynolds of Johns Hopkins University, a board-certified veterinary pathologist, was consulted on this phenomenon and she noted that mild necrosis and apoptosis might be difficult to distinguish using light microscopy. She further stated, “There is, as you say, single cell necrosis within the epidermis and calicodermis. Within the gastrodermis, there is segmental necrosis. The single cells undergoing necrosis have not lost membrane integrity, but have several nuclear blebs which are each 1/3 to 1/10 the size of an intact nucleus. These cells are sometimes also swollen. Typically, an apoptotic cell has nuclear blebbing, and is also shrunken, instead of swollen. It is very difficult to make the distinction between apoptosis and necrosis using light microscopy. I would indicate in the report that there are individual cells undergoing necrosis / apoptosis, within otherwise normal epidermal and calicodermal layers. Within the segments of necrotic gastrodermis, there is loss of cells and scant evidence of nuclear remnants, i.e. the necrosis in this region is more pronounced.”

The calicodermis, the epithelium forming the aragonite exoskeleton of the coral on the basal body wall of the polyps and gastrovascular canals, should appear as a squamous, but substantial layer (Figure 8.6a) and the calicoblasts should contain fine acidophilic granules (believed to be proteins for the organic matrix of the skeleton) (Figure 8.6b). It should be cuboidal to short columnar in areas of more active calcification (Figure 8.6c). In multiple samples, the calicoblasts were attenuated, lacked the acidophilic granules, or were lysing (Figure 8.6d). Sometimes the epithelium had small circular spots of basophilic material on cell surfaces (possibly staining this way due to an associa-

tion with calcium and a degenerative process); in some cases darker basophilic-staining coccoid masses were associated with the surface of this epithelium (possibly bacteria). Dr. Reynolds noted that the cells were undergoing necrosis. Because of the high levels of boring endolithic fungi in the skeleton, and suspect apicomplexan parasites burrowing in the epithelium with associated vacuolation and necrosis within the gastrodermis, the calicodermal condition might have been related to these stressors. Although the mechanism of action is uncertain, any susceptibility of cells to mechanical or toxic stress could lead to necrosis, and then lysing of the other epithelium (the gastrodermis) could occur as enzymes were released by the degrading cells.

As has been noted in recent literature, the coral holobiont—all of the associated organisms living on, within tissues, or boring through the skeleton—must be taken into consideration (Wegley *et al.*, 2007). In this study, zooxanthellae, bacterial aggregates (Figure 8.7a and b) (Rohwer *et al.*, 2002), endolithic pigmented or nonpigmented fungi or filamentous algae (Figure 8.7c and d) (e.g., *Ostreobium quekettii*), boring sponges, suspect apicomplexan protozoan parasites (Upton and Peters 1986), and other suspect bacteria were seen in tissue sections. The role of most of these organisms is unknown. Although the bacterial aggregates of *Porites* spp. have not been associated with any tissue damage (Peters, unpub. observ.), observations made during this study and others indicated that the protozoans, fungi, and suspect RLOs can be detrimental to the coral (Upton and Peters, 1986; Bents *et al.*, 2000; Peters unpub. observ.). The samples provided additional histological data to assist in understanding this species of coral, since most studies have been on its biochemistry or microbiology. In particular, the susceptibility of the coral to biotic factors can be mediated by exposure to abiotic factors when levels

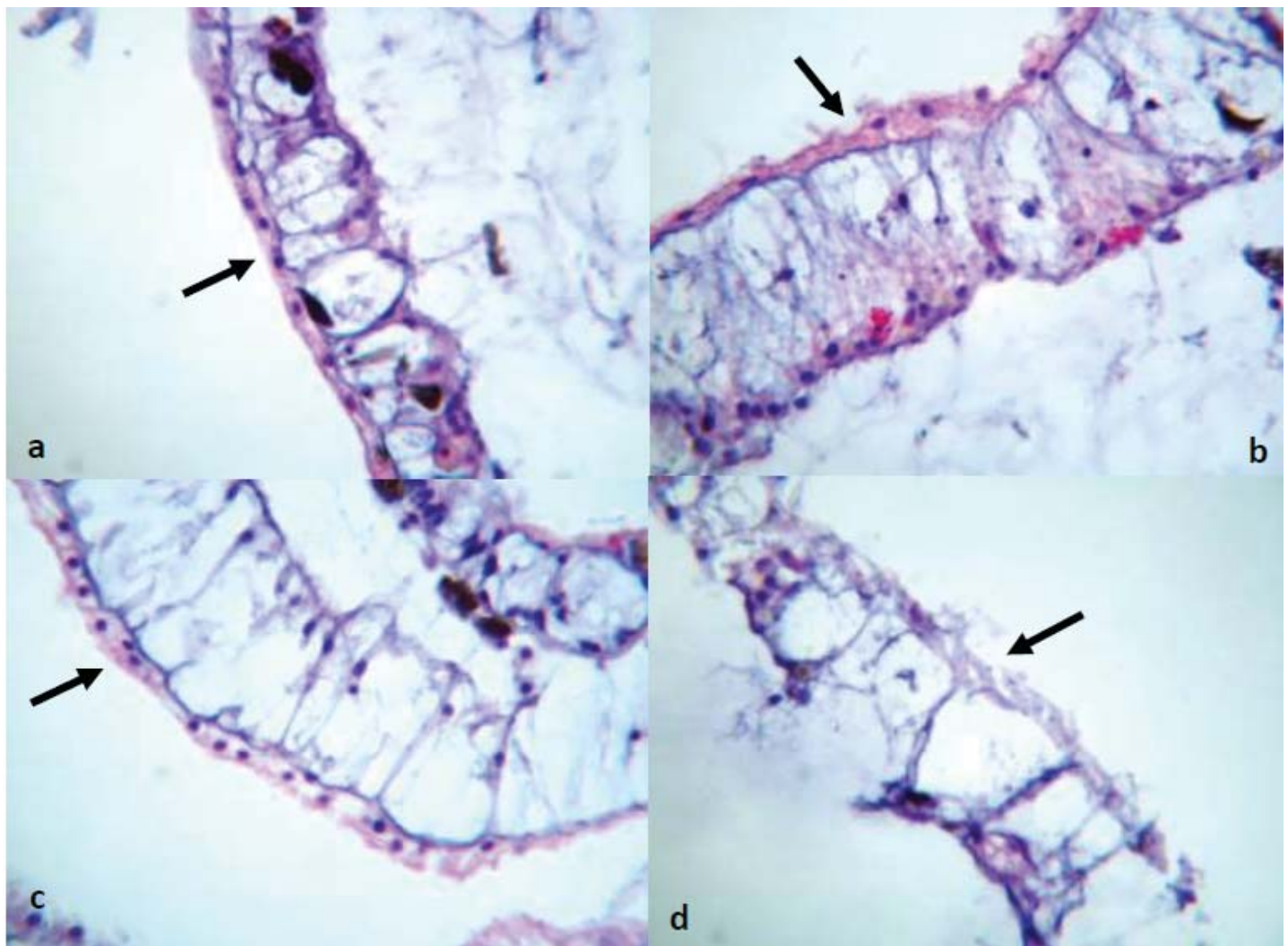


Figure 8.6. Examples of calicodermis condition. (a) Squamous calicoblasts (arrow), 40x. (b) Deeper calicodermis filled with fine acidophilic granules of calcification proteins (arrow), 40x. (c) Apical extensions on calicoblasts in more actively calcifying region (arrow), 40x. (d) Necrosis and lysing calicodermis (arrow), 40x.

exceed the limits to which the organism is adapted, causing stress (Stebbing, 1981). In addition, it is possible that some of the biotic commensals or parasites may succumb to abiotic stressors sooner than the coral (e.g., Cook *et al.*, 1990; Williams and MacKenzie, 2003). This study provided new data that will be useful in understanding these inter-relationships, although caution in their interpretation is warranted at this time because so little is known.

The first apicomplexan parasite discovered in the scleractinian corals was described by Peters (1984b). Upton and Peters (1986) identified one of the tissue parasites as a coccidian and this included oocysts found in the lobes of the mesenterial filaments of *P. astreoides*. Later, sporozoites that hatched from the oocysts were detected in the gastrodermis of other samples of this species and in other corals. Dr. Upton had proposed a life cycle in that paper for this parasite, suggesting that it might continue to cycle within the coral host and not have an alternate host, although

whether this is true is unknown. What has been identified in the current STEER *P. astreoides* samples appears to be a different endoparasite. Oocysts similar to the ones originally found were present in some of the samples in the lobes of the mesenterial filaments, but might be larger in size. A single-celled elongated organism with eosinophilic cytoplasm was detected in the basal body wall gastrodermis of some samples, never seen in the surface body wall. The most unusual related stage also found in the basal body wall was not single-celled, but was a chain of four cells, with one end cell resembling the single-celled elongated organism. The other three cells had prominent nuclei (same size and nuclear details as the single-celled organism's nucleus) and appeared to be filled with circular vacuoles, possibly containing lipid. These were identified as "sporozoan" for data collection. Although gregarines, also apicomplexan parasites, have a two-cell stage, they also have a thick-walled sporocyst stage, which was not seen in these samples.

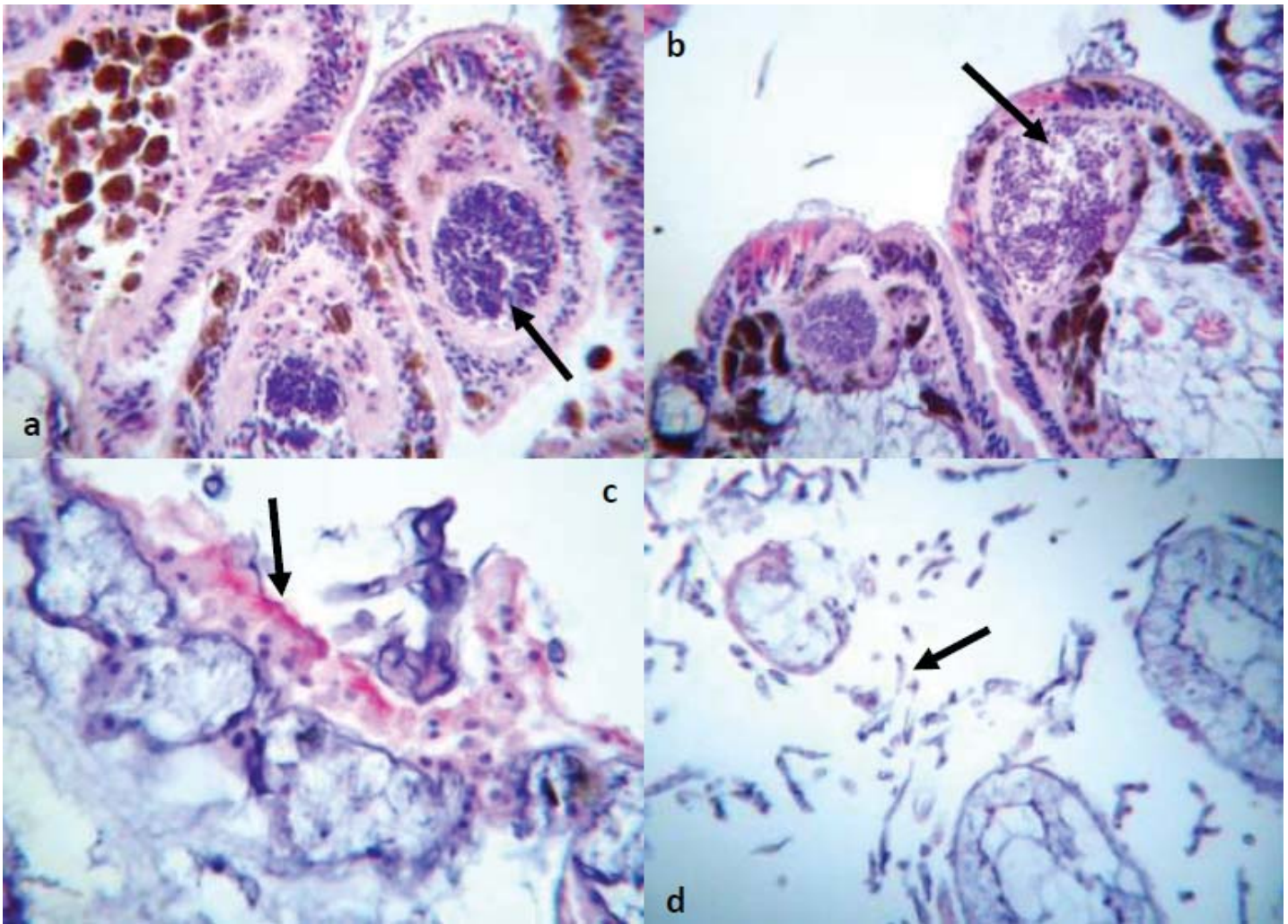


Figure 8.7. Examples of bacterial aggregates in mesoglea of tentacles (a and b), 25x; calicodermis reaction to suspect fungal hyphae (c), 40x; endolithic algae or fungi where skeleton was removed along basal gastrovascular canals (d), 10x.

A photomicrograph was sent to Dr. Nathan Kirk, Oregon State University, who has done extensive studies of the coral coccidia for his dissertation. He researched this question and found a couple of references to septate *Eugregarines* that form chains while in syzygy. He noted that “The two taxa are *Hirmacystis* and *Nematopsis* spp. *Hirmacystis polymorpha* can form chains of 10–15 individuals in a row in syzygy. Interestingly, *Nematopsis* (a genus that you detected in *Porites* samples in the 1984 paper) can also form chains (straight or forked) of up to 10 individual trophozoites prior to reproduction. However, the cartoon pictures from the illustrated guide do NOT look like the image you sent me. However, the zoite in the far left looks almost exactly like the pictures from your 1986 paper with Steve Upton. There are very few cases of coccidians undergoing syzygy (where they form chains) so that is an extremely interesting result as well if these are truly coccidians.” The molecular analyses he performed of numerous coral species tissue samples “suggested that some of these apicomplexans are quite different from each other.”

However, due to their numbers and movement through the basal body wall gastrodermis (Figure 8.8c), these organisms are capable of causing damage to the coral. It is possible that they, and the fungi, help to break down the basal body wall cells that the coral can no longer support nutritionally (Figure 8.9). Although how the coral tissue remains on the surface of the skeleton—a proposed mechanism is that it detaches from the skeleton and moves up—perhaps in the perforate corals the deepest cells simply die as those of the surface epithelia proliferate to continue forming the polyps and coenenchyme. Bacteria may contribute to this (Figure 8.10a). Some of the brooded planula larvae contained one or a few of the sporozoites in their lipid-filled gastrodermis, indicating that these may be acquired at the same time as the zooxanthellae from the parent’s tissue. Dr. Kirk noted he had found evidence of larval uptake of coccidians in his molecular studies, but he has not done histopathological examinations. More work will need to be done on this, of course, but this sample set is valuable for apicomplexan research.

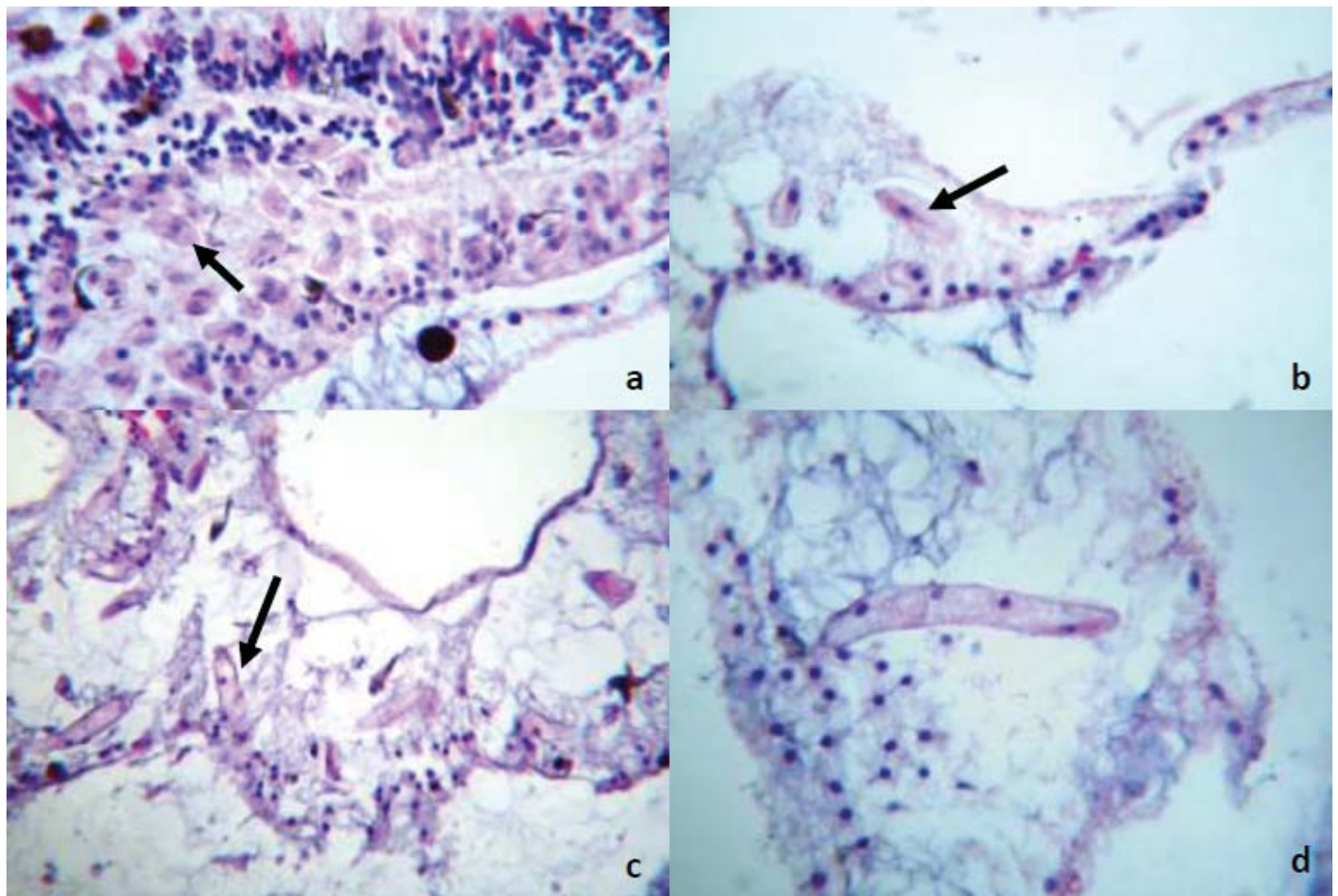


Figure 8.8. Examples of apicomplexan parasites of *P. astreoides*. (a) Suspect oocysts in lobes of mesenterial filaments (arrow), 40x). (b) Suspect sporozoite stages in basal body wall gastrodermis, 40x). (c) Numerous sporozoan stages in degraded basal body wall gastrodermis. (d) Suspect 4-cell sporozoan stage, 40x.

