



TECHNICAL REPORT 2038
March 2013

Marine Ecologic Index Survey of San Diego Bay

Dr. Kara Sorensen
Brandon Swope
Victoria Kirtay
SSC Pacific

Approved for public release.

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San Diego, CA 92152-5001

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ADMINISTRATIVE INFORMATION

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Taxonomist specialties were as follows:

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- Gretchen Lambert, MS – University of Washington, Ascidians
- John Ljubenkov – Senior Scientist at Dancing Coyote Environmental, Mollusca, Cnidaria, and other minor phyla
- Leslie Harris – collections manager at Natural History Museum Los Angeles county, Polychaetes
- Melissa Blando – University of California, San Diego marine ecology student – minor phyla.

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EXECUTIVE SUMMARY

BACKGROUND

San Diego Bay is the largest naturally occurring embayment in the Southern California Bight (SCB), consisting of approximately 11,000 acres of marine habitat and home to a diverse aquatic population that helps sustain various fauna that use the bay for breeding, rearing young, and migratory respite. It is also one of California's five major ports and is an important hub for industry and commerce and home to the U.S. Navy Pacific Fleet. As a result, the anthropogenic influences can at times be at odds with the ecological needs. Many researchers have indicated that the introduction and spread of non-indigenous marine organisms is probably one of the greatest threats to the sustainability of complex marine ecosystems, including the San Diego Bay. To mitigate this threat, both native and non-indigenous species populations need to be monitored routinely. In response to this, Navy Region Southwest (NRSW) conducted a Marine Ecological Index Study to establish a directory of current benthic conditions within San Diego Bay's marine environment.

REPORT FOCUS

The purpose of this study was to conduct an ecological index, early detection survey to identify and catalog native and non-indigenous species near naval facilities within the four hydrographic regions in the bay. Work was similar to a Rapid Assessment Survey (RAS) methodology and a team of taxonomists identified live specimens for 5 days. The focus of this study was to identify native, introduced, and cryptogenic species present on multiple natural and artificial habitats within the four hydrographic regions. This overall goal was broken down into four key objectives: (1) to understand and summarize historic data on species distribution, including presence of exotic species within the four management regions of the San Diego Bay; (2) to plan and execute a Rapid Assessment (5-day) Survey using a random sampling strategy to identify species in each of the four hydrographic regions of the Bay (marine, thermal, seasonally hypersaline, and estuarine); (3) to assess feasibility of using DNA barcoding as a tool for augmenting species identification in a rapid assessment platform; and (4) to provide summary of data relative to the four management regions, including species distribution and relative abundance relative to U.S. Navy facilities.

RESULTS SUMMARY

The team collected and identified 6,477 organisms, with 299 species represented from 13 phyla. Species identified in this study were similar to those reported in previous studies; however, there were some differences in distribution within the bay. In addition, two previously unreported species were identified. Results presented will include the distribution of native and non-indigenous species identified from natural and artificial habitats within the four hydrographic regions.

ACRONYMS AND ABBREVIATIONS

BOLD	Barcode of Life Data Systems
BRI	Benthic Response Index
CATEX	Categorical Exclusion
CBOL	Consortium for the Barcode of Life
DNA	Deoxyribonucleic Acid
EIS	Environmental Impact Statement
ETOH	Ethanol
GPS	Global Positioning System
MgSO ₄	Magnesium Sulfate
NBSD	Naval Base San Diego
NAVFAC	Naval Facilities Engineering Command Southwest
NAVSTA	Naval Station San Diego
NRSW	Navy Region Southwest
PSU	Practical Salinity Units
RAS	Rapid Assessment Survey
SDRWQCB	Regional Water Quality Control Board, San Diego Region
INRMP	San Diego Bay Integrated Natural Resource Management Plan
SCB	Southern California Bight
SCCWRP	Southern California Coastal Water Research Project
SSC Pacific	Space and Naval Warfare System Center Pacific
SP	Species (abridged name given)
BPTCP	Bay Protection Toxic Cleanup Program
SWRCB	State Water Resources Control Board's
TMDL	Total Maximum Daily Load
WOE	Weight of Evidence

