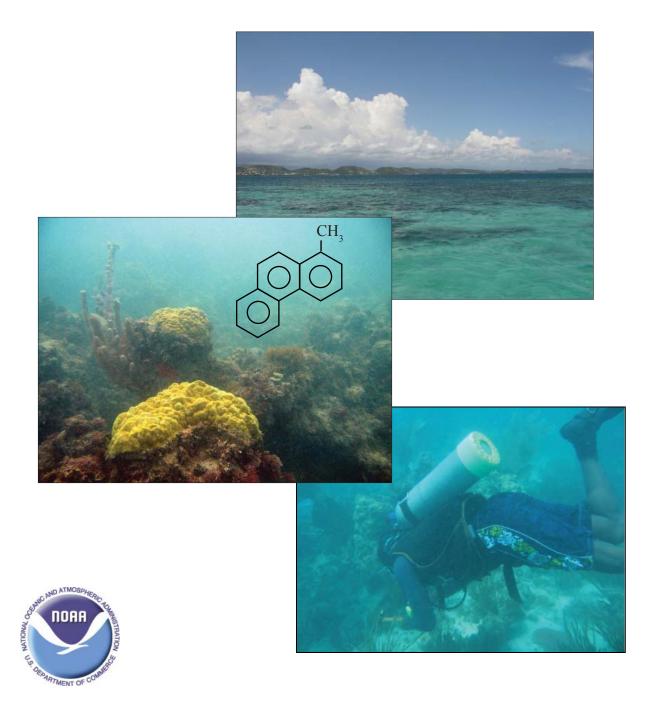
# Chemical Contaminants in the Coral *Porites astreoides* from Southwest Puerto Rico



NOAA Technical Memorandum NOS NCCOS 91

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# Chemical Contaminants in the Coral *Porites astreoides* from Southwest Puerto Rico

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# Table of Contents

List	of Tables	iii
List	of Figures	iii
I.	Introduction	1
II.	Materials and Methods. Study Area Sampling Design Coral Tissue Collection Coral Tissue Processing Chemical Contaminant Analysis Statistical Analysis	2 2 4 4 4
III.	Results and Discussion Field Data Chemical Contaminants Polycyclic Aromatic Hydrocarbons Polychlorinated Biphenyls DDT Other Organochlorine Pesticides Butyltins Trace Elements Chromium Copper Nickel Zinc	7 8 .11 .13 .15 .15 .16 .16 .16
IV.	Summary and Conclusions	. 20
V.	Acknowledgments	. 22
VI.	Literature Cited	. 23
VII	<ul> <li>Appendices</li> <li>A. Polycyclic aromatic hydrocarbons (PAHs) in coral (<i>Porites astreoides</i>) samples from southwest Puerto Rico (ng/dry g)</li> <li>B. Polychlorinated biphenyls (PCBs) in coral (<i>Porites astreoides</i>) samples from southwest Puerto Rico (ng/dry g)</li> </ul>	. 28
	C. Organochlorine pesticides in coral ( <i>Porites astreoides</i> ) samples from southwest Puerto Rico (ng/dry g)	. 30

D.	Butyltins in coral (Porites astreoides) samples from southwest Puerto Rico (ng/dry g)
E.	Trace and major elements in coral (Porites astreoides) samples from southwest
	Puerto Rico (ng/dry g)

## List of Tables

1.	Chemical contaminants analyzed in the coral tissues from southwest Puerto Rico	5
2.	Structures of selected organic compounds.	6
	Field data from the August 2005 collection of Porites astreoides	
4.	Mean and standard error (SE) of trace and major and elements in coral tissues	16

# List of Figures

1.	Coral sites sampled in southwest Puerto Rico	3
2.	Total PAHs in the coral Porites astreoides and in the sediments	9
3.	Comparison of total PAHs in the coral Porites astreoides normalized to lipid content, and	
	PAHs in the sediment normalized to percent fines (% silt + % clay)	10
4.	Total PCBs in the coral Porites astreoides and in the sediments	12
5.	Comparison of total PCBs in the coral Porites astreoides normalized to lipid content, and	
	PCBs in the sediment normalized to percent fines (% silt + % clay)	13
6.	Total DDT in the coral Porites astreoides and in the sediments	14
7.	Comparison of total DDT in the coral Porites astreoides normalized to lipid content, and	
	DDT in the sediment normalized to percent fines (% silt + % clay)	15
8.	Copper in the coral <i>Porites astreoides</i> and in the sediments	17
9.	Nickel in the coral Porites astreoides and in the sediments	19
	Zinc in the coral Porites astreoides and in the sediments	

iv

# Chemical Contaminants in the Coral *Porites astreoides* from Southwest Puerto Rico

This report presents an initial characterization of chemical contamination in coral tissues (<u>Porites</u> <u>astreoides</u>) from southwest Puerto Rico. It is the second technical report from a project to characterize chemical contaminants and assess linkages between contamination and coral condition. The first report quantified chemical contaminants in sediments from southwest Puerto Rico. This document summarizes the analysis of nearly 150 chemical contaminants in coral tissues. Although only eight coral samples were collected, some observations can be made on

the correlations between observed tissue and sediment contaminant concentrations. The concentrations of polycyclic aromatic hydrocarbons (PAHs), typically associated with petroleum spills and the combustion of fossil fuels, and polychlorinated biphenyls (PCBs) in the coral tissues were comparable to concentrations found in adjacent sediments. However,



Fishermen returning to La Parguera.

the concentration of a chemical contaminant (e.g., PAHs) in the coral tissues at a particular site was not a good predictor of what was in the adjacent sediments. In addition, the types of PAHs found in the coral tissues were somewhat different (higher ratios of alkylated PAHs) than in sediments. The levels of PCBs and DDT in coral tissues appeared higher just outside of Guanica Bay, and there was evidence of a downstream concentration gradient for these two contaminant classes. The trace elements copper, zinc and nickel were frequently detected in coral tissues, and the concentration in the corals was usually comparable to that found in adjacent sediments. Chromium was an exception in that it was not detected in any of the coral tissues analyzed. Additional work is needed to assess how spatial patterns in chemical contamination affect coral condition, abundance and distribution.

#### Introduction

Occurring in tropical and subtropical waters, coral reefs are among the most biologically rich ecosystems on earth, providing humans with living resources and services that have been valued at \$375 billion per year (Costanza *et al.*, 1997). Bryant *et al.* (1998) have estimated, however, that 58% of the world's coral reefs are at medium to high risk from overexploitation, coastal development, and pollution from inland and marine based sources. Bryant *et al.* (1998) concluded that pollution (both inland and

> marine based) is responsible for 34% of the coral reefs rated at medium to high risk. Although pollution has frequently been cited as a major factor in the decline of coral reefs (Burke and Maidens, 2004; Fabricius, 2005; Edinger *et al.*, 1998), the concentration of chemical contaminants present and their effects on corals is for the most part, unknown.

The US Coral Reef Task

Force (USCRTF) identified pollution as a key threat to coral reef resources in Puerto Rico, the US Virgin Islands, and southeast Florida (FDEP, 2004). Quantifying the types and concentrations of chemical contaminants present in reef areas is an important step towards understanding their impacts on corals and other reef organisms. Characterizing chemical contaminants present in coral reefs can also provide important baseline information needed to assess subsequent changes in environmental quality, as might result from the implementation of management actions (e.g., best management practices) to preserve or restore affected areas.

This report contains an initial characterization of coral contaminants in southwest Puerto Rico, and is the second report from a project to characterize chemical contaminants in the coral reefs in southwest Puerto Rico, and assess linkages between contaminants and coral condition. The first report (Pait *et al.*, 2007) detailed chemical contamination in sediments and included an assessment of the correlations between chemical contaminants and coral species richness. The analysis of chemical contaminants in coral tissues represents an extension of the capabilities of NOAA's National Status and Trends (NS&T) Program which has monitored the Nation's coastal waters for over 20 years.

The project in southwest Puerto Rico utilized an assessment framework developed within NOAA's National Centers for Coastal Ocean Science (NCCOS) that integrates environmental chemistry, biogeography, and geographic information system (GIS) analysis (both terrestrial and marine), to understand how anthropogenic inputs affect coral reef ecosystems. The

integration and concurrent analysis of these data types maximizes the likelihood of quantifying linkages between stressors (*e.g.*, chemical contaminants) and coral condition. This information provides a valuable baseline and enables managers to make better informed decisions on the use of finite resources (e.g., funds and personnel) to conserve and restore coral reef habitats. A longer term goal of the assessment framework is to incorporate a series of coral biomarkers, so that linkages between chemical contaminants and coral condition can be assessed at both the community and molecular levels.

Partners within NCCOS included the Center for Coastal Monitoring and Assessment (CCMA), the Center for Coastal Environmental Health and Biomolecular Research (CCEHBR) and the Hollings Marine Laboratory (HML). CCMA provided the chemical contaminant analysis along with the interpretation, and CCEHBR/HML provided laboratory space for the processing of coral tissues. The University of Puerto Rico-Mayagüez provided



Mustard hill coral, <u>Porites astreoides</u>. Image courtesy of NOAA CCMA Biogeography Branch.

expertise in local coral reef ecology, interpretation of data and a base of operations during field work.

#### *Materials and Methods Study Area*

The study area in southwest Puerto Rico was approximately 85 sq. km, and included the nearshore waters adjacent to the town of La Parguera and then east to Guanica Bay (Figure 1). The area is within a NOAA-funded Coral Reef Ecosystem Study (CRES) site, and has been designated as a Natural Reserve

> by the Puerto Rico Department of Natural and Environmental Resources (DNER). In this part of Puerto Rico, the shelf area extends approximately 10 km from shore, then drops off at about 20 m. The shelf has both emergent and submerged reefs and contains a variety of hard and soft corals, along with extensive seagrass beds, coastal fringing mangroves and a series of mangrove islands (Kendall *et al.*, 2001; Christensen *et al.*, 2003). The predominant

longshore flow in the study area is from east to west (CFMC, 1998). The town of La Parguera has a population of approximately 26,000 and is a popular weekend and vacation destination. The Guanica Bay watershed in the eastern portion of the study area contains the towns of Ensenada and Guanica, and over the years several industrial operations including sugar processing, fertilizer mixing, and textile manufacture have operated there. In addition, the Lajas Valley which has a significant amount of agricultural production, drains to Guanica Bay through a series of manmade canals (Figure 1).

