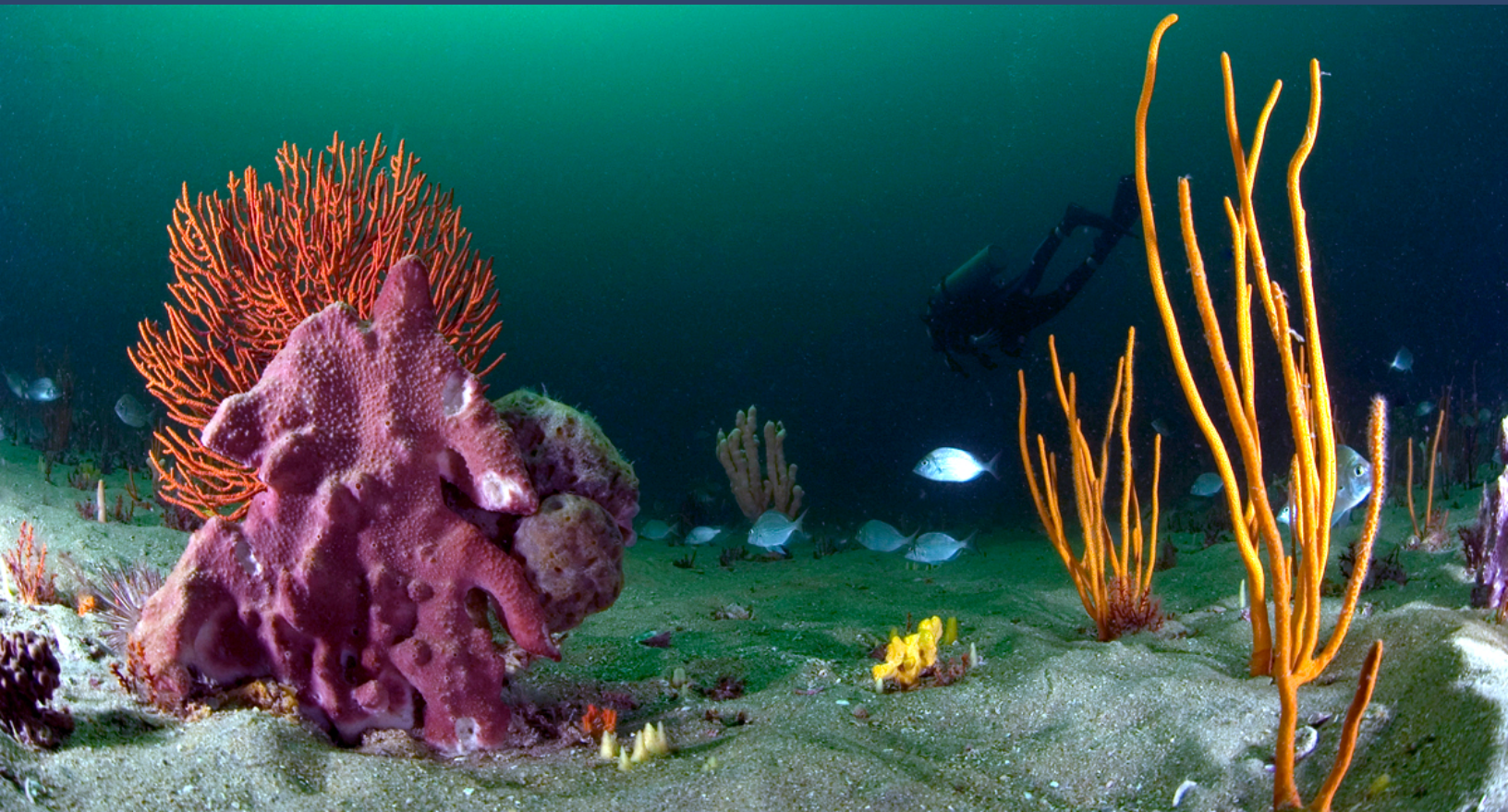


Long-Term Monitoring of Ecological Conditions In Gray's Reef National Marine Sanctuary: Soft-Bottom Benthic Assemblages and Contaminant Levels in Sediments and Biota (2000, 2005, and 2012/13)



NOAA Technical Memorandum NOS NCCOS 206

Long-Term Monitoring of Ecological Conditions In Gray's Reef National Marine Sanctuary: Soft-Bottom Benthic Assemblages and Contaminant Levels in Sediments and Biota (2000, 2005, and 2012/13)

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November 2015

NOAA Technical Memorandum NOS NCCOS 206



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Commerce

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The appropriate citation for this report is:

Balthis, W. L., J. L. Hyland, C. Cooksey, M. H. Fulton, E. F. Wirth. 2015. Long-Term Monitoring of Ecological Conditions In Gray's Reef National Marine Sanctuary: Soft-Bottom Benthic Assemblages and Contaminant Levels in Sediments and Biota (2000, 2005, and 2012/13). NOAA Technical Memorandum NOS NCCOS 206, NOAA National Ocean Service, Charleston, SC 29412-9110. 35 pp. doi:10.7289/V5/TM-NOS-NCCOS-206

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Executive Summary

In July 2012 and June 2013, NOAA's National Centers for Coastal Ocean Science (NCCOS) and Gray's Reef National Marine Sanctuary (GRNMS) conducted a study to assess the status of ecological condition and stressor impacts in GRNMS, with a primary focus on the soft-bottom benthos and sediment quality. The study is a continuation of an ongoing partnership, initiated in 2000, between NCCOS and GRNMS to provide periodic assessments of the status of condition as a quantitative basis for tracking potential changes through time, using common indicators and sampling protocols.

As in previous studies completed in 2000 and 2005, the 2012/13 study used a probabilistic design consisting of 20 sampling sites selected randomly from each of the 20 cells of a rectangular grid bounded by the study region (GRNMS). Eleven of the 20 sites were sampled in July 2012. Mechanical issues with the research vessel precluded sampling at the remaining nine sites during the 2012 effort, and sampling was completed in June 2013.

Water depths at the stations sampled in 2012/13 ranged from 14.1 – 20.8 m; near-bottom salinities occupied a narrow range of 34.7 – 36.1 psu; and near-bottom dissolved oxygen (DO) levels were consistently high (6.1 – 6.8 mg L⁻¹), well above a benthic hypoxic effect threshold of about 2 mg L⁻¹. Sediments were composed mainly of sands (84 – 97% sand) and sediment total organic carbon (TOC) content averaged 0.17% and ranged from 0 – 1%, which is well below the range (> 5% TOC) reported to be associated with potential risks to benthic fauna from organic over-enrichment.

Concentrations of chemical contaminants in sediments were at low background levels or below the limit of detection at all sites sampled. There were no exceedances of published sediment quality guidelines (SQG) for the higher ERM or lower ERL bioeffect levels for any individual contaminant at any of the 20 sites. Similarly, mean ERM-Q values at all sites fell within the range of concentrations associated with a low incidence of benthic impacts in southeastern estuaries. No significant sediment toxicity (as measured by the Microtox[®] solid-phase assay) was observed in any of the samples analyzed from the 20 sites.

None of the collected specimens of black sea bass (*Centropristis striata*) or turkey wing arks (*Arca zebra*) had contaminant levels in excess of FDA guideline values for fish and/or molluscan shellfish. However, in one black sea bass (out of 10 specimens), concentrations of inorganic arsenic (estimated as 2% of total arsenic) fell within the range of concentrations for which EPA recommends limiting fish consumption to four meals per month. Concentrations of cadmium in six (of 10) analyzed samples of arks fell within EPA's four-meal-per-month endpoints; the corresponding upper endpoint for cadmium was exceeded in two (of 10) ark samples.

A total of 421 benthic taxa (319 identified to species level) were enumerated from 60 replicate 0.04m² grab samples (3 per site) collected at the 20 GRNMS sites. Total number of taxa (3 grabs combined), mean number of taxa per grab, H diversity, and density (# m⁻²) averaged 105, 60.1, 4.8, and 7,190 m⁻² respectively. Dominant phyla included annelids, arthropods, and molluscs. Collectively, annelids were by far the most abundant taxonomic group, representing 56% of total density. The most abundant individual taxon was the lancelet, *Branchiostoma caribaeum* (a cephalochordate) – this single species made up 9.1% of total density of all taxa collected at the 20 sites, and is an important prey item of fish species at GRNMS.

Among the 2000, 2005, and 2012/13 surveys, ranges of key abiotic environmental factors (near-bottom DO, TOC, sediment contaminants) were similar. In all years, DO and TOC concentrations were at normal levels outside of reported bioeffect ranges. Levels of chemical contaminants in sediments measured in the three surveys were well below sediment quality guideline values, although one site sampled in 2000 had a Cu concentration exceeding the lower-threshold ERL guideline value of $34 \mu\text{g g}^{-1}$ but below the higher ERM value of $270 \mu\text{g g}^{-1}$.

No chemical contaminant concentrations in tissues of fish or shellfish were found to exceed FDA human-health guidelines in any of the three surveys. However, as noted above for the 2012/13 survey, concentrations of inorganic arsenic (estimated as 2% of total arsenic) in specimens of black sea bass collected in 2000 fell within the range of concentrations for which EPA recommends limiting fish consumption to four meals per month. In all three surveys, concentrations of cadmium fell within or exceeded the four-meals-per-month guideline levels in ark samples.

Percent composition of benthic assemblages (major phyla) was similar across the three sampling periods, with episodic or seasonal blooms of taxa (*Ervilia* sp. A and *Branchiostoma caribaeum* respectively) noted in 2000 and 2012/13. Abundance and diversity of taxa in the three surveys also were similar, but the higher level of taxonomic resolution achieved in identification of benthic taxa in 2012/13 resulted in higher calculated metrics of species richness.

Surveys of benthic infaunal assemblages in Gray's Reef to date, including re-sampling of six sites each in 2001 and 2002, have resulted in the identification of a total of 790 taxa, with the largest number of individual taxa identified in 2012/13 samples, as noted above. Species accumulation curves generated from the combined data suggest an estimated maximum of nearly 900 benthic taxa present in the sanctuary. This estimate is substantially higher than an earlier estimate of 800 taxa, and is likely due to the higher degree of taxonomic resolution achieved in taxonomic identifications in 2012/13. Additional sampling of infauna at GRNMS is needed to test and validate the most recent estimate.

1.0 Introduction

Since April 2000, NOAA's National Centers for Coastal Ocean Science (NCCOS) and the Office of National Marine Sanctuaries (ONMS) have been engaged in a collaborative partnership to augment the management of National Marine Sanctuaries (NMS) through increased scientific understanding of sanctuary sites (ONMS/NCCOS 2004). NCCOS scientists have been working with Gray's Reef NMS (GRNMS) to assess the status of ecological condition and stressor impacts throughout the sanctuary and surrounding shelf areas, with a major focus on the soft-bottom benthos and sediment quality. Such activities are important to fulfill key research and monitoring goals for GRNMS in supporting scientific research and long-term monitoring to enhance the understanding of the Sanctuary environment and to improve management decision-making (NOAA 2014).

GRNMS is a marine protected area located 32 km offshore of Sapelo Island, Georgia (Figure 1), encompassing 57.2 km² of "live bottom" habitat and associated sandy substrate. Named in recognition of Milton B. Gray, a taxonomist and curator who studied the area in the 1960s to obtain collections of reef-associated fauna for the University of Georgia Marine Institute (Hunt 1974), Gray's Reef was designated a National Marine Sanctuary in January, 1981. It is one of the largest near-shore rocky reefs off the southeastern United States, and lies in a transition zone between temperate and tropical waters. Located at the boundary between the inner and mid-continental shelf portion of the South Atlantic Bight (SAB), which extends from Cape Hatteras, North Carolina to Cape Canaveral, Florida, Gray's Reef is influenced by freshwater inflow, winds, and tides, with some influence from the Gulf Stream, which flows along the shelf edge (NOAA 2014). Water depths throughout the sanctuary average approximately 20 m. The bottom consists of extensive but discontinuous rock outcroppings of moderate height (2-3 m) interspersed with unconsolidated sediments of sand and shell hash (ONMS/NCCOS 2004).