Several other structures that have been found previously or were new observations were also present in these samples, including suspect rickettsia-like organisms (RLOs) that were identified by sequencing only as 90% similar to an organism in the Rickettsiales (Casas *et al.*, 2004), a circular cell containing up to four nuclei (“unknown cell”) (Figure 8.10c), a metazoan that has been consistently seen next to the calicodermis deep in the coral tissue (Figure 8.10c) (Peters, unpub. observ. of other *Porites* from the Caribbean), and clusters of multiple single cells having an eosinophilic cytoplasm and minute basophilic particles but no definitive nucleus apparently residing in spaces where septal skeleton should have been (Figure 8.10d). The latter cells have been found in other corals (Peters 1978, Renegar *et al.*, 2008), but their true nature has not been investigated. Gonad development and brooding of planula larvae varied among the sites (Figure 8.11). However, this observation must also be cautiously interpreted. Gonad development might be affected by exposure to toxicants (e.g., Peters *et al.*, 1981), but location

of the sample in the colony can also affect whether gonads will be present. Chornesky and Peters (1987) noted that colony age and size can affect this, and younger polyps on the margins of colonies or small colonies can lack gonad development. Thus, the results must be compared with the size of the colonies and location of tissue sampled to be sure they represent what was happening at each site. Only females and hermaphrodites were found in the STEER, consistent with previous research on this species. Numbers of planulae larvae present varied with the site.

Summary statistics for all of the parameters examined in this study grouped by stratum are presented in Table 8.2. A summary of observations for each sampling site is presented below:

Stratum 1: Site MLC 0

Samples 12-180, 12-181, 12-182: 12-180 had algae growing underneath the coral margin, 12-181 had larger polyps and more space between the polyps on the larger piece,

12-182A was broken in sampling, 12-182B had boring sponge. This location was closest to the St. Thomas municipal dump. The general condition of these colonies was good to fair, with minimal to moderate surface body wall ruptures and moderate to severe lysing of gastrodermal mucocytes along the oral surface. These colonies also had mild to marked hypertrophy and numbers of epidermal mucocytes. One colony had diffuse single cell necrosis or apoptosis characterized by fine basophilic particles in

the bases of epidermal and gastrodermal cells. This same colony had minimal changes in the calicodermis, whereas the other two had moderate to marked reduced acidophilic

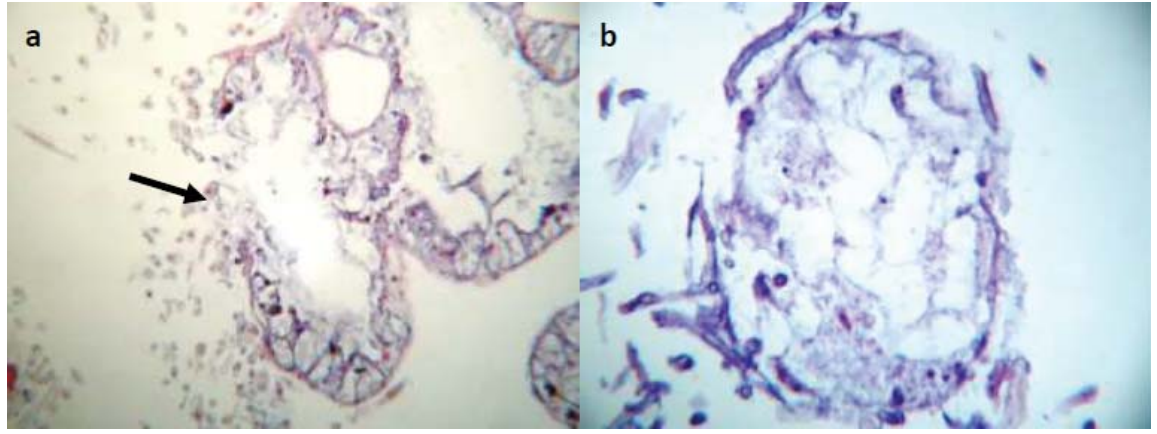


Figure 8.9. Examples of apparent destruction of the basal body wall of most aboral gastrovascular canals in the tissue layer of the perforate coral, *P. astreoides*. (a) Fungal hyphae next to break in basal body wall (arrow), 10x. (b) Necrosis and lysing of calicodermis, mesoglea, and gastrodermis adjacent to suspect fungal hyphae, 40x.

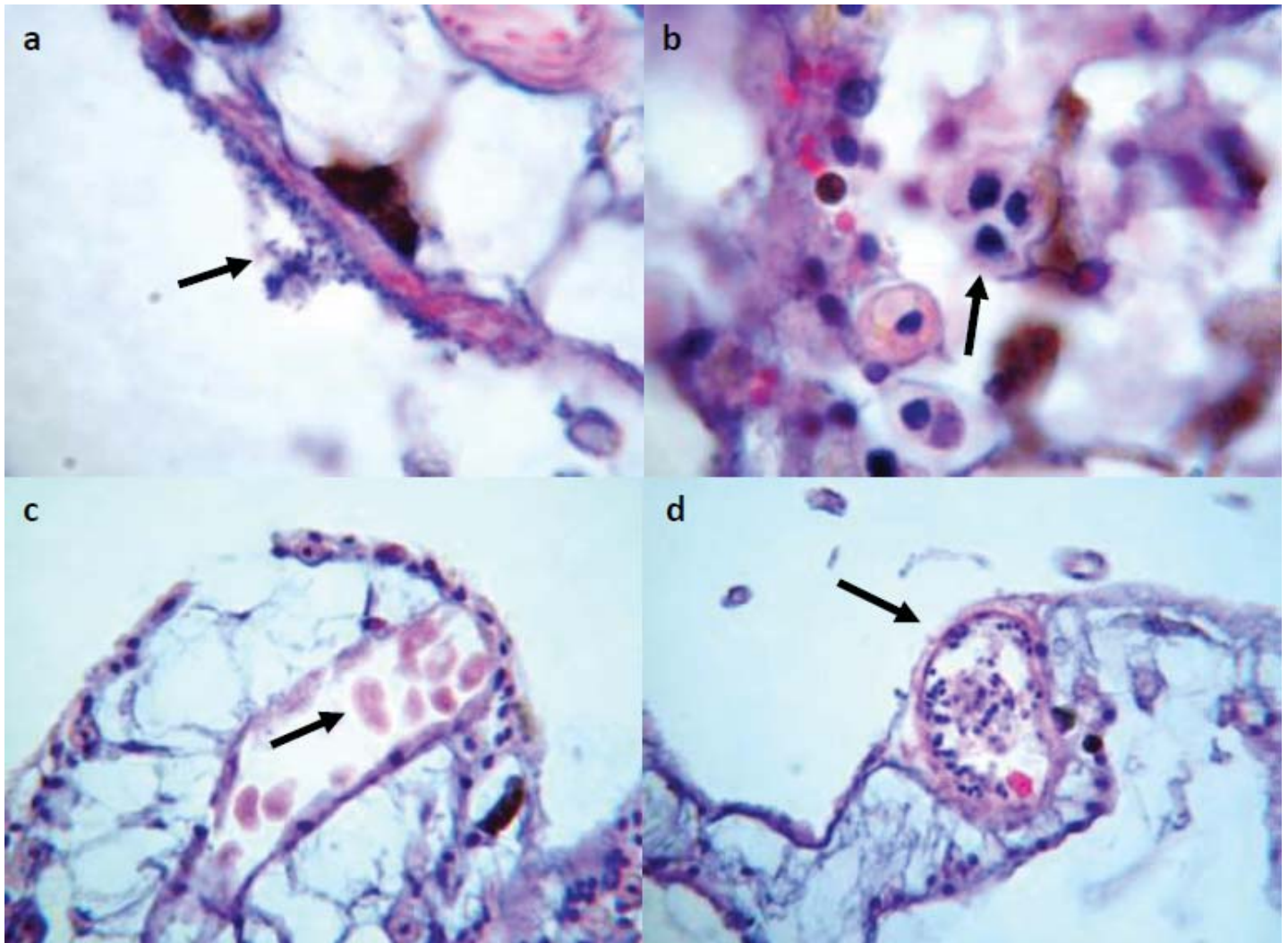


Figure 8.10. Examples of other unknowns present in the STEER samples. (a) Suspect bacteria on surface of calicodermis, 100x. (b) Unknown multinucleated cell, 100x. (c) "Blobs" in skeletal space of septal ridge on oral surface, 40x. (d) Metazoan adjacent to basal body wall, 40x.

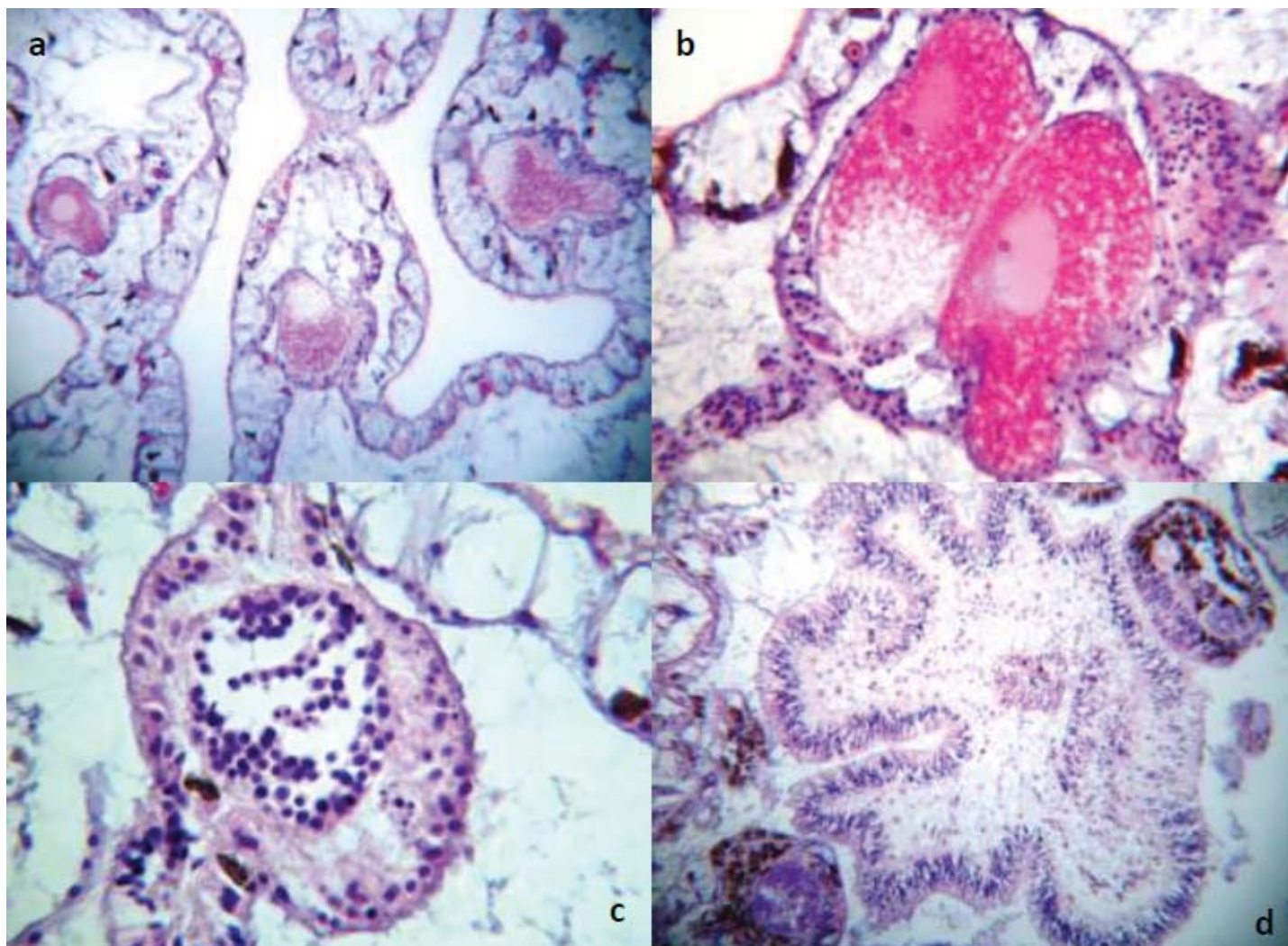


Figure 8.11. Examples of reproductive structures present in the STEER samples. (a) Multiple oocytes developing in mesenteries, 10x. (b) Two mature oocytes in mesentery, 25x. (c) Spermary in mesentery, 40x. (d) Planula larva in cross section through oral surface of polyp, 10x.

granules, lysing, and suspect bacteria or necrosis of this epithelium, but this was not correlated with the amount of endolithic suspected fungi or algae. None of the samples contained suspect apicomplexan oocysts, sporozoites, or sporozoans. Only one sample had a single early developing oocyte, none had spermaries or planulae. These samples were collected from a shallow site (4 ft depth) subjected to high temperatures, salinity changes, and land-based sources of pollution and the species was difficult to find at this site. The lack of gonad development and apicomplexan parasites (found at all other sites) is intriguing, but may not be related to contaminant exposures, as colony size or age (Chornesky and Peters, 1987), and isolation might also control these factors.

Stratum 2: Site HBI28P

Samples 12-198, 12-199, 12-200, 12-201, 12-202: All samples were grossly apparently healthy but with some

surface mucus and sediment, except 12-201A, which broke into fragments and was not included in the calculations. This location was farther east away from the Municipal Landfill, in shallow water of Cas Cay Mangrove Lagoon, close to a developed shoreline, seaward of the Compass Point Marina corridor. The general condition of these colonies was fair to poor, with minimal to severe, diffuse surface body wall ruptures and marked to severe, diffuse lysing of gastrodermal mucocytes along the oral surface. These colonies also had mild to severe hypertrophy and numbers of epidermal mucocytes. All subsamples had minimal to moderate single cell necrosis or apoptosis characterized by fine basophilic particles in the bases of epidermal and gastrodermal cells. 12-201 had small clusters of variably sized focal to multifocal black particles of unknown nature in the mucus of the surface gastrovascular canals (Figure 8.12). The calicodermis of all the subsamples had minimal to mild attenuation and reduced acidophilic

Table 8.2. Summary statistics for histopathological observations for each parameter for each stratum.

Parameter	Stratum 1			Stratum 2			Stratum 3			Stratum 4			Stratum 5		
	Mean	St.Dev.	Range	Mean	St.Dev.	Range	Mean	St.Dev.	Range	Mean	St.Dev.	Range	Mean	St.Dev.	Range
0 = Excellent, 1 = Very Good, 2 = Good, 3 = Fair, 4 = Poor, 5 = Very Poor															
General Microscopic Condition Coral (100x)	2.7	0.6	2.0–3.0	3.5	0.5	3.0–4.0	3.4	0.4	3.0–4.0	3.1	0.1	3.0–3.3	3.2	0.4	3.0–4.0
General Microscopic Condition Zooxanthellae (100x)	0.0	0.0	0.0–0.0	0.0	0.0	0.0–0.0	0.0	0.0	0.0–0.0	0.0	0.0	0.0–0.0	0.4	0.5	0.0–1.0
Relative Quantity (0 = None to 5 = Maximum)															
Melanin-like Granular Amoebocytes, Surface Body Wall	2.3	0.8	1.5–3.0	3.3	0.8	2.0–3.0	3.7	0.7	2.5–4.0	2.8	0.6	2.0–3.7	4.2	0.7	3.0–5.0
Melanin-like Granular Amoebocytes, Mesenteries and Mesenterial Filaments	2.5	1.5	1.0–4.0	3.2	0.8	2.0–4.0	1.6	0.8	1.0–2.5	1.9	0.2	1.5–2.0	2.2	1.3	1.0–4.0
Score = Severity or Intensity (1–5) x Extent (1–3)															
Suspect Sampling Damage, Surface Body Wall Ruptures	4.5	2.8	2.0–7.5	11.3	4.3	4.0–15.0	6.3	4.6	0.5–12.0	9.0	2.7	6.0–12.0	4.2	1.8	2.0–6.0
Suspect Sampling Damage, Lysing Gastrodermal Cells	12.0	3.0	9.0–15.0	13.2	1.6	12.0–15.0	9.3	0.7	9.0–10.5	11.0	1.4	9.0–12.0	10.5	1.5	9.0–12.0
Epidermis, Epidermal Mucocytes Hypertrophy	9.6	2.1	8.0–12.0	11.5	3.2	6.3–15.0	5.8	3.1	2.0–9.0	7.5	3.4	3.0–12.0	6.2	2.8	3.0–9.0
Epidermis, Necrosis or Apoptosis?	1.3	2.2	0.0–3.8	5.7	1.6	3.0–7.5	0.1	0.1	0.0–0.3	0.0	0.1	0.0–0.2	1.8	4.0	0.0–9.0
Gastrodermis, Black Particles in Released Mucus, Gastrovascular Canals	0.5	0.9	0.0–1.5	0.2	0.4	0.0–1.0	1.2	2.7	0.0–6.0	0.1	0.1	0.0–0.3	0.2	0.4	0.0–1.0
Gastrodermis, Necrosis or Apoptosis?	3.2	3.9	0.0–7.5	4.2	2.0	1.5–6.0	1.5	3.1	0.0–7.0	1.8	2.9	0.0–6.7	3.0	4.2	0.0–9.0
Calicodermis, Attenuation and Lack of Acidophilic Granules	6.8	3.8	2.5–9.0	3.2	1.6	2.0–6.0	7.9	5.9	0.5–15.0	8.3	1.8	6.0–10.5	12.9	2.5	9.0–15.0
Calicodermis, Suspect Bacteria on Apical Surface or Necrosis?	8.0	4.0	4.0–12.0	5.5	1.6	3.8–7.5	6.8	1.3	6.0–9.0	6.6	3.6	2.0–12.0	9.1	4.4	2.0–12.0
Skeleton, Endolithic Algae and Fungi	6.8	5.1	1.0–10.5	2.7	3.1	0.0–6.0	10.8	2.0	9.0–13.5	8.2	1.7	6.0–10.5	8.5	4.1	2.0–12.0
Apicomplexa, Oocysts	0.0	0.0	0.0–0.0	0.0	0.0	0.0–0.0	1.3	1.7	0.0–3.9	3.2	3.0	0.2–8.0	0.0	0.0	0.0–0.0
Apicomplexa, Sporozoites	0.0	0.0	0.0–0.0	1.2	1.3	0.0–3.0	6.1	3.1	2.0–9.0	1.6	0.9	0.0–2.0	0.5	0.9	0.0–2.0
Apicomplexa, Sporozoa	0.0	0.0	0.0–0.0	3.3	1.9	2.0–6.3	9.8	3.0	6.3–13.5	5.5	3.4	2.0–11.0	10.2	3.4	6.0–15.0
Bacterial Aggregates in Tentacles	0.3	0.6	0.0–1.0	3.0	1.0	2.0–4.0	4.7	1.2	3.0–6.0	2.8	1.0	1.5–4.0	4.0	2.9	2.0–9.0
Other Bacteria (rickettsia-like organisms in cnidoglandular bands)	0.0	0.0	0.0–0.0	1.8	1.7	0.3–4.0	0.3	0.7	0.0–1.5	0.8	0.8	0.1–2.0	0.0	0.0	0.0–0.0
Percent Containing															
Unknown Cells	0	0	0–0	0	0	0–0	0	0	0–0	0	0	0–0	20	45	0–100
Calicodermis/Skeleton Unknown Metazoan	0	0	0–0	0	0	0–0	0	0	0–0	80	45	0–100	0	0	0–0
Skeletal Space Containing "Blobs"	0	0	0–0	0	0	0–0	20	45	0–100	40	55	0–100	0	0	0–0
Gonads, Oocytes (Stages)	33	58	0–100	100	0	100–100	80	45	0–100	100	0	100–100	60	55	0–100
Gonads, Spermatocytes (Stages)	0	0	0–0	0	0	0–100	60	55	0–100	40	55	0–100	20	45	0–100
Planulae, Count	0	0	0–0	0.4	0.5	0–1.0	0.2	0.4	0.0–1.0	11.0	9.6	0.0–22.0	0.6	0.9	0.0–2.0

granules, and diffuse mild suspect bacteria or necrosis of this epithelium. Endolithic suspected fungi or algae were not detected in three of the subsamples and minimal to moderate levels in the others with limited in extent. Suspect apicomplexan developmental stages were found in all of the subsamples, but no oocysts, and minimal to mild focal infections of sporozoites and sporozoans; moderate, diffuse amounts of the latter in 12-199A. All of the subsamples contained oocytes in middle to mature development, two contained small spermaries, and none had planulae. These results suggest the colonies were under some stress leading to the single cell necrosis, but were still producing gonads.