#### Sampling Design

For this study, a stratified random sampling design based on mapped habitat was used to select the sampling points. Strata included lagoon, backreef and bankshelf areas. Determining the location of coral and sediments in the study area was made possible by the extensive benthic habitat mapping completed by CCMA's Biogeography Branch (Kendall *et al.*, 2001). Using ArcGIS®, sampling





points where both sediments and corals could be collected were identified.

## Coral Tissue Collection

Mustard hill coral, *Porites astreoides*, was collected and analyzed from eight sites for this project (Figure 1). *P. astreoides* was chosen as it is a common species of coral in Florida, the Bahamas and the Caribbean (Humann and DeLoach, 2002), and occurs throughout the study area. Colonies of *P. astreoides* are generally massive but can also be found as encrusting forms, particularly in shallow, surging waters (Veron, 2000).

Samples were collected from the vessel *Aquanauta* using a GPS programmed with the station coordinates. The coral samples were taken by NCCOS SCUBA divers (see inset) in August 2005 using a hammer and an acetone-rinsed punch. The punch produced a coral core with a diameter of 1.2 cm and a

similar core length. Approximately 20 cores were taken at each site and placed in a 50ml Teflon<sup>®</sup> centrifuge tube underwater. The tube was then brought to the surface, drained of water and placed immediately in a dewar charged with liquid nitrogen. At the end of the mission, the dewars were shipped to the Hollings Marine Laboratory (HML) in Charleston, South Carolina and the samples were stored at -80° C prior to processing.



The coral tissues were processed using the protocols found in Downs *et al.* (2005). Briefly, the corals were ground in an acid washed, solvent-rinsed (acetone) mortar and pestle. Liquid nitrogen was added before the grinding commenced to cool the mortar, and during the grinding process to prevent the tissues from thawing and to keep the coral matrix brittle to facilitate the grinding process. The ground coral was placed into a precleaned jar, and then frozen.

### Chemical Contaminant Analysis

TDI-Brooks International, and the Trace Element Research Laboratory (TERL) at Texas A&M University, both in College Station, conducted the organic and inorganic analyses, respectively. A list of the nearly 150 compounds analyzed in the coral samples is shown in Table 1, which included polycyclic aromatic hydrocarbons (PAHs), polychlorinated biphenyls (PCBs), organochlorine pesticides, butyltins, and trace and major elements. All coral cores from a site were composited in order to have enough material to detect the contaminants of interest. Each reported coral contaminant value in this report represents the analysis of one composite sample from a site.

PAHs are typically associated with the use and combustion of fossil fuels such as oil and gasoline. PAHs are toxic to both aquatic and terrestrial organisms. In corals, the PAHs fluoranthene and pyrene have been shown to be toxic to adult corals, particularly in



were stored at -80° C prior to Coral tissue collection using hammer and punch.

the presence of increased ultraviolet radiation (phototoxicity) (Peachey and Crosby, 1996; Guzman-Martinez *et al.*, 2007). PAHs were analyzed using gas chromatography/ mass spectrometry in the selected ion monitoring mode.

Polychlorinated biphenyls or PCBs (Table 2) are a class of synthetic compounds that were used in numerous applications

ranging from electrical transformers and capacitors, to hydraulic and heat transfer fluids, to pesticides and paints. PCBs readily accumulate in the environment and are toxic (reproductive and developmental toxicity) to biota. Solbakken *et al.* (1984) investigated the bioconcentration of radiolabeled hexaPCB (2,4,5,2',4',5'-hexachlorobiphenyl) in coral. The PCB was rapidly accumulated in *Diploria strigosa* and *Madracis decatis*, however, depuration proceeded at a slow rate; after 275 days nearly 33 percent of the original radioactivity from the PCB remained in the coral. Most uses of PCBs were banned in the US in

Naphthalene 1-Methylnaphthalene	PAHs - High Molecular Weight	<b>Organochlorine Pesticides</b>	PCBs	Major and Trace Elements
1-Methylnaphthalene	Fluoranthene	Aldrin	PCB8/5	Aluminum (Al)
	Pyrene	Dieldrin	PCB18	Arsenic (As)
2-Methylnaphthalene	C1-Fluoranthenes/Pyrenes	Endrin	PCB28	Cadmium (Cd)
2,6-Dimethylnaphthalene	C2-Fluoranthenes/Pyrenes	Heptachlor	PCB29	Chromium (Cr)
1,6,7-Trimethylnaphthalene	C3-Fluoranthenes/Pyrenes	Heptachlor-Epoxide	PCB31	Copper (Cu)
C1-Naphthalenes	Naphthobenzothiophene	Oxychlordane	PCB44	Iron (Fe)
C2-Naphthalenes	C1-Naphthobenzothiophenes	Alpha-Chlordane	PCB45	Lead (Pb)
C3-Naphthalenes	C2-Naphthobenzothiophenes	Gamma-Chlordane	PCB49	Manganese (Mn)
C4-Naphthalenes	C3-Naphthobenzothiophenes	Trans-Nonachlor	PCB52	Mercury (Hg)
Benzothiophene	Benz[a]anthracene	Cis-Nonachlor	PCB56/60	Nickel (Ni)
C1-Benzothiophenes <sup>1</sup>	Chrysene	Alpha-HCH	PCB66	Selenium (Se)
C2-Benzothiophenes <sup>1</sup>	C1-Chrysenes	Beta-HCH	PCB70	Silicon (Si)
C3-Benzothiophenes <sup>1</sup>	C2-Chrysenes	Delta-HCH	PCB74/61	Silver (Ag)
Biphenyl	C3-Chrysenes	Gamma-HCH	PCB87/115	Tin (Sn)
Acenaphthylene	C4-Chrysenes	2,4'-DDD	PCB95	Zinc (Zn)
Acenaphthene	Benzo[b]fluoranthene	4,4'-DDD	PCB99	
Dibenzofuran	Benzo[k]fluoranthene	2,4'-DDE	PCB101/90	
Fluorene	Benzo[e]pyrene	4,4'-DDE	PCB105	
C1-Fluorenes	Benzo[a]pyrene	2,4'-DDT	PCB110/77	
C2-Fluorenes	Perylene	4,4'-DDT	PCB118	
C3-Fluorenes	Indeno[1,2,3-c,d]pyrene	1,2,3,4-Tetrachlorobenzene	PCB128	
Anthracene	Dibenzo[a,h]anthracene	1,2,4,5-Tetrachlorobenzene	PCB138/160	
Phenanthrene	C1-Dibenzo[a,h]anthracenes	Hexachlorobenzene	PCB146	
1-Methylphenanthrene	C2-Dibenzo[a,h]anthracenes	Pentachloroanisole	PCB149/123	
C1-Phenanthrene/Anthracenes	C3-Dibenzo[a,h]anthracenes	Pentachlorobenzene	PCB151	
C2-Phenanthrene/Anthracenes	Benzo[g,h,i]perylene	Endosulfan II	PCB153/132	
C3-Phenanthrene/Anthracenes		Endosulfan I	PCB156/171/202	
C4-Phenanthrene/Anthracenes	Butyltins	Endosulfan Sulfate	PCB158	
Dibenzothiophene	Monobutyltin	Mirex	PCB170/190	
C1-Dibenzothiophenes <sup>1</sup>	Dibutyltin	Chlorpyrifos	PCB174	
C2-Dibenzothiophenes <sup>1</sup>	Tributyltin		PCB180	
C3-Dibenzothiophenes	Tetrabutyltin		PCB183	
			PCB18/	
			PCD194	
			PCD201/157/172	
			PCB206	
			DCB200	

<sup>1</sup>The benzothiophenes and dibenzothiophenes are thio-PAHs; Abbreviations: PAHs, polycyclic aromatic hydrocarbons; HCH, hexachlorocyclohexane; DDT, DDD, dichlorodiphenyldichloroethane; DDE, dichlorodiphenyldichloroethylene; PCB, polychlorinated biphenyl

the late 1970s. PCBs were analyzed using gas chromatography/ electron capture detection.

A number of organochlorine pesticides, including DDT (Table 2) and chlordane were used as insecticides in the past. Organochlorine pesticides are typically neurotoxins, environmentally persistent and readily

Table 2. Structures of selected organic compounds.

_	Compound Class	Compound	Structure	Use
	Polycyclic aromatic hydrocarbon (PAH)	Benzo[a]pyrene		Byproduct of use and combustion of fossil fuels
	Polychlorinated Biphenyl	2,2', 5,5'-Tetra Chlorinated Biphenyl		Former widespread use in transformers, capacitors and hydraulic and heat transfer applications
	Organochlorine Pesticide	DDT	$Cl \rightarrow Cl \rightarrow Cl$	Insecticide, banned in US in 1972
;	Butyltin	Tributyltin (TBT) (X = anion or anionic group)	$C_{4}H_{9}$ $H_{9}C_{4}-Sn-X$ $C_{4}H_{9}$	Biocide; banned on smaller vessels (<25 m) in the US in 1988

accumulate in biota. Both PCBs and DDT have been shown to interfere with the endocrine system. Wang *et al.* (2008) showed that DDT accumulates in the coral *Porites evermanni*. Organochlorine pesticides were also analyzed using gas chromatography/electron capture detection.

Butyltins have a range of uses from biocides to catalysts to glass coatings. Tributyltin or TBT was widely used in antifoulant paints for boat hulls beginning in the late 1960s. Banned on smaller vessels in the US beginning in 1988, the use of TBT was associated with endocrine disruption, specifically an imposex condition in marine gastropod molluscs. Negri et al. (2002) investigated the effects of TBT in sediments on the coral Acropora microphthalma. Sediments were collected from an area where a cargo ship had grounded on Australia's Great Barrier Reef. Sediments contained approximately 160 µg/g TBT, and when diluted to 5 percent of the original concentration, prevented successful settlement of coral larvae in the laboratory on preconditioned terracotta tiles. In the project in southwest Puerto Rico, butyltins were quantified using gas chromatography/flame photometry.