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BACKGROUND

The San Diego Bay is the largest naturally occurring embayment in the Southern California Bight (SCB), consisting of approximately 11,000 acres of marine habitat and home to a diverse aquatic population (San Diego Unified Port, 1990, Integrated Natural Resource Management Plan [INRMP], 2013). The bay is approximately 15 miles long and varies in width from 0.4 to 3.6 miles. Its average depth is approximately 21 feet but ranges from 59 feet (mouth of bay) to ~ 3 feet (south end of bay) (Wang et al., 1998). There is some freshwater contribution to the bay that comes primarily from the Sweetwater and Otay Rivers that feed into the south end of the bay and from various inputs related to surface runoff. However, freshwater input is low for 9 months of the year (INRMP, 2013). Salinity levels at the mouth of the bay are similar to the nearby ocean (31.2 to 31.4 practical salinity units [psu]) (Largier, Hollibaugh, and Smith, 1997). However, the south bay region may exhibit hypersaline conditions during the summer and more estuarine conditions (as low as 22 psu) following heavy winter rains (Largier, 1997). Due to the depth gradient and air temperature changes in this region, water temperatures are seasonal, with the highest temperature occurring in July and August and the lowest in January and February (Smith, 1972). Temperature ranges from a 1993 survey report indicated the warmest temperature at 84.7 °F (south bay in July and August) to 59.2 °F north of Coronado Bridge in January (Lapota et al., 1993). As winds in this region are typically mild, circulation in the San Diego Bay is primarily related to tidal exchange (Wang et al., 1998). Tidal patterns consist of two highs and two lows each day and range from 5.6 feet, with an extreme high of 9.8 feet (Largier, 1997). Overall, a combination of circulation of ocean currents outside the bay and the ebb and flood of tides within the bay dictate access and transport of organisms into the bay along with influencing mixing, dispersion of pollutants, maintaining water quality for marine life, and moderating water temperature (INRMP, 2013; Largier, 1995; Chadwick 1997).

The San Diego Bay is one of California's five major ports and is an important hub for industry and commerce, having ties to international shipping trade (i.e., automotive and various commodities) with the two main cargo facilities located south of Coronado bridge. It also is home to two cruise ship operations and the U.S. Pacific Fleet. As a result, San Diego Bay has and always will coexist with military and other non-cargo maritime activities (San Diego Unified Port District, 1990). At the same time, the bay is home to a diverse population of fish and wildlife with several migrant species using the bay to breed, raise young, or as a migratory staging area (INRMP, 2013). As a result of these diverse functions, the anthropogenic utilities can be at times at odds with the environmental resources. Thus, the primary goal of the U.S. Navy, San Diego Unified Port authorities, and other stakeholders in developing the INRMP was to "ensure the long-term health, restoration, and protection of San Diego Bay's ecosystem in concert with the bay's economic, naval, navigational, recreational, and fisheries needs" (INRMP, 2013). One of many key objectives under the INRMP is to "Minimize the harmful ecological, economic, and human health impacts of aquatic invasive species in San Diego Bay" (INRMP, 2013).

Many researchers have indicated that the introduction and spread of non-indigenous marine organisms is probably one of the greatest threats to the sustainability of complex marine ecosystems, including the San Diego Bay (Pederson et al., 2005; Cohen et al., 2001; Cohen et al., 2005; Zedler, 1992; Crooks, 1997; INRMP, 2013). These non-native organisms can impact an ecosystem not only through habitat loss and environmental degradation, but also through economic and public health problems (Wilcove et al., 1998; Pimental, Lach, Zuniga, and Morrison, 2000; Cohen et al., 2005; Ruiz et al., 2000; CDFG, 2006; McCarthy and Khambaty, 1994). For example, some aquatic invasive species have been found to disrupt fisheries and

aquaculture production, clog waterways (CDFG, 2006), compete with natural species altering natural ecosystem, and serve as vectors for parasites and disease that can harm both native species and humans (Wilcove et al., 1998; Ruiz et al., 2000). A strain of cholera never before seen in the United States, for example, was believed to have been transported into the Chesapeake Bay in the ballast water of 14 to 15 vessels (Ruiz et al., 2000) while toxic red tide causing dinoflagellates were also believed to have been introduced by ballast water or shellfish imports (Hallegraeff and Bolch, 1991). As the global movement of goods and services continue to grow, the potential threat of non-native species on the San Diego Bay will increase.