Stratum 3: Site HBI23P

Samples 12-193, 12-194, 12-195, 12-196, 12-197: All samples were grossly apparently healthy (uniform covering of pigmented tissue and polyps) but with mild to moderate surface mucus and sediment, except 12-195B, which broke into fragments and was not included in the calculations. This location was nearshore in the St. James Reserve, downstream of the developed Jersey Bay watershed. The general condition of these colonies was good to poor, with no to marked severity of surface body wall ruptures and moderate to marked lysing of gastrodermal mucocytes along the oral surface. These colonies also had minimal to marked hypertrophy and numbers of epidermal mucocytes. Only one colony had focal minimal single cell necrosis or apoptosis of the epidermal cells and two had minimal, diffuse to mild, multifocal single cell necrosis of the gastrodermal cells. All of the colonies had degradation of the calicodermis, including marked to severe, diffuse attenuation, lack of acidophilic granules, and cell lysing in 12-194A, 12-195, and 12-197; and all had mild to moderate, multifocal to diffuse suspect bacteria or necrosis of this epithelium. They all had more severe and extensive proliferation of endolithic organisms in the skeleton. All of the samples contained suspect apicomplexan oocysts, sporozoites, or sporozoans; the latter were marked to severe, diffuse infections of the deep basal body wall gastrodermis. Two colonies lacked gonads, the others had developing oo-

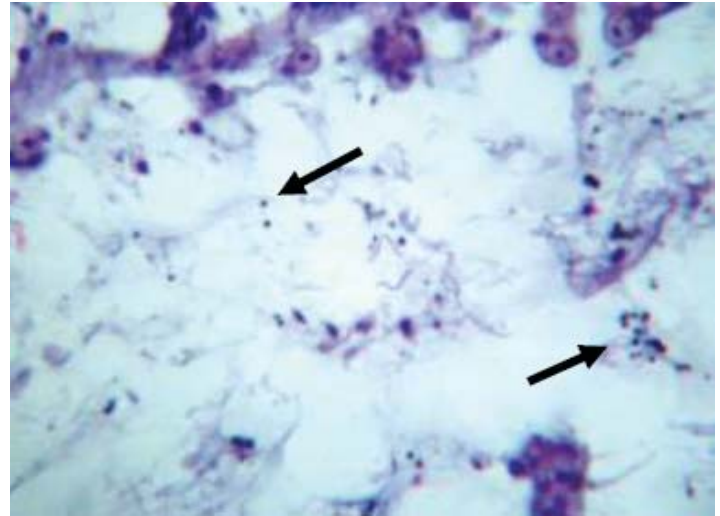


Figure 8.12. Example of black particles trapped in mucus secretions (arrows), 40x.

cytes or spermaries, but none contained planula larvae. The presence of hypertrophied mucocytes and minimal necrosis of the surface body wall suggests the sampled colonies were stressed.

Stratum 4: Site HBI15P

Samples 12-188, 12-189, 12-190, 12-191, 12-192: The cores from this site grossly appeared to be healthy except for mild to moderate mucus or sediment particles on the surfaces, but three subsamples (12-189B, 12-190B, 12-191A-2) were broken in fragments and not included in the calculations. Samples were collected from a hardbottom site farther offshore and deeper (42 ft) than the other stations (4–8 ft), at the entrance to Jersey Bay. The general condition of these colonies was fair to poor, with multifocal to diffuse, mild to marked surface body wall ruptures and moderate to marked lysing of gastrodermal mucocytes along the oral surface. One sample had minimal hypertrophy of epidermal mucocytes, the others had mild to marked hypertrophy and numbers of epidermal mucocytes. One colony had diffuse, severe necrosis or apoptosis of the gastrodermis, with minimal changes noted in two other colonies, and only one had multifocal minimal single cell

Table 8.3. Summary of results of the histopathological examinations, means of Specific Condition Scores Sum and percent of colonies with selected condition at each site.

Stratum	Mean Condition Scores \pm St. Dev.	Sampling Damage %	Epidermal Necrosis or Apoptosis %	Gastrodermal Necrosis or Apoptosis %	Calicodermis Degeneration %	Gonads and Larvae %	Apicomplexan Parasites %
1 (n=3)	36.2 \pm 3.6	100	33	67	100	33	0
2 (n=5)	37.3 \pm 5.5	100	100	100	100	100	100
3 (n=5)	51.1 \pm 10.5	100	20	40	100	80	100
4 (n=5)	42.8 \pm 5.8	100	20	40	100	100	100
5 (n=5)	35.0 \pm 5.8	100	20	40	100	60	100

necrosis of the epidermis. The calicodermis of all the colonies had diffuse, moderate to marked attenuation and lack of acidophilic granules, and most also had mild to marked suspect bacteria or necrosis, except the calicodermis had only minimal changes in 12-191. Mild to marked levels of endolithic organisms were noted in these samples throughout the deep tissue. All of the samples contained suspect apicomplexan oocysts, sporozoites, or sporozoans, with marked levels of oocysts in 12-190 and of sporozoans in 12-192. All of the colonies were developing gonads, but only one with spermaries was found; all but one colony also were brooding larvae—more larvae here than at any of the other sites. The sample with severe necrosis of the gastrodermis also had marked epidermal mucocyte hypertrophy, numerous suspect sporozoans in the deep gastrodermis and endolithic organisms in the skeleton, and one area of black particles in the gastrovascular canal mucus, but still contained developing oocytes and planula larvae. This illustrates the challenges in determining the health of a colony.

Stratum 5: Site HBI7P

Samples 12-183, 12-184, 12-185, 12-186, 12-187: Sampling damage was present on all of the cores collected at this site, with mucus and sediment on their surfaces, but otherwise were grossly apparently healthy. Subsamples 12-189B, 12-190B, and 12-191A-2 were broken and not included in the analyses. This sampling location was near shore, but the farthest east from MLC 0, in Great Bay. The general condition of these colonies was good to poor, with minimal to moderate surface body wall ruptures and moderate to marked lysing of gastrodermal mucocytes along the oral surface. These colonies also had mild to moderate hypertrophy and numbers of epidermal mucocytes. One colony (12-186) had moderate single cell necrosis or apoptosis characterized by fine basophilic particles in the bases of epidermal and gastrodermal cells (also seen in 12-183, mild). Two subsamples had black particles in released mucus. The calicodermis of all of the subsamples had marked to severe attenuation, lack of acidophilic granules, and lysing of the epithelium, and multifocal to diffuse, minimal to marked bacteria or necrosis occurring. All of the subsamples had mild to marked amounts of endolithic algae and fungi. Suspect apicomplexan sporozoans were found in

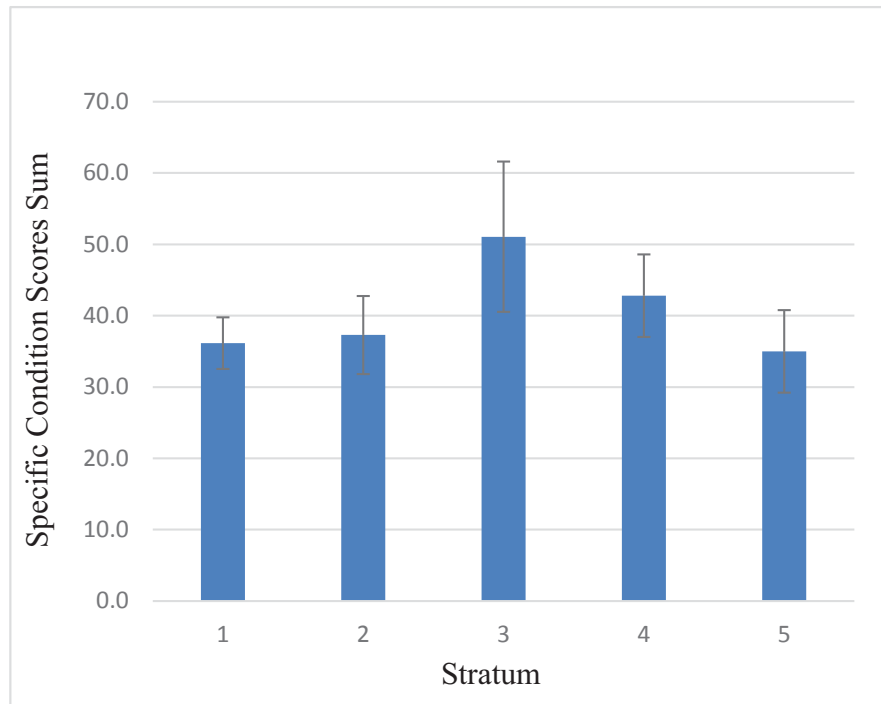


Figure 8.13. Comparison of means of Specific Condition Scores Sum with standard deviations among the five strata.

all the subsamples, most numerous in 12-186 and 12-187; no or minimal amounts of oocysts and sporozoites. Only 60% of the samples had developing gonads, one colony with spermaries, and 40% had planulae in their polyps. The sporozoan infections of the gastrodermis might be most responsible for the condition of these colonies.

The means of the Specific Condition Scores Sums of the samples, which ranged from 30–61.8 (out of a possible total of 180 points for worst severity of 5 and most extensive distribution of 3 for all 12 parameters) are presented by site and observations of percentage of affected colonies for selected data are summarized in Table 8.3. A comparison of the mean Specific Condition Scores Sums by Stratum is presented in Figure 8.13. Based on these summed scores, the samples from Stratum 3 were in worse condition than the other sites. A single-factor analysis of variance for those data indicated significant differences among the strata ($p = 0.011$), with t-tests revealing the differences to be between Stratum 1 or 5 and 3. However, the Kruskal-Wallis analysis barely revealed these differences ($p = 0.057$). Stratum 2 samples were significantly different from all other strata for epithelial necrosis or apoptosis, but at low intensities.

8.4. CONCLUSIONS

In conclusion, the most widely seen effect across all of the sites was the lysing of the gastrodermis along the oral gastrovascular canals and cavities, filling these lumens with mucus and also epidermal ruptures releasing mucus and

other lysed cells and zooxanthellae onto the subsample surface. Similar lesions have been observed in other species, but not to such a level of severity and extent.

The impact of hammering the stainless steel coring tube into the coral colony to remove the tissue and skeleton biopsy probably needs to be reconsidered and a method with less vibration used (e.g., more manual pressing of the coring tube, less intense pounding, or a diamond-tipped rock coring device on a pneumatic drill). The presence of hypertrophied mucocytes and single cell or segmental necrosis or apoptosis in some of the colonies, and reproduction capabilities indicate that some of the sampled corals might be experiencing stress, but associating the effects with exposures will require additional observations and measurements of environmental parameters. The presence of suspect apicomplexan parasites, particularly the sporozoan stage that was burrowing through the gastrodermis of the deep basal body wall, could be contributing to colony condition, as well, because that epithelium was becoming vacuolated and necrotic, potentially in concert with proliferation of fungi in the skeleton.

Because of lack of knowledge of how the polyps remain on the surface of the skeleton in these nonperforate corals, perhaps a cyclical breakdown of deep tissue occurs as the surface tissues continue to grow. More observations of growth patterns in the scleractinian corals, including the histopathology of the tissues, is needed to examine this question.

The STEER *P. astreoides* colony samples are valuable because the effects of other environmental stressors, especially biotic associations of parasites and endolithic organisms, were detected by light microscopy and will assist in interpreting the results of toxicology analyses.

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CHAPTER 9: AN ASSESSMENT OF NUTRIENTS, SEDIMENTATION, AND TOTAL SUSPENDED SOLIDS (TSS) IN THE ST. THOMAS EAST END RESERVES (STEER)

Anthony S. Pait¹, Francis R. Galdo, Jr.², S. Ian Hartwell¹, Andrew L. Mason¹, Dennis A. Apeti¹, Christopher F. G. Jeffrey^{1,3}, Anne M. Hoffman⁴, and Simon J. Pittman^{1,5}

¹NOAA/NOS/National Centers for Coastal Ocean Science, Center for Coastal Monitoring and Assessment (CCMA), 1305 East/West Highway, Silver Spring, MD 20910

²The University of the Virgin Islands, St. Thomas, USVI

³CSS-Dynamac Consolidated Safety Services, Fairfax, VA 22030, under CSS-Dynamac Contract #EA-133C-14-NC-1384

⁴The Nature Conservancy, St. Thomas, USVI

⁵The Marine Institute, Plymouth University, United Kingdom

9.1. INTRODUCTION

This chapter contains the results of the monthly monitoring of nutrients and total suspended solids (TSS) in the water column, along with sedimentation using sediment traps, as part of the project in the STEER. The monthly monitoring was carried out by project partners at the University of the Virgin Islands (UVI) from 2012 through 2013.

Nutrients were identified as a major environmental concern in the STEER Management Plan (STEER, 2011). While dissolved nutrients are essential to productivity in aquatic systems, an overabundance of nutrients can help trigger macroalgae and phytoplankton blooms, resulting in degradation of water quality and habitat. In coral reef systems, algal blooms can lead to algae out-competing and then smothering juvenile and adult corals, ultimately resulting in the loss of those corals (Fabricius, 2005; D'Angelo and Wiedenmann, 2014; Box and Mumby, 2007).

It has also been shown that nutrients can have direct effects on corals. Ammonium and phosphate in parts per billion ($\mu\text{g/L}$ or ppb) concentrations can impact fertilization success (Harrison and Ward, 2001), while nitrate has been shown to decrease calcium deposition in corals (Marubini and Davis, 1996).

Sediments can be deposited on coral reefs as a result of surface water transporting eroded soils, as might occur during a storm, or through the resuspension of bottom sediments. Sedimentation has been shown to impact coral reefs (Fabricius, 2005; Burke *et al.*, 2011; Waddell *et al.*, 2005). The deposition of sediments in reef areas can act to smother corals and physically abrade coral tissues. At the

very least, sediments “raining” down on corals results in the organisms having to expend energy to remove sediment particles, meaning there is less energy available for other functions including growth and reproduction. Elevated sedimentation has been linked to less coral cover, lower coral diversity and recruitment, along with lower growth and calcification rates (Fabricius, 2005; Weber *et al.*, 2012;

ISRS, 2004; Rogers, 1990). Like nutrients, the input of sediment was identified as a major threat in the STEER Management Plan (STEER, 2011).

The presence of total suspended solids or TSS in the water column acts to decrease the amount of light available for corals as well as seagrasses. Nemeth and Nowlis (2001) suggested that excessive levels of suspended sediments led to increased coral bleaching, specifically along the north shore of St. Thomas.



Diver installing a sediment trap in the STEER.

9.2. METHODS

The locations for the monitoring sites (Figure 9.1) were selected non-randomly by project partners at the University of the Virgin Islands (UVI) and the USVI Department of Planning and Natural Resources (DPNR), in order to target potential sources of land based pollution. These sites were used to assess terrestrial inputs from the surrounding watershed, including residential areas surrounding Benner Bay, and the Bovoni Landfill, which is adjacent to Mangrove Lagoon. Samples for nutrient analysis, TSS and sedimentation were collected monthly from five targeted sites from January 2012 to November 2013. A sixth site, at Little St. James (LSJ, Figure 9.1) Island, was added and then sampled beginning in March 2012. The methods used for the collection and analysis are described below.

Nutrients

At each site, duplicate samples were collected for the analysis of dissolved nutrients. Nitrile gloves were worn by personnel, in order to prevent contamination of the water samples. In the field, samples were kept in the dark and then filtered into acid-washed 125 ml high density polyethylene (HDPE) bottles, using 60 ml syringes equipped with disposable Millipore Sterivex 0.22 μm filter units. The syringe was first rinsed with several full volumes of site water. Next, the syringe was refilled, the Sterivex filter attached, and a full volume of site water was pushed

column for reduction of nitrate to nitrite. Orthophosphate was measured using the methodology of Bernhardt and Wilhelms (1967), with the modification of hydrazine as a reductant. Silicate determination was accomplished following the methods of Armstrong *et al.* (1967) using stannous chloride.

Ammonium analysis was based on the method of Harwood (Harwood and Kuhn, 1970) using dichloroisocyanurate as an oxidizer. Urea was measured using diacetyl-monoximine



Figure 9.1. Sampling sites for nutrients, sedimentation, and TSS along with the STEER boundary.

through the filter. The sample bottles were rinsed three times with filtered site water. The filter was removed and the syringe refilled with site water. After replacing the filter, the syringe/filter process was repeated until the bottles were filled, leaving adequate headspace to allow for expansion during freezing. Samples were then immediately packed on ice for transport to UVI. On return to the laboratory, the water samples were frozen until shipped.