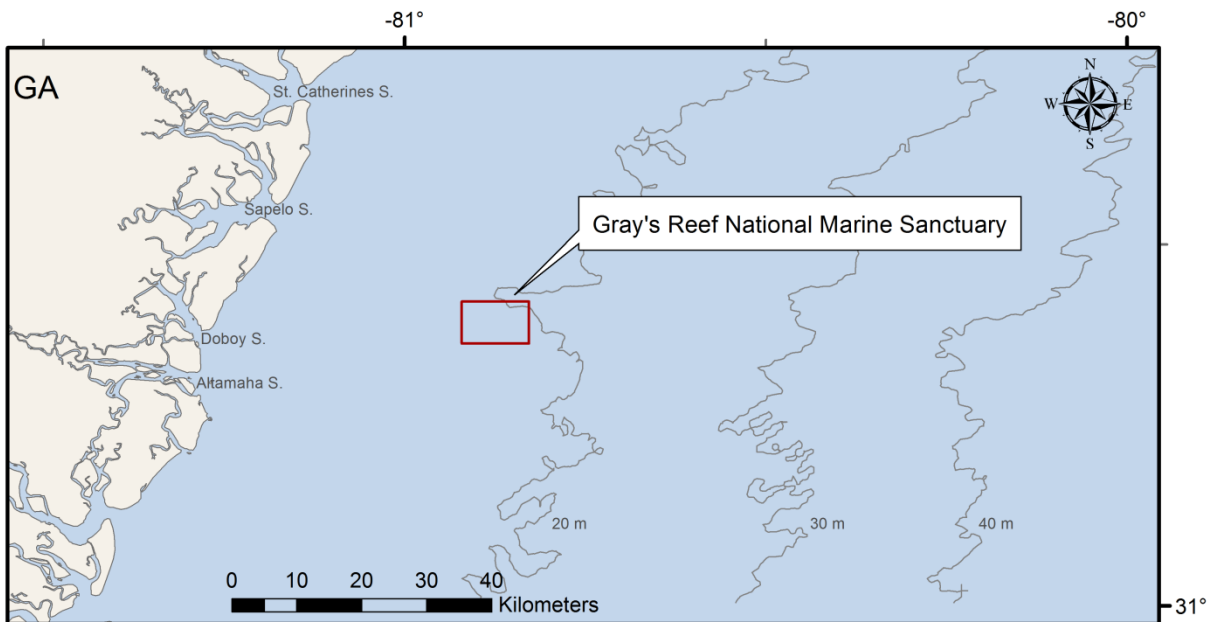


Figure 1. Location of Gray's Reef National Marine Sanctuary off the coast of Georgia, USA.

An initial characterization of GRNMS was conducted in spring 2000 to evaluate the condition and distribution of benthic macroinfauna, including sediment-associated stressors, to provide a quantitative benchmark for tracking any future changes due to natural or human disturbances. The results of the spring 2000 survey suggested that soft-bottom habitats associated with GRNMS support highly diverse infaunal assemblages, thereby challenging a commonly-held assumption that these relatively featureless expanses of substrate surrounding “live bottom” habitat are biological voids (Cooksey et al 2004, Hyland et al. 2006). Levels of man-made, chemical contaminants were found at low, but detectable, concentrations in sediments and edible tissues of target species (black sea bass, turkey wing arks), suggesting background conditions while highlighting the need for future monitoring to track potential changes in levels of these substances.

A follow-up survey in spring 2005 (Balthis et al. 2007) found similarly low levels of contaminants in sediments and fish and shellfish tissues, with measured concentrations being well below corresponding sediment quality and human health guidelines. Similar patterns in species richness and diversity of soft-bottom benthic assemblages also were observed in 2000 and 2005.

This report presents the results of a follow-up survey, conducted in summer 2012/spring 2013, which provides additional data points for the long-term monitoring record as a basis for tracking status and trends in sanctuary conditions and supports important management products such as the GRNMS management plan (NOAA 2014) and GRNMS Condition Reports (ONMS 2008).

2.0 Methods

Sampling sites (Figure 2) were selected using the spatially-stratified design developed and applied in previous studies in 2000 (Cooksey et al. 2004) and 2005 (Balthis et al. 2007). The sampling grid was created from a rectangular tessellation of the study region (GRNMS), yielding 20 grid cells, each covering 2.9 km². Sites were generated by selecting one random point (latitude/longitude) from each grid cell (tessellation-stratified sampling, one point per stratum; Stevens 1997, Barabesi et al 2012).

At each site, surface-to-bottom profiles of physical properties of water (depth, temperature, salinity, conductivity, pH, and dissolved oxygen) were taken using a Sea-Bird SBE 19 SEACAT CTD profiler (Sea-Bird Electronics, Inc. 2006).

Three replicate sediment samples for macroinfaunal analysis were collected at each site using a 0.04 m² Young grab sampler. Each replicate was sieved in the field through a 0.5-mm mesh screen and preserved in 10% buffered formalin. Sediment samples were shipped to the laboratory, where they were rinsed through a 0.5-mm mesh sieve to remove preservatives and sediment, stained with Rose Bengal, and stored in 70%-isopropanol until processing. Infauna were sorted from sample debris under a Wild M-5A dissecting microscope and identified to the lowest practical identification level, usually species.

The upper 2-3 cm of sediment from additional multiple grabs taken at each station were combined into a single composite sample, then sub-sampled for analysis of metals, organics (PAHs, pesticides, PCBs), total organic carbon (TOC), and grain size. Samples for grain size analysis were prepared by sieve separation followed by timed pipette extractions as described in Plumb (1981). TOC analysis followed USEPA Method 9060. A minimum of 5g (wet weight) of sediment was initially dried for 48 h. Weighed subsamples were ground to fine consistency and acidified to remove sources of inorganic carbon (e.g.,

shell fragments). The acidified samples were ignited at 950°C and the carbon dioxide evolved was measured with an infrared gas analyzer.

Black sea bass (*Centropristis striata*) and turkey wing arks (*Arca zebra*) were collected by hook-and-line fishing and scuba diving, respectively, at targeted stations for analysis of chemical contaminants in tissues. Each specimen was wrapped individually in heavy aluminum foil, placed in two nested plastic bags, sealed, and frozen at -20° C until transferred on ice to the laboratory for analysis. Arks were collected by divers, rinsed with ambient seawater, wrapped in heavy aluminum foil, placed in double plastic bags, and frozen (-20° C) until analysis. Analyses of chemical contaminants were performed on tissue homogenate prepared from individual skin-on fillets of black sea bass and composited arks (aggregates consisting of 3 – 15 individuals per station).

Methods for analysis of chemical contaminants followed those of Sanders (1995), Fortner et al. (1996), Kucklick et al. (1997), and Clum et al. (2002). Although matrix-specific extraction methods were required for some chemicals (e.g., all metals except Hg), subsequent instrumental analyses were the same for both sediments and tissues. Trace metal analyses were performed using inductively coupled plasma mass spectrometry (ICP/MS) for the following suite of metals: Al, Cr, Cu, Fe, Mn, Ni, Sn, and Zn. Additional trace metals (Ag, As, Cd, Pb, Se) were analyzed using graphite furnace atomic absorption (GFAA). Cold vapor atomic absorption (CVAA) was used for analysis of total Hg. Two classes of organic compounds (PCBs and pesticides) were analyzed by dual-column gas chromatography with electron capture detection (GC-ECD). An ion-trap mass spectrometer equipped with a gas chromatograph (GC/MS-IT) was used for analysis of PAHs.

The biological significance of observed chemical contaminant concentrations in sediments was evaluated in relation to Effects Range-Low (ERL) and Effects Range-Median (ERM) sediment quality guidelines (SQG) values from Long et al. (1995), where available ([Appendix A](#)). ERL values are lower-threshold bioeffect limits, below which adverse effects on sediment-dwelling organisms are not likely to occur. ERM values, in contrast, represent mid-range concentrations of chemicals above which adverse effects are likely to occur. Overall sediment contamination from multiple chemicals was expressed using the mean ERM quotient (mean ERM-Q), which is calculated as the mean of the ratios of individual chemical concentrations in a sample relative to corresponding published ERM values (Long et al. 1998, 2000; Long and MacDonald 1998; Hyland et al. 1999). Sediment toxicity was assessed using the Microtox[®] solid-phase assay (Microbics 1992).

Levels of chemical contaminants in tissues of black sea bass (*Centropristis striata*) and arks (*Arca zebra*) were assessed in comparison to human health guidelines, where available. Some guidelines, such as action levels, tolerances, or levels of concern ([Appendix B](#)) set by the US FDA (FDA 1984, 1993a-e, 1994, 2007), establish limits that are not to be exceeded in food (e.g., fish and shellfish) shipped or marketed in interstate commerce. Others, such as those developed by the US EPA, represent risk-based guidelines for use in issuing fish consumption advisories for non-commercially caught fish and shellfish. Whereas the FDA guidelines are intended to be protective of general consumers of seafood products, EPA health endpoints ([Appendix C](#)) target recreational and subsistence fishers who typically consume more fish than the general population and often harvest the fish they consume from the same local waterbodies over many years (USEPA 2000).

Population and community-level indices were used to characterize the benthic infaunal assemblages inhabiting the soft-bottom sediments of Gray's Reef. These included numbers of species, H' diversity (Shannon and Weaver 1949) derived with base-2 logarithms, density (m⁻²) of total fauna (all species combined), and density of numerically dominant fauna. Ordination methods (non-metric multidimensional scaling, NMDS) were used to assess similarity of benthic assemblages within and among the three survey periods (2000, 2005, 2012/13).

Species accumulation curves (SACs) were generated using R statistical software (R Core Team 2015) and the *vegan* community ecology extension to R (Oksanen et al 2015). Additional data collected in 2001 and 2002 at a subset of six stations within the sanctuary (see Hyland et al. 2006, Cooksey et al. 2004) were included in this analysis. SACs were used to illustrate the cumulative number of taxa encountered with increasing sample size, beginning in spring 2000 and continuing with each successive sampling event in 2001, 2002, 2005, and 2012. Because the order in which samples are added to the total affects the shape of the curve (Colwell and Coddington 1994), SACs were generated as the mean of random permutations of the combined data (samples added in random order) or subsampling without replacement (Gotelli and Colwell 2001). A two-parameter, asymptotic model (Clench 1979) was fit to the resulting species accumulation values, providing an estimated maximum number of taxa. The purpose of this particular exercise was to help further our understanding and predictions of marine biodiversity within the sanctuary.

3.0 Results

3.1 Summer 2012 / Spring 2013 Survey

Sampling was successful at 11 of 20 sites within Gray's Reef from July 26 - 28, 2012. Due to mechanical problems with the research vessel, the remaining nine sites could not be sampled during the summer 2012 effort, and were completed the following spring (June 2 - 3, 2013). Black sea bass were collected by hook-and-line fishing at five of the 20 sites; arks were collected by diver at 10 of the 20 sites (Figure 2). Thirteen of the 20 sites were represented by at least one of these two species.