Several studies show that the number and extent of non-native species is correlated to commercial shipping transporting these exotics in ballast water and as hull fouling (Cohen and Carlton, 1995, 1998; Cohen et al., 2005; Ruiz et al., 2000). Non-native species have also been shown to preferentially attach to artificial man-made structures (Lambert and Lambert, 2003; Chapman and Carlson, 1991; Pederson et al., 2005). As a major industrial and recreational port, San Diego Bay has many potential vector sources, including numerous ships, dry docks, navigation buoys and marina floats, recreational boats, and fisheries and marine aquaculture. In a study by Foss, Pode, Sowby, and Ashe (2007) indicated that the primary introduction vectors for the San Diego Bay most likely were hull/ship fouling, followed by ballast water and aquaculture. As a result, several studies show that non-indigenous species has arrived into the San Diego Bay, and will continue to arrive and spread without early detection and mitigation (Cohen et al., 2005; Lambert and Lambert, 2003; Lambert and Lambert, 1998; Maloney et al., 2007). Managing introduced species requires knowing what is present and identifying potential vector sources.

As a result, one of the listed implementation goals of the 2013 INRMP is to conduct research on invertebrates, including density, abundance, diversity, and critical function, and to better understand proportions abundance of invasive species, habitat alterations, and invasive role in the food chain. In response to these goals, Navy Region Southwest (NRSW) was interested in conducting a field study to establish an ecological index of existing benthic conditions within San Diego Bay's marine environment. The focus of this index survey was to conduct a rapid assessment early detection survey (RAS) of San Diego Bay to identify and catalog native and invasive species. While there are several forms of surveys, a proposed methodology that several researchers use is a RAS approach (Cohen et al., 1998, 2001, and 2003; Pederson et al., 2005). A RAS survey entails bringing a team of taxonomic experts to a specific location to sample 15 to 20 sites over 5 days (Cohen, 2004). It is a qualitative approach of visual searches within a fixed area and/or time frame focused on identification of native, non-native, and cryptogenic species, expansion on data collected in past surveys, assessment of invasion status, and documentation of new introductions (Cohen et al., 2005; Pederson et al., 2005). This report describes a RAS approach used to identify native, introduced, and cryptogenic species present as fouling and infaunal communities in multiple natural and artificial habitats within the four hydrographic regions of San Diego Bay. Methodology described in this report is similar to surveys conducted in the Northeast (Pederson et al., 2005), Puget Sound, Washington, and Tillamook Bay, San Francisco (Cohen et al., 2001; Cohen, 2004).

OBJECTIVES/PRIMARY TASKING

The overall objective for this project was to conduct a rapid assessment early detection survey of the San Diego Bay to identify native species and characterize presence, including identification of any invasive species. This overall goal was broken down into four key objectives:

1. To understand and summarize historic data on species distribution, including presence of exotic species, within the four hydrographic regions of the San Diego Bay. To that end the following tasking was executed:
 - a. Currently available historic benthic data were evaluated along with survey data. The data were evaluated to determine species distribution, and relative abundance within the four hydrographic regions. Evaluation included graphical/mapping and data comparisons.
2. To plan and execute a Rapid Assessment Survey (5 days) using a random sampling strategy to identify species in each of the four hydrographic regions of the bay. To that end, the following tasking was executed:
 - a. A comprehensive study workplan was developed that included specific sampling locations, a sampling plan, and taxonomic expertise was identified and submitted and approved by NAVFAC-SW in June 2011 (see Appendix A). The workplan includes both an initial categorical exclusion (CATEX) approval for settling plate deployment in June 2010, and an amendment to the work plan for the RAS specimen collections in June 2011 (See Appendix B).
 - b. A minimum of four sampling sites were identified within each of the four hydrographic regions, and 16 settling plates were deployed 12 months prior to conducting the rapid survey.
 - c. A rapid assessment survey (5 days) was conducted by a team of five taxonomic experts (predominantly in the lab), along with a team of SPAWAR Systems Center Pacific (SSC Pacific) scientists and volunteer/student help to survey the 16 identified sites within the four hydrographic regions of the San Diego Bay. Sampling was conducted during low tide and live and/or fresh samples were brought back to the laboratory at SSC-Pacific for examination and identification by the team of taxonomic experts.
3. To assess feasibility of use of DNA barcoding as a tool for augmenting species identification in a rapid assessment platform. The following tasking was executed:
 - a. A subset of sample vouchers collected in the field and stored in 95% ETOH (alcohol), and were submitted for DNA barcoding analysis.
 - b. DNA species identification analysis was then compared to taxonomic expert identification to assess sensitivity and accuracy, as it compares to expert taxonomist identification.
4. To provide a summary of data relative to the four hydrographic regions, including species distribution and abundance relative to Navy facilities.