Water samples were analyzed by the Geochemical and Environmental Research Group (GERG) at the Texas A&M University, through a subcontract with TDI-Brooks, International. The following is a brief summary of methods used for the analysis of nutrients in the water samples. Nitrate and nitrite analyses were based on the methodology of Armstrong *et al.* (1967) and use a ground cadmium

and micarbozide. Total nitrogen and total phosphorus concentrations were determined after an initial decomposition step. This method involves persulfate oxidation while heating the sample in an autoclave (Hansen and Koroleff 1999). After oxidation of the samples, nutrient determinations were conducted on a Technicon® II analyzer.

As the water samples were filtered for this project, reported values are for dissolved nutrient species. Proposed nutrient thresholds for coral reefs for dissolved organic nitrogen (DIN), and for orthophosphate (also referred to as soluble reactive phosphorus or SRP), are compared with the results from the STEER.

Table 9.1. Location of sites and mean water quality values from STEER sites.

Site Name	Site Code	Latitude (DD)	Longitude (DD)	Salinity (ppt)	Turbidity (NTU)	pH	Depth (m)	Chlorophyll a (µg/l)	DO (mg/l)	Temperature (°C)
Mangrove Lagoon	ML	18.31385	-64.87988	35.6 ±0.3	4.07 ±0.71	8.09 ±0.03	1.3 ±0.1	1.86 ±0.17	6.79 ±0.13	29.3 ±0.3
Benner Bay	BB	18.31670	-64.8674	36.0 ±0.1	10.7 ±1.4	8.13 ±0.03	1.2 ±0.1	1.12 ±0.08	6.18 ±0.12	29.3 ±0.2
Rotto Cay	RC	18.31331	-64.86423	35.8 ±0.1	1.41 ±0.20	8.13 ±0.02	2.1 ±0.1	0.37 ±0.03	6.37 ±0.16	28.6 ±0.2
Cowpet Bay	CB	18.31487	-64.84267	35.9 ±0.1	0.91 ±0.13	8.17 ±0.02	1.7 ±0.1	0.32 ±0.02	6.54 ±0.03	28.3 ±0.2
Saint James	SJ	18.30302	-64.83671	35.9 ±0.1	1.03 ±0.16	8.16 ±0.02	1.8 ±0.1	0.28 ±0.03	6.42 ±0.02	28.0 ±0.1
Little St. James	LJ	18.30069	-64.83003	35.8 ±0.1	1.52 ±0.13	8.15 ±0.02	1.6 ±0.1	0.28 ±0.03	6.51 ±0.02	28.4 ±0.1

Values are ± standard error. Abbreviations: DD, decimal degrees; ppt, parts per thousand; NTU, nephelometric turbidity units; m, meters; DO, dissolved oxygen; mg/l, milligrams/ liter; °C, degrees Celsius. Note that depth is the water depth at which the water quality measurements were made, and not the depth of the site.

Sedimentation

Sediment trap arrays were deployed at five sites located throughout the STEER, and included Great St. James (SJ), Cowpet Bay (CB), Rotto Cay (RC), Benner Bay (BB), and Mangrove Lagoon (ML). In March 2012, at the request of DPNR, an additional station was added near Little St. James Island (LSJ), to address concerns related to major ongoing construction activities on this island. GPS coordinates were recorded for each station, and are shown in Table 9.1.

Each sediment trap consisted of a two-inch diameter PVC cylinder, sealed at one end, with a minimum height-to-diameter ratio of 4:1, the same as those already in use by the University of the Virgin Islands (Smith *et al.*, 2008). This height:diameter ratio has been shown to minimize the resuspension and escape of trapped sediments under normal flow conditions (Smith *et al.*, 2008). The cylinders were deployed vertically, affixed to metal posts at a height of approximately 60 cm above the seafloor, and approximately 30 cm for very shallow sites (Benner Bay and Mangrove Lagoon). The cylinders were affixed to the posts with combinations of Velcro and plastic zip-ties, allowing for monthly exchange of the traps. Three replicate cylinders were spaced approximately one meter apart, in order to minimize the effects of water flow disturbances between replicates, thus helping to ensure independence of replicate samples (Butman, 1984).

At four sites (Little St. James, St. James, Cowpet Bay, and Rotto Cay), traps were exchanged by divers using SCUBA. For trap arrays located in the shallowest sites (Benner Bay and Mangrove Lagoon), the exchange was generally accomplished by wading, taking care to minimize disturbance to bottom sediments. During sediment trap exchanges, each trap was carefully removed from its post, and sealed with a tight-fitting cap. Due to the fact that a variety of small fish and invertebrates were attracted to the refuge provided by

the sediment traps, cylinders were checked for the presence of marine organisms prior to being capped.

Replacement traps were cleaned thoroughly between deployments, and rinsed just prior to deployment at each site. Newly deployed traps were visually inspected just before divers departed a site, in order to ensure that no sediment entered during the exchange process. If sediment was observed in a newly deployed trap, the cylinder was removed from the post, cleaned, and replaced.

Beginning in April 2012, Pettit (Kop-Coat, Inc.) inflatable boat anti-fouling paint was applied to the upper 5 cm of the traps due to concerns that fouling of trap apertures at several sites, could significantly affect trap performance.

Efforts were taken to ensure that traps were retrieved and replaced at all sites within a period of no more than 24 hours, however on several occasions, extenuating circumstances (e.g., storms), resulted in delays of several days between sites. Such occasions have been noted and were taken into account when calculating sedimentation rates. Following retrieval, the sealed sediment traps were stored upright until decanted and filtered.

Sediment samples were decanted, filtered, rinsed with freshwater to remove salts, then dried completely at 70 °C. Samples were then cooled to room temperature in a desiccator, and weighed to the nearest 0.001 g to obtain total dry weight of accumulated sediments for each trap.

An analysis of sediment composition (organic/carbonate/terrigenous fractions) was carried out using loss on ignition (LOI) at 550 °C and 950 °C, following the techniques described by Heiri *et al.* (2001). To determine organic content, homogenized sediment sub-samples were placed into clean, labeled, and pre-weighed porcelain crucibles. Crucibles and sediments were then dried at 105 °C, cooled,

and weighed to the nearest 0.001 g prior to combustion at 550 °C in a muffle furnace (Fisher Scientific Isotemp 550 series). Samples were held at 550° C for four (4) hours, then cooled to room temperature. Samples were again dried in a desiccator, and re-weighed post-combustion. To determine carbonate content, the subsamples were returned to the muffle furnace and combusted at 950 °C for two (2) hours. After cooling to room temperature, the weighing process was repeated a final time to obtain dry weight post-combustion. Organic and carbonate contents of samples were calculated by the following equations, respectively:

$$\% \text{LOI } 550^{\circ} \text{ C} = ((\text{DW } 105^{\circ} \text{ C} - \text{DW } 550^{\circ} \text{ C}) / \text{DW } 105^{\circ} \text{ C}) * 100$$

$$\% \text{LOI } 950^{\circ} \text{ C} = ((\text{DW } 550^{\circ} \text{ C} - \text{DW } 950^{\circ} \text{ C}) / \text{DW } 105^{\circ} \text{ C}) * 100$$

Total Suspended Solids

At each site, a one (1) liter water sample was collected for analysis of total suspended solids (TSS) each month. A clean, pre-labeled bottle was uncapped and submerged approximately one half meter below the water surface. The bottle was filled and rinsed out twice with site water, then filled, capped, and placed on ice for transport back to UVI. The water samples for TSS were stored refrigerated for a period of no longer than one month from collection until analysis. Each one liter sample was filtered through a pre-weighed glass fiber filter mounted in a suction flask apparatus. (if less than one liter was used, the exact volume of water filtered was noted). The filtered sample was rinsed several times with deionized water in order to remove salts, then dried at 105° C, and weighed to the nearest 0.001 g. Calculation of TSS was as follows:

$$\text{TSS (mg/L)} = ([A-B] * 1000) / C$$

Where A = End weight of the filter in grams (g)
 B = Initial weight of the filter in grams (g)
 C = Volume of water filtered in liters (l).

Statistical Analysis

The data collected were analyzed using JMP® statistical software. Because the data were not normally distributed, nonparametric tests (e.g. Spearman's nonparametric multivariate correlation) were used. When needed, pairwise comparisons were made using the Wilcoxon Rank-Sum Method.

9.3. RESULTS AND DISCUSSION

Water Parameters

In addition to the site location information for the monthly monitoring, Table 9.1 also contains a summary of the water parameters measured. Note that depth refers to the depth made for the water quality measurements, and not the depth of the water at the site. More detailed results for these and other parameters measured in this part of the project can be found in Pait *et al.* (2015).

The mean (average) turbidity was highest at the Benner Bay site (10.7 NTU). A nonparametric (Wilcoxon Rank-Sum) comparison between sites

indicated that the turbidity at the Benner Bay site was significantly ($p < 0.0001$) higher than all the other sites sampled monthly in the STEER, including Mangrove Lagoon.



Sediment traps deployed in the STEER. Larger cylinder is a passive water chemistry sampler used in another part of the project (see Chapter 6).

For chlorophyll a, the mean for Mangrove Lagoon (1.86 µg/L) was significantly ($p < 0.0001$) higher than all other sites, including Benner Bay ($p = 0.0005$), using a Wilcoxon nonparametric pairwise comparison test. Nonpoint source inputs from surrounding residential areas, along with inputs from the horse racetrack, which are adjacent to the northern border of Mangrove Lagoon, are likely contributing nutrients, leading to increased phytoplankton growth (and correspondingly higher chlorophyll a concentrations) seen in Mangrove Lagoon. For pH, salinity, and temperature, the average readings between sites were fairly similar.

Nutrients

A summary of the results from the nutrient monitoring can be seen in Table 9.2, and in Figures 9.2 to 9.8. The intervals in the maps were derived using the Jenks Natural Breaks Classification Method. In general, higher nutrient concentrations were found in the western portion of the study area, specifically in Mangrove Lagoon and Benner Bay.

Differences by Site

Nonparametric analyses indicated that ammonium (NH_4^+), nitrite (NO_2^-), and dissolved inorganic nitrogen (DIN) varied significantly by site. The variation of the combined nitrate/nitrite (sum of NO_3^- and NO_2^-), however, was not significant. Mangrove Lagoon and Benner Bay had significantly higher ($p < 0.05$) mean concentrations of NH_4^+

Table 9.2. Summary statistics for nutrients measured in the STEER. All units are expressed as mg/L N or mg/L P.

Nutrient	Mangrove Lagoon			Benner Bay			Rotto Cay		
	Mean	Median	Max	Mean	Median	Max	Mean	Median	Max
NO ₃ ⁻ + NO ₂ ⁻	0.04 ±0.023	0.016	0.422	0.013 ±0.004	0.008	0.075	0.007 ±0.002	0.004	0.022
NH ₄ ⁺	0.020 ±0.003	0.017	0.055	0.016 ±0.003	0.011	0.042	0.007 ±0.002	0.002	0.039
Urea	0.004 ±0.001	0.003	0.017	0.007 ±0.003	0.005	0.056	0.004 ±0.001	0.003	0.023
DIN	0.061 ±0.024	0.031	0.442	0.029 ±0.005	0.022	0.08	0.014 ±0.003	0.005	0.059
DON	0.305 ±0.048	0.312	0.652	0.280 ±0.040	0.239	0.62	0.258 ±0.037	0.289	0.467
Total N	0.366 ±0.060	0.347	1.094	0.309 ±0.040	0.286	0.701	0.272 ±0.037	0.305	0.526
HPO ₄ ⁼	0.004 ± 0.001	0.006	0.012	0.006 ±0.001	0.006	0.012	0.005 ±0.001	0.004	0.011
Total P	0.012 ±0.001	0.011	0.027	0.012 ±0.001	0.011	0.022	0.017 ±0.006	0.009	0.116

Nutrient	Cowpet Bay			Saint James			Little Saint James		
	Mean	Median	Max	Mean	Median	Max	Mean	Median	Max
NO ₃ ⁻ + NO ₂ ⁻	0.007 ±0.002	0.005	0.021	0.009 ±0.002	0.004	0.024	0.007 ±0.002	0.003	0.024
NH ₄ ⁺	0.006 ±0.002	0.003	0.022	0.006 ±0.001	0.004	0.017	0.006 ±0.001	0.004	0.015
Urea	0.003 ±0.001	0.003	0.01	0.003 ±0.001	0.004	0.007	0.003 ±0.001	0.002	0.008
DIN	0.014 ±0.003	0.011	0.034	0.015 ±0.003	0.008	0.036	0.013 ±0.003	0.007	0.036
DON	0.251 ±0.032	0.257	0.502	0.255 ±0.037	0.267	0.438	0.297 ±0.036	0.36	0.45
Total N	0.265 ±0.031	0.262	0.504	0.270 ±0.036	0.276	0.469	0.310 ±0.036	0.385	0.463
HPO ₄ ⁼	0.005 ±0.001	0.005	0.011	0.005 ±0.001	0.007	0.013	0.005 ±0.002	0.003	0.018
Total P	0.013 ±0.003	0.009	0.047	0.011 ±0.002	0.008	0.05	0.009 ±0.001	0.008	0.022

Abbreviations: NO₃⁻, nitrate; NO₂⁻, nitrite; NH₄⁺, ammonium; DIN, dissolved inorganic nitrogen; DON, dissolved organic nitrogen; HPO₄⁼, orthophosphate. Mean is ±SE.

and DIN than at Cowpet Bay, Rotto Cay, Saint James, and Little St. James. For nitrite, Benner Bay had significantly higher mean values than all other sites, except for Mangrove Lagoon. Finally, Mangrove Lagoon and Benner Bay did not differ significantly from each other for any of the nutrient species analyzed, indicating elevated levels of the nutrients analyzed at both of these locations. Figures 9.2 to 9.8 show the mean and maximum values recorded at the sites during the monthly sampling. From these figures as well, it can be seen that the Mangrove Lagoon and Benner Bay sites, had higher concentrations (both the mean and maximum values), for a number of the nutrients analyzed. The watershed surrounding the western portion of the study area includes the higher density residential areas of Estate Bovoni and Anna's Retreat, along with the active Bovoni Landfill, and are likely contributing nutrients to the STEER. In addition, there appear to be a number of live-aboard boats in Benner Bay, which are also potential sources of nutrients. The east to west gradients seen with nitrogen compounds was not seen with orthophosphate and total phosphate, which indicates a different matrix of sources and/or forcing functions that drive distributions of these nutrients.

Approximately 70% of the residential housing adjacent to Mangrove Lagoon and Benner Bay are on septic systems, many of which are failing (Horsley Witten, 2013). There is also a deteriorating sewer infrastructure for those connected to the public sewer system (Horsley Witten, 2013). The STEER Management Plan (STEER, 2011) cited nutrients as a high to very high threat in the STEER. An assessment of fecal coliform bacteria (FCB) in Mangrove Lagoon and Benner Bay (Tetra Tech, 2005), indicated a variety of sources including improperly functioning septic systems, urban runoff and discharges from recreational vessels in the area. The highest concentrations of the sewage marker *Clostridium perfringens* was found in Mangrove Lagoon, adjacent to where Turpentine Gut empties into the lagoon (Chapter 4).

Variation by Latitude and Longitude

Nutrient concentrations were assessed to determine if they varied by longitude (east/west orientation) and latitude (onshore/offshore orientation). A non parametric analysis (Spearman's) revealed a significant ($p < 0.05$) and negative correlation between longitude and ammonium, nitrite, DIN, and total phosphorus, indicating that moving east to west,

concentrations of these nutrients tended to increase. For latitude, there was a significant ($p < 0.05$) positive correlation for these same nutrients, indicating that as latitude increased (i.e., moving towards the more populated shores), there was an associated increase in concentrations. These results provide some evidence that land-based activities may be sources of the elevated nutrient concentrations in the STEER. The sites further offshore tended to have lower levels for a number of the nutrient species. Although there were construction activities observed on Little St. James Island during the field work, nutrient concentrations around this island were lower than in the Mangrove Lagoon and Benner Bay areas.

Comparison with Other U.S. Caribbean Studies

A number of projects in the U.S. Caribbean have examined the concentration of various nutrients in nearshore waters.

Turpentine Gut appears to have ended in 2008. Because of this, rainfall data was used in place of streamflow, as a proxy to assess the relationship between the amount of rainfall (precipitation) and variations in nutrient concentrations in the STEER. The rationale for using this data is that higher rainfall would be expected to result in additional runoff and nutrients entering streams that flow into the STEER, and also directly into the STEER through runoff from streets and hillsides, which would not be accounted for from the stream gauge data. The closest rainfall gauge to the STEER is located at Redhook Bay in St. Thomas. Unfortunately, nearly 40% of the data were missing from the records for the time period that the nutrient monitoring was conducted for this project. On those days when water samples were collected for nutrient analysis, missing data for rainfall was over 80%. As a result, precipitation records from the Cyril E. King Airport in West Charlotte Amalie,

Table 9.3. Comparison of STEER nutrient results with those from other U.S. Caribbean locations.

Nutrient	St. Thomas			St. John ¹						St. Croix ¹			Puerto Rico ²		
	STEER			Lameshur Bay			Coral Bay			Teague Bay			Guanica		
	Min	Mean	Max	Min	Mean	Max	Min	Mean	Max	Min	Mean	Max	Min	Mean	Max
NO ₃ + NO ₂	0	0.014	0.426	ND	0.001	2.664	ND	0.001	3.936	ND	0.001	0.289	0	0.14	3.68
NH ₄	0	0.01	0.055	ND	0.004	0.175	ND	0.0039	0.2335	ND	0.010	0.978	0	0.05	6.45
Orthophosphate	0	0.005	0.018	ND	0.002	0.158	ND	0.0016	1.8176	ND	0.002	1.234	0	0.05	1.48

¹Data from Smith *et al.*, 2013. ²Data from Whittall *et al.*, 2013.

Abbreviations: NO₃, nitrate; NO₂, nitrite. NH₄, ammonium; ND, not detected.

Summarized results from these studies, along with those from the current work in the STEER, are presented in Table 9.3. From this table, it can be seen that for most of the nutrient species, the mean concentrations appeared to be somewhat higher in the STEER compared to the locations monitored in St. John and St. Croix (Smith *et al.*, 2013), although lower than that found in and around Guanica Bay, Puerto Rico. Data from Guanica Bay includes both coastal embayments (similar to the STEER), and an enclosed estuary. The maximum concentrations found in these other studies, however, were almost always higher than that recorded in the STEER (Table 9.3). These data indicate that the nutrient concentrations in the STEER were similar to what has been found in other parts of the USVI and also in southwest Puerto Rico.

Nutrient Concentrations and Rainfall

Ideally, streamflow gauge data would be used to help assess nutrient delivery to the STEER. There are, however, no active streamflow gauges in the Turpentine Gut watershed, the largest watershed that drains directly to the STEER. The record of the USGS stream gauge data in

with a more complete data set, were used. Daily rainfall records for the Cyril E. King Airport were accessed through NOAA's National Climatic Data Center. Daily rainfall estimates were plotted against concentrations for each nutrient species at each of the six sites in the STEER.