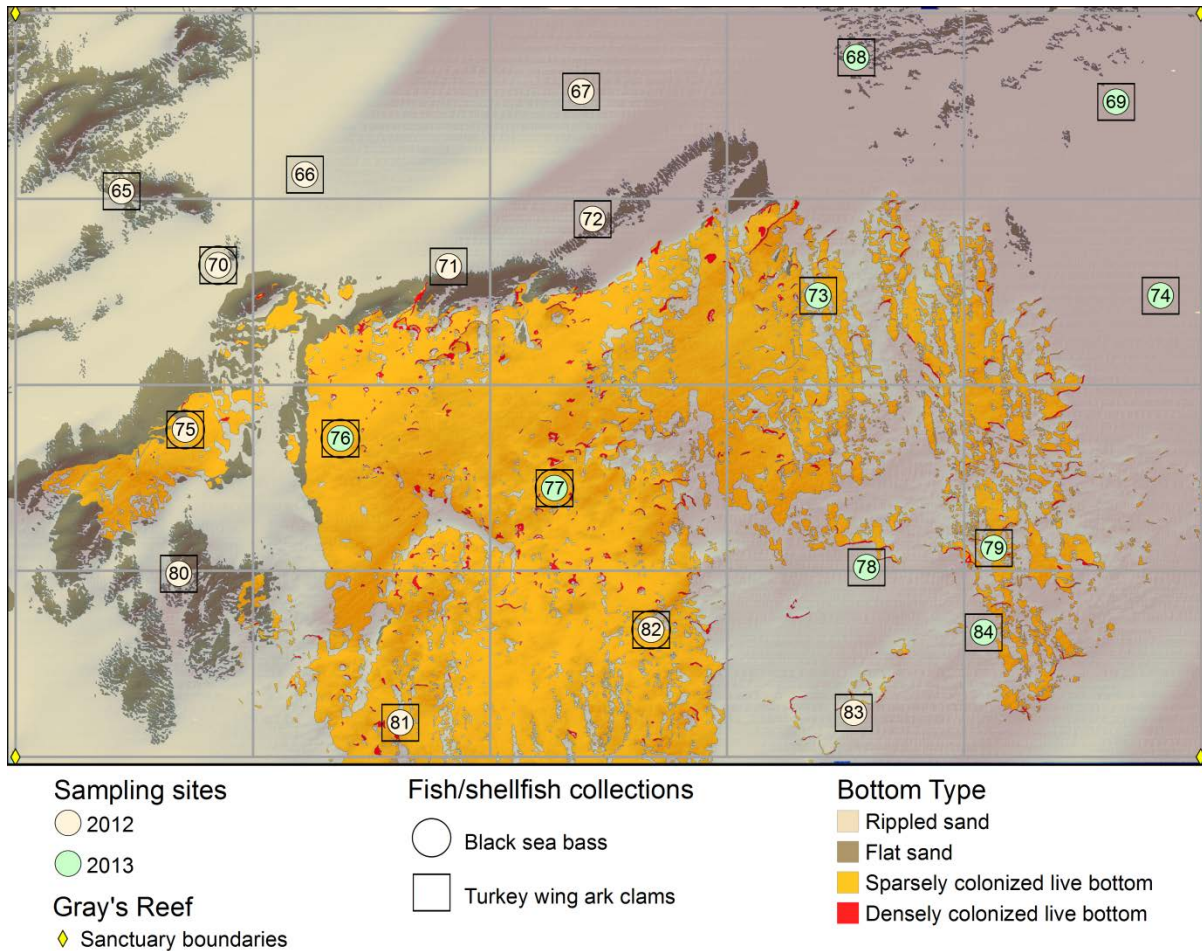


Figure 2. Map showing the location of 20 sampling stations in Gray's Reef National Marine Sanctuary (inset of Figure 1). Fish and shellfish collection sites are indicated by open circles (black sea bass) and open boxes (turkey wing arks). Bottom type classifications are based on those described in Kendall et al. 2005.

Measured depths averaged 18.0 m and ranged from 14.1 – 20.8 m at the 20 stations sampled throughout the sanctuary in 2012 and 2013. Near-bottom salinities were typical of an offshore environment (34.7 – 36.1 psu) and dissolved oxygen (DO) levels were within a narrow range of 6.1 – 6.8 mg L⁻¹ (Figure 3), well above a benthic hypoxic effect threshold of about 2 mg L⁻¹ (Diaz and Rosenberg 1995). Sediments were composed mostly of sand (84 – 97 %), with a correspondingly low proportion of fine-grained sediments. Sediment total organic carbon content (TOC) averaged 0.17% and ranged from 0 – 1%, which is well below a reported range (> 5%) used in prior EPA National Coastal Condition Reports (e.g., EPA 2012) to flag potential risks to benthic fauna from organic over-enrichment. TOC concentrations were also well below the slightly more conservative cutpoint of > 3.5% identified by Hyland et al. (2005) as an upper range associated with a high risk of degraded benthic condition based on data from multiple coastal areas around the world. A summary of the values of each of these abiotic environmental variables by station is provided in Appendix D.

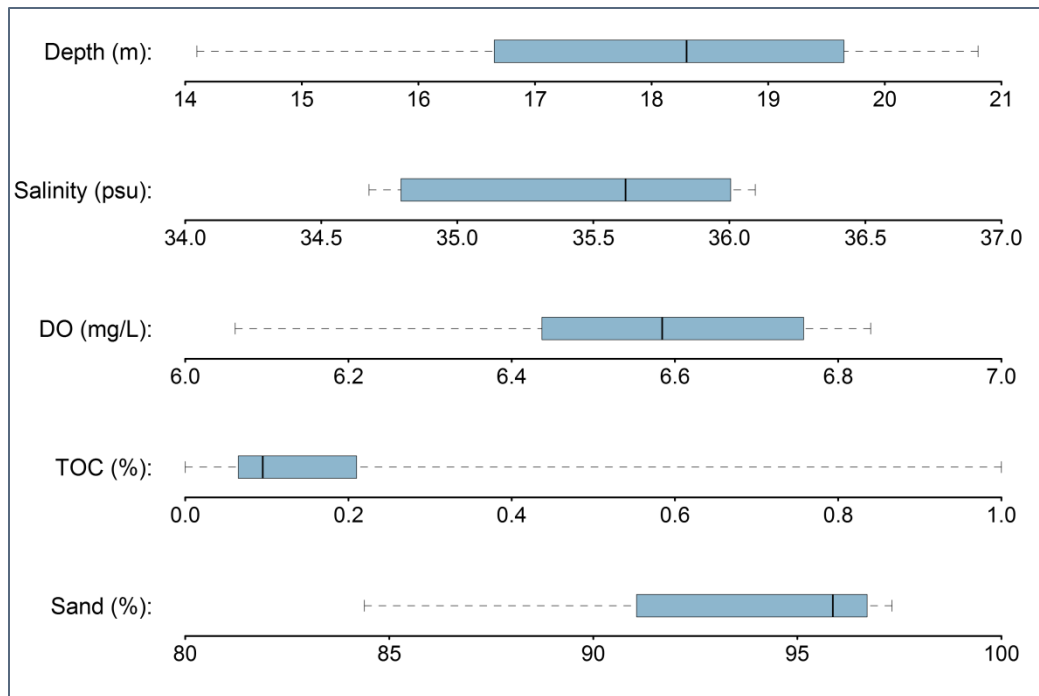


Figure 3. Medians, quartiles, and ranges (min, max) for oceanographic and sediment characteristics measured at GRNMS.

Sediment contaminants

Concentrations of chemical contaminants in sediments were at low background levels or below the limit of detection at all sites sampled (Appendix E). There were no exceedances of published sediment quality guidelines (SQG) for the higher ERM or lower ERL bioeffect levels for any individual contaminant at any of the sites (Figure 4). The low number (zero) of SQG exceedances at GRNMS sites is similar to that of stations surveyed in the surrounding South Atlantic Bight (SAB) in 2004 (Cooksey et al. 2010), and lower in comparison to sites in southeastern estuaries, where some locations (representing 4% of the survey area) had sediment contaminant concentrations in excess of the lower ERL or upper ERM guideline values (Figure 4). Comparisons based on calculated mean ERM-Q values also suggest low levels of sediment-associated contaminants at Gray's Reef, with values at all sites falling within the range of concentrations associated with a low incidence of benthic impacts in southeastern estuaries (Hyland et al. 2003).

Sediment Toxicity

No sediment samples from any of the 20 stations sampled in this study were found to be toxic, based on criteria for the Microtox[®] assay (Silt-clay < 20 %: Toxic if EC50 < 0.5 %; Ringwood et al. 1997).

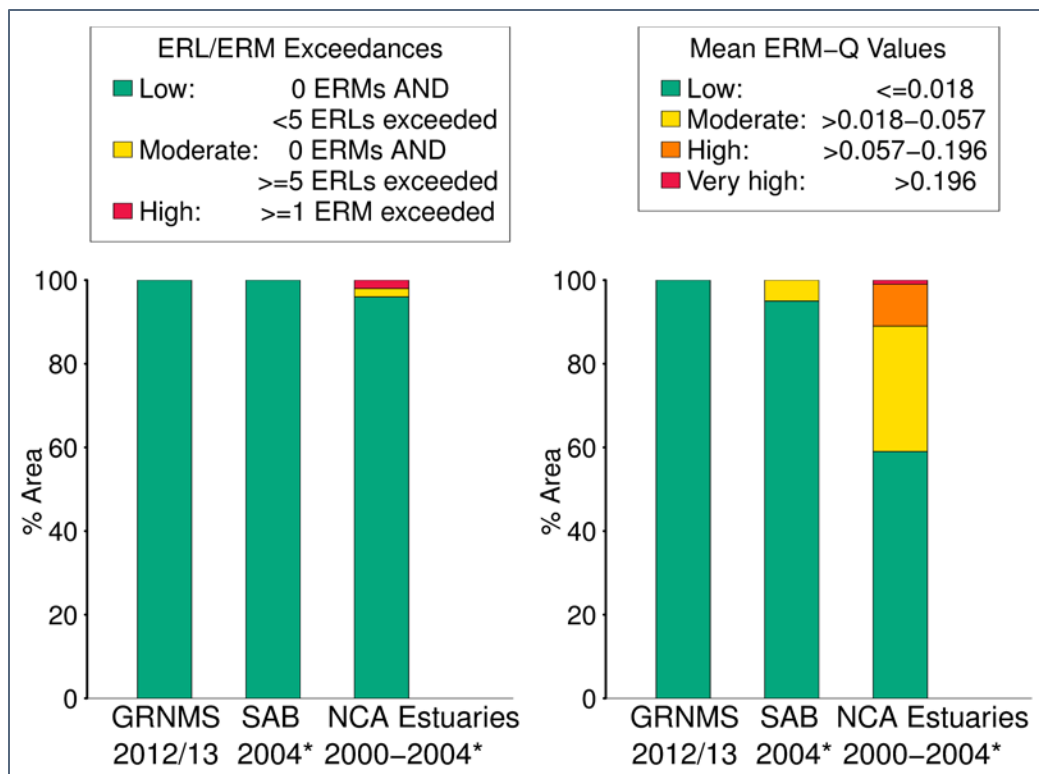


Figure 4. Proportion of survey region (% area) classified according to ranges of sediment quality guidelines. Left graph: ERM and ERL values used to determine number of exceedances are from Long et al. (1995) and cutpoints for defining range categories are based on methods used in previous EPA National Coastal Condition Reports (e.g., EPA 2012) and southeastern ecological assessments (e.g., Cooksey et al. 2010). Right graph: Mean ERM-Q cutpoints are derived from Hyland et al. (2003; also see Cooksey et al. 2010 for similar applications). (*Data from Cooksey et al. 2010.)

Tissue Chemistry

Samples of tissue from black sea bass (individual skin-on fillets) and arks (composite samples of multiple individual specimens) were collected successfully at stations indicated in Figure 2 and listed in Table 1. Concentrations of most metals, PAHs, pesticides, and PCBs were at low background levels or below the limit of detection in tissues. None of the specimens of black sea bass or arks had contaminant levels in excess of FDA guideline values (see Appendix B) for fish and/or molluscan shellfish. However, concentrations of inorganic arsenic (estimated as 2% of total arsenic) measured in black sea bass (Appendix F) fell within the range of concentrations (see Appendix C) for which EPA recommends limiting fish consumption to four meals per month. EPA health endpoints also were exceeded for cadmium in samples of arks (Appendix G). It is uncertain whether these trace metals (arsenic and cadmium) are of natural or anthropogenic origin; however, it is known that fish accumulate arsenic in their tissues, and the degree of bioaccumulation depends on trophic level (Kirby and Maher 2002). Similarly, molluscan shellfish bioaccumulate contaminants in their tissues and are known to have a high affinity for cadmium (Frazier 1979). Although both natural and anthropogenic sources could contribute to elevated levels, Valette-Silver et al. (1999) found that most of the high concentrations of arsenic measured in bivalves and sediments in the southeastern U.S. coincide spatially with natural phosphate deposits.

Table 1. Locations of fish and shellfish collections for the GRNMS 2012/13 survey.