Fifteen trace and major elements (Table 1) were analyzed in the coral tissues. Unlike a number of

the organic compounds, all of the trace and major elements are found naturally to some extent in the environment. Mining and manufacturing processes, along with the use and disposal of products can result in elevated concentrations, particularly for trace elements, in the environment. A number of trace elements are toxic at low concentrations. Cadmium, and chromium for example, can impair development and reproduction in several invertebrate species.

Reichelt-Brushett and Harrison (2005) found that a copper concentration of 20  $\mu$ g/L and higher significantly reduced the fertilization success in brain coral *Goniastrea aspera*. At copper concentrations at or above 75  $\mu$ g/L, fertilization success was one percent or less. Fertilization success was also significantly reduced in *Acropora longicyathus* at 24  $\mu$ g/L, similar to *G. aspera*.

Victor and Richmond (2005) found that copper concentrations of 10  $\mu$ g/L and higher significantly affected fertilization in the reef building coral *Acropora surculosa* in the laboratory after five hours. They also found that embryo development was affected when gametes were exposed to copper at concentrations of 12  $\mu$ g/L. At concentrations of 58  $\mu$ g/L or higher, no embryo development was observed.

Site	Collection	Latitude	Longitude	Water Depth	Dissolved Oxygen	Temperature	Salinity	Conductivity	Secchi Depth
	Date	(DD)	(DD)	(m)	(mg/L)	(°C)	(ppt)	(mS)	(m)
Coral 1	8/27/05	17.94219	-66.91481	9.1	4.63	30.0	33.9	56.8	3.7
Coral 2	8/27/05	17.93012	-66.93719	7.0	4.61	29.6	33.8	56.1	6.7
Coral 3	8/27/05	17.96335	-67.02396	10.7	5.32	30.1	34.4	57.5	5.8
Coral 4	8/28/05	17.93594	-67.04942	18.9	5.04	29.6	34.3	57.0	7.9
Coral 5	8/27/05	17.96351	-67.01735	9.1	5.31	30.1	34.5	52.5	bottom
Coral 6	8/27/05	17.96334	-67.04679	13.7	4.81	29.6	34.5	57.3	7.3
Coral 8	8/28/05	17.93339	-67.10513	4.3	4.89	30.1	34.5	57.7	bottom
Coral 9	8/28/05	17.9362	-67.11609	4.6	4.62	30.1	34.6	57.9	3.7

Table 3. Field data from the August 2005 collection of Porites astreoides.

Abbreviations: DD, decimal degrees; m, meters; mg/L, milligrams/liter; ppt, parts per thousand; mS, milli-Siemans.

Reichelt-Brushett and Michalek-Wagner (2005) investigated the effects of copper on the soft coral *Lobophytum compactum*. A significant difference in fertilization success was found at a copper concentration of 117  $\mu$ g/L.

Goh and Chou (1997) found that a copper concentration of 40  $\mu$ g/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.

Almost all of the work to assess the toxicity of chemical contaminants to corals, including the toxicity of trace and major elements, has been carried out using aqueous concentrations of contaminants. Very little work has been carried out to assess the toxicity of contaminants to corals which are adsorbed to sediments. As a result, the concentration of contaminants in the aqueous phase that cause effects in corals, can't be directly compared to what was found in the sediments in southwest Puerto Rico or in the tissues of *P. astreoides*, in terms of effects. However, the work that has been carried out by researchers shows that a variety of chemical contaminants can have toxic effects in corals.

Silver, cadmium, copper, lead, and tin were analyzed using inductively coupled plasma - mass spectrometry. Aluminum, arsenic, chromium, iron, manganese, nickel, silicon and zinc were analyzed using inductively coupled plasma - optical emission spectrometry. Mercury was analyzed using cold vapor - atomic absorption spectrometry. Selenium was analyzed using atomic fluorescence spectrometry. Total metal concentration was analyzed for this project. Additional information on the protocols used for the analysis of the organic and the inorganic contaminants can be found in Kimbrough *et al.* (2006), and Kimbrough and Lauenstein (2006), respectively.

#### Statistical Analysis

All contaminant data were analyzed using JMP® statistical software. None of the coral data were normally distributed, and transformations (log10) were not effective. As a result, nonparametric tests including Wilcoxon rank-sum, Spearman's correlation, and Wilcoxon matched-pairs signed rank tests were run to examine differences, for example, in total PAH concentration between coral and sediment, and concentration differences between coral and sediment at a site. Because the coral cores sampled at a site were composited for one analysis, between site comparisons for chemical contaminants in corals were not possible. Results are presented at the 0.05 level of significance.

### **Results and Discussion**

#### Field Data

Data collected in the field in August 2005 are shown in Table 3. The water depth ranged from 4 to nearly 19 meters. *P. astreoides* was abundant at the sites sampled, and occurred mainly as the encrusting form. The largest colony diameter measured approximately 40 cm.

Dissolved oxygen near the surface (< 1 m) ranged between 4 and 5 mg/L (ppm). Secchi depth was used

as a crude indicator of water clarity. The Secchi depth was lowest (3.7 m) at the Coral 1 and Coral 9 stations. Coral 1 is at the mouth of Guanica Bay, and may be influenced by the outflow from this waterbody. A slight sediment plume is visible at the mouth of Guanica Bay in Figure 1. The erosion of clayey soils in the watershed from cleared upland areas, inputs from the Lajas Valley agricultural areas, and scouring along the Rio Loco have been suggested as sources of the turbidity (P. Sturm, pers. comm.)

#### Chemical Contaminants

There are relatively few publications on the presence of chemical contaminants in coral tissues; most of

these appear to be for metals. To the best of our knowledge, this is the first publication that quantifies chemical contaminants present in corals collected from southwest Puerto Rico. When possible, the concentrations of chemical contaminants found in the coral tissues are compared to those found in corals elsewhere. In addition, coral contaminant levels are compared to contaminant levels found in the sediments, as both sediments and living tissue tend to accumulate



Encrusting form of <u>Porites astreoides</u>, similar to the size of the colonies sampled. Diameter of colony shown is approximately 25 cm.

hydrophobic organic compounds as well as trace elements. The results of the chemical contaminant analysis in the coral tissues are reported on a dry weight basis. More detailed information on the results (e.g., individual contaminants) can be found in Appendices A - E.

*Polycyclic Aromatic Hydrocarbons.* Total PAHs in *P. astreoides* and in adjacent sediments are shown in Figure 2. More detailed information on the concentration of individual PAHs can be found in Appendix A. Although there was substantial variation in total PAHs in corals and sediments across the study area, a nonparametric (Wilcoxon) analysis indicated the concentration of total PAHs in the corals was not significantly different (P > 0.05, ChiSquare = 0.5404) from the concentration in the sediments. The concentration

tion of total PAHs in the sediment at a particular site, however, did not covary with the concentration in the coral (P > 0.05, Spearman Rho = -0.1429), and the Wilcoxon matched-pairs test indicated no significant correlation (P = 0.4609) between the paired measurements over the study area. Because a limited number of coral samples were taken as part of this study, additional work is needed to assess the relationship between PAHs in *P. astreoides* and in sediments.

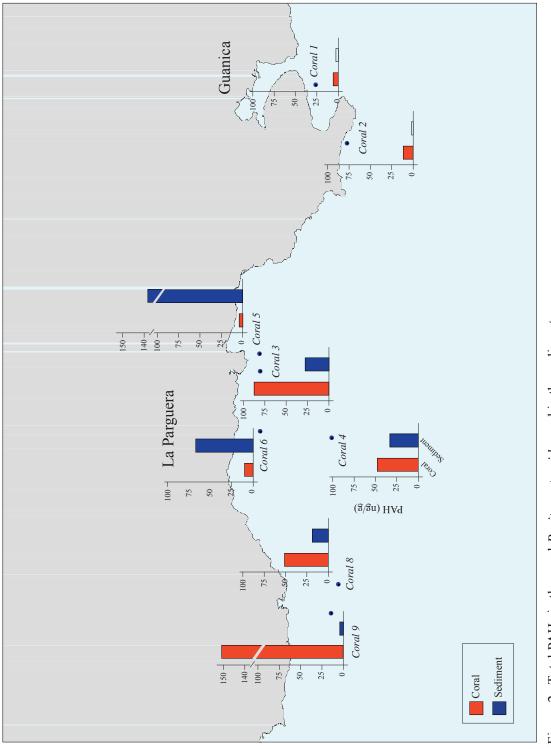
Total PAHs in the coral tissues were somewhat elevated at Coral 3 and Coral 4, near the town of La Parguera. There is a significant amount of recreational boat traffic in and out of La Parguera, and this

> along with stormwater runoff and local atmospheric deposition from the town (e.g., from automobile traffic), may be some of the sources for PAHs.

The highest level of total PAHs in coral tissues were found at Coral 9 (Figure 2), in the western portion of the study area, which is away from the town of La Parguera. Coral 9 is adjacent to the emergent reefs of El Palo and Margarita (Figure 1). The reef area is somewhat

offshore, but could be subjected to recreational boating activities. As will be seen below, the PAHs found in the coral tissues at Coral 9 were more indicative of uncombusted petroleum products than combustion byproducts. In Figure 2, it can also be seen that the sediment concentration of PAHs at Coral 9 was substantially lower than the concentration found in the coral tissues. It is unclear why there was a difference in the concentration of PAHs in coral versus sediments at Coral 9. Kennedy *et al.* (1992) showed that benzo[a]pyrene is rapidly taken up by the boulder coral *Montastrea annularis*, while Knap *et al.* (1982) showed that the brain coral *Diploria strigosa* rapidly concentrated the PAH phenanthrene.