Nonparametric tests (Spearman's) failed to show a significant positive correlation between rainfall and nutrients in the STEER. An example of the data for nitrite + nitrate (NO₂ + NO₃) plotted against rainfall for Benner Bay is presented in Figure 9.9. The highest concentration of nitrite + nitrate was found in the May 2013 sampling, however, rainfall for that day along with the day before was 0 mm. During the period of highest rainfall (1,364 mm) in September 2013, there were unfortunately no water samples taken for nutrient analysis. Additional graphs for nutrients and rainfall can be seen in Pait *et al.* (2015).

It's unclear why there wasn't a positive correlation between nutrient concentrations and rainfall in the STEER, although there are a number of possibilities. One of these is the lack of more complete rain gauge data adjacent to the

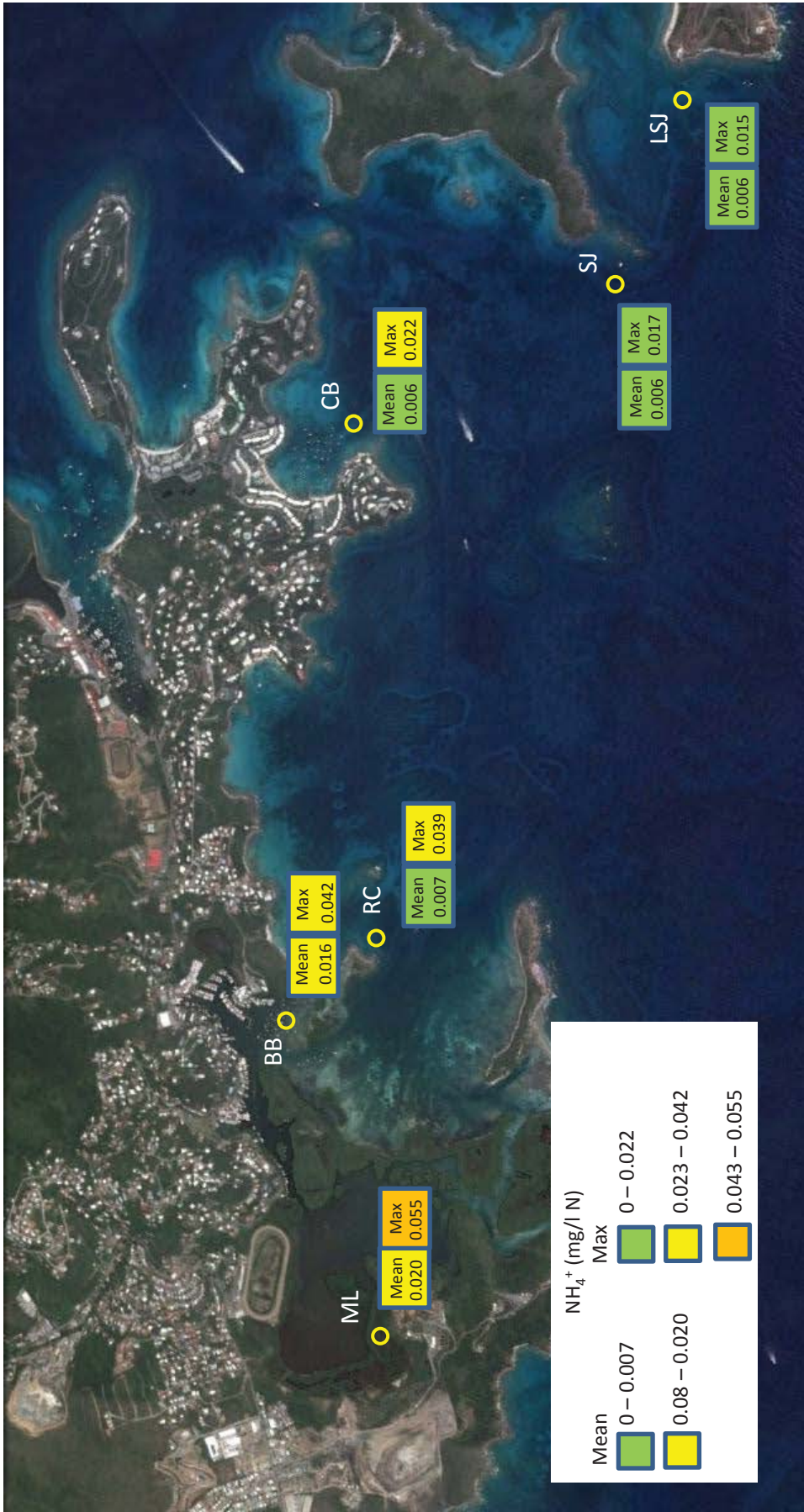


Figure 9.2. Mean and max concentrations of ammonium (NH_4^+) detected in water samples from the STEER.



Figure 9.3. Mean and max concentrations of nitrate (NO₃⁻) and nitrite (NO₂⁻) detected in water samples from the STEER.



Figure 9.4. Mean and max concentrations of urea detected in water samples from the STEER.

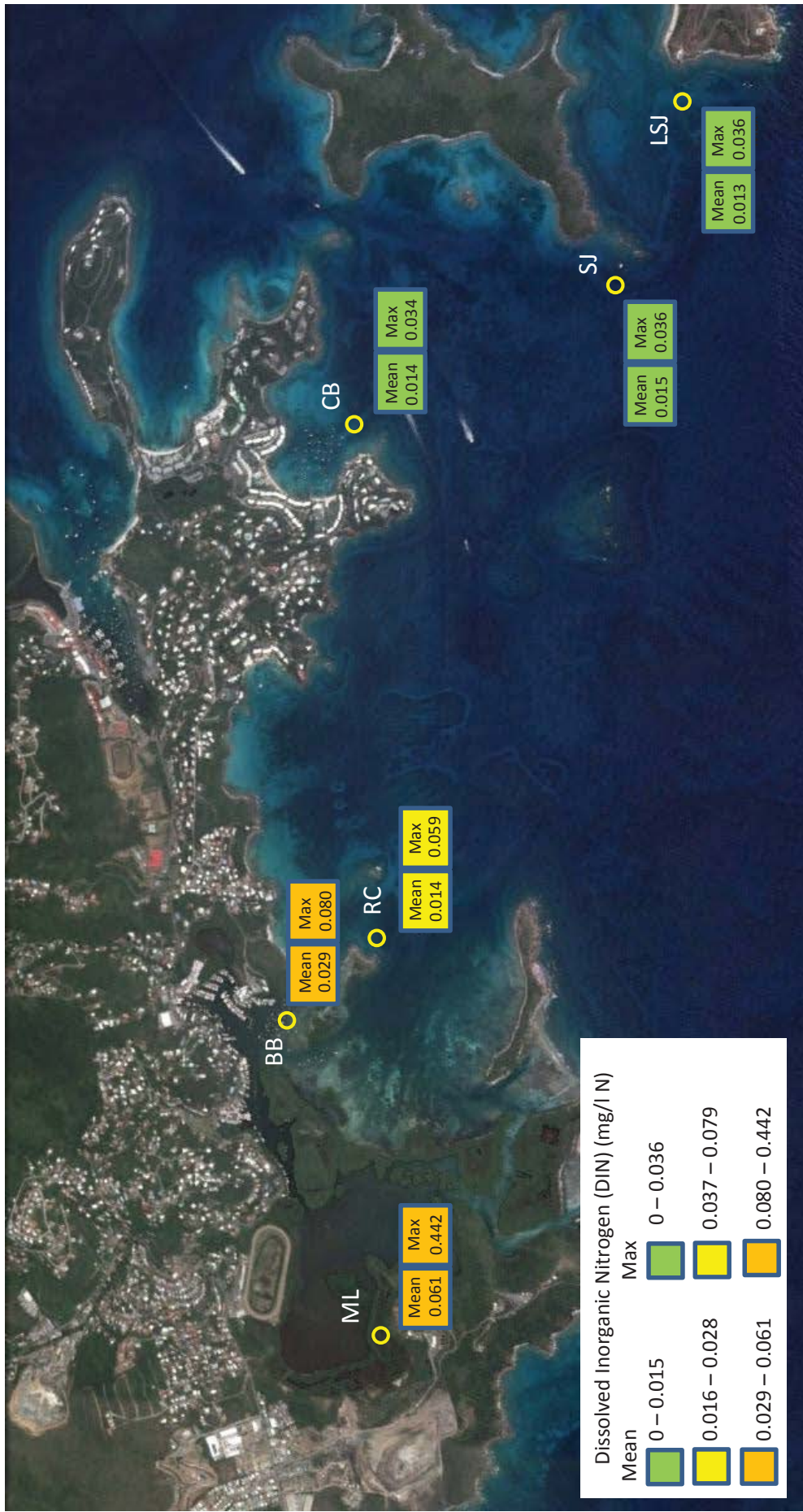


Figure 9.5. Mean and max concentrations of dissolved inorganic nitrogen (DIN) detected in water samples from the STEER.

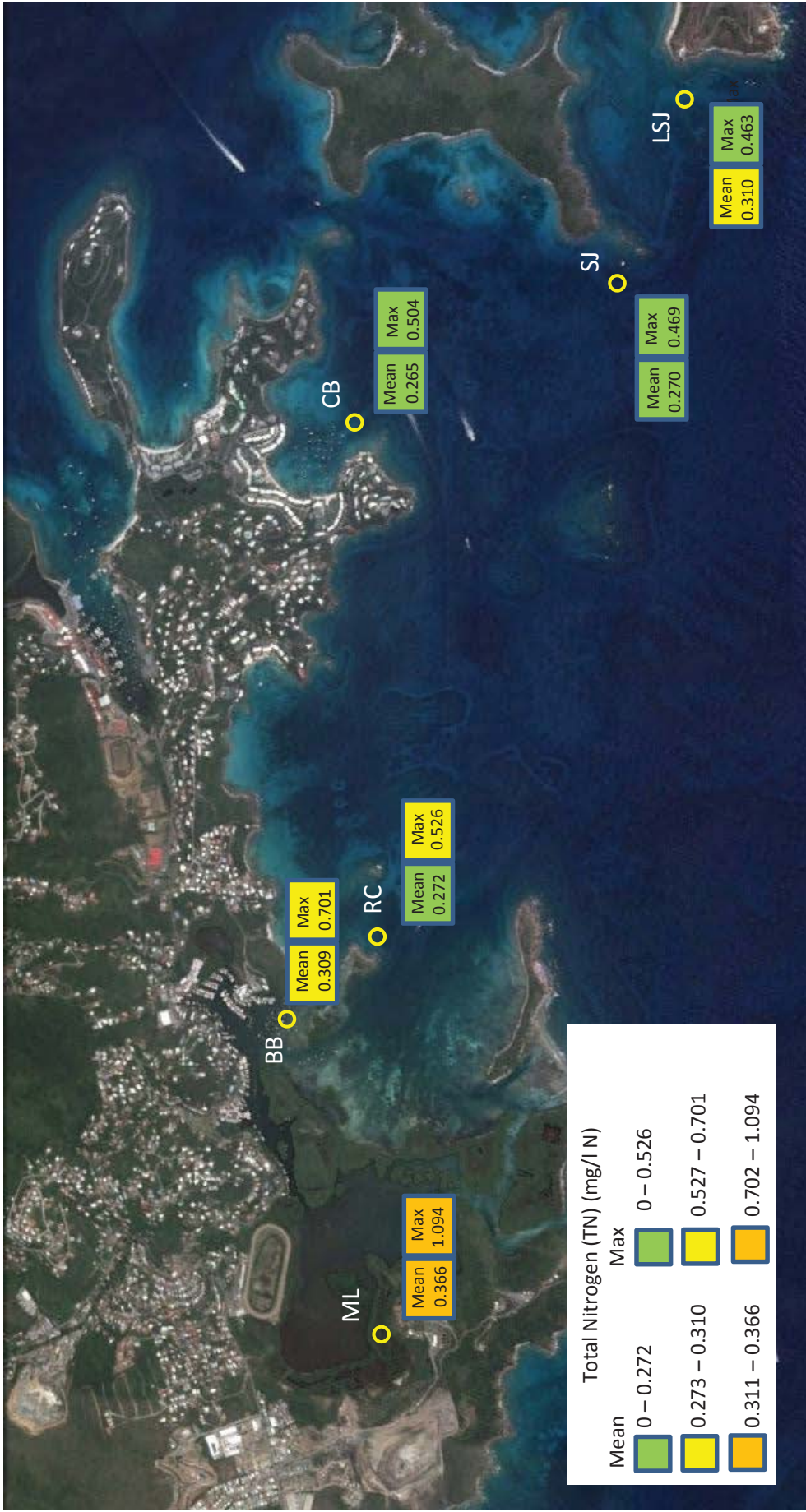


Figure 9.6. Mean and max concentrations of total nitrogen (TN) detected in water samples from the STEER.

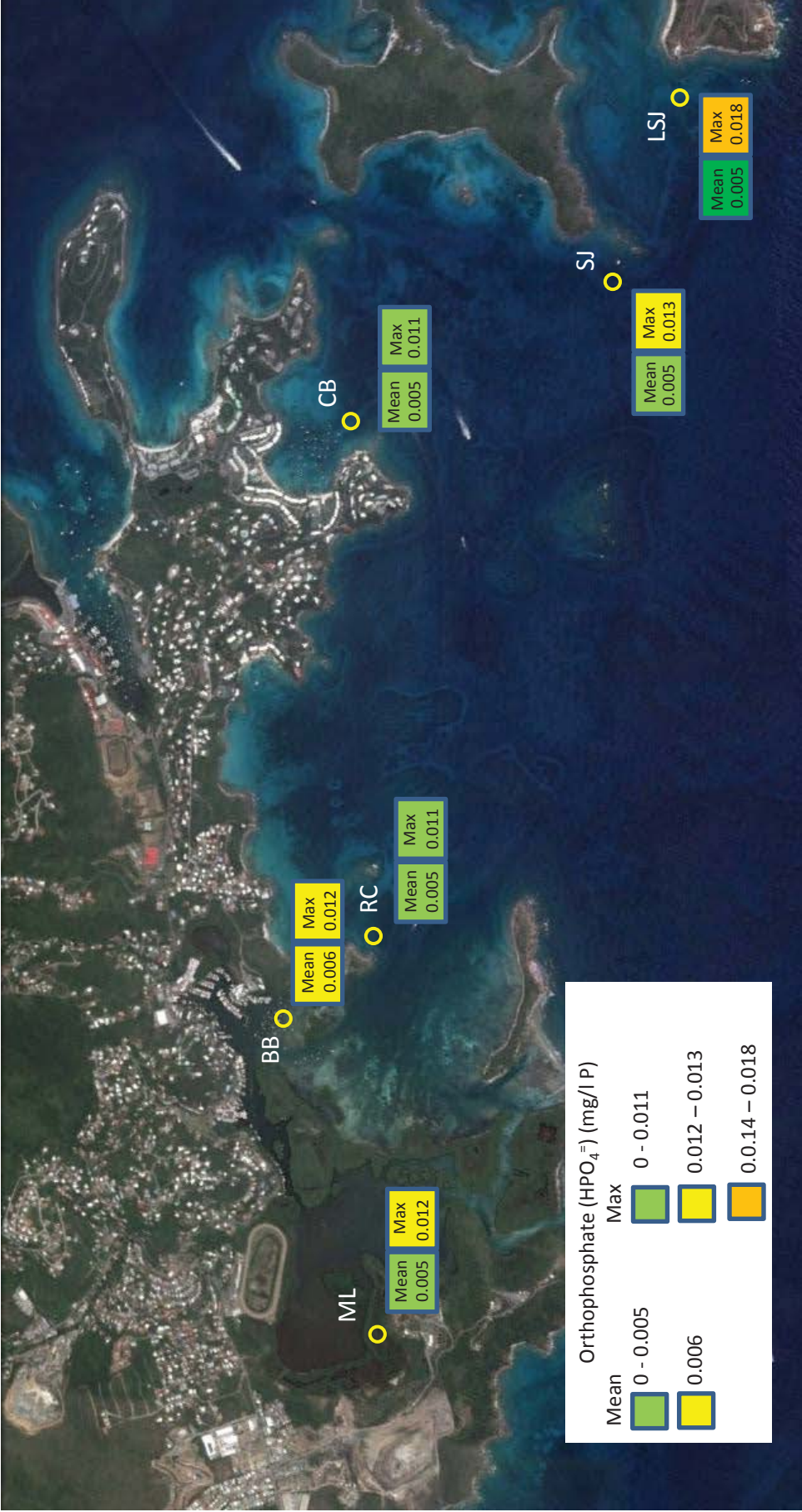


Figure 9.7. Mean and max concentrations of orthophosphate (HPO_4^-) detected in water samples from the STEER.



Figure 9.8 Mean and max concentrations of total phosphorus (TP) detected in water samples from the STEER.

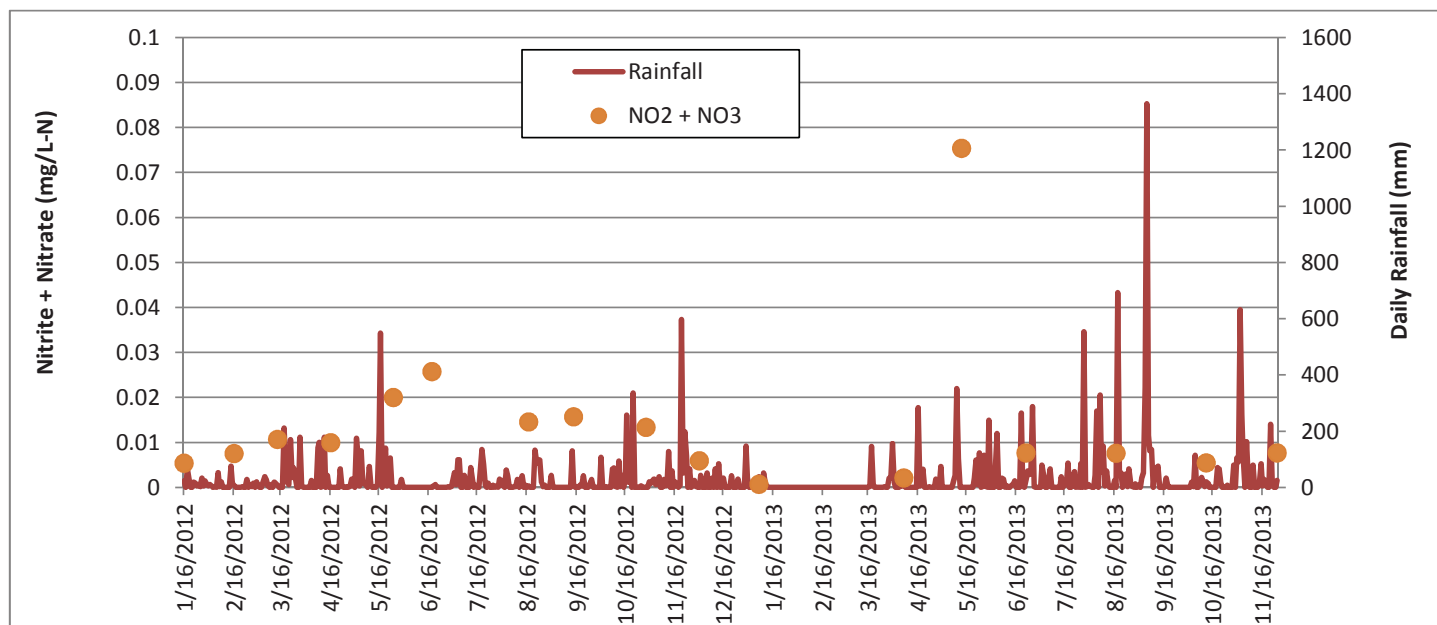


Figure 9.9. Nitrate and nitrite concentrations versus rainfall in Benner Bay during the study period.