Station	Common Name	Scientific Name	No. of specimens
70	Black sea bass	<i>Centropristis striata</i>	2
72	Turkey wing ark	<i>Arca zebra</i>	10
73	Turkey wing ark	<i>Arca zebra</i>	15
74	Turkey wing ark	<i>Arca zebra</i>	4
75	Black sea bass	<i>Centropristis striata</i>	2
76	Black sea bass	<i>Centropristis striata</i>	2
	Turkey wing ark	<i>Arca zebra</i>	7
77	Black sea bass	<i>Centropristis striata</i>	2
	Turkey wing ark	<i>Arca zebra</i>	11
78	Turkey wing ark	<i>Arca zebra</i>	4
79	Turkey wing ark	<i>Arca zebra</i>	11
81	Turkey wing ark	<i>Arca zebra</i>	14
82	Black sea bass	<i>Centropristis striata</i>	2
83	Turkey wing ark	<i>Arca zebra</i>	3
84	Turkey wing ark	<i>Arca zebra</i>	12

Benthos

A total of 421 taxa (319 identified to species level) were enumerated from 60 replicate 0.04m² grab samples (3 per site) collected at the 20 GRNMS sites. Diverse and abundant macroinfaunal assemblages were present in these samples. Total number of taxa (3 grabs combined), mean number of taxa per grab, H diversity, and density (# m⁻²) averaged 105, 60.1, 4.8, and 7,190 m⁻² respectively and ranged from 65 – 138, 33.3 – 79.7, 4.0 – 5.4, and 1,525 – 12,167 m⁻² respectively across the 20 stations (Table 2). Measures of mean species richness and diversity at Gray’s Reef were comparable to, and somewhat higher than, those reported for the larger surrounding SAB (38 and 4.2, respectively; Cooksey et al. 2010). High values of H’ reflect both the high numbers of taxa present in soft-bottom sediments at Gray’s Reef and the evenness of the assemblages (i.e., low dominance of any given species, with abundance of taxa relatively evenly distributed among species present).

Table 2. Summary measures of taxonomic richness, diversity, and abundance for 20 stations sampled in the GRNMS 2012/13 survey.

	Overall	Overall	Percentiles		
	Mean	Range	10 th	50 th	90 th
Total Richness ^a (3 grabs)	105	65–138	74.1	107.5	125
Mean Richness ^b (per grab)	60.1	33.3–79.7	43.4	62.8	70.9
Mean Density (# m ⁻²)	7,190	1,525–12,167	4,101	6,696	10,871
Mean H’ (per grab)	4.8	4–5.4	4.2	4.9	5.4

^a Pooled # of taxa in three replicate grab samples combined.

^b Number of taxa averaged over three replicate grabs.

Relative proportions of dominant phyla (% of total # of taxa, % of total density) are shown in Figure 5. Members of phylum Annelida were the most numerous taxa, both by numbers of taxa (45% of total # of taxa) and density (56% of total density). Arthropod crustaceans and molluscs were the next most frequently encountered taxonomic groups (as % of total taxa). Phylum Mollusca consisted of roughly equal numbers of gastropod and bivalve taxa, though bivalves were slightly more abundant (58% of molluscan abundance). Together, annelids, arthropods, and molluscs accounted for ~93% of total # of taxa and ~75% of total density. Although only two chordate taxa were collected - the lancelet, *Branchiostoma caribaeum*, and tunicates (Class Ascidiacea) – *B. caribaeum* was highly abundant at several sites, making up 9.1% of total density of all taxa. Lancelets are an important prey item of fish species (e.g., tomtate, *Haemulon aurolineatum*) common to Gray’s Reef. In a study of food and feeding of tomtate in the South Atlantic Bight (SAB), Sedberry (1985) found that *B. caribaeum* made up the largest volume (41.6%) of prey in the diet of *H. aurolineatum*. ‘Other’ taxa in Figure 5 consisted mainly of two phyla, Sipuncula and Nemertea, of which the latter made up 3.2% of total density of all taxa.

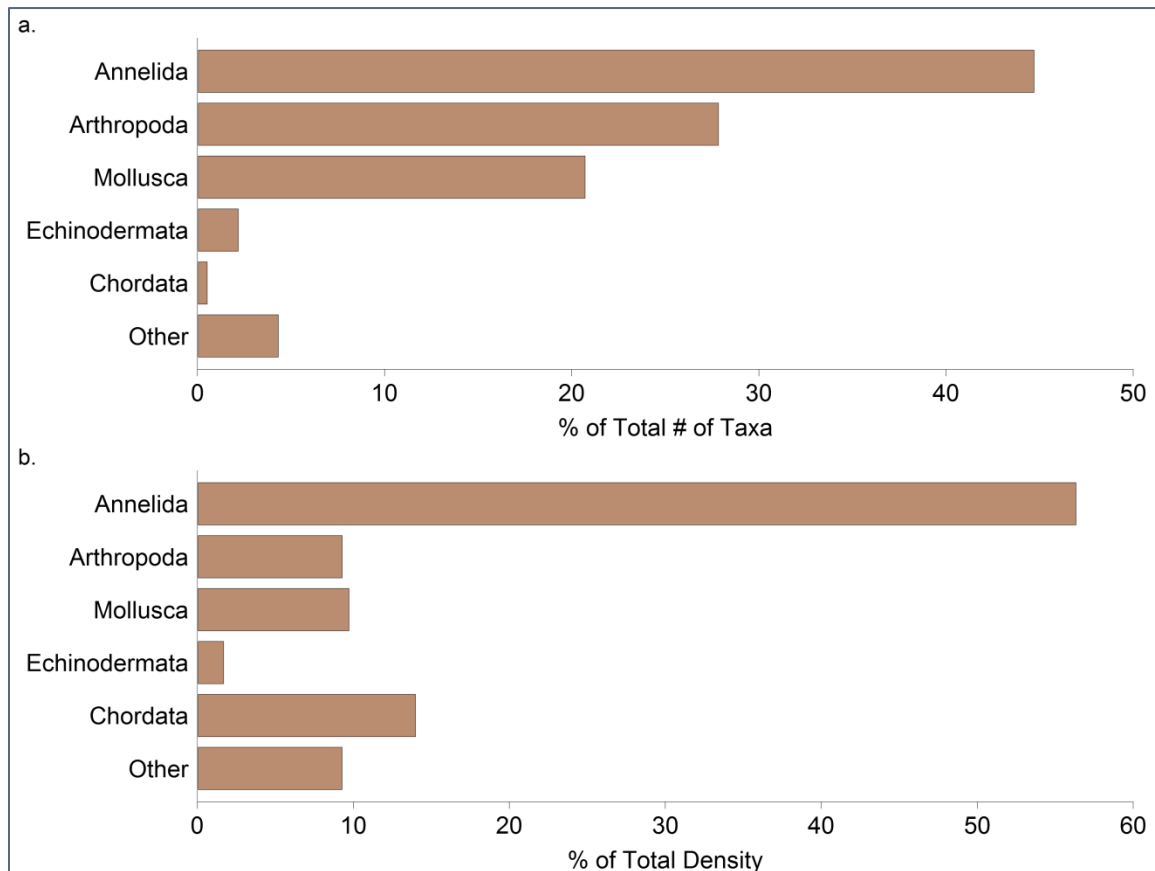


Figure 5. Relative proportions of dominant phyla by a) percent of total number of taxa and b) percent of total density for taxa identified in the GRNMS 2012/13 survey.

Table 3 lists the upper 10 dominant (most abundant) taxa collected at Gray’s Reef sites during the 2012/13 survey. *Branchiostoma caribaeum* was the most abundant species, making up 9.1% of total density of all taxa and occurring at all of the 20 stations sampled (53 of 60 grabs). Oligochaeta represented 6.4% of total density, followed by tunicates (Class Ascidiacea) at 4.8% of total density. Six of the 7 remaining taxa in the list of 10 dominants were polychaetes (19.4% of total density combined). Phylum Nemertea was the 8th most abundant taxon at 3.2% of total density. Collectively, these 10 dominant taxa represented only 44% of total density, which demonstrates the high level of species diversity at Gray’s Reef.

Table 3. Ten dominant (most abundant) taxa identified in the GRNMS 2012/13 survey.

Taxon	Group	Average Density (m ⁻²)	% of Total Density	Cumulative % Density	% of Station Occurrence
<i>Branchiostoma caribaeum</i>	Other	654	9.1	9.1	100
Oligochaeta	Oligochaeta	462	6.4	15.5	100
Ascidiacea (LPIL)	Other	348	4.8	20.4	90
<i>Pomatoceros americanus</i>	Polychaeta	310	4.3	24.7	85
<i>Protodorvillea kefersteini</i>	Polychaeta	284	4.0	28.6	95
<i>Sphaerosyllis piriferopsis</i>	Polychaeta	269	3.7	32.4	90
<i>Spio pettiboneae</i>	Polychaeta	249	3.5	35.8	85
Nemertea	Other	233	3.2	39.1	100
<i>Pionosyllis gesae</i>	Polychaeta	185	2.6	41.6	90
<i>Goniadides carolinae</i>	Polychaeta	173	2.4	44.0	85

3.2 Among-Year Comparisons

Data from the 2012/13 survey were compared to earlier data from 2000 and 2005 to look for trends in the measured variables and any evidence of changes in the quality of sanctuary condition including potential signals of anthropogenic stress. Ranges of key abiotic environmental factors (sediment contaminants, TOC, near-bottom DO) were similar among the various surveys (Table 4). In all years, DO and TOC concentrations were at normal levels outside of reported bioeffect ranges. Also, as in the two previous surveys, levels of chemical contaminants in sediments were well below sediment quality guideline values. One site sampled in 2000 had a Cu concentration (103 µg g⁻¹) which exceeded the lower-threshold ERL guideline value of 34 µg g⁻¹ but was below the higher ERM value of 270 µg g⁻¹ (Cooksey et al. 2004). As a result, this same site had a mean ERM-Q value of 0.024, which falls within the range (mean ERM-Q of 0.018 – 0.057) found to be associated with a ‘moderate’ risk of impacts to benthic fauna (Hyland et al. 2003). Hence, only one of 27 contaminants having corresponding sediment quality guidelines was found at a concentration exceeding the lower ERL guideline value, and this was observed at only one of 60 sites sampled over the three survey periods.

Similar to sediments, no chemical contaminant concentrations in edible tissues of fish and shellfish were found to exceed FDA human-health guidelines (FDA 1984, 1993a-e, 1994, 2007) used in prior reports on ecological condition of GRNMS (Cooksey et al. 2004, Balthis et al. 2007). However, when compared to

guidelines targeting recreational and subsistence fishers (EPA 2000), concentrations of inorganic arsenic (estimated as 2% of total arsenic) in specimens of black sea bass (*C. striatus*) collected in the 2000 and 2012/13 surveys fell within the range of concentrations (see [Appendix C](#)) for which EPA recommends limiting fish consumption to four meals per month. In all three surveys (2000, 2005, 2012/13), concentrations of cadmium fell within or exceeded the four-meals-per-month guideline levels in samples of arks (see [Appendix G](#) for 2012/13). As noted previously, molluscan shellfish are known to bioaccumulate contaminants in their tissues and have a high affinity for cadmium (Frazier 1979).

Table 4. Among-year comparisons of key abiotic environmental variables. Cutpoints for defining range categories are based on methods used in previous EPA National Coastal Condition Reports (e.g., EPA 2012) and southeastern ecological assessments (e.g., Cooksey et al. 2010).

	2000	2005	2012/13
Sediment Contamination (% area):			
• None to Low: No ERMs and < 5 ERLs exceeded	100%	100%	100%
• Moderate: ≥ 5 ERLs exceeded	0%	0%	0%
• High: ≥ 1 ERM exceeded	0%	0%	0%
Sediment TOC:			
• Mean (% dry wt.)	0.27	1.0	0.17
• Range (% dry wt.)	0.08 – 1.97	0 – 1.9	0 – 1.0
• % area with TOC < 2% (low)	100%	100%	100%
• % area with TOC 2 – 5% (moderate)	0%	0%	0%
• % area with TOC > 5% (high)	0%	0%	0%
Near-Bottom DO levels:			
• Mean (mg L ⁻¹)	8.0	7.6	6.5
• Range (mg L ⁻¹)	7.6 – 8.2	7.5 – 7.6	6.1 – 6.8
• % area with DO > 5 mg L ⁻¹ (high)	100%	100%	100%
• % area with DO 2 – 5 mg L ⁻¹ (moderate)	0%	0%	0%
• % area with DO < 2 mg L ⁻¹ (low)	0%	0%	0%

Percent composition of benthic assemblages (major phyla) across the three sampling periods is displayed in [Figure 6](#). Similar numbers of taxa were found in each survey, but different patterns of percent density are apparent from the figure. A strong seasonal bloom of the bivalve mollusc *Ervilia* sp. A was observed in 2000, with this single species accounting for 56% of the density of all taxa. A similar, but less pronounced, pulse of *Branchiostoma caribaeum* ('Other' bar, [Figure 6b](#)) was noted in 2012. In a previous study in shelf waters off the Georgia coast, Frankenberg (1971) also documented a summer pulse in density of *B. caribaeum*. Both *Ervilia* and *B. caribaeum* are important prey items of fish species (e.g. tomtrate, Sedberry 1985) common to Gray's Reef.