The ratio of phenanthrene to anthracene has been used as an indicator of petrogenic (e.g., petroleum





product from fuel spills or other discharges) versus pyrogenic (high temperature combustion) sources of PAHs (Neff *et al.*, 2005; Budzinski *et al.*, 1997). The mean phenanthrene:anthracene (P/A) ratio for the sediments at the coral sites was  $4.6 \pm 0.8$ . For the coral tissues,

Organic compounds such as PAHs, PCBs and DDT tend to accumulate in sediments with higher silt and clay fractions. Pait *et al.* (2007) normalized the sediment concentration of PAHs from southwest Puerto Rico to the percent fines (% silt plus the % clay frac-

however, the P/A ratio was  $11.7 \pm$ 2.1. Budzinski et al. (1997) defined a P/A ratio <10 as being indicative of pyrogenic sources, and greater than 10 for petrogenic inputs, possibly indicating higher accumulation of petrogenic-type PAHs in the corals collected from southwest Puerto Rico.

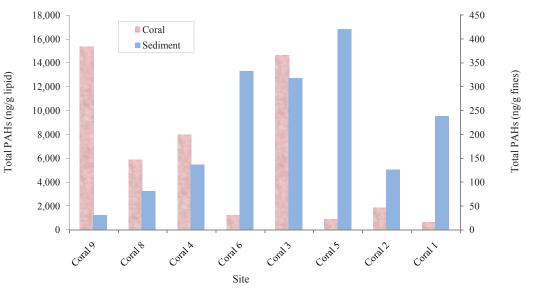


Figure 3. Comparison of total PAHs in the coral *Porites astreoides* normalized to lipid content, and PAHs in the sediment normalized to percent fines (% silt + % clay). Sites are organized from west (left side) to east (right side).

The ratio of nonalkylated or "parent" PAHs

to alkylated forms has also been used to distinguish petrogenic from pyrogenic sources (Neff et al., 2005; Columbo et al., 1989). In general, alkylated PAHs are more abundant than nonalkylated in petroleum products, relative to combustion byproducts (Neff et al., 2005). In the sediments adjacent to the coral sites, the nonalkylated/ alkylated ratio was  $2.78 \pm$ 0.39. The ratio in the coral tissues was  $1.06 \pm 0.49$ , and that difference was significant (P < 0.05). At Coral 9, the nonalkylated to alkylated PAH ratio indicated that 95% of the PAHs detected in the coral tissues were alkylated. It appears that the corals collected in this limited sampling accumulated greater amounts of those PAHs that would be associated with petrogenic sources such as oil or gasoline. Additional work is needed to assess whether corals are exposed to or preferentially accumulate greater levels of alkylated PAHs, or if the elevated levels of alkylated PAHs represent more recent accumulation of PAHs, compared to the sediments.

tion), an approach which has been used to help identify sources of contaminants such as PAHs (Birch, 2003; Burgess *et al.*, 2001). An analogous approach can also be used for biological tissues, by dividing the organic contaminant concentration by the percent lipid of the tissues, to normalize the contaminant concentration and help identify possible sources (Elskus *et al.*, 2005). The results of normalizing PAHs by coral lipid content and sediment by the fines can be seen in Figure 3. The results for the corals and sediments were similar to what was seen in Figure 2, i.e., higher PAH levels in corals in the central and western portions of the study area, and higher PAH levels in sediments around the town of La Parguera.

Only two published papers were found that quantified PAHs in corals. Thomas and Li (2000) used supercritical extraction of ground and air dried samples of *Porites compressa* followed by immunoaffinity chromatography, and analysis by gas chromatography and mass spectrometry. Corals were collected at Kaneohe Bay in Oahu, Hawaii, which is impacted by industrial, military and civilian use and wastewater discharges. Thomas and Li (2000) analyzed for a number of PAHs including fluorene, phenanthrene, anthracene, fluoranthene, pyrene, chrysene, benzo[e] pyrene, and benz[a]pyrene. The total concentration for these PAHs in the coral from Kaneohe Bay was 220 ng/g. Total PAH, as defined for the work in southwest Puerto Rico is the sum of the 53 PAHs in Appendix A. In the corals from southwest Puerto Rico, the mean total PAH concentration was 41.9

 $\pm 17.1$  ng/g. However, when the same PAHs that Thomas and Li (2000) analyzed are summed for southwest Puerto Rico, the mean total was lower, 2.2  $\pm 0.48$  ng/g.

Readman *et al.* (1996) analyzed for PAHs (including phenanthrene, fluoranthene, and pyrene) in the coral *Porites lutea* from Kuwait. In sections of the coral corresponding to the 1988/1989 timeframe (prior to the Gulf War), the concentration of these PAHs was approxi-



Grinding of coral tissues in preparation for analysis of chemical contaminants.

mately 0.3 ng/g. It should be noted, however, that Readman *et al.* (1996) sectioned and analyzed coral skeleton for PAHs.

*Polychlorinated Biphenyls (PCBs).* Although no longer manufactured in the U.S., the accumulative and persistent nature of a number of the 209 congeners (configurations) of PCBs (Table 2) have resulted in their becoming ubiquitous contaminants in the environment. Some use (e.g., certain types of transformers) of PCBs is still permitted in the US.

The results of total PCB analysis in samples of *P. astreoides* and in adjacent sediments is shown in Figure 4. A summary by individual PCB congener can be found in Appendix B. Total PCBs were significantly higher in the sediments compared to coral tissues (P<0.05). Overall, the concentration of total PCBs in the corals did not covary (P > 0.05, Spearman Rho =

-0.3095) with total PCBs in the sediments, however, the Wilcoxon matched-pairs test indicated a significant correlation (P < 0.05) between individual paired measurements.

At the two sites just outside of Guanica Bay (Coral 1 and Coral 2), the concentrations of total PCBs were similar in both coral tissues and sediments. At these sites, there was also some evidence of a concentration gradient in the tissues moving downstream from the mouth of Guanica Bay. As part of the overall project, two sites within Guanica Bay were sampled for sedi-

> ment contaminants (Pait et al., 2007). The analysis of the sediment samples from inside Guanica Bay revealed substantially elevated (> 1,000 ng/g) and toxicologically relevant levels of PCBs (Pait et al., 2008a). The results presented here may indicate that PCBs are being transported out of Guanica Bay and into surrounding waters. The small number of corals collected as part of this project, however, highlights the need for additional work to assess not only the

relationships between PCBs in coral tissues and sediments, but also whether a gradient of PCBs in coral tissues and in sediments moving downstream from Guanica Bay exists.

In Figure 5, the concentration of total PCBs is normalized to percent lipid (coral) and % fines (sediment). The sites are plotted in a west to east direction to be comparable with the map. Figure 5 shows a decreasing total PCB normalized concentration in sediments moving downstream from Guanica Bay.

Unfortunately, there do not appear to be many published papers on the accumulation of PCBs in corals. Additional information is needed not only on the presence of PCBs in coral tissues but also on their effects in coral and other coral reef organisms. Two articles were located that contained an analysis of PCBs in coral tissues. El Nemr *et al.* (2004) analyzed

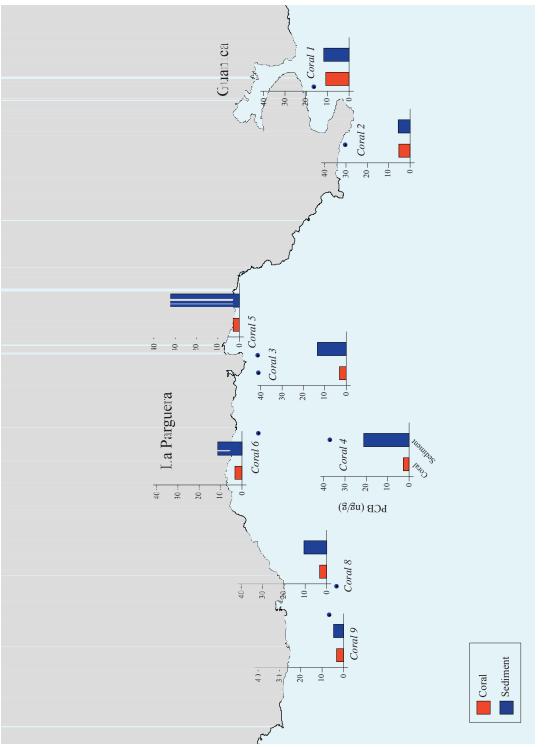


Figure 4. Total PCBs in the coral *Porites astreoides* and in the sediments.

Acropora sp. tissues from a number of sites along the Egyptian Red Sea Coast. The average concentration of seven congeners in the coral tissues was 18 ng/g (El Nemr et al., 2004) The highest concentration at the sites sampled in southwest Puerto Rico for these same congeners in P. astreoides was only 0.64 ng/g.

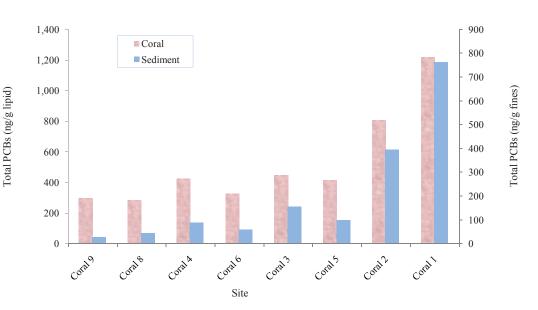


Figure 5. Comparison of total PCBs in the coral *Porites astreoides* normalized to lipid content, and PCBs in the sediment normalized to percent fines (% silt + % clay). Sites are organized from west (left side) to east (right side).