STEER. Rainfall data near the STEER might reveal better correlations between rainfall and nutrient concentrations. Rainfall patterns on St. Thomas can be highly localized, and more complete rainfall data near the STEER might help better elucidate any relationships between rainfall, the mountainous terrain, and nutrients. Another possibility is that discharges from septic systems into the STEER either from streams like Turpentine Gut, or via groundwater to the STEER, are providing a continuous source of nutrients, independent of rainfall. Another factor may be the influence of winds or tides on the resuspension of sediments into the water column containing nutrients. There is also significant boating activity including some liveaboards, particularly in the Benner Bay area, which are a likely source of nutrients to the STEER. There did not appear to be any significant differences in the nutrient concentrations between the wet and dry seasons. Finally, sampling for this project was not designed to be during or just after heavy rainfall events, which might have provided a better correlation between rainfall and nutrient levels.

Relationships Between Nutrients

Whitall *et al.* (2013) examined correlations between nutrient species, leaving out those that are autocorrelated (e.g., orthophosphorus and total phosphorus), and found that ammonium, urea and orthophosphorus were associated with runoff, while the oxidized forms of nitrogen tended to percolate into soils during rainfall events, and eventually into groundwater. Spearman's rank correlation coefficient analysis for the STEER also indicated significant correlations between ammonium, urea and orthophosphorus, and that these species were not correlated with the more oxidized forms of nitrogen, particularly nitrate plus nitrite.

Nutrients and Coral Reefs

Lapointe (1997) suggested nutrient thresholds for marine waters, above which macroalgae and phytoplankton were more likely to flourish on coral reefs, in the greater Caribbean. While it needs to be acknowledged that coral species respond differently to elevated nutrient concentrations, and that there has been some controversy regarding the use of thresholds (Hughes and Connell, 1999; Szmant, 2002) proposed by Lapointe (1997) and others, these thresholds provide an opportunity to assess nutrient levels that may be of concern. Thresholds were proposed for orthophosphate (0.003 mg/L as P), and dissolved inorganic nitrogen (DIN, 0.014 mg/L as N). DIN is the summation of nitrate, nitrite and ammonium.

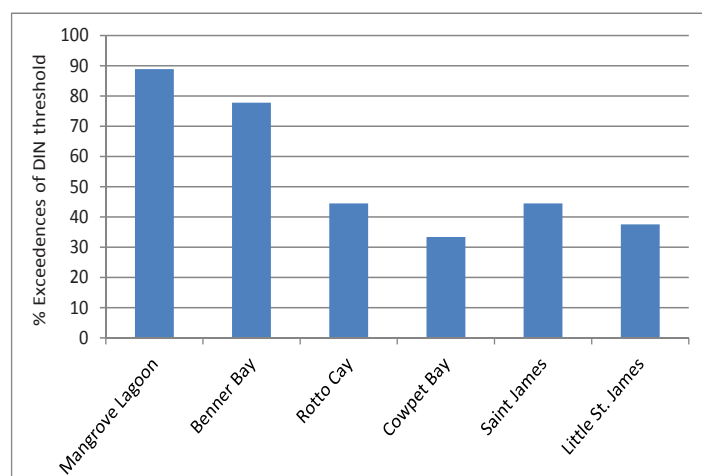


Figure 9.10. Percent exceedances of DIN threshold by site in the STEER.

Using the values proposed by Lapointe (1997), 58% of the samples analyzed from the STEER were above the threshold for orthophosphate, with a slightly higher number (61%) within the western portion of the study area. For DIN (Figure 9.10), the mean exceedance across the STEER was 55%. A nonparametric analysis using the Wilcoxon method indicated that DIN varied by sampling site, with DIN at Mangrove Lagoon and Benner Bay significantly higher ($p < 0.01$)

In Figure 3.13 from Chapter 3, it appears that macroalgae may be somewhat abundant in Mangrove Lagoon. In addition, Figure 9.11 shows herbivore biomass at the sites sampled in the STEER. There does not appear to be an overabundance of herbivores, particularly in Mangrove Lagoon, and perhaps in the western portion of Benner Bay. Some areas of the STEER may be at risk, to blooms of macroalgae which could outcompete and eventually smother corals in the area. There is also concern regarding the direct effects of nutrients on corals, including reduced fertilization and reduced calcification rates (Harrison and Ward, 2001; Marubini and Davis, 1996). The likely source of these nutrients are inputs from the developed areas around the STEER.

Sedimentation

A summary of the results from the monthly monitoring of sediment traps in the STEER (January 2012 to November 2013) can be seen in Figures 9.12 to 9.17.

Differences by Site

The average percent deposition of sediments by weight is presented in Figure 9.12. For all sites, terrigenous material was dominant, accounting for 60 to 70% of total sediment in the traps, highlighting the role of terrestrial inputs to the STEER. Conversely, it can also be seen that carbonate inputs appeared somewhat smaller in Mangrove Lagoon and Benner Bay, compared to the more offshore sites, as might be expected. Also, the organic fraction tended to be higher at the Mangrove Lagoon, Benner Bay and perhaps the Rotto Cay sites. Organic inputs, however, can be from terrestrial or marine sources.

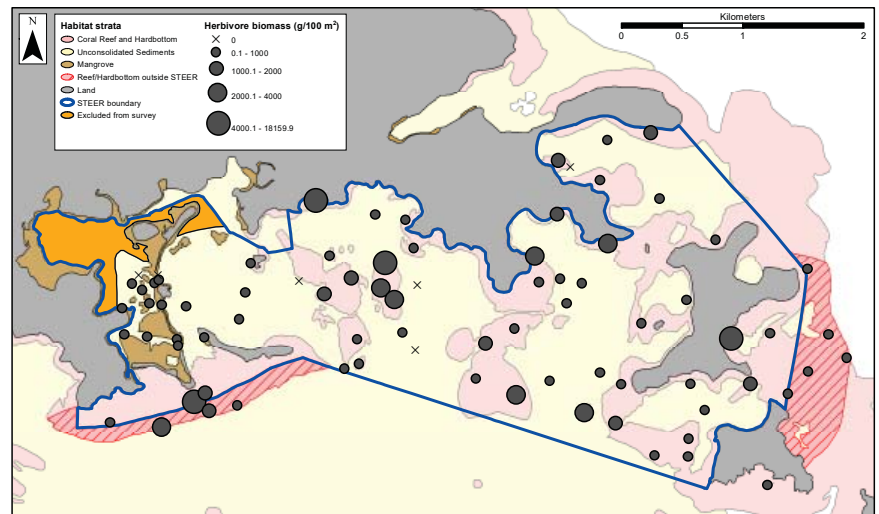


Figure 9.11. Herbivore biomass in the STEER.

The percent terrigenous material in the sediment traps in the STEER was significantly higher ($p < 0.05$) in Mangrove Lagoon, Benner Bay, and Rotto Cay compared to the other locations in the STEER. In addition to fresh inputs from the surrounding watershed, resuspended bed sediments from within the STEER can enter the traps as well, as a result of storms (wave and wind activity), or from boat traffic in Benner Bay. The contribution from new versus resuspended materials can not be determined using the traps deployed.

The percent carbonate deposition in the sediment traps was significantly higher ($p < 0.05$) at Rotto Cay, Cowpet Bay and Saint James, further away from the land-based inputs in the Benner Bay and Mangrove Lagoon areas. Carbonate sediments are derived primarily from marine skeletons (e.g., coral) and shells.

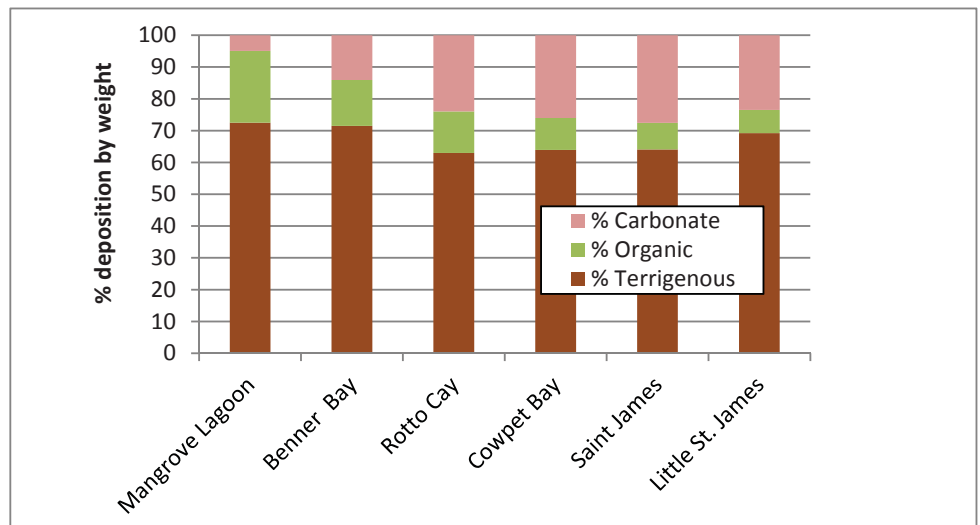


Figure 9.12. Bulk deposition of sediment in traps placed in the STEER.

The sediment trap site adjacent to Little St. James Island (Figure 9.1), arguably the site furthest away from Benner Bay and Mangrove Lagoon, appeared to have a somewhat higher terrigenous fraction (Figure 9.12), and a correspondingly lower carbonate input. The difference in the percent carbonate fraction between the Little St. James and Saint James sites was significant. During the course of the sediment trap work, substantial earth moving activities were observed on Little St. James Island (Figure 9.13), which would likely be a significant source of terrigenous sediments entering the adjacent waters.



Figure 9.13. Excavation activity on Little St. James Island, a possible source of terrigenous sediments found in the sediment traps at this site.

A comparison of the mean daily accumulation rates in the sediments traps by site is presented in Figure 9.14. The mean accumulation rate (9.02 mg/cm²/day) in the Benner Bay sediment traps was significantly ($p < 0.001$) higher than the trap accumulation rates at all other sites in the STEER. Similar to the bulk deposition of sediments, the highest accumulation rates were seen for the terrigenous fraction. The mean terrigenous accumulation rate (6.45 mg/cm²/day) was also higher ($p < 0.0001$) in Benner Bay than the other sediment traps in the STEER, including Mangrove Lagoon. Although Mangrove Lagoon receives input from Turpentine Gut, the only perennial stream on St. Thomas, the highest rate of terrigenous sediment input occurred in Benner Bay.

There are likely two sources of input to the traps placed at Benner Bay: terrestrial materials washed off the roads and hill-

sides from the surrounding landscape, and resuspended sediments from within Benner Bay. The hillsides in the watershed surrounding Benner Bay are fairly steep, and in some areas bare soils are exposed. These factors, along with widespread construction activities, and periodic torrential downpours, can lead to the transport of soils into the STEER (Horsley Witten, 2013). Pait *et al.* (2013) found that sediments in northern Benner Bay had some of the highest silt and clay fractions (31 - 56%) in the STEER. Silt and clay sediments are typically derived from terrestrial sources. Sediment coring in northern Benner Bay by NCCOS in 2013 (see Chapter 5), revealed a thick layer of silt and clay over a deeper layer of shell hash,

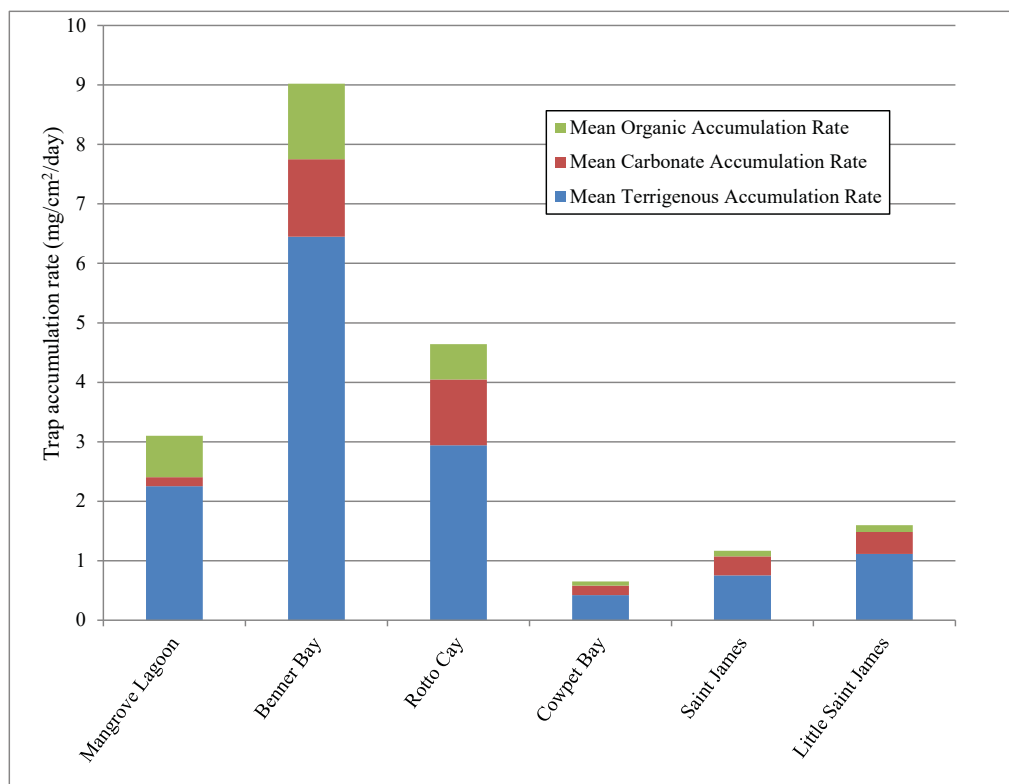


Figure 9.14. Average accumulation rates at the sediment trap sites in the STEER.

indicating that changes in land use over time have resulted in higher amounts of terrigenous materials transported to the STEER.

Variation by Latitude and Longitude

A nonparametric analysis revealed a significant negative correlation (Spearman's $Rho = -0.6686$, $p < 0.0001$) between longitude and mean trap accumulation rate, and between the terrigenous accumulation rate and longitude (Spearman's $Rho = -0.6625$, $p < 0.0001$), indicating that moving east to west, the deposition rate of terrigenous sediments tended to increase.

There was also a significant (Spearman's $Rho = 0.5047$, $p < 0.001$) positive correlation between latitude and mean trap accumulation rate, along with the terrigenous accumulation rate, indicating that deposition tended to increase moving from the more offshore sites to the nearshore locations.

Comparison with Other U.S. Caribbean Studies

The mean terrigenous accumulation in sediments traps found by Sherman *et al.* (2013) within Guanica Bay Puerto Rico, was roughly 58%, less but somewhat similar to what was found in the STEER. Outside of Guanica Bay, the percent terrigenous was only 24%. As noted, the mean percent terrigenous sediment in the traps in the STEER was higher, approximately 66% terrigenous and 20% carbonate. Within the STEER, Mangrove Lagoon (73%) and Benner Bay (71%) were even higher. Working in St. John, Gray *et al.* (2012), found that terrigenous input into the sediment traps in Coral Bay located near the shore accounted for 56%. Out on the reefs in Coral Bay, that value fell to roughly 21% (Gray *et al.*, 2012). In Lameshur Bay, terrigenous sediment accounted for approximately 32% of the material in the nearshore sites, and fell to roughly 15% out on the reefs in this Bay. Terrigenous input into the sediment traps in the STEER was quite high. Additional work is needed to better characterize the inputs of fresh material from the surrounding watershed versus resuspended sediments.

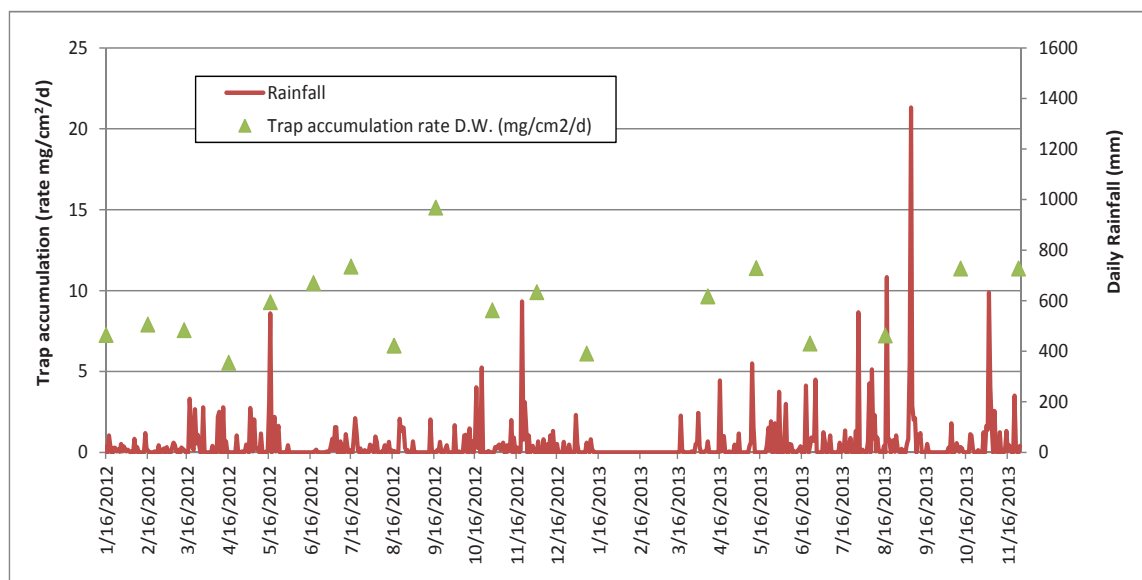


Figure 9.15. Bulk deposition in Benner Bay sediment traps plotted against rainfall.