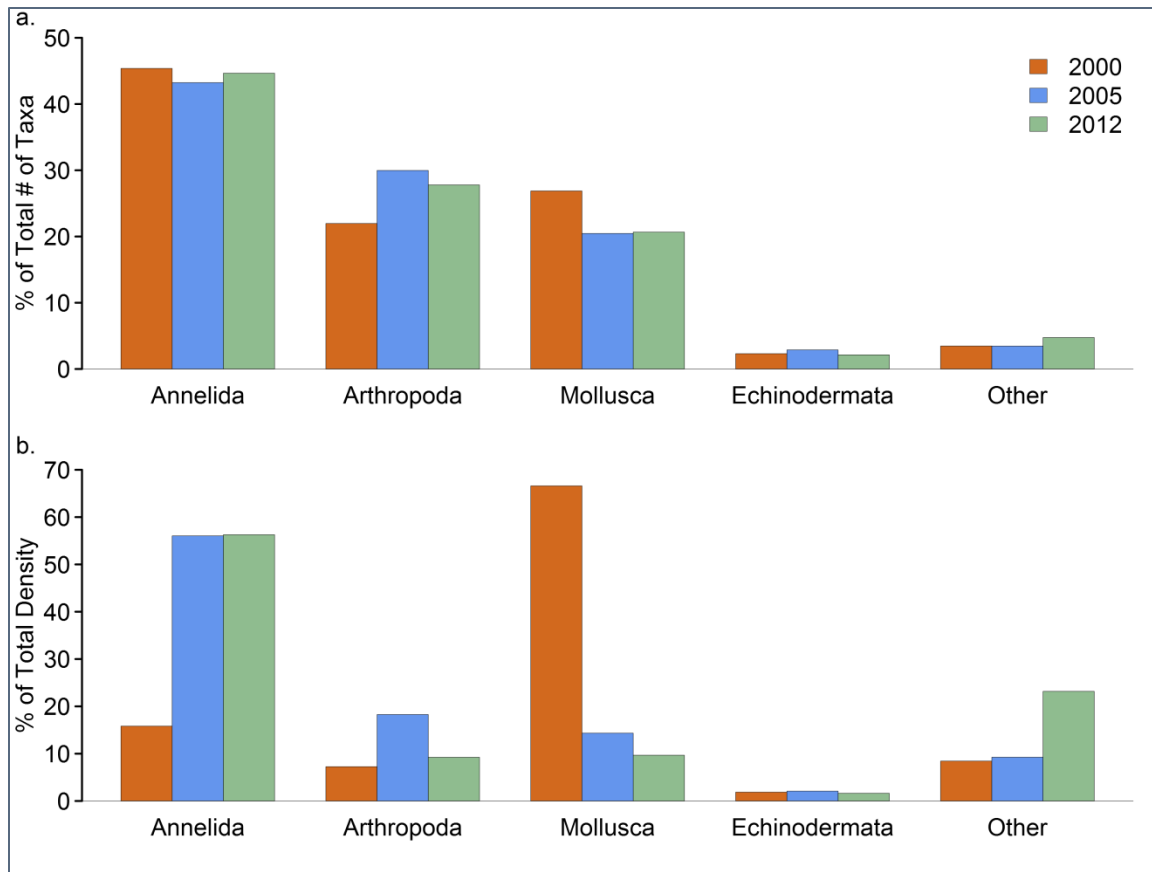


Figure 6. Comparison of percent composition (major phyla) by a) percent of total number of taxa and b) percent of total density of taxa identified in GRNMS surveys in 2000, 2005, and 2012/13.

Comparison of measures of benthic abundance and diversity (Figure 7) illustrate the influence of large numbers of *Ervilia* sp. A seen in the 2000 survey and noted above. Two stations had densities of *Ervilia* sp. A in the range of 40,000 – 50,000 individuals m⁻², resulting in Shannon (H') diversity being lower for those samples due to the unequal (uneven) proportions of taxa. The apparent increase in mean number of taxa suggested by Figure 7, however, is likely due to an entirely different phenomenon. Beginning in 2012, identifications of benthic infauna were performed at a higher level of taxonomic resolution than in previous years. Hence, while infaunal abundance and diversity (H') were similar in 2012/13 compared to 2000 and 2005, the number of unique taxa resolved were greater in 2012/13 samples since more taxa were identified to species, rather than to a higher taxonomic level (e.g., Family, Order, etc.). Hence, species was the 'lowest practical identification level' (LPIL) more often for samples analyzed in the 2012/13 survey than for those analyzed in 2000 and 2005.

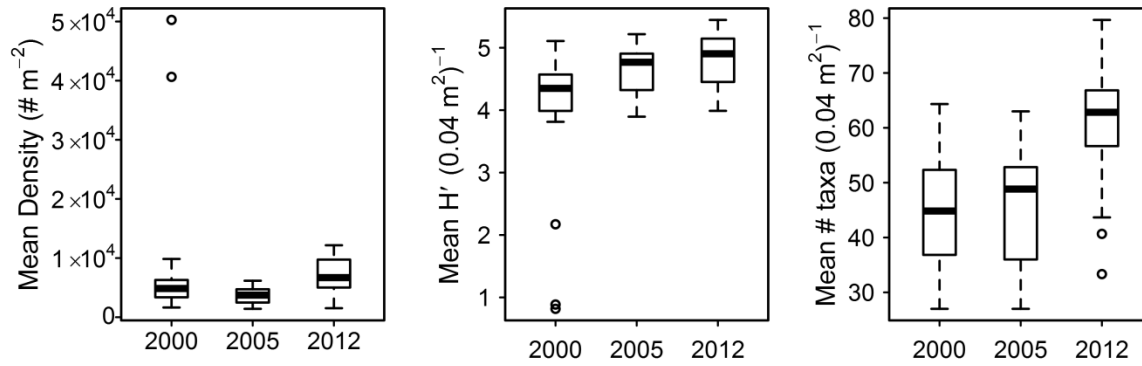


Figure 7. Comparison of measures of mean density, diversity, and taxonomic richness for GRNMS surveys in 2000, 2005, and 2012/13.

The influence of taxonomic resolution is further illustrated in Figure 8, which displays the result of non-metric multidimensional scaling (NMDS) of benthic community data from all three survey periods. Sites sampled in 2012/13 occupy a region of the ordination space that is fairly distinct from the 2000 and 2005 sites. Ordination of the benthic data after aggregating taxa to Family level (Figure 8b), however, reveals that the three datasets are much more similar (as expected) when benthic assemblages are viewed from a higher taxonomic level. The distribution of taxa by survey period is also reflected in the figure. One can see, for instance, that a fairly large number of taxa (blue filled circles in Figure 8a) were identified only in 2012/13 samples, but after aggregating the data to Family level there is a higher degree of similarity of taxa when compared with the other two survey periods (Figure 8b). Although there are more families common among the three surveys, a number of families were only encountered in any one effort; a few examples are labeled in Figure 8b.

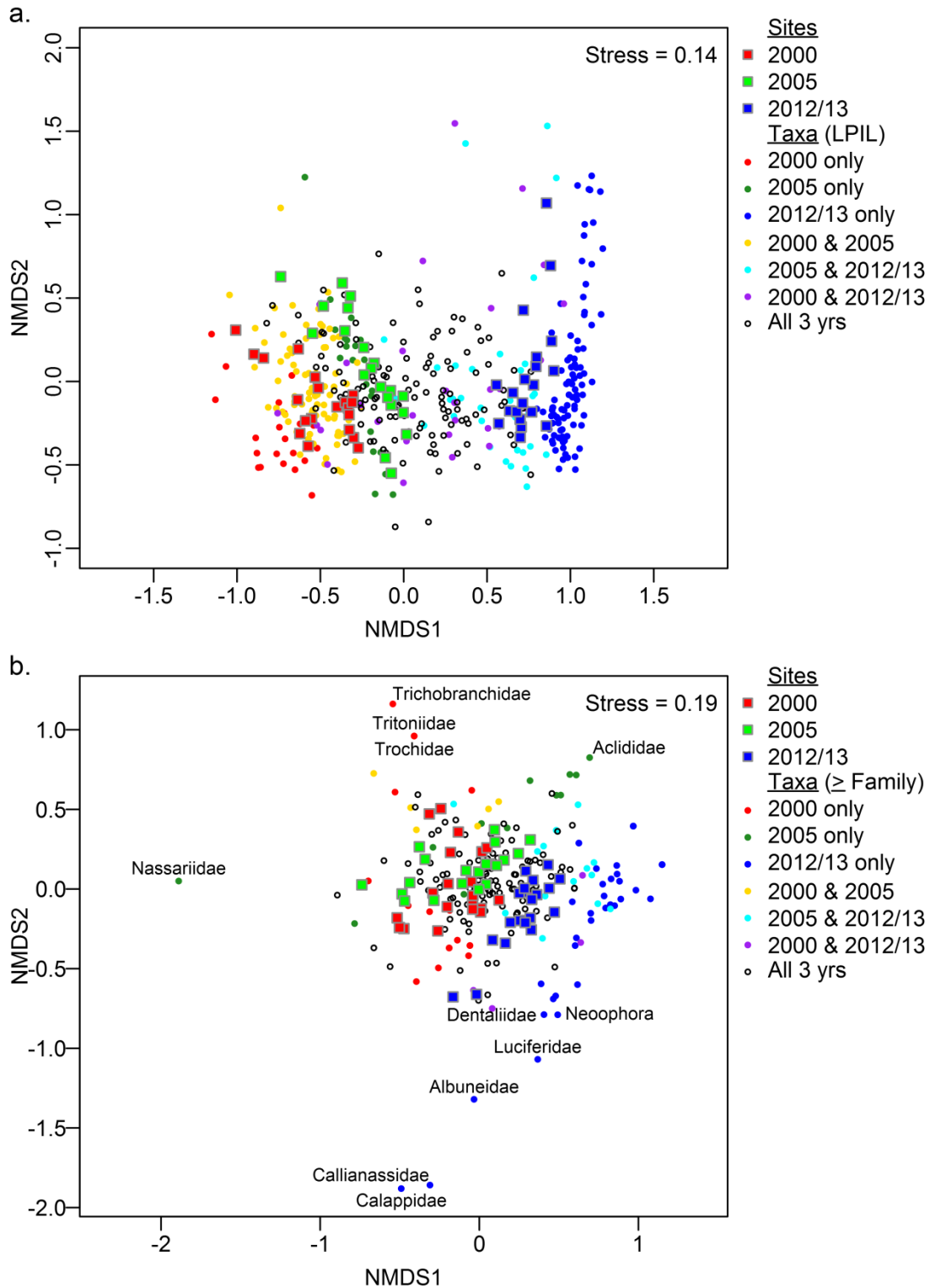


Figure 8. Results of non-metric multidimensional scaling (NMDS) performed on benthic taxa a) identified to lowest practical taxonomic level (LPIL) and b) aggregated to Family level from GRNMS surveys in 2000, 2005, and 2012/13. Filled boxes represent sampling sites, circles indicate taxa, different colors correspond to survey period(s).

Surveys of benthic infaunal assemblages in Gray’s Reef to date, including re-sampling of six sites each in 2001 and 2002, have resulted in the identification of a total of 790 taxa (Table 5), with the largest number of individual taxa identified in 2012/13 samples, as noted above. Based on species accumulation curves developed from the 2000-2005 data, it appeared previously that the number of taxa likely to be encountered in Gray’s Reef may have been approaching an estimated maximum of ~800 taxa (Balthis et al. 2007). When updated with samples collected in 2012/13, the species accumulation plot suggests that the maximum may be somewhat higher than the previous estimate. Figure 9 shows the number of taxa by sample, generated as the mean of random permutations of the combined 2000 – 2012/13 data (samples added in random order), as described previously. A curve generated similarly as for the 2000 – 2005 data is shown for comparison. The dashed curve is the result of fitting a two-parameter, asymptotic nonlinear model (Clench 1979, Colwell and Coddington 1994) to the combined 2000 – 2012/13 species accumulation data. The dotted line corresponds to an asymptotic maximum number of taxa predicted by the model, which yields a rough estimate of ~890 taxa. With the higher-resolution taxonomic identifications achieved in the 2012/13 survey, a total of 202 new taxa were added to the list of benthic fauna collected in Gray’s Reef to date, more than twice the number of taxa added in the 2005 survey.