Miao *et al.* (2000) analyzed coral

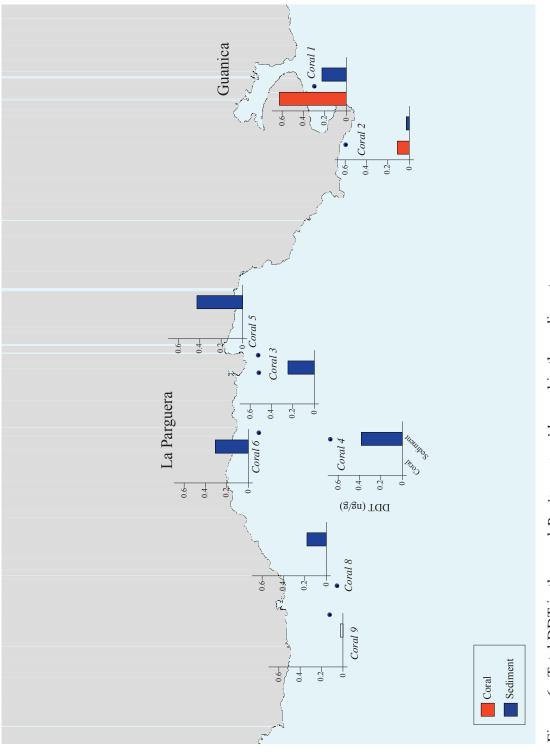
(Porites lobata) and sediment samples for a number of PCB congeners from four sites in the French Frigate Shoals in the Northwest Hawaiian Islands. Seventeen of the congeners analyzed by Miao et al. (2000) were also analyzed in the coral samples from southwest Puerto Rico. The approximate mean concentration for these congeners in P. lobata was 110 ng/g. In P. astreoides from southwest Puerto Rico, the mean for the same 17 congeners was less than 1 ng/g. Miao et al. (2000) noted that over the years, both the military (1942-1946) and the US Coast Guard (1952-1979) have had airfields and refueling stations on the adjacent Tern Island. In addition, large quantities of unidentified debris have been landfilled on the island, and elevated levels of PCBs have been detected in the soils (Miao et al., 2000). The approximate mean concentration for the 17 congeners in sediments at the sites in the French Frigate Shoals was 100 ng/g. At the coral sites in southwest Puerto Rico, the mean concentration for these same PCB congeners in the sediments was lower,  $5.70 \pm 1.67$ ng/g, indicating a lower level of PCB contamination at the coral sites sampled.

*DDT.* The map of total DDT detected in *P. astre-oides* and in the sediments at the coral sites is shown

in Figure 6. A number of organochlorine pesticides, including DDT were used extensively as insecticides in the past. Organochlorine pesticides are typically neurotoxins, environmentally persistent, and readily accumulate in biota.

Total DDT refers to the sum of the two parent compounds, 2,4'-DDT and 4,4'-DDT, along with metabolites DDD and DDE. Detailed information on the concentration of parent and degradation products of DDT along with the other organochlorine pesticides measured in the coral tissues can be found in Appendix C. Total DDT in coral was only detected at the two sites just outside of Guanica Bay. Total DDT concentration at Coral 1 was 0.62 ng/g and at Coral 2, the total DDT concentration was 0.11 ng/g. At both sites, the concentration of total DDT in the coral tissues was higher than in the sediments. As with the PCBs, there was some evidence of a concentration gradient, indicating that DDT transported out of Guanica Bay could be a source in the coral tissues just outside the Bay. Additional analyses of samples would help confirm if a concentration gradient exists.

Within Guanica Bay, total DDT was elevated in the two sediment samples taken as part of this project (Pait *et al.*, 2007). At one of the two sites, total





DDT in the sediment (46.9 ng/g) slightly exceeded the Effects Range-Median (ERM) concentration (46.1 ng/g),indicating a greater likelihood of biological effects on biota inhabiting this area. The ERM is the concentration above which toxicity in test organisms (typically amphipods) is frequently (50th percentile) observed (Long et al., 1998).

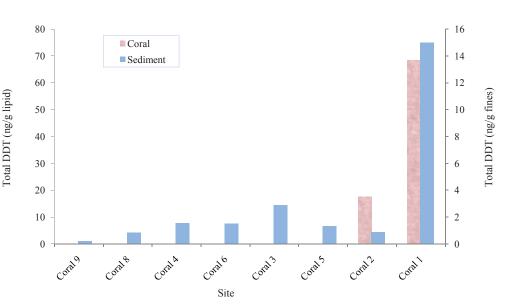


Figure 7. Comparison of total DDT in the coral *Porites astreoides* normalized to lipid content, and DDT in the sediment normalized to percent fines (% silt + % clay). Sites are organized from west (left side) to east (right side).

The Lajas Valley (Figure 1) is situated inland from a series of coastal hills known as the Sierra Bermeja which separates the narrow coastal plain containing La Parguera from the valley. Historically, the Lajas Valley was used for extensive sugarcane production, but now has significant pastureland along with the production of selected vegetable and melon crops (USDA, 2004). Although now banned in the US, the insecticide DDT was likely used in the Lajas Valley in the past to control insects on crops such as sugarcane and pineapple.

As with PCBs, a comparison can be made between total DDT found in the coral tissues and in the sediments, normalized to percent lipids and percent fines, respectively. Figure 7 shows higher normalized total DDT in the coral tissues, and a similar pattern in the sediments in the area adjacent to Guanica Bay, indicating that the DDT transported out of the Bay may be a source in corals and adjacent sediments.

A few studies were found that quantified DDT and its metabolites in coral tissues. Glynn *et al.* (1995) collected and analyzed sediment and *P. astreoides* for residues of total DDT from the Florida Keys National Marine Sanctuary, in the area near Key Largo. The highest concentration detected was 0.01 ng/g. Glynn *et al.* (1989) also looked at total DDT in *P. astreoides* further north, in Biscayne National Park, in an area known as Alina's Reef, and detected higher concentrations, ranging from 3.41 - 43.56 ng/g. El Nemr *et al.* (2004) detected a mean total DDT concentration of  $5.7 \pm 0.8$  ng/g (dry weight) in coral tissues from the Egyptian Red Sea Coast.

*Other Organochlorine Pesticides*. None of the other organochlorine pesticides (e.g., dieldrin, chlordane, and endosulfan) analyzed in the coral tissues were detected (Appendix C). Glynn *et al.* (1995) found concentrations of other organochlorine pesticides near or below the level of detection in samples of *P. astreoides* in Florida. Glynn *et al.* (1989) and El Nemr *et al.* (2004) were able to detect a series of other organochlorine pesticides in coral tissues from Florida and along the Egyptian Red Sea Coast, respectively.

*Butyltins*. Tributyltin (TBT), the compound formerly used in antifoulant paints for boat hulls and associated with endocrine disruption in gastropods, was not found in any of the coral tissue samples analyzed,

or in any of the adjacent sediments. The degradation products of TBT, namely dibutyltin and monobutyltin were found at low levels ( $\leq 2$  ng/g) in coral tissues (Appendix D) and sediments ( $\leq 1$  ng/g).

Trace Elements. A total of 15 elements were analyzed in the tissues of P. astreoides. The mean and standard errors for the elements analyzed are shown in Table 4. More detailed information can be found in Appendix E. Unlike a number of the organic compounds analyzed, all of the trace and major elements are found naturally to some extent in the environment. When viewing the results, it should be noted that the concentration units for the trace and major elements ( $\mu g/g$ ), are different from the organics (ng/g).

Chromium. Chromium was not detected in any of the coral tissues analyzed, although this metal was detected in the sediments from all eight of the coral sites. Although there were only eight coral samples taken as part of this project, it was expected that chromium would have been quantifiable in P. astreoides at some of the sites, particularly adjacent to Guanica Bay. At Coral 1, just outside of Guanica Bay, the chromium concentration in the sediment was 11.3  $\mu$ g/g. As part of the overall project, two sediment samples were taken within Guanica Bay. The concentration of chromium at these two sites was 394 and 440  $\mu$ g/g (Pait et al., 2008b). Both of these values are above the ERM of 370  $\mu$ g/g, indicating that chromium may be having an adverse effect on the benthic organisms at these sites.

It is not clear why chromium was not found in any of the coral tissues sampled. It may be related to the small number of samples taken as part of this project. Alternatively, it could be related to the uptake characteristics of *P. astreoides*. Esslemont (2000) noted

Table 4. Mean and standard error (SE) of trace and major elements in coral tissues.

Element	Mean (µg/g)	SE
Silver (Ag)	< 0.01	< 0.01
Aluminum (Al)	37.8	7.31
Arsenic (As)	0.00	_
Cadmium (Cd)	0.00	_
Chromium (Cr)	0.00	_
Copper (Cu)	2.06	0.30
Iron (Fe)	90.8	37.7
Mercury (Hg)	0.00	_
Manganese (Mn)	3.01	0.58
Nickel (Ni)	1.32	0.30
Lead (Pb)	< 0.01	< 0.01
Selenium (Se)	0.05	0.05
Silicon (Si)	62.8	8.27
Tin (Sn)	0.02	0.02
Zinc (Zn)	6.09	1.82

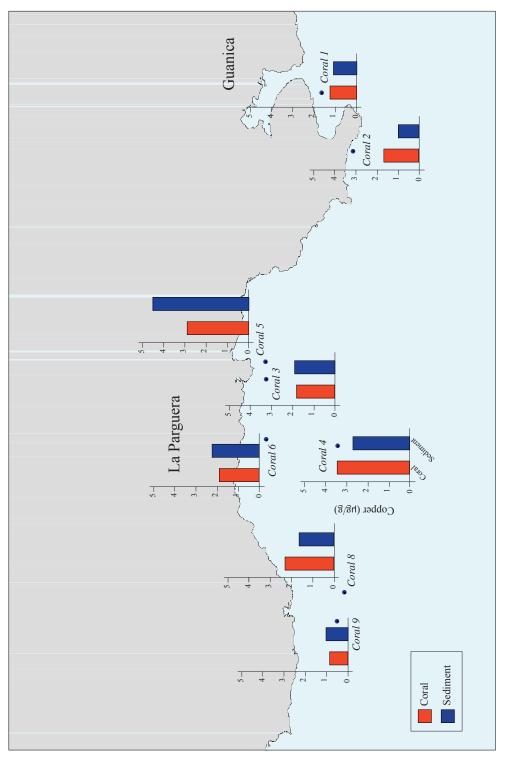
SE, standard error

differences in the uptake of certain metals between coral species, and concluded that some species (*G. aspera* and *Pocillopora damicornis*) showed evidence of being able to regulate metals in more contaminated areas.