Sediment Deposition and Rainfall

As with nutrients, there did not appear to be a good correlation between sediment deposition and rainfall. A plot of the sediment trap accumulation rate and rainfall for Benner Bay can be seen in Figure 9.15. A plot of the terrigenous accumulation rate showed similar results. Nonparametric (Spearman's) analyses of the results between sedimentation and daily and monthly rainfall (Cyril E. King Airport) failed (Spearman's $Rho = 0.0747$, $p = 0.4491$) to show any significant relationship, either across the STEER or for each site.

On the northwestern coast of St. Thomas, Nemeth and Nowlis (2001) found that sedimentation closely tracked rainfall during a period of early construction at a new resort near Caret Bay, however, once the earth-moving phase had been completed, that relationship disappeared, resulting in an overall lack of significance between rainfall and sedimentation during their study. In the STEER, it would likely take sustained rainfall over a period of days in order to alter the amount of sediment accumulating in the traps from surface water runoff, and so correlations with daily rainfall might not be particularly useful. However, monthly rainfall amounts did not correlate with the sediment deposition rates either. It may be that other factors are involved with increases in sedimentation, including increased wave action resuspending sediments already within the STEER.

Sediment Deposition Over Time

Plots of sediment deposition over time, both in terms of percent deposition and by sedimentation rate for three of the sites in the STEER are shown in Figure 9.16. Graphs for Rotto Cay, Cowpet Bay, and Saint James can be seen

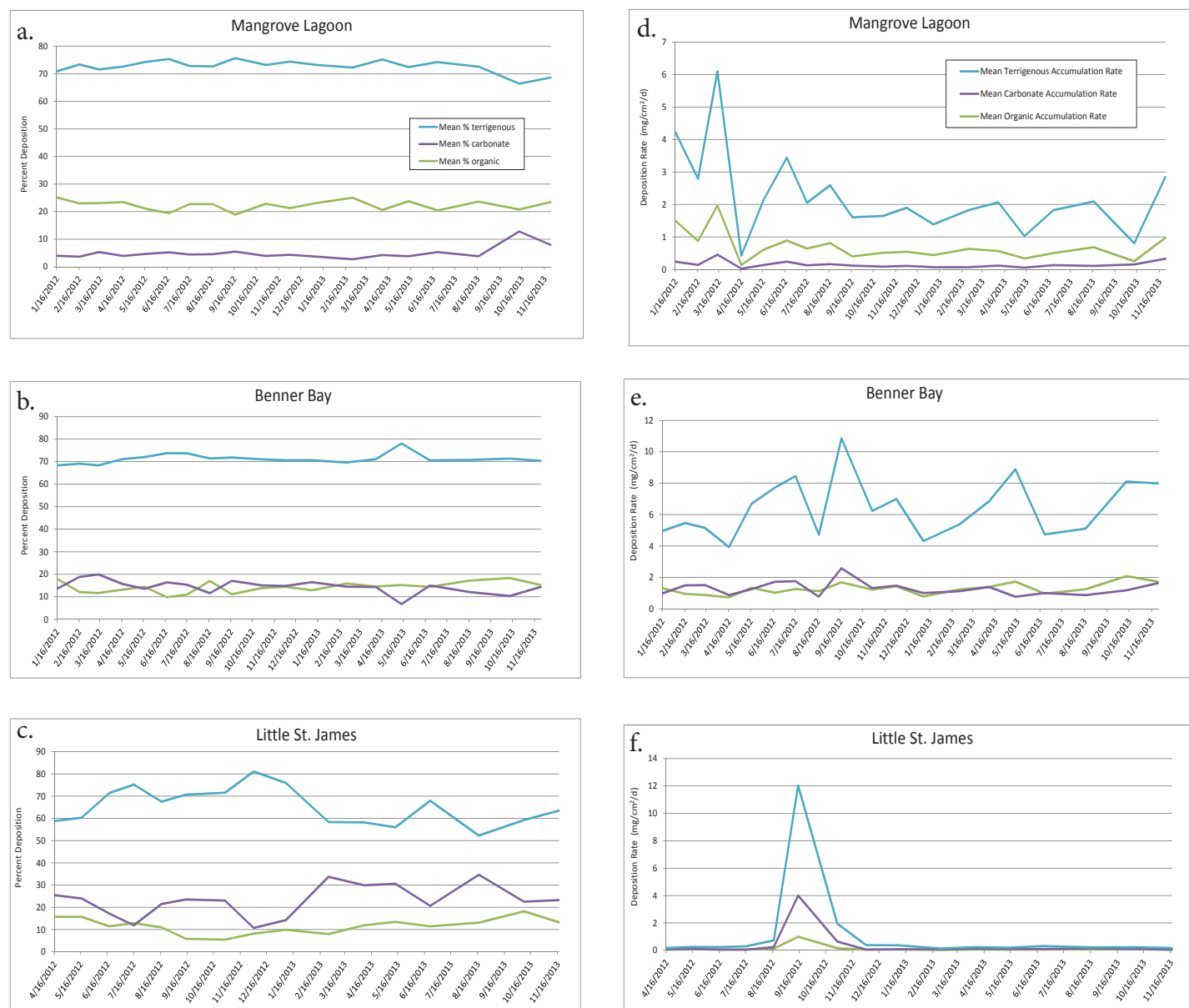


Figure 9.16. Percent and rates of deposition for terrigenous, carbonate and organic materials in the sediment traps in the STEER over time for Mangrove Lagoon (a, d), Benner Bay (b, e), and Little St. James (c, f).

in the appendices in Pait *et al.* (2015). In Figure 9.16, the contributions from terrestrial, carbonate and organic sources were fairly constant over time for both Mangrove Lagoon (a) and Benner Bay (b). In both cases, terrigenous material was the major component (~70%) found in the sediment traps. The lack of substantial variation in the sources (terrestrial versus carbonate) in Mangrove Lagoon and Benner Bay sediment traps could indicate a continuous input of terrestrially-derived sediments at these two sites as a result of new input, resuspension of sediments into the water column from storms, tides and boats (in Benner Bay), or a combination of these phenomena. On the right hand side of Figure 9.16, for Mangrove Lagoon (d) and Benner Bay (e), there is substantial variation in the daily rate of terrigenous and carbonate deposition (organic material can

be derived from terrestrial or marine sources) over time. In March 2012, there was a sharp increase in the terrigenous fraction found in the sediment traps in Mangrove Lagoon (Figure 9.16d). Specifically, the rate increased from 2.79 mg/cm²/day to 6.10 mg/cm²/day, a factor of 2.18. However, at the same time, the carbonate deposition rate went from 0.141 to 0.456 mg/cm²/day, a factor of 3.23, higher than the rate increase for the terrigenous fraction. While the increase in the terrigenous sediment deposition rate may have been related to rainfall during this period, other factors such as the movement of tides or winds that could have led to a resuspension of sediments cannot be ruled out. In addition, there does not appear to be a corresponding increase in the rate of sediment deposition in the adjacent Benner Bay site in March 2012, as might be expected if rainfall had resulted

in higher surface water runoff from the steep hillsides, carrying terrestrially derived materials to the STEER.

For Little St. James (Figure 9.16c), deposition appeared more variable, and may be related to weather and wave patterns. In Figure 9.16f, it can be seen that there was a spike in terrigenous, organic and carbonate deposition into the Little St. James sediment traps sampled in September 2012. In August 2012, sediment traps in the STEER were sampled around the middle of the month. In late August 2012, Hurricane Isaac passed to the south of the USVI, and appears likely responsible for the increased rates of sedimentation found when the sediment traps were retrieved in September. Further evidence of this is presented in Figure 9.17. Total monthly deposition of sediment in the traps is plotted from January 2012 through October 2012. The average deployment of the sediment traps during this period was 30 days. Daily rainfall records from the Cyril E. King Airport are shown in Figure 9.17b. Wave height data (Figure 9.17c) were obtained from the Caribbean Coastal Ocean Observing System (CariCOOS), for a site south of St. John (Station 41052), and accessed online through NOAA's National Data Buoy Center data base (http://www.ndbc.noaa.gov/station_page.php?station=41052). Although the station is not within the boundaries of the STEER (there appear to be no wave stations in the STEER), it does provide an indication of likely increases in wave height, at least entering the southern portion of the STEER.

In Figure 9.17a, there was increased deposition in the traps found during the September 2012 sampling, particularly in the more offshore traps at Little St. James and Saint James (identified in red circle). The amount of sediment in the traps at Little St. James (17.3 g) in September 2012, was actually higher than in the traps at Benner Bay. Also, the amount of sediment in the traps at Little St. James in September 2012 was higher by a factor of 14 from the previous month at this site. At Saint James, the amount of sediment in the traps was 12.7 g, an increase in sediment deposition by a factor of 12 from the previous month. As mentioned, Hurricane Isaac passed to the south of the USVI in late August 2012. While there did not appear to be a substantial amount of rainfall (Figure 9.17b) in St. Thomas associated with this system, wave height increased to nearly 4 meters (Figure 9.17c) at the CariCOOS wave station just south of St. John. It seems likely that waves from Hurricane Isaac, moving into the southern portion of the STEER, resulted in the remobilization of bed sediments that were subsequently deposited into the sediment traps at both Saint James and Little St. James. As can be seen in Figure 9.16, the percent contribution from terrigenous versus carbonate during this time did not appear to change appreciably. During the

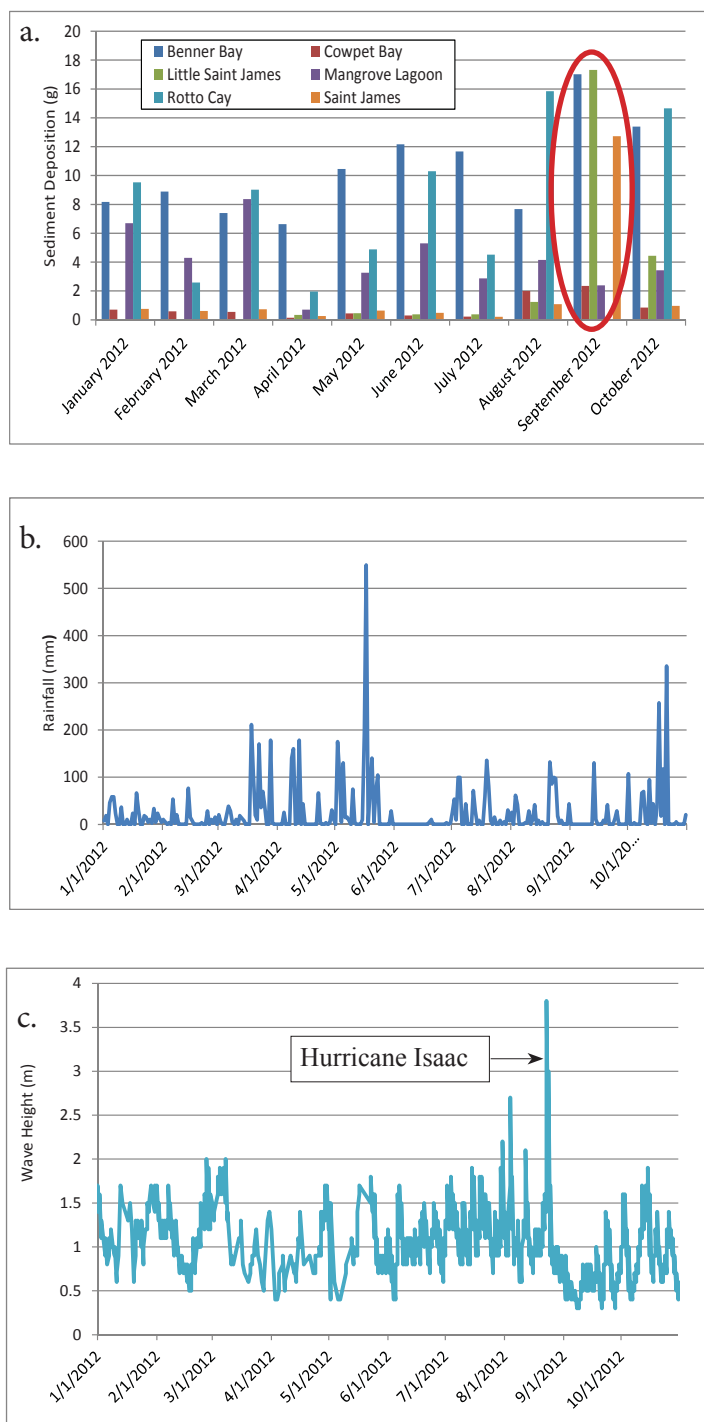


Figure 9.17. Plots of mean sediment deposition (a), rainfall (b) and wave height at Station 41052 (c).

same period, the Benner Bay traps also accumulated more sediment, however, the percent terrigenous contribution also did not change.

Sedimentation and Coral Reefs

Rogers (1990) suggested that sedimentation rates above 10 mg/cm²/day on coral reefs may be excessive, and that heavy sedimentation on reefs has been associated with

fewer coral species, less live coral, reduced recruitment and slower accretion rates among other effects. Research by Smith *et al.* (2008) and Nemeth and Nowlis (2001) have shown this value may be useful at least in the USVI, for determining when coral reefs are at risk to being impacted by chronic sedimentation.

Using the rate of 10 mg/cm²/day, none of the sites had a mean rate above this, although Benner Bay (Figure 9.14) (9.02 mg/cm²/day) was fairly close. There were nine occasions, however, during the course of the monthly monitoring when a site exceeded this threshold. Benner Bay exceeded the threshold on six occasions, and Rotto Cay, St. James (September 2012) and Little St. James (September 2012) each exceeded the threshold once. Although none of the sites had a mean sedimentation rate above the suggested threshold, sedimentation in Benner Bay appears to be high enough to put the corals in this area at some risk. Additional work is needed to better describe sedimentation throughout Benner Bay. In addition, it would also be useful to understand the association between coral species richness/diversity and sedimentation in Benner Bay and throughout the Reserves, in order to assess how sedimentation and perhaps the chemical contaminants present may be affecting the distribution of individual coral species.

Total Suspended Solids (TSS)

A summary of the results from TSS monitoring in the STEER is presented in Figure 9.18. TSS includes silts which can settle out onto reefs impacting the health of corals, along with plankton and other materials, some naturally occurring while others are derived from human activities. TSS also impacts corals and seagrasses by decreasing the amount of light available for photosynthesis. In northern Benner Bay (in addition to northern Mangrove Lagoon), the biological survey by SCUBA divers could not be conducted, due in part to poor visibility.

Differences by Site

Nonparametric analysis (Wilcoxon Rank-Sum) revealed that the mean TSS value (8.32 ±0.88 mg/L) in Benner Bay

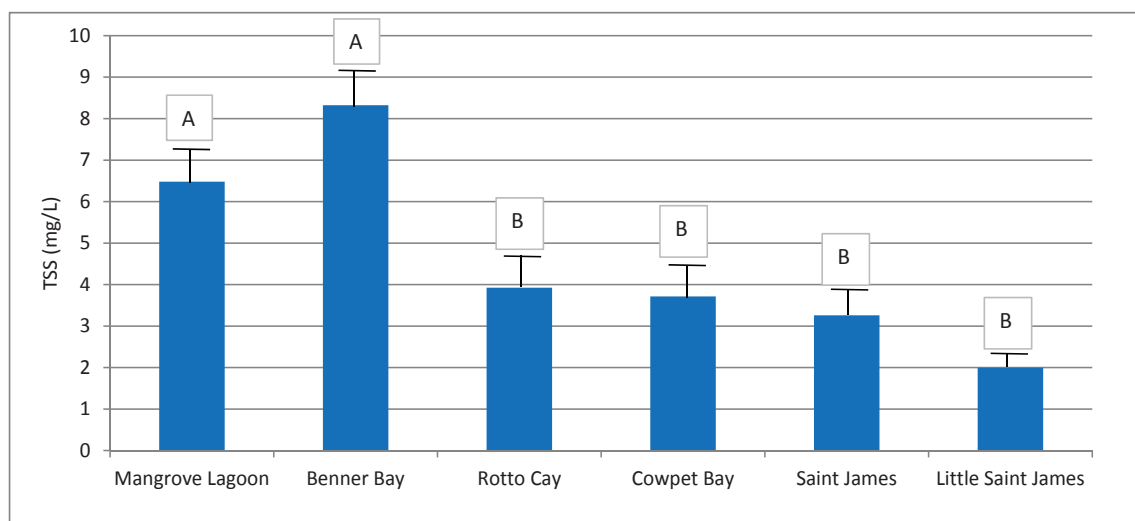


Figure 9.18. Total suspended solids (TSS) means (±SE) at sampling sites in the STEER. Sites with the same letter were not statistically different.

was significantly ($p < 0.01$) higher than the other sites, with the exception of Mangrove Lagoon. As was seen earlier, the Benner Bay site had a significantly different (higher) mean turbidity than the other sites in the STEER (Table 9.1). Finally, TSS at Rotto Cay, Cowpet Bay, Saint James, and Little Saint James were not significantly different from each other.

Variation by Latitude and Longitude

Like the results from the sediment traps, a nonparametric analysis indicated there was a significant and negative (Spearman's Rho = -0.5466, $p < 0.0001$) correlation between longitude and TSS, indicating that moving east to west, concentrations of TSS tended to increase. In addition, there was a positive and significant (Spearman's Rho = 0.4725, $p < 0.0001$) correlation between latitude and TSS, indicating that TSS was higher in the more nearshore sites.

Table 9.4. Comparison of TSS in the STEER with other locations in the USVI.

	TSS (mg/l)		
	Minimum	Mean	Maximum
St. Thomas			
STEER	0.5	4.7	18.6
St. John ¹			
Lameshur Bay	0.4	6.5	22.0
Coral Bay	1.0	6.5	22.0
Fish Bay	0.8	6.8	24.0
St. Croix ¹			
Teague Bay	1.0	6.4	28.0

¹Data from Smith *et al.*, 2013.

Comparison with Other U.S. Caribbean Studies

Table 9.4 presents a comparison of the TSS results from work in the STEER, with that of measurements made in St. John and St. Croix; (Smith *et al.*, 2013). From this table, it can be seen that the STEER sites were similar to the sites sampled on St. John and St. Croix, for both the mean and maximum values. The highest TSS value recorded in the STEER was 18.6 mg/L, in Benner Bay in January 2012.

TSS Concentrations and Rainfall

A nonparametric correlation analysis (Spearman Rank Correlation) was first run on TSS versus daily rainfall. The results (Spearman's $Rho = -0.0844$, $p = 0.3535$,) indicated no significant relationship between daily rainfall and TSS in the STEER throughout the course of the study. The lack of a significant relationship with rainfall was similar to that found for the sediment traps. As with those results, it is not clear why there wasn't a significant relationship between TSS and rainfall in the STEER. Although stream flow data was not available, higher rainfall might be expected to result in higher levels of TSS. As with sedimentation, it is possible that at least some of the TSS was the result of resuspension of sediments. It could also be that there is a lag time between a large rain event and when that water empties into the STEER. This second possibility seems unlikely though, given the steep slopes in much of the watershed. Additional work is needed to assess the relationship between rainfall, stream-flow and sediment delivery to the STEER.