Table 5. Numbers of taxa identified at GRNMS by survey period.

Sampling Year	Number of Sites	# of 0.04 m ² Grab Samples Collected	Total # of taxa across all sites	# of new taxa added	Mean # of taxa per site
2000	20	60	348	348	45
2001	6	18	305	96	60
2002	6	18	265	64	45
2005	20	60	353	80	47
2012/13	20	60	421	202	60
Overall	72	216		790	51

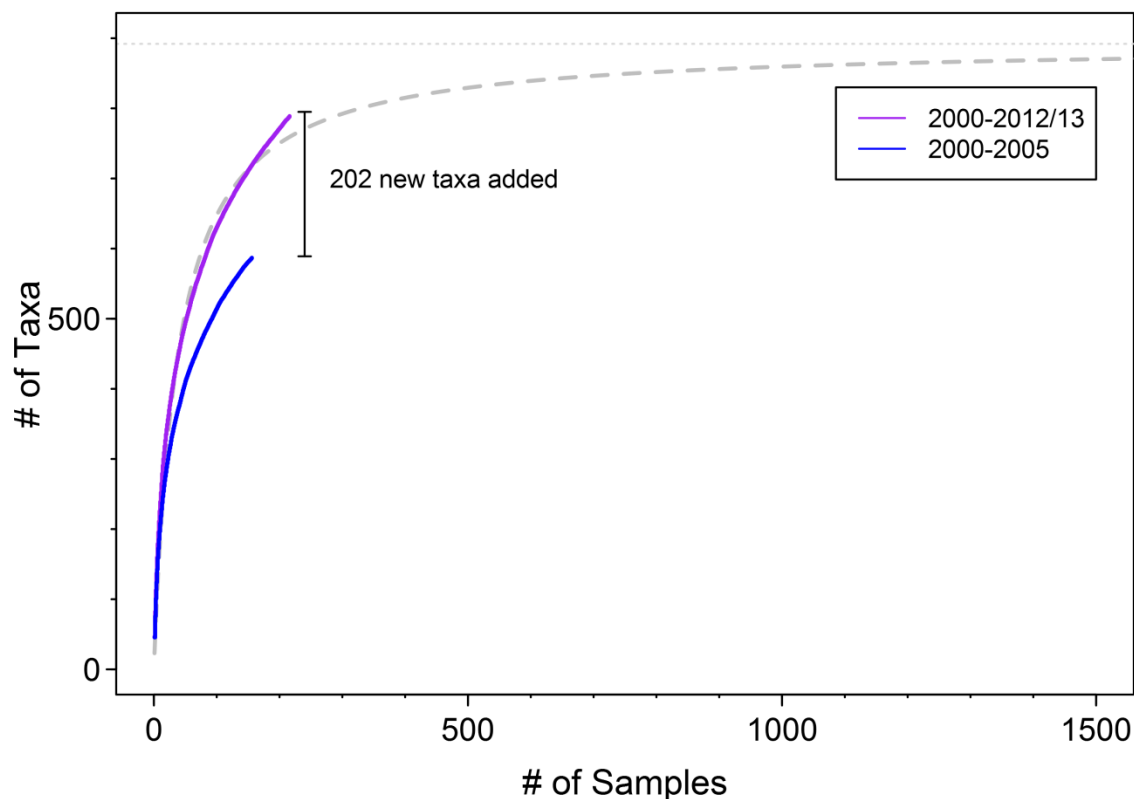


Figure 9. Comparison of species accumulation curves developed for 2000-2005 and 2000-2012/13 benthic data from GRNMS. Dashed line shows the result of fitting a non-linear asymptotic model to the 2000-2012/13 benthic data, extrapolating the species accumulation curve to additional potential sample collections.

4.0 Conclusions

Surveys of GRNMS in 2000, 2005, and 2012/13 lead to the following broad observations:

- Most chemical contaminants in soft-bottom sediments in GRNMS were at consistently low, background levels or below the limit of detection. Concentrations of copper exceeded the corresponding lower sediment quality guideline (ERL) at one station in 2000. No other exceedances of any sediment quality guidelines were observed in 2000, 2005, or 2012/13.
- Chemical contaminants in tissues of black sea bass (*Centropristis striata*) and turkey wing arks (*Arca zebra*) were present in detectable concentrations for most classes of contaminants in the three survey periods. No contaminants exceeded FDA human health guideline values. Levels of inorganic arsenic (estimated as 2% of total arsenic) measured in edible tissues of black sea bass in 2000 and 2012/13, however, fell within the range of concentrations for which EPA recommends limiting consumption to four meals per month. Levels of cadmium in arks fell within (2000, 2012/13) or exceeded (2000, 2005, 2012/13) the recommended four-meal-per-month concentration range for that contaminant.

- The extensive areas of soft-bottom sediments throughout the sanctuary support highly diverse assemblages of benthic infauna. The mean number of taxa averaged over the three survey efforts in 2000, 2005, and 2012/13 (51 taxa per sample) is comparable to, but somewhat higher than, that reported for offshore substrates of the surrounding SAB (mean of 38 taxa per sample, Cooksey et al. 2010) and much higher than at sites of comparably-high salinity (> 30 psu) sampled with the same methods in estuaries throughout the southeastern U. S. (mean=22 for 34 sites sampled 1994-1995; Hyland et al. 1996, Hyland et al. 1998). Although many of the same taxa were encountered in all three surveys, the relative abundance of individual species and patterns of dominance were variable and less repeatable, reflecting the temporally-dynamic nature of infaunal assemblages within the sanctuary and surrounding SAB.
- Cumulatively, approximately 700 taxa were identified in 2000, 2005, and 2012/13. The inclusion of six sites that were re-sampled in 2001 and 2002 brought the total number of infaunal taxa to 790. Species accumulation curves generated from these combined data suggest an estimated maximum of nearly 900 benthic taxa present in the sanctuary. This estimate is substantially higher than an earlier estimate of 800 taxa, and is likely due to the higher degree of taxonomic resolution achieved in taxonomic identifications in 2012/13. Additional sampling of infauna at GRNMS is needed to test and validate the most recent estimate.

5.0 Acknowledgments

This work was sponsored by NOAA's National Ocean Service/National Centers for Coastal Ocean Science/ Center for Coastal Environmental Health and Biomolecular Research (NOS/NCCOS/CCEHBR) and the Gray's Reef National Marine Sanctuary (GRNMS) Office. Various institutions and individuals were involved in field collections and sample processing and analysis. These included Todd Recicar (GRNMS) for research vessel operations; JD Dubick and Blaine West (NOS/NCCOS/CCEHBR) for field operations, sample collection, and processing; JD Dubick and Stephen Roth (NOS/NCCOS/CCEHBR) for analysis of sediment grain size (2012 samples); Alpha Scientific, Inc. for identification and enumeration of benthic infauna and analysis of sediment grain size (2013 samples) and TOC; LouAnn Reed, Brian Shaddrix, and Lynn Thorsell (NOS/NCCOS/CCEHBR) for sediment and tissue chemistry analysis; and Katy Chung for sediment toxicity analysis.

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Appendices

Appendix A. ERM and ERL guideline values in sediments (Long et al. 1995).

Chemical	ERL	ERM
Metals ($\mu\text{g g}^{-1}$)		
Arsenic	8.2	70
Cadmium	1.2	9.6
Chromium	81	370
Copper	34	270
Lead	46.7	218
Mercury	0.15	0.71
Nickel	20.9	51.6
Silver	1	3.7
Zinc	150	410
Organics (ng g^{-1})		
Acenaphthene	16	500
Acenaphthylene	44	640
Anthracene	85.3	1,100
Fluorene	19	540
2-Methylnaphthalene	70	670
Naphthalene	160	2,100
Phenanthrene	240	1,500
Benzo[a]anthracene	261	1,600
Benzo[a]pyrene	430	1,600
Chrysene	384	2,800
Dibenz[a,h]Anthracene	63.4	260
Fluoranthene	600	5,100
Pyrene	665	2,600
Low molecular weight PAHs	552	3,160
High molecular weight PAHs	1,700	9,600
Total PAHs	4,020	44,800
4,4-DDE	2.2	27
Total DDT	1.58	46.1
Total PCBs	22.7	180

Appendix B. Action levels, tolerances, and levels of concern established by the Food and Drug Administration (FDA 1984, 1993a-e, 1994, 2007) for poisonous or deleterious substances in human food.

	FDA Guideline Value
Metals ($\mu\text{g g}^{-1}$)	
Arsenic	86
Cadmium	4
Chromium	13
Lead	1
Mercury (methylmercury)*	1
Nickel	80
Organics (ng g^{-1})	
Aldrin	300
Chlordane	300
DDD*	5000
DDE*	5000
DDT*	5000
Dieldrin	300
Heptachlor	300
Heptachlor epoxide	300
Mirex	100
PCB (total)	2000

Appendix C. Risk-based EPA advisory guidelines for recreational fishers (USEPA 2000).

	EPA Advisory Guidelines Concentration Range	
Metals ($\mu\text{g g}^{-1}$)		
Arsenic (inorganic) ^c	>0.35	– 0.70 ^a
Cadmium	>0.35	– 0.70 ^a
Mercury (methylmercury) ^d	>0.12	– 0.23 ^a
Selenium	>5.90	– 12.00 ^a
Organics (ng g^{-1})		
Chlordane	>590	– 1,200 ^a
Chlorpyrifos	>350	– 700 ^a
DDT (total)	>59	– 120 ^a
Dieldrin	>59	– 120 ^a
Endosulfan	>7,000	– 14,000 ^a
Heptachlor epoxide	>15	– 31 ^a
Hexachlorobenzene	>940	– 1,900 ^a
Lindane	>350	– 700 ^a
Mirex	>230	– 470 ^a
Toxaphene	>290	– 590 ^a
PAHs (benzo[a]pyrene)	>1.6	– 3.2 ^b
PCB (total)	>23	– 47 ^a

^a Range of concentrations for non-cancer health endpoints; based on the assumption that consumption over a lifetime of four 8-oz meals per month would not generate a chronic, systemic health risk.

^b Range of concentrations for cancer health endpoints; based on the assumption that consumption over a lifetime of four 8-oz meals per month would yield a lifetime cancer risk no greater than an acceptable risk of 1 in 100,000.

^c Inorganic arsenic, the form considered toxic, estimated as 2% of total arsenic.

^d Because most mercury in fish and shellfish tissue is present primarily as methylmercury and because of the relatively high cost of analyzing for methylmercury, the conservative assumption was made that all mercury is present as methylmercury (U.S. EPA, 2000).

Appendix D. Locations (latitude, longitude), depths, bottom-water, and sediment characteristics of 2012/13 GRNMS sampling stations.