A number of authors have detected chromium in coral skeletons and tissues. Esslemont (2000) analyzed samples of G. aspera, Acropora formosa and P. damicornis on the Great Barrier Reef in Australia for trace elements. Using a technique that analyzed both the tissue and coral skeleton, Esslemont (2000) detected chromium in concentrations from 15 - 65  $\mu$ g/g. In a second technique that analyzed only the living tissue, chromium

concentrations were even higher, ranging from 52 - 110  $\mu$ g/g. In sediments, Esslemont (2000) noted that total chromium was lower than in corals, the highest was around 18  $\mu$ g/g, similar to the 11.3  $\mu$ g/g found in the sediments just outside of Guanica Bay. Scott and Davies (1997) detected chromium in coral skeleton from the genus *Porites* (species not identified) in Indonesia, at concentrations up to 1.5  $\mu$ g/g. Additional work is needed to assess whether chromium is present in *P. astreoides* or perhaps other corals in the study area, particularly since elevated chromium concentrations were found in the sediments within Guanica Bay (Pait *et al.*, 2008b).

*Copper*. The results of the analysis of copper in the tissues of *P. astreoides* is shown in Figure 8. The average copper concentration in the coral tissues was  $2.06 \pm 0.30 \ \mu\text{g/g}$  (Table 4). The highest concentration of copper detected in the coral tissues sampled was  $2.94 \ \mu\text{g/g}$  at Coral 5 (Figure 8). This was also the site where the highest copper was found in the adjacent sediments (4.51  $\mu$ g/g). A nonparametric (Wilcoxon) analysis indicated that copper in corals was not sig-





nificantly different (P > 0.05, ChiSquare = 0.6744) from the concentration in adjacent sediments, and a Spearman's test indicated that the concentration of copper in the sediment covaried with the concentration in the coral (P < 0.05). The Wilcoxon matched-pairs test, however, indicated no significant correlation (P = 0.7422) between paired measurements.

A number of other studies have found that copper can accumulate in coral tissues. David (2003) investigated the concentration of trace elements using a pneumatic drill to obtain cores of *P. lobata* from the western coast of Marinduque Island in the Philippines. Sites were established adjacent to a stockpile of old mine tailings, as well as a site that was not exposed (reference site) to the contamination. The mean copper concentration in the coral cores from

the area that had received input from the stockpile was  $3.1 \ \mu g/g$ . The concentration in the corals taken from the reference site was  $0.7 \ \mu g/g$ .

Glynn *et al.* (1989) investigated the presence of a series of heavy metals in coral tissues within Biscayne Bay National Park off southeast Florida. Several coral species including *P. astreoides* were collected and analyzed. Both living tissue and associated coral skeleton were analyzed. The copper

concentrations typically ranged from 5 to 12  $\mu$ g/g, although the copper in one sample from Alina's Reef was 90  $\mu$ g/g.

Scott and Davies (1997) detected copper in *Porites spp.* at concentrations up to 18  $\mu$ g/g in Indonesia. The higher concentrations of copper were found in reefs adjacent to Male, the capital city of the Republic of Maldives.

Esslemont (2000) also analyzed for copper in the tissue and skeleton of four species of coral at sites on the Great Barrier Reef, and detected copper in the 2 to 5  $\mu$ g/g range, similar to what was found in *P. astreoides*. The highest copper concentration, roughly

28  $\mu$ g/g was found in *G. aspera*, in the Townsville Harbor area.

Miao *et al.* (2001) analyzed both tissue and skeleton of *P. evermanni* from the French Frigate Shoals in the Northwest Hawaiian Islands, and also from the island of Oahu in Hawaii. Concentrations of copper in *P. evermanni* were on the order of 5  $\mu$ g/g in the French Frigate Shoals and somewhat higher (8  $\mu$ g/g) near Oahu.

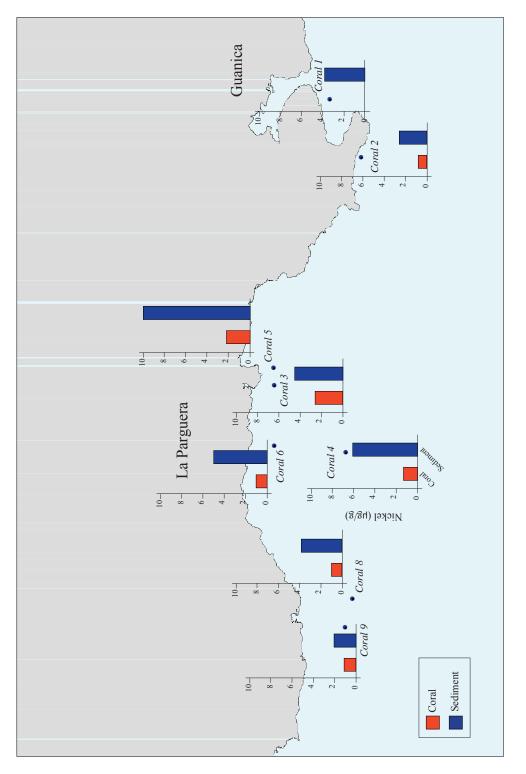
*Nickel*. The concentration of nickel detected in the tissues of *P. astreoides* and in adjacent sediments is shown in Figure 9. The mean concentration of nickel in the coral tissues was  $1.32 \pm 0.30 \ \mu g/g$ ; and in the adjacent sediments,  $4.78 \pm 0.94 \ \mu g/g$ . At all the coral sites, the concentration of nickel in the sediments was

higher than in the coral tissues. A Wilcoxon test indicated that the nickel concentration in the corals was different from that in the sediments (P < 0.05). In addition, the concentration of nickel in the sediment at a particular site did not covary with the concentration in the coral (P > 0.05, Spearman Rho)= 0.4048), and a Wilcoxon matched-pairs test indicated a significant difference (P < 0.05) between paired measurements.

At Coral 1, just outside of Guanica Bay, nickel was not detected in the sample of *P. astreoides*. At the two sediment sites in Guanica Bay (Pait *et al.*, 2008b), the concentration of nickel was elevated, 434 and 442  $\mu$ g/g, substantially above the ERM value of 52  $\mu$ g/g. As with chromium, additional work is needed to better assess the presence of nickel in corals in the area adjacent to Guanica Bay. Esslemont (2000) characterized trace elements in sediments and corals in the Townsville section of the Great Barrier Reef Marine Park. As part of that work, the bioavailability of trace elements with a series of solvents to determine the readily bioavailable fraction



Abandoned industrial facility on the shores of Guanica Bay.





versus the fraction that was more recalcitrant. Esslemont (2000) found that trace elements such as lead, copper and zinc were more biologically available in the sediments from the Townsville Harbor area, while chromium, nickel and cadmium were less bioavailable.

Because only one sample was taken in the area outside of the mouth of Guanica Bay, it is not possible to conclude that elevated levels of nickel are not present in *P. astreoides* in this area. It would be useful to further characterize the extent of nickel in sediments inside and outside of Guanica Bay and in coral tissues, to better assess whether nickel is being transported out of the Bay.

A number of papers have been published on nickel in coral tissues. Reichelt-Brushett and McOrist (2003) analyzed both living and nonliving components of the coral *Acropora tenuis* on the Great Barrier Reef. Skeletal concentrations of nickel ranged from 0. 3 to 0.7  $\mu$ g/g, while tissues ranged from 0.7 to 3.8  $\mu$ g/g, similar to the concentrations found in *P. astreoides* in southwest Puerto Rico.

Esslemont (2000) analyzed tissue and skeleton in *G. aspera*, and found concentrations ranging from 10 to 20  $\mu$ g/g, higher than what was found in *P. astreoides* tissues in southwest Puerto Rico. Scott and Davies (1997) detected nickel in coral skeleton from *Porites spp.*, ranging from 0.25 to 5  $\mu$ g/g.

*Zinc*. The results of the zinc analysis in P. *astreoides* is shown in Figure 10 and in Appendix E. Zinc was detected in all of the coral samples analyzed.

The average concentration of zinc in *P. astreoides* was  $6.09 \pm 1.82 \ \mu g/g$ . A nonparametric (Wilcoxon) analysis indicated that the concentration of zinc in corals was not significantly different (P > 0.05, ChiSquare = 2.8235) from the concentration in the adjacent sediments. However, the concentration of zinc in the sediment at a particular site was not a good predictor of the concentration of zinc in coral tissue (P > 0.05, Spearman Rho = -0.1905). This can also be seen in Figure 10. At Coral 9 (18.3  $\mu g/g$ ) for example, the concentration of zinc in the tissues was nearly an order of magnitude higher than what was found in the sediments (1.88  $\mu g/g$ ).

Zinc has been quantified in coral tissues from a number of areas. Miao *et al.* (2001) analyzed whole samples of *P. evermanni* from the French Frigate Shoals in the Northwest Hawaiian Islands. The highest zinc concentration detected in the coral was 4.3  $\mu g/g$ , similar to the mean zinc found in *P. astreoides*. At Disappearing Island in the French Frigate Shoals, zinc was measured in both the coral and sediment. The concentration of zinc in the coral was 2.3  $\mu g/g$ ; the concentration of zinc detected in the sediment was 7  $\mu g/g$ . David (2003) detected zinc concentrations between 1 and 2  $\mu g/g$  in cores taken from *P. lobata* in the Philippines.

A number of other assessments have been done to quantify zinc in coral skeleton. In their analysis of *A*. *tenuis* from the Great Barrier Reef, Reichelt-Brushett and McOrist (2003) detected skeletal concentrations ranging from 0.2 to 0.5  $\mu$ g/g, while zinc in the tissues separated from the coral matrix contained approximately 17  $\mu$ g/g.