TSS and Coral Reefs

Rogers (1990) suggested that suspended sediment concentrations above 10 mg/L on coral reefs may be excessive, and that heavy sedimentation on reefs has been associated with fewer coral species, less live coral, reduced recruitment and slower accretion rates among other effects. None of the sites in the STEER had a mean TSS value above 10 mg/L, however, the mean for Benner Bay during the study was 8.32 mg/L.

There were 19 occasions during the monthly monitoring when TSS exceeded 10 mg/L. The highest TSS value recorded was 18.6 mg/L in Benner Bay. Forty-seven percent of the exceedances of the threshold occurred in Benner Bay, followed by Mangrove Lagoon with 26%. Similar to the results for sedimentation, corals in the Benner Bay area would appear at some risk as a result of suspended solids.

While there were a number of exceedances in Mangrove Lagoon, there appear to be very few corals in this area; corals were only found in the extreme southern portion of the lagoon.

Relationships Between Parameters

Finally, a series of nonparametric analyses (Spearman Rank Correlation) were carried out to assess relationships between parameters measured in this part of the project in the STEER. The mean sediment trap accumulation rate was highly correlated with TSS. This is perhaps not surprising, as some of the TSS eventually settles out of the water column. Areas with higher levels of TSS might also be areas of higher sediment deposition, as a result of transport from the watershed, or from resuspension within the STEER. A number of the nutrients, particularly the non-oxidized forms (e.g., ammonium, urea, and orthophosphate) were correlated ($p < 0.05$) with both TSS and sediment deposition, again suggesting areas with higher

inputs of sediments were associated with higher inputs of specific nutrient classes



Apparatus for filtering water samples for measuring TSS in the STEER.

9.4. CONCLUSIONS

The monitoring of nutrients, sedimentation and total suspended solids (TSS) for 23 months was part of the project in the STEER to assess the presence and effects of land-based sources of pollution, and a characterization of the biological communities within the STEER. The results indicated elevated levels of nutrients in the western portion (Mangrove Lagoon and Benner Bay) of the STEER. These areas contain higher densities of residential housing, most of which have septic systems, and many that are likely failing. There is also a horse racetrack along with the Bovoni Landfill which border Mangrove Lagoon on the north and west sides, as well as live-aboard boats in Benner Bay, all of which have the potential to contribute nutrients, leading to higher concentrations in this part of the STEER. Ammonium, nitrite and dissolved inorganic nitrogen (DIN) were higher in Benner Bay and Mangrove Lagoon than the other sites in the STEER. For DIN, both Mangrove Lagoon and Benner Bay had significantly higher levels than the other four sites.

A significant relationship between rainfall and nutrients, although expected, was not found. Because there are no active stream gauges in the watershed draining to the STEER,

rainfall was used a proxy for streamflow. The reason for the lack of correlation between rainfall and nutrients was not clear. It would be useful to have an active stream gauge in the watershed that could be used to assess the relationship between stream flow and concentrations of nutrients. If a stream gauge is not possible, then more complete rainfall data at Redhook would be helpful.

The concentrations of the nutrients measured in the STEER were similar to other sites recently monitored in the U.S. Caribbean. Nutrient values in the STEER appeared slightly higher than in St. John and St. Croix, USVI, but lower than in Guanica Bay in Puerto Rico. Approximately 60% of the nutrient samples analyzed from the STEER were above a proposed threshold for coral reefs for orthophosphate, with a slightly higher number within the western portion of the study area.

Results from the monitoring of sedimentation indicated that terrigenous material accounted for roughly 60 - 70% of the material found in the sediment traps, highlighting the role of terrestrial inputs to the STEER. Terrigenous inputs to the sediment traps in the STEER appeared higher than in studies that have been conducted in nearby St. John, USVI, and also in the Guanica Bay area in Puerto Rico, highlighting the magnitude of terrigenous input to the STEER. The percent terrigenous material in the sediment traps was higher in both Mangrove Lagoon and Benner Bay compared to the other locations in the STEER. It is likely that in addition to inputs of sediment from the surrounding watershed, resuspended sediments as a result of storm (wave and wind activity) or boat traffic (Benner Bay) are likely contributing to the material found in the traps as well.

The average daily accumulation rates in the sediments traps were higher in Benner Bay than at all the other sites in the STEER, including Mangrove Lagoon. The mean accumulation rate and the terrigenous accumulation rate were negatively correlated with longitude, and positively correlated with latitude, indicating rates tended to be higher in the western and nearshore portions of the study area.

There are steep hillsides in the area around northern Benner Bay, that contribute terrigenous materials to the STEER through surface water runoff. Follow up work in 2013 for another part of the STEER project (Chapter 5), in which sediment cores were taken, revealed a thick layer of silt and clay over older shell hash in sediment cores from northern Benner Bay, testifying to historic changes in land use that have resulted in increased transport of terrigenous material to the STEER. In addition, this area contains numerous marinas and boat yards. The boat traffic in and out of this

portion of the STEER may be resuspending sediments into the water column as well.

As with nutrients, there did not appear to be a good correlation between sediment deposition and rainfall. A stream gauge in the watershed would help with understanding the relationship between streamflow, rainfall and sediment delivery.

The percent deposition of sediment over time by source (i.e., terrigenous, carbonate and organic) was fairly constant, particularly for Mangrove Lagoon and Benner Bay, with terrigenous material being the major component (~70%) found in the sediment traps. It would appear that the lack of substantial variation in the sources (terrestrial versus carbonate) in the Mangrove Lagoon and Benner Bay traps could indicate a continuous input of terrestrially-derived sediments at these two sites as a result of runoff, resuspension of sediments from storms, tides or boats, or a combination of these phenomena.

A comparison of the sedimentation rates in the STEER with a proposed threshold for coral reefs (10 mg/cm²/day), indicated that none of the sites monitored in the STEER had an average above this value, although Benner Bay (9.02 mg/cm²/day) was close.

Results from the monthly monitoring indicated that like the nutrient and sedimentation monitoring, the western and more nearshore portions of the STEER had higher levels of TSS. Benner Bay and Mangrove Lagoon had higher TSS levels than the other sites monitored in the STEER. Results indicated that the TSS levels in the STEER were similar to those found in studies in St. John and St. Croix. As with sedimentation, there was no correlation between TSS and rainfall.

None of the sites in the STEER had a mean value that exceeded a proposed TSS threshold for coral reefs of 10 mg/L. The highest was Benner Bay with a mean of 8.32 mg/L.

The results from this part of the project in the STEER show that nutrients in particular, along with sedimentation are frequently elevated. The Mangrove Lagoon and Benner Bay areas are of particular concern and in need of additional management efforts to reduce the input of these environmental stressors in order to ensure a healthy, functioning ecosystem.

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CHAPTER 10: SUMMARY AND CONCLUSIONS

NOAA's National Centers of Coastal Ocean Science along with local partners, and with funding from NOAA's Coral Reef Conservation Program (CRCP), conducted a three year chemical and biological assessment of the St. Thomas East End Reserves or STEER, in St. Thomas, USVI. The STEER, located on the southeastern end of the island, is a collection of four reserves and sanctuaries, and was established in order to manage the area as one comprehensive ecological unit, rather than separate management areas. The project was requested by local partners in order to assess the presence of anthropogenic stressors suspected of impacting living resources within the STEER, characterize the biological communities within the STEER, and establish a baseline that can be used to measure progress in restoring affected habitats.

Components of the project included: 1) development of an updated high resolution benthic habitat map for the STEER; 2) characterization of fish communities and associated benthic habitats throughout the STEER; 3) quantification of a suite of 185 organic (e.g., hydrocarbons) and inorganic (i.e., metals) chemical contaminants in sediment, coral, conch and fish; 4) follow-up field work to further quantify contaminants in surface sediments and in sediment cores from areas in Benner Bay found to have elevated levels of contaminants in the initial survey; 5) assessment of water-soluble chemical contaminants using passive water samplers; 6) determination of sediment toxicity using a set of established sediment toxicity bioassays; 7) characterization of the benthic infaunal community (organisms living within the sediments); 8) histologic examination of tissues from the coral *Porites astreoides* and finally; 9) monitoring of nutrients, sedimentation and total suspended solids (TSS) throughout the STEER.

The STEER contains a variety of habitats including coral reefs, seagrass beds and mangroves. Patch reefs are scattered throughout the STEER, interspersed with areas of unconsolidated sediments consisting primarily of sand, along with areas of gravel and of silt. Seagrasses are abundant throughout the STEER, and the largest remaining stand of mangroves in St. Thomas can be found in the STEER.

The STEER watershed contains high-density residential areas, the Bovoni Landfill that serves both St. Thomas and St. John, marinas and boat yards, various commercial/industrial activities, a number of resorts, and an EPA Superfund site, all of which have the potential to contribute a variety of land-based sources of pollution or LBSP, including chemical contaminants, nutrients and sediments. Live-

aboard boats moored within the STEER are also a likely source of pollution.

For the biological survey (fish communities and benthic habitats) of the STEER, a total of 80 sites were surveyed. Unfortunately, due to low visibility (high turbidity) and concerns for the health of divers related to water quality, a survey of the upper third of Mangrove Lagoon and also northern Benner Bay could not be carried out. These areas appear to be particularly impacted by LBSP. Results from other areas in the STEER indicated that coral cover and coral community structure were similar to that of coral reef areas in St. John and St. Croix in the USVI, and with selected locations in Puerto Rico. The low coral cover (5.2%) observed in the STEER is in line with values from similar studies and surveys in St. Croix (2.9%) and St. John (4.5%) found by NOAA NCCOS, and likely reflects the decline of coral reefs in the Caribbean as a whole. The high density urban population along with other activities within the watershed contributing LBSP, are additional stressors in the STEER.

Fish metrics in the STEER were similar to other U.S. Caribbean monitoring locations, using the same methodology. Biomass for grunts and snapper were higher in St. Croix than in the STEER, and may reflect the role that the extensive mangroves in the Mangrove Lagoon/Benner Bay area play, as nursery areas for juvenile fishes. The functioning of this part of the STEER as an important nursery area highlights the need to conserve and restore Mangrove Lagoon, which along with northern Benner Bay, appears to be the area in the STEER most impacted by LBSP. Had the divers been able to conduct their surveys within northern Mangrove Lagoon and northern Benner Bay, it seems likely that the impacts of LBSP in these areas on fish metrics and benthic habitats would have been seen.

Chemical contaminants in sediments sampled in the STEER were elevated in the Mangrove Lagoon and northern Benner Bay areas. A number of contaminants, particularly copper and tributyltin (TBT), were found at higher concentrations in the western part of the STEER, particularly in northern Benner Bay. Copper at one site in northern Benner Bay was above a NOAA sediment quality guideline indicating that effects on benthic organisms were likely. Tributyltin, banned for use as a boat hull antifoulant in the US, was found at the third highest concentration in the history of NOAA's National Status and Trends (NS&T) Program. Levels of several other metals were also elevated in Mangrove Lagoon. In addition, the benthic infaunal

community in both of these areas appeared to be severely diminished. Finally, the results of the bioassays indicated significant sediment toxicity in Mangrove Lagoon and Benner Bay using multiple tests.

From these results, the western portion of the STEER appears to be the most impacted by the chemical contaminants present. During the planning phase of the project, partners felt that Mangrove Lagoon would likely have high or very high concentrations of chemical contaminants in sediments, particularly metals, due to the proximity of the unlined Bovoni Landfill, located on the shores of Mangrove Lagoon. Somewhat surprisingly, however, northern Benner Bay had concentrations for a number of contaminants (e.g., copper, chromium, lead, zinc, TBT, total DDT, and total PCBs) that were as high if not higher than in Mangrove Lagoon. As noted in Chapter 4, other research has found evidence that the mangroves and the clay sediments in this area may be acting as a buffer, somewhat protecting Mangrove Lagoon for now from contaminants, particularly trace elements, originating from Bovoni Landfill, making their way into the lagoon. It should also be noted, that there may be areas within Mangrove Lagoon with high chemical contaminant concentrations (above sediment quality guidelines, for example), but because of the stratified random sampling design, were not sampled. Additional work within Mangrove Lagoon would help address inputs and impacts from the landfill. In particular, it would be useful to target areas within Mangrove Lagoon that appear to be receiving inputs of runoff directly from the landfill. It would be useful to sample not only sediments in these areas, but also deploy passive water samplers capable of sequestering trace elements (e.g., Chemcatcher®) from the water column, to see if they are being transported into Mangrove Lagoon and beyond from the landfill.

Following the initial assessment of chemical contaminants in sediments from the STEER, additional work was requested by the USVI Department of Planning and Natural Resources (DPNR), to further assess levels of TBT and trace elements in northern Benner Bay, particularly in the areas adjacent to the marinas and boat yards, which were found to be elevated during the initial assessment. DPNR noted that there had been a request to dredge the navigation channels leading to the marinas and boat yards in northern Benner Bay, in order to accommodate larger vessels. NC-COS scientists recommended that in addition to surface samples, sediment cores also be taken and analyzed to assess concentrations of contaminants in deeper, older sediments. Results from the follow-up chemical contaminant analysis indicated that some of the sediment cores were highly contaminated. Copper, for example, was found as

high as 1,540 µg/g in a sediment core, which is more than five times higher than the NOAA sediment quality guideline (SQG) concentration associated with toxic effects on benthic organisms. In addition, TBT was found at greater than 5,000 ng Sn/g, nearly an order of magnitude higher than the highest concentration (550 ng Sn/g) ever recorded in NOAA's NS&T Program, and over an order of magnitude higher than an upper screening value established to help determine when additional testing or assessments would be advisable at EPA Superfund sites (see Chapter 4). In sediments, TBT degrades to dibutyltin (DBT) and then monobutyltin (MBT), and finally to elemental tin, primarily through microbial action. The sum of TBT, DBT, and MBT is sometimes referred to as total butyltins. The total butyltins concentration was over 9,000 ng Sn/g in one sample from a sediment core in northern Benner Bay, eight times higher than the highest total butyltins concentration recorded by NOAA's NS&T Program. In addition, in those sediment cores from Benner Bay containing high levels of butyltins, the highest contribution was from TBT, which may indicate that in the deeper sediments from these cores, environmental conditions are such (e.g., anoxic sediments) that degradation of the TBT is proceeding very slowly.

The results from the follow-up contaminants work raises serious concerns regarding the effects that would occur from dredging the navigation channels or pile driving in the northern Benner Bay area. If the channels are dredged, the resuspension of highly contaminated sediments into the water column would likely impact biota in the area, particularly those in the adjacent mangroves, which has been shown through this project and others to be a valuable nursery area for juvenile fishes. In addition, once the sediments are dredged, the question becomes where and how to dispose of the highly contaminated dredge spoil, without impacting other environments, either marine or terrestrial.

The results of the deployment of the passive water samplers in the STEER indicated a fairly typical mix of water soluble contaminants, including those associated with detergents, personal care products such as DEET, indole and menthol, and plasticizers such as DEHP and DEP. The only compound found that exceeded water quality criteria was the plasticizer DEHP. In 1988, an EPA Superfund site was established in the STEER watershed, due to contamination of groundwater and wells by chlorinated volatile organic compounds (CVOCs), including tetrachloroethylene. As part of the remediation effort, groundwater treatment systems were put in place to remove CVOCs, including tetrachloroethylene. Significantly, there were no detections of tetrachloroethylene in the passive water samplers deployed in Turpentine Gut, Mangrove Lagoon or Benner Bay.

The results of the analysis of biota indicated that levels of chemical contaminants in conch were below FDA action guidelines for molluscan shellfish consumption. However, PCBs in one fish analyzed exceeded an EPA guideline for recreational fishers. Given the elevated contaminant levels found in sediments in portions of the STEER, more work is needed to assess contaminant levels in fish and also in invertebrates, in order to better understand the effects the contaminants in the STEER are having, not only in terms of human health, but also the effects on larvae and juvenile fish and other biota, particularly those that inhabit the western portion of the STEER. Trace and major elements along with concentrations of TBT would be candidate contaminants for additional investigations.

The histological analysis of coral tissue (*Porites astreoides*) in the STEER showed some pathologies (e.g., necrosis or apoptosis) in colonies from Benner Bay. However, there was no clear pattern between pathologies and sites. Also, there were almost no corals in Mangrove Lagoon, and the only ones found there were at the extreme southern end of the lagoon. The effects of low visibility, pollution, and slightly lower salinities likely precludes coral from existing in most of Mangrove Lagoon.

The monitoring of nutrients and sedimentation in the STEER indicated, as with a number of the other stressors measured during this project, that the western portion had higher inputs than the eastern and southern portions of the study area. Nutrient levels were significantly higher at the sites in the western portion, specifically in Benner Bay and Mangrove Lagoon. Comparisons with proposed coral reef criteria for nutrients indicated that approximately 50-60% of the samples collected during the course of the study, particularly in Mangrove Lagoon and Benner Bay, had levels above a suggested threshold for orthophosphate and dissolved inorganic nitrogen (DIN). It has been suggested that levels above the threshold can sustain macroalgal blooms on Caribbean coral reefs, which increases the risk of corals being outcompeted and ultimately overgrown by macroalgae. In addition, sedimentation rates appeared to be highest in the area of Benner Bay, and were close to a suggested rate, above which impacts to the structure and function of coral reefs have been associated.

The STEER contains multiple anthropogenic stressors. The evidence was strongest in the western portion of the STEER, where elevated concentrations of chemical contaminants in sediments, higher concentrations of water-soluble chemical contaminants, elevated nutrient and sedimentation rates and total suspended solids were found. Effects included evidence of reduced reproductive fitness

in corals from Mangrove Lagoon and increased necrosis in coral tissue in Benner Bay, and reduced and in some cases, severely reduced benthic infaunal communities (Mangrove Lagoon), all of which point to the impacts of these stressors on the STEER. Although fish metrics in the STEER were similar to other U.S. Caribbean monitoring locations in the STEER, that was for the areas where divers could conduct surveys. Had the divers been able to conduct the biological surveys in these areas, it seems likely that the biological communities there would have been diminished. All of these results point to the effects that anthropogenic stressors are having in the STEER. The impacts of LBSP on the western portion of the STEER is of particular concern as the mangroves and associated habitats serve as important nursery areas for a variety of vertebrate and invertebrate marine species. Loss of suitable habitat further impacts the ability of the STEER to be a source of new recruits of marine resource species for St. Thomas and beyond.



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