Station	Latitude	Longitude	Station Depth (m)	Temperature (°C)	Salinity (psu)	DO (mg L ⁻¹)	pH	Gravel (%)	Sand (%)	Silt+Clay (%)	TOC (%)
65	31.4071	-80.9129	16.9	29.0	35.8	6.1	8.1	0.9	96.5	2.6	0.2
66	31.4084	-80.8985	19.8	28.9	35.9	6.2	8.1	0.7	97.3	2.0	0.2
67	31.4149	-80.8768	20.8	28.5	36.0	6.3	8.2	0.7	97.0	2.4	1.0
68	31.4176	-80.8552	19.1	23.9	34.7	6.5	7.8		93.9	6.2	0.1
69	31.4141	-80.8348	17.7	23.5	34.8	6.5	7.8		90.7	9.3	0.1
70	31.4013	-80.9053	20.0	28.9	35.9	6.1	8.1	1.9	96.4	1.7	0.1
71	31.4012	-80.8872	19.5	28.8	35.9		8.1	0.7	97.3	2.0	0.0
72	31.4049	-80.8759	20.8	29.1	35.4		8.2	0.4	96.6	3.0	0.0
73	31.3989	-80.8582	15.2	24.4	34.7	6.8	7.8		91.5	8.5	0.1
74	31.3989	-80.8313	16.5	24.0	34.8	6.6	7.8		91.6	8.4	0.1
75	31.3884	-80.9079	17.4	28.8	36.1	6.4	8.2	1.8	95.8	2.4	0.1
76	31.3877	-80.8957	14.1	24.7	34.7	6.8	7.7		88.0	12.0	0.1
77	31.3838	-80.8789	16.3	24.7	34.8	6.7	7.7		84.4	15.6	0.2
78	31.3776	-80.8544	16.8	24.8	34.9	6.8	7.7		90.8	9.2	0.1
79	31.3791	-80.8444	15.3	24.7	34.9	6.8	7.7		88.7	11.3	0.1
80	31.3771	-80.9084	19.0	28.8	36.0	6.5	8.2	0.9	95.9	3.2	0.0
81	31.3654	-80.8910	18.9	28.6	36.1	6.6	8.2	0.6	97.1	2.3	0.5
82	31.3727	-80.8713	19.4	28.5	36.1	6.6	8.2	1.6	96.4	2.0	0.0
83	31.3662	-80.8554	19.9	28.1	36.1	6.7	8.2	1.1	96.8	2.1	0.2
84	31.3725	-80.8452	16.8	24.8	35.0	6.8	7.7		91.4	8.7	0.2

Appendix E. Summary of sediment contaminant concentrations measured at Gray's Reef National Marine Sanctuary in July 2012 and June 2013 in relation to sediment quality guidelines (SQG). Concentrations below method detection limits are reported as < MDL; in these cases, a value of zero was used for data computations (e.g., averaging across stations).

Analyte	Average	Range		SQG ^a		# Sites > SQG	
		Min.	Max.	ERL	ERM	ERL	ERM
<i>Metals (µg g⁻¹ dry wt., unless indicated otherwise)</i>							
Aluminum (%)	0.99	0.49	1.97	–	–	–	–
Antimony	0.22	< MDL	1.49	–	–	–	–
Arsenic	2.60	1.38	4.61	8.2	70	0	0
Barium	71.43	42.76	125.48	–	–	–	–
Beryllium	0.11	< MDL	0.15	–	–	–	–
Cadmium	0.33	0.18	0.59	1.2	9.6	0	0
Chromium	7.95	3.92	11.85	81	370	0	0
Cobalt	0.94	< MDL	1.42	–	–	–	–
Copper	1.05	< MDL	3.98	34	270	0	0
Iron (%)	0.24	0.19	0.29	–	–	–	–
Lead	3.41	1.93	5.60	46.7	218	0	0
Lithium	0.77	0.57	1.12	–	–	–	–
Manganese	54.30	24.47	112.57	–	–	–	–
Mercury	0.00	0.00	0.01	0.15	0.71	0	0
Nickel	1.43	0.98	1.79	–	–	–	–
Selenium	0.33	< MDL	0.53	–	–	–	–
Silver	< MDL	< MDL	< MDL	1	3.7	0	0
Thallium	< MDL	< MDL	< MDL	–	–	–	–
Tin	0.51	0.37	0.66	–	–	–	–
Uranium	2.91	1.41	4.75	–	–	–	–
Vanadium	7.77	3.88	13.31	–	–	–	–
Zinc	22.44	12.51	85.49	150	410	0	0
<i>PAHs (ng g⁻¹ dry wt.)</i>							
1,6,7-Trimethylnaphthalene	< MDL	< MDL	< MDL	–	–	–	–
2,6-Dimethylnaphthalene	< MDL	< MDL	< MDL	–	–	–	–
Benzo[g,h,i]perylene	< MDL	< MDL	< MDL	–	–	–	–
Dibenz[a,h]anthracene	< MDL	< MDL	< MDL	63.4	260	0	0
Indeno[1,2,3-c,d]pyrene	< MDL	< MDL	< MDL	–	–	–	–
1-Methylnaphthalene	< MDL	< MDL	< MDL	–	–	–	–
1-Methylphenanthrene	< MDL	< MDL	< MDL	–	–	–	–
2-Methylnaphthalene	< MDL	< MDL	< MDL	70	670	0	0

Acenaphthene	< MDL	< MDL	< MDL	16	500	0	0
Acenaphthylene	< MDL	< MDL	< MDL	44	640	0	0
Anthracene	< MDL	< MDL	< MDL	85.3	1100	0	0
Benz[a]anthracene	< MDL	< MDL	< MDL	261	1600	0	0
Benzo[a]fluoranthene	< MDL	< MDL	< MDL	–	–	–	–
Benzo[a]pyrene	< MDL	< MDL	< MDL	430	1600	0	0
Benzo[b]fluoranthene	< MDL	< MDL	< MDL	–	–	–	–
Benzo[e]pyrene	< MDL	< MDL	< MDL	–	–	–	–
Benzo[j]fluoranthene	< MDL	< MDL	< MDL	–	–	–	–
Benzo[k]fluoranthene	< MDL	< MDL	< MDL	–	–	–	–
Biphenyl	< MDL	< MDL	< MDL	–	–	–	–
Chrysene+Triphenylene	< MDL	< MDL	< MDL	384 ^b	2800 ^b	0	0
Dibenzothiophene	< MDL	< MDL	< MDL	–	–	–	–
Fluoranthene	< MDL	< MDL	< MDL	600	5100	0	0
Fluorene	< MDL	< MDL	< MDL	19	540	0	0
Naphthalene	< MDL	< MDL	< MDL	160	2100	0	0
Perylene	< MDL	< MDL	< MDL	–	–	–	–
Phenanthrene	< MDL	< MDL	< MDL	240	1500	0	0
Pyrene	< MDL	< MDL	< MDL	665	2600	0	0
Retene	< MDL	< MDL	< MDL	–	–	–	–
HMW_PAH	< MDL	< MDL	< MDL	1700	9600	0	0
LMW_PAH	< MDL	< MDL	< MDL	552	3160	0	0
TOT_PAH ^c	< MDL	< MDL	< MDL	4022	44792	0	0
<i>PCBs (ng g⁻¹ dry wt.)</i>							
Total PCBs	0.04	< MDL	0.23	22.7	180	0	0
<i>Pesticides (ng g⁻¹ dry wt.)</i>							
2,4'-DDD (o,p'-DDD)	< MDL	< MDL	< MDL	–	–	–	–
2,4'-DDE (o,p'-DDE)	< MDL	< MDL	< MDL	–	–	–	–
2,4'-DDT (o,p'-DDT)	< MDL	< MDL	< MDL	–	–	–	–
4,4'-DDD (p,p'-DDD)	< MDL	< MDL	< MDL	–	–	–	–
4,4'-DDE (p,p'-DDE)	< MDL	< MDL	< MDL	2.2	27	0	0
4,4'-DDT (p,p'-DDT)	< MDL	< MDL	< MDL	–	–	–	–
Aldrin	< MDL	< MDL	< MDL	–	–	–	–
alpha-Chlordane	< MDL	< MDL	< MDL	–	–	–	–
alpha-Hexachlorocyclohexane (alpha-BHC)	< MDL	< MDL	< MDL	–	–	–	–
beta-Hexachlorocyclohexane (beta-BHC)	< MDL	< MDL	< MDL	–	–	–	–
Chlorpyrifos	< MDL	< MDL	< MDL	–	–	–	–
cis-Nonachlor	< MDL	< MDL	< MDL	–	–	–	–

Dieldrin	< MDL	< MDL	< MDL	–	–	–	–
Endosulfan I	< MDL	< MDL	< MDL	–	–	–	–
Endosulfan II (Beta-Endosulfan)	< MDL	< MDL	< MDL	–	–	–	–
Endosulfan sulfate	< MDL	< MDL	< MDL	–	–	–	–
Endrin	< MDL	< MDL	< MDL	–	–	–	–
gamma-Chlordane	< MDL	< MDL	< MDL	–	–	–	–
gamma-Hexachlorocyclohexane (gamma-BHC = Lindane)	< MDL	< MDL	< MDL	–	–	–	–
Heptachlor	< MDL	< MDL	< MDL	–	–	–	–
Heptachlor epoxide	< MDL	< MDL	< MDL	–	–	–	–
Hexachlorobenzene (HCB)	< MDL	< MDL	< MDL	–	–	–	–
Mirex	< MDL	< MDL	< MDL	–	–	–	–
Oxychlordane	< MDL	< MDL	< MDL	–	–	–	–
trans-Nonachlor	< MDL	< MDL	< MDL	–	–	–	–
DDD ^d	< MDL	< MDL	< MDL	–	–	–	–
DDE ^e	< MDL	< MDL	< MDL	–	–	–	–
DDT ^f	< MDL	< MDL	< MDL	–	–	–	–
Total DDT ^g	< MDL	< MDL	< MDL	1.58	46.1	0	0

^a SQGs are the ERL and ERM values from Long et al. (1995).

^b ERL/ERM values are for chrysene only.

^c Without perylene.

^d DDD = 2,4'-DDD + 4,4'-DDD.

^e DDE = 2,4'-DDE + 4,4'-DDE.

^f DDT = 2,4'-DDT + 4,4'-DDT.

^g Total DDTs = DDD + DDE + DDT.

Appendix F. Summary of contaminant concentration ranges observed in edible tissues of black sea bass (*Centropristis striata*) at GRNMS sites in 2012/13. Concentrations are reported on a wet-weight basis and compared with FDA^a and EPA^b human-health guideline values, where available. Concentrations below method detection limits are reported as < MDL; in these cases, a value of zero was used for data computations (e.g., averaging across all stations). Comparisons to FDA guidelines are expressed as the number of specimens (out of 10 individual fish fillets, total) having tissue concentrations that exceeded the respective guideline. Comparisons to EPA guidelines are expressed as the number of specimens with tissue concentrations falling within the range for which EPA recommends limiting consumption to four meals per month (“# Specimens”) or the number exceeding the upper limit of the corresponding four-meals-per-month range (“# Specimens Exceeding”).

Black sea bass (n=10)					
Analyte	Average	Range		EPA Human-Health Guidelines	
		Min.	Max.	# Specimens Exceeding	# Specimens Exceeding
<i>Metals (ug g⁻¹ dry wt., unless indicated otherwise)</i>					
Silver	< MDL	< MDL	< MDL	–	–
Aluminum (%)	8.14	1.67	30.71	–	–
Arsenic	13.05	7.75	19.88	0	–
AS_inorganic	0.26	0.15	0.40	–	1
Barium	0.07	< MDL	0.19	–	–
Beryllium	< MDL	< MDL	< MDL	–	–
Cadmium	< MDL	< MDL	< MDL	0	0
Cobalt	< MDL	< MDL	< MDL	–	–
Chromium	0.43	0.13	0.71	0	–
Copper	0.15	< MDL	0.40	–	–
Iron (%)	25.28	23.64	26.24	–	–
Mercury	0.06	0.04	0.10	0	0
Lithium	< MDL	< MDL	< MDL	–	–
Manganese	0.05	< MDL	0.29	–	–
Nickel	0.05	< MDL	0.27	0	–
Lead	< MDL	< MDL	< MDL	0	–
Antimony	< MDL	< MDL	< MDL	–	–
Selenium	0.58	0.39	0.76	–	0
Tin	0.02	< MDL	0.08	–	–
Thallium	< MDL	< MDL	< MDL	–	–
Uranium	< MDL	< MDL	< MDL	–	–
Vanadium	0.22	0.14	0.32	–	–
Zinc	8.02	3.78	16.88	–	–
<i>PAHs (ng g⁻¹ dry wt.)</i>					
Acenaphthene	< MDL	< MDL	< MDL	–	–
Acenaphthylene	< MDL	< MDL	< MDL	–	–