Esslemont (1999) detected a mean zinc concentration in the skeleton of the coral *G. aspera* of 1.87  $\pm 0.75 \ \mu g/g$  at a site in the Great Barrier Reef (Heron Island). In corals collected in Darwin Harbor (Northern Territory), the mean zinc concentration in the coral skeleton was higher,  $4.07 \pm 0.93 \ \mu g/g$ . In a second species, *Acropora annuligera*, the mean zinc concentration in the coral skeleton was  $1.59 \pm 0.62 \ \mu g/g$ .

Bastidas and Garcia (1999) analyzed skeletal sections in *P. astreoides* from the northwest coast of Venezuela. Two sites were selected within the Parque Nacional Morrocoy, an area noted by the authors as being impacted by industrial activities, tourism and sedimentation from terrestrial inputs. Tissues were removed prior to extraction and analysis. The mean concentrations of zinc in the coral skeleton at the two sites sampled was  $10.67 \pm 0.84$  and  $9.12 \pm 0.71 \mu g/g$ .

### Summary and Conclusions

This is the second technical report from a project to characterize chemical contaminants in sediments and in the coral *Porites astreoides* from southwest Puerto Rico, and assess linkages between chemical contaminants and coral condition. The study area for the

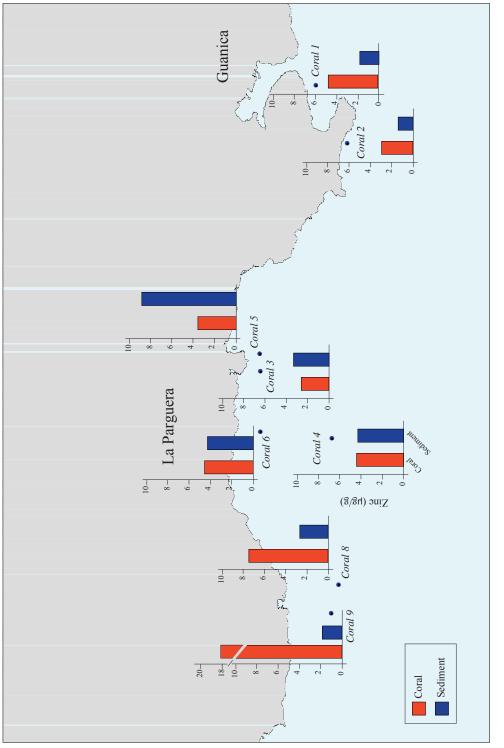


Figure 10. Zinc in the coral Porites astreoides and in the sediments.

project extended from Guanica Bay west to the area around the town of La Parguera.

The results of the chemical contaminant analysis indicated accumulation of organic contaminants such as PAHs and PCBs, and trace elements such as copper and zinc. Similar concentrations of contaminants were found in coral tissues and sediments although sediment concentrations were sometimes higher.

An interesting phenomenon was seen for PAHs (polycyclic aromatic hydrocarbons). At some sites, the sediments contained higher levels of PAHs, while at others, coral tissues were higher. The reason for this difference is unknown, although the low number of coral samples analyzed for this project may be responsible. In addition, the corals appeared to contain higher levels of PAHs associated with petrogenic, or uncombusted hydrocarbon sources compared to the sediments. This was particularly true in the western portion of the study area, and could be an indicator of boat traffic through the area, or perhaps other unidentified sources. The higher levels of petrogenic PAHs in the living coral tissues could also be an indicator of more recent deposition, or perhaps differences in bioavailability of certain PAHs to coral tissues. The ability of coral to metabolize PAHs is unknown. Additional sampling and analysis of corals is needed to better understand the distribution of this contaminant class.

There was also some, although limited, evidence of a contaminant concentration gradient in the coral tissues moving away from Guanica Bay. Levels of total PCBs and total DDT were higher at the sampling site just outside of Guanica Bay, and lower at the site further away and downstream of the outflow from Guanica Bay. The same was true when the coral were normalized to % lipid. Pait *et al.* (2008a) found that two sites sampled within Guanica Bay for sediments had elevated levels of total DDT and total PCBs. A similar pattern was seen in the sediments, particularly when the sediments were normalized for the smaller (silt and clay) grain sizes.

For the trace elements, copper and zinc appeared to be accumulated in *P. astreoides* at levels similar to what was found in the adjacent sediments. Chromium, however, was not detected in any of the coral samples analyzed for this project. This included the site (Coral 1) just outside of Guanica Bay. Pait *et al.* (2008b) found substantially elevated levels of chromium in the sediments at the two sites sampled within the Bay. This is also true for nickel which appears elevated within Guanica Bay sediments, but which was not detected in the coral sample taken just outside the Bay.

Additional sampling and analysis in the study area would increase the number of data points and allow a better understanding of the accumulation of chemical contaminants in *P. astreoides*, the relationship between contaminants in coral tissues and in sediments, and the transport of contaminants within and out of Guanica Bay to the surrounding coral reef areas.

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Appendices

Appendix A. Polycyclic aromatic hydrocarbons (PAHs) in coral (*Porites astreoides*) samples from south-west Puerto Rico (ng/dry g).

Compound	Coral 1	Coral 2	Coral 3	Coral 4	Coral 5	Coral 6	Coral 8	Coral 9	Blank
Decalin	0.0 U								
C1-Decalin	0.0 U								
C2-Decalin C3-Decalin	0.0 U 0.0 U								
C4-Decalin	0.0 U 0.0 U								
Naphthalene	1.5 J	1.2 J	1.8 J	1.1 J	1.3 J	1.2 J	1.4 J	1.7 J	0.6 J
C1-Naphthalenes	0.8 J	0.9 J	1.1 J	0.8 J	1.0 J	0.8 J	0.8 J	1.0 J	0.3 J
C2-Naphthalenes	1.1 J	1.1 J	1.2 J	1.2 J	1.2 J	1.2 J	1.5 J	0.0 U	0.0 U
C3-Naphthalenes	0.0 U								
C4-Naphthalenes	0.0 U								
Benzothiophene	0.0 U								
C1-Benzothiophene	0.0 U	0.0 U	0.0 U	0.0 U 0.0 U	0.0 U 0.0 U	0.0 U	0.0 U	0.0 U	0.0 U
C2-Benzothiophene C3-Benzothiophene	0.0 U 0.0 U								
Biphenyl	0.0 U 0.9 J	0.0 U 0.9 J	1.0 J	0.0 U 0.9 J	1.0 J	0.0 U 0.9 J	1.1 J	1.1 J	0.6 J
Acenaphthylene	0.0 U								
Acenaphthene	0.0 U								
Dibenzofuran	0.5 J	0.3 J	0.6 J	0.4 J	0.4 J	0.6 J	0.6 J	0.5 J	0.3 J
Fluorene	0.0 U	0.0 U	0.3 J	0.2 J	0.0 U	0.3 J	0.4 J	0.2 J	0.0 U
C1-Fluorenes	0.0 U	0.0 U	1.6 J	1.3 J	0.0 U	0.0 U	1.3 J	1.4 J	0.0 U
C2-Fluorenes	0.0 U	0.0 U	7.8	3.3	0.0 U	0.0 U	4.0	7.7	0.0 U
C3-Fluorenes	0.0 U	0.0 U	9.3	5.0	0.0 U	0.0 U	5.7	15.4	0.0 U
Anthracene Phenanthrene	0.0 U 0.5 J	0.1 J 0.6 J	0.2 J 2.4 J	0.1 J 1.5 J	0.1 J 0.6 J	0.2 J 1.4 J	0.1 J 2.0 J	0.2 J 2.0 J	0.0 U 0.3 J
C1-Phenanthrenes/Anthracene	0.5 J 0.0 U	0.6 J 0.8 J	2.4 J 9.9	1.5 J 5.2	0.6 J 0.0 U	1.4 J 0.0 U	2.0 J 6.4	2.0 J 14.5	0.3 J 0.0 U
C2-Phenanthrenes/Anthracene	0.0 U 0.0 U	0.8 J 2.7 J	9.9 16.1	9.0	0.0 U 0.0 U	0.0 U 0.0 U	8.9	31.6	0.0 U 0.0 U
C3-Phenanthrenes/Anthracene	0.0 U	2.2 J	10.1	6.2	0.0 U	0.0 U	6.0	21.6	0.0 U
C4-Phenanthrenes/Anthracene	0.0 U	12.3	0.0 U						
Dibenzothiophene	0.0 U	0.0 U	0.5 J	0.1 J	0.0 U	0.0 U	0.3 J	0.2 J	0.0 U
C1-Dibenzothiophenes	0.0 U	0.0 U	4.6	2.1	0.0 U	0.0 U	2.8	5.7	0.0 U
C2-Dibenzothiophenes	0.0 U	0.0 U	8.6	4.7	0.0 U	0.0 U	4.9	16.5	0.0 U
C3-Dibenzothiophenes	0.0 U	0.0 U	6.8	4.4	0.0 U	0.0 U	4.2	19.2	0.0 U
Fluoranthene Pyrene	0.2 J 0.2 J	0.1 J 0.1 J	0.4 J 0.2 J	0.2 J 0.2 J	0.3 J 0.2 J	1.2 J 0.9 J	0.3 J 0.2 J	0.5 J 0.4 J	0.0 U 0.0 U
C1-Fluoranthenes/Pyrenes	0.2 J 0.0 U	0.1 J 0.0 U	0.2 J 1.8 J	0.2 J 0.0 U	0.2 J 0.0 U	0.9 J 0.0 U	0.2 J 0.0 U	0.4 J 0.0 U	0.0 U 0.0 U
C2-Fluoranthenes/Pyrenes	0.0 U 0.0 U	0.0 U	1.8 J	0.0 U					
C3-Fluoranthenes/Pyrenes	0.0 U								
Naphthobenzothiophene	0.0 U								
C1-Naphthobenzothiophene	0.0 U								
C2-Naphthobenzothiophene	0.0 U								
C3-Naphthobenzothiophene	0.0 U								
Benz(a)anthracene	0.0 U	0.0 U	0.1 J	0.0 U	0.0 U	0.6 J	0.0 U	0.0 U	0.0 U
Chrysene C1-Chrysenes	0.0 U 0.0 U	0.0 U 0.0 U	0.2 J 0.0 U	0.0 U 0.0 U	0.0 U 0.0 U	0.4 J 0.0 U	0.0 U 0.0 U	0.0 U 0.0 U	0.0 U 0.0 U
C2-Chrysenes	0.0 U 0.0 U								
C3-Chrysenes	0.0 U								
C4-Chrysenes	0.0 U								
Benzo(b)fluoranthene	0.0 U								
Benzo(k)fluoranthene	0.0 U								
Benzo(e)pyrene	0.0 U								
Benzo(a)pyrene	0.0 U								
Perylene	0.0 U								
Indeno(1,2,3-c,d)pyrene Dibenzo(a,h)anthracene	0.0 U 0.0 U								
C1-Dibenzo(a,h)anthracene	0.0 U 0.0 U								
C2-Dibenzo(a,h)anthracene	0.0 U 0.0 U								
C3-Dibenzo(a,h)anthracene	0.0 U								
Benzo(g,h,i)perylene	0.0 U								
Total PAHs	5.7	11.0	87.8	47.9	6.1	9.7	52.9	154	2.1
Individual Alkyl Isomers*									
2-Methylnaphthalene	0.9 J	0.9 J	1.0 J	0.8 J	1.0 J	0.9 J	0.9 J	1.0 J	0.4 J
1-Methylnaphthalene	0.5 J	0.5 J	0.7 J	0.5 J	0.6 J	0.4 J	0.5 J	0.6 J	0.2 J
2,6-Dimethylnaphthalene	0.3 J	0.4 J	0.5 J	0.4 J	0.6 J	0.6 J	0.7 J	0.2 J	0.0 U
1,6,7-Trimethylnaphthalene	0.0 U								
1-Methylphenanthrene	0.0 U	0.3 J	2.5	1.4 7.7 I	0.0 U	0.0 U	1.9	3.4	0.0 U
C30-Hopane	0.0 U	7.8 J	13.9	7.7 J	0.0 U	0.0 U	5.5 J	37.9	0.0