Anthracene	< MDL	< MDL	< MDL	—	—	—
Benz[a]anthracene	< MDL	< MDL	< MDL	—	—	—
Benzo[a]pyrene	0.42	< MDL	0.95	—	0	0
Benzo[e]pyrene	< MDL	< MDL	< MDL	—	—	—
Benzo[a]fluoranthene	< MDL	< MDL	< MDL	—	—	—
Benzo[b]fluoranthene	< MDL	< MDL	< MDL	—	—	—
Benzo[j]fluoranthene	< MDL	< MDL	< MDL	—	—	—
Benzo[j+k]fluoranthene	< MDL	< MDL	< MDL	—	—	—
Benzo[g,h,i]perylene	< MDL	< MDL	< MDL	—	—	—
Biphenyl	< MDL	< MDL	< MDL	—	—	—
Chrysene+Triphenylene	< MDL	< MDL	< MDL	—	—	—
Dibenz[a,h]anthracene	< MDL	< MDL	< MDL	—	—	—
Dibenzothiophene	< MDL	< MDL	< MDL	—	—	—
2,6-Dimethylnaphthalene	< MDL	< MDL	< MDL	—	—	—
Fluoranthene	< MDL	< MDL	< MDL	—	—	—
Fluorene	< MDL	< MDL	< MDL	—	—	—
Indeno[1,2,3-c,d]pyrene	< MDL	< MDL	< MDL	—	—	—
1-Methylnaphthalene	< MDL	< MDL	< MDL	—	—	—
2-Methylnaphthalene	< MDL	< MDL	< MDL	—	—	—
1-Methylphenanthrene	< MDL	< MDL	< MDL	—	—	—
Naphthalene	< MDL	< MDL	< MDL	—	—	—
Perylene	< MDL	< MDL	< MDL	—	—	—
Phenanthrene	< MDL	< MDL	< MDL	—	—	—
Pyrene	< MDL	< MDL	< MDL	—	—	—
Retene	< MDL	< MDL	< MDL	—	—	—
1,6,7-Trimethylnaphthalene	< MDL	< MDL	< MDL	—	—	—
TOT_PAH	0.42	< MDL	0.95	—	—	—
<i>PCBs (ng g⁻¹ dry wt.)</i>						
TOT_PCB	1.98	0.42	6.82	0	0	0
<i>Pesticides (ng g⁻¹ dry wt.)</i>						
2,4'-DDD (o,p'-DDD)	0.06	< MDL	0.64	—	—	—
4,4'-DDD (p,p'-DDD)	0.23	< MDL	1.00	—	—	—
2,4'-DDE (o,p'-DDE)	0.00	< MDL	0.01	—	—	—
4,4'-DDE (p,p'-DDE)	0.10	< MDL	0.33	—	—	—
2,4'-DDT (o,p'-DDT)	< MDL	< MDL	< MDL	—	—	—
4,4'-DDT (p,p'-DDT)	< MDL	< MDL	< MDL	—	—	—
Aldrin	< MDL	< MDL	< MDL	0	—	—
alpha-Hexachlorocyclohexane	< MDL	< MDL	< MDL	—	—	—

alpha-Chlordane	0.01	< MDL	0.03	–	–	–
beta-Hexachlorocyclohexane	< MDL	< MDL	< MDL	–	–	–
Chlorpyrifos	< MDL	< MDL	< MDL	–	–	–
cis-Nonachlor	0.02	< MDL	0.07	–	–	–
Dieldrin	0.02	< MDL	0.14	0	0	0
Endosulfan I	< MDL	< MDL	< MDL	–	–	–
Endosulfan II (Beta-Endosulfan)	< MDL	< MDL	< MDL	–	0	0
Endrin	< MDL	< MDL	< MDL	–	–	–
Endosulfan sulfate	< MDL	< MDL	< MDL	–	–	–
gamma-Chlordane	< MDL	< MDL	< MDL	–	–	–
Hexachlorobenzene (HCB)	0.01	< MDL	0.06	–	0	0
Heptachlor	< MDL	< MDL	< MDL	0	–	–
Heptachlor epoxide	0.00	< MDL	0.04	0	0	0
Lindane	< MDL	< MDL	< MDL	–	0	0
Mirex	0.01	< MDL	0.06	0	0	0
Oxychlordane	0.01	< MDL	0.05	–	–	–
Total Chlordane	0.12	< MDL	0.29	–	0	0
trans-Nonachlor	0.07	< MDL	0.22	–	–	–
DDD ^c	0.30	< MDL	1.00	0	–	–
DDE ^d	0.11	< MDL	0.33	0	–	–
DDT ^e	< MDL	< MDL	< MDL	0	–	–
Total DDT ^f	0.40	< MDL	1.06	0	0	0

^a FDA (1984, 1993a-e, 1994).

^b EPA (2008).

^c DDD = 2,4'-DDD + 4,4'-DDD.

^d DDE = 2,4'-DDE + 4,4'-DDE.

^e DDT = 2,4'-DDT + 4,4'-DDT.

^f Total DDTs = DDD + DDE + DDT.

Appendix G. Summary of contaminant concentration ranges observed in edible tissues of turkey wing arks (*Arca zebra*) at GRNMS sites in 2012/13. Concentrations are reported on a wet-weight basis and compared with FDA^a and EPA^b human-health guideline values, where available. Concentrations below method detection limits are reported as < MDL; in these cases, a value of zero was used for data computations (e.g., averaging across all stations). Comparisons to FDA guidelines are expressed as the number of specimens (out of 10 composite samples, each consisting of 3 – 15 individual arks) having tissue concentrations that exceeded the respective guideline. Comparisons to EPA guidelines are expressed as the number of specimens with tissue concentrations falling within the range for which EPA recommends limiting consumption to four meals per month (“# Specimens”) or the number exceeding the upper limit of the corresponding four-meals-per-month range (“# Specimens Exceeding”).

Turkey wing arks (n=10 ^c)						
Analyte	Average	Range		FDA Action Levels # Specimens	EPA Human-Health Guidelines	
		Min	Max		# Specimens	# Specimens Exceeding
<i>Metals (ug g⁻¹ dry wt., unless indicated otherwise)</i>						
Silver	0.27	0.11	0.51	–	–	–
Aluminum	12.80	4.88	21.72	–	–	–
Arsenic	8.73	4.07	14.14	0	–	–
AS_inorganic	0.17	0.08	0.28	–	0	0
Barium	2.81	1.06	8.02	–	–	–
Beryllium	< MDL	< MDL	< MDL	–	–	–
Cadmium	0.55	0.17	0.98	0	6	2
Cobalt	0.03	< MDL	0.13	–	–	–
Chromium	1.16	0.25	2.53	0	–	–
Copper	0.77	0.58	0.96	–	–	–
Iron	39.09	19.47	48.22	–	–	–
Mercury	0.01	0.01	0.02	0	0	0
Lithium	0.15	< MDL	0.24	–	–	–
Manganese	3.12	1.63	3.99	–	–	–
Nickel	0.11	< MDL	0.23	0	–	–
Lead	0.01	< MDL	0.03	0	–	–
Antimony	< MDL	< MDL	< MDL	–	–	–
Selenium	0.96	0.42	1.92	–	0	0
Tin	0.01	< MDL	0.07	–	–	–
Thallium	< MDL	< MDL	< MDL	–	–	–
Uranium	< MDL	< MDL	< MDL	–	–	–
Vanadium	1.22	0.65	2.61	–	–	–
Zinc	8.37	5.10	11.06	–	–	–
<i>PAHs (ng g⁻¹ dry wt.)</i>						
Acenaphthene	< MDL	< MDL	< MDL	–	–	–

Acenaphthylene	< MDL	< MDL	< MDL	—	—	—
Anthracene	< MDL	< MDL	< MDL	—	—	—
Benz[a]anthracene	< MDL	< MDL	< MDL	—	—	—
Benzo[a]pyrene	< MDL	< MDL	< MDL	—	0	0
Benzo[e]pyrene	< MDL	< MDL	< MDL	—	—	—
Benzo[a]fluoranthene	< MDL	< MDL	< MDL	—	—	—
Benzo[b]fluoranthene	< MDL	< MDL	< MDL	—	—	—
Benzo[j]fluoranthene	< MDL	< MDL	< MDL	—	—	—
Benzo[j+k]fluoranthene	< MDL	< MDL	< MDL	—	—	—
Benzo[g,h,i]perylene	< MDL	< MDL	< MDL	—	—	—
Biphenyl	< MDL	< MDL	< MDL	—	—	—
Chrysene+Triphenylene	< MDL	< MDL	< MDL	—	—	—
Dibenz[a,h]anthracene	< MDL	< MDL	< MDL	—	—	—
Dibenzothiophene	< MDL	< MDL	< MDL	—	—	—
2;6-Dimethylnaphthalene	< MDL	< MDL	< MDL	—	—	—
Fluoranthene	< MDL	< MDL	< MDL	—	—	—
Fluorene	< MDL	< MDL	< MDL	—	—	—
Indeno[1;2;3-c;d]pyrene	< MDL	< MDL	< MDL	—	—	—
1-Methylnaphthalene	< MDL	< MDL	< MDL	—	—	—
2-Methylnaphthalene	< MDL	< MDL	< MDL	—	—	—
1-Methylphenanthrene	< MDL	< MDL	< MDL	—	—	—
Naphthalene	< MDL	< MDL	< MDL	—	—	—
Perylene	< MDL	< MDL	< MDL	—	—	—
Phenanthrene	< MDL	< MDL	< MDL	—	—	—
Pyrene	< MDL	< MDL	< MDL	—	—	—
Retene	< MDL	< MDL	< MDL	—	—	—
1;6;7-Trimethylnaphthalene	< MDL	< MDL	< MDL	—	—	—
TOT_PAH	< MDL	< MDL	< MDL	—	—	—
<i>PCBs (ng g⁻¹ dry wt.)</i>						
Total PCB	0.72	< MDL	1.43	0	0	0
<i>Pesticides (ng g⁻¹ dry wt.)</i>						
2;4'-DDD (o;p'-DDD)	< MDL	< MDL	< MDL	—	—	—
4;4'-DDD (p;p'-DDD)	0.13	< MDL	0.65	—	—	—
2;4'-DDE (o;p'-DDE)	0.04	0.02	0.05	—	—	—
4;4'-DDE (p;p'-DDE)	0.02	< MDL	0.05	—	—	—
2;4'-DDT (o;p'-DDT)	< MDL	< MDL	< MDL	—	—	—
4;4'-DDT (p;p'-DDT)	< MDL	< MDL	< MDL	—	—	—
Aldrin	< MDL	< MDL	< MDL	0	—	—

alpha-Hexachlorocyclohexane (alpha-BHC)	< MDL	< MDL	< MDL	—	—	—
alpha-Chlordane	0.00	< MDL	0.02	—	—	—
beta-Hexachlorocyclohexane (beta-BHC)	< MDL	< MDL	< MDL	—	—	—
Chlorpyrifos	< MDL	< MDL	< MDL	—	—	—
cis-Nonachlor	< MDL	< MDL	< MDL	—	—	—
Dieldrin	0.01	< MDL	0.05	0	0	0
Endosulfan I	< MDL	< MDL	< MDL	—	—	—
Endosulfan II (Beta-Endosulfan)	< MDL	< MDL	< MDL	—	0	0
Endrin	< MDL	< MDL	< MDL	—	—	—
Endosulfan sulfate	< MDL	< MDL	< MDL	—	—	—
gamma-Chlordane	< MDL	< MDL	< MDL	—	—	—
Hexachlorobenzene (HCB)	< MDL	< MDL	< MDL	—	0	0
Heptachlor	< MDL	< MDL	< MDL	0	—	—
Heptachlor epoxide	< MDL	< MDL	< MDL	0	0	0
gamma-Hexachlorocyclohexane (gamma-BHC = Lindane)	< MDL	< MDL	< MDL	—	0	0
Mirex	< MDL	< MDL	< MDL	0	0	0
Oxychlordane	< MDL	< MDL	< MDL	—	—	—
trans-Nonachlor	0.02	< MDL	0.03	—	—	—
Total Chlordane	0.02	< MDL	0.05	—	0	0
DDD ^d	0.13	< MDL	0.65	0	—	—
DDE ^e	0.06	0.02	0.09	0	—	—
DDT ^f	< MDL	< MDL	< MDL	0	—	—
Total DDT ^g	0.19	0.02	0.73	0	0	0

^a FDA (1984, 1993a-e, 1994).

^b EPA (2008).

^c Total of 10 specimens, each consisting of 3 - 15 individual arks.

^d DDD = 2,4'-DDD + 4,4'-DDD.

^e DDE = 2,4'-DDE + 4,4'-DDE.

^f DDT = 2,4'-DDT + 4,4'-DDT.

^g Total DDTs = DDD + DDE + DDT.



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