Notes: J, below method detection level, MDL; U, not detected; Blank was ground glass processed in the same way as the samples \*Individual isomers contained in alkylated (C1-C4 sums)

Appendix B. Polychlorinated biphenyls (PCBs) in coral (*Porites astreoides*) samples from southwest Puerto Rico (ng/dry g).

Compound	Coral 1	Coral 2	Coral 3	Coral 4	Coral 5	Coral 6	Coral 8	Coral 9	Blank
PCB8/5	0.00 U	0.00 U							
PCB18	0.00 U	0.00 U							
PCB28	0.00 U	0.00 U							
PCB29	0.00 U	0.00 U							
PCB31	0.00 U	0.00 U							
PCB44	0.00 U	0.00 U							
PCB45	0.00 U	0.00 U							
PCB49	0.00 U	0.00 U							
PCB52	0.21 J	0.00 U	0.00 U	0.00 U	0.08 J	0.00 U	0.00 U	0.00 U	0.00 U
PCB56/60	0.00 U	0.00 U							
PCB66	0.00 U	0.00 U							
PCB70	0.00 U	0.00 U							
PCB74/61	0.00 U	0.00 U							
PCB87/115	0.00 U	0.00 U							
PCB95	0.00 U	0.00 U							
PCB99	0.17 J	0.00 U	0.00 U						
PCB101/90	0.33	0.00 U	0.00 U						
PCB105	0.00 U	0.05 J	0.00 U	0.00 U					
PCB110/77	0.00 U	0.00 U							
PCB118	0.00 U	0.08 J	0.00 U	0.00 U					
PCB128	0.00 U	0.00 U							
PCB138/160	0.00 U	0.00 U							
PCB146	0.26	0.09 J	0.00 U	0.00 U					
PCB149/123	0.00 U	0.00 U							
PCB151	0.00 U	0.00 U							
PCB153/132	1.75	0.69	0.22 J	0.16 J	0.25	0.19 J	0.18 J	0.35	0.00 U
PCB156/171/202	0.15 J	0.00 U	0.00 U						
PCB158	0.00 U	0.00 U							
PCB170/190	0.46	0.14 J	0.00 U	0.00 U					
PCB174	0.11 J	0.00 U	0.00 U						
PCB180	0.58	0.00 U	0.00 U						
PCB183	0.13 J	0.00 U	0.00 U						
PCB187	0.67	0.25	0.00 U	0.00 U					
PCB194	0.18 J	0.00 U	0.00 U						
PCB195/208	0.00 U	0.00 U							
PCB199	0.39	0.00 U	0.00 U						
PCB201/157/173	0.00 U	0.00 U							
PCB206	0.00 U	0.00 U							
PCB209	0.00 U	0.00 U							
Total PCBs	10.97	4.84	2.68	2.55	2.91	2.60	2.57	2.97	2.19

Notes: J, below method detection level, MDL; U, not detected; Blank was ground glass processed in the same way as the samples.

Compound	Coral 1	Coral 2	Coral 3	Coral 4	Coral 5	Coral 6	Coral 8	Coral 9	Blank
Aldrin	0.00 U	0.00 U							
Dieldrin	0.00 U	0.00 U							
Endrin	0.00 U	0.00 U							
Heptachlor	0.00 U	0.00 U							
Heptachlor-Epoxide	0.00 U	0.00 U							
Oxychlordane	0.00 U	0.00 U							
Alpha-Chlordane	0.00 U	0.00 U							
Gamma-Chlordane	0.00 U	0.00 U							
Trans-Nonachlor	0.00 U	0.00 U							
Cis-Nonachlor	0.00 U	0.00 U							
Alpha-HCH	0.00 U	0.00 U							
Beta-HCH	0.00 U	0.00 U							
Delta-HCH	0.00 U	0.00 U							
Gamma-HCH	0.00 U	0.00 U							
2,4'-DDD	0.15 J	0.06 J	0.00 U	0.00 U					
4,4'-DDD	0.00 U	0.00 U							
2,4'-DDE	0.00 U	0.00 U							
4,4'-DDE	0.24	0.05 J	0.00 U	0.00 U					
2,4'-DDT	0.22 J	0.00 U	0.00 U						
4,4'-DDT	0.00 U	0.00 U							
1,2,3,4-Tetrachlorobenzene	0.00 U	0.00 U							
1,2,4,5-Tetrachlorobenzene	0.00 U	0.00 U							
Hexachlorobenzene	0.00 U	0.00 U							
Pentachloroanisole	0.00 U	0.00 U							
Pentachlorobenzene	0.00 U	0.00 U							
Endosulfan II	0.00 U	0.00 U							
Endosulfan I	0.00 U	0.00 U							
Endosulfan Sulfate	0.00 U	0.00 U							
Mirex	0.00 U	0.00 U							
Chlorpyrifos	0.00 U	0.00 U							
Total HCH	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Total Chlordane	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Total DDTs	0.62	0.11	0.00	0.00	0.00	0.00	0.00	0.00	0.00

Appendix C. Organochlorine pesticides in coral (*Porites astreoides*) samples from southwest Puerto Rico (ng/dry g).

Notes: J, below method detection level, MDL; U, not detected; Blank was ground glass processed in the same way as the samples.

Annendix D	Butyltins in coral	(Porites astreoides	) samples from	southwest Puerto Rico	(ng/dry g)
Appendix D.	Durynins in corar	(1 ornes usireolues	<i>j</i> samples nom	southwest I delto Rico	(ing/uny g).

Compound	Coral 1	Coral 2	Coral 3	Coral 4	Coral 5	Coral 6	Coral 8	Coral 9	Blank
Monobutyltin	1.83 J	1.67 J	0.00 U	2.08 J	1.40 J	1.66 J	1.24 J	1.97 J	1.83 J
Dibutyltin	0.00 U	1.18 J	1.46 J	1.23 J	1.32 J	1.23 J	1.09 J	1.56 J	0.69 J
Tributyltin	0.00 U	0.00 U							
Tetrabutyltin	0.00 U	0.00 U							

Notes: J, below method detection level, MDL; U, not detected; Blank was ground glass processed in the same way as the samples.

Compound	Coral 1	Coral 2	Coral 3	Coral 4	Coral 5	Coral 6	Coral 8	Coral 9	Blank
Ag	0 U	0 U	0 U	0 U	0 U	0 U	0.0227	0.016	0.0939
AĬ	40.6	82.2	40.5	34.1	44.3	21.6	23.3	15.9	41.1
As	0 U	0 U	0 U	0 U	0 U	0 U	0 U	0 U	0 U
Cd	0 U	0 U	0 U	0 U	0 U	0 U	0 U	0 U	0 U
Cr	0 U	0 U	0 U	0 U	0 U	0 U	0 U	0 U	0 U
Cu	1.24	1.76	1.82	3.54	2.94	1.94	2.34	0.922	0 U
Fe	59.8	68.5	59.9	50.4	61.9	37.4	353	35.2	2.83
Hg	0 U	0 U	0 U	0 U	0 U	0 U	0 U	0 U	0 U
Mn	4.69	4.12	2.09	1.63	5.36	1.55	3.58	1.05	0 U
Ni	0 U	0.832	2.7	1.25	2.31	0.939	1.32	1.17	0 U
Pb	0 U	0.0734	0 U	0 U	0 U	0 U	0 U	0 U	0 U
Se	0 U	0 U	0 U	0 U	0 U	0.374	0 U	0 U	0 U
Si	71.1	105	69.6	56.3	79.6	45.5	43.2	32.3	119
Sn	0 U	0 U	0 U	0 U	0 U	0.143	0 U	0 U	0 U
Zn	4.81	2.93	2.62	4.55	3.5	4.53	7.46	18.3	0.961

Appendix E. Trace and major elements in coral (*Porites astreoides*) samples from southwest Puerto Rico ( $\mu$ g/dry g).

Qualifiers: J=Below the MDL, U=Not detected

United States Department of Commerce

Gary F. Locke Secretary

National Oceanic and Atmospheric Administration

Jane Lubchenco Under Secretary of Commerce for Oceans and Atmospheres

National Ocean Service

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