METHODOLOGY

This methodology section is divided into seven main subsections: (A) development of a historic background profile, (B) identification of sampling locations, (C) settling plate study, (D) rapid assessment survey, (E) grain-size analysis, (F) DNA barcoding, and (G) statistical analysis-community metrics. General methodologies for each subsection are provided below.

A comprehensive study work plan was developed for the field aspects of this study. The work plan included information on sampling locations, a sampling plan, and general methodologies. This was vetted through Naval Facilities Engineering Command (NAVFAC)-Southwest prior to executing these study components, with final revision approval for rapid assessment survey component given in June 2011 (See Appendix A). Thus, an abbreviated version of general methodology will be provided for the field collection aspects of this effort here. For a more detailed methods description, please refer to Appendix A.

A. Development of a Historic Background Profile

The historical background profile of benthic infaunal data in San Diego Bay was developed by reviewing the literature, collection of records, and unpublished benthic infaunal data from San Diego Bay. These historical data were used to assess the current status of benthic species in San Diego Bay and to compare the historical data in relation to the results from the current Marine Ecological Index Study within the four hydrographic regions of San Diego Bay.

Biological community metric data (e.g., abundance, total numbers of taxa, Shannon-Wiener Diversity Index, and the Benthic Response Index (BRI)) were queried from each of the data sets obtained from historical studies. The principal goals in developing a historical background profile were: (1) to provide a synopsis of the total species abundance and types (species) of benthic organisms found in San Diego Bay over the past 15 years, and (2) evaluate the current results in context with the historical data. While the primary goal was to provide an overview of the relative abundance and diversity of species found in San Diego Bay, other biological community metrics, such as Shannon-Wiener Diversity Index and the BRI were reported as well.

B. San Diego Bay Sampling Locations

Based on hydrodynamic factors and previously identified hydrographic regions, the bay was divided into four sampling regions: marine, thermal, seasonally hypersaline, and estuarine. An estimated boundary of these four regions based on prior studies (Largier, 1996; Largier, Hollibaugh, and Smith, 1997; INRMP, 2013) is shown in Figure 30. This study focused specifically on defining these four areas as distinct hydrographic regions. A map of the four regions is in Appendix A. Four or more sample locations were selected in each of the four regions. Sampling location selection was based on sediment characteristics (SCCWRP, 2013; SSC Pacific, 2009; SSC San Diego, 2007; U.S. Navy, 2005), proximity to naval facilities, and also consider the range of sediment types within the four regions. Within each hydrographic regions, various sediment types/mental conditions, and land use. Predominant habitat types included intertidal shoreline structures, intertidal mudflats, and benthic habitat within sub-tidal regions. Sampling locations were selected in part to include the various habitat types. Sample locations relating to hydrographic regions are provided in Figure 2 through Figure 5.



Figure 1. San Diego Bay with four management regions identified.



Figure 2. Marine Region: study sites S1–S5.



Figure 3. Thermal Region: study sites S6–S9.



Figure 4. Hypersaline Region: study sites S10–S14. Figure 5. Estuarine Region: study sites S15–S18.

To ensure sufficient coverage of the diversity of communities within the San Diego Bay 18 prospective sites (4 to 5 within each region) were initially identified for sampling. Sampling from each of the locations then involved two primary elements, a settling plate study and a 5-day rapid assessment survey, which are described below. Two of the prospective sites (S13 and S15) were not used during the study due to availability of the locations. Station S13 is the nesting area for California Least Terns and the location was off limits, and S15 was located within the Chula Vista Nature Reserve and unavailable.

C. Settling Plate Study and Deployment

Settling plates. Settling plates measured 25 x 25 cm and consisted of either a wood or acrylic. To allow for the varying conditions of the different sampling locations, two types of settling plate setups were assembled, a floating plate and a line tie-off plate setup. A minimum of two settling plates (wood and acrylic) were deployed at each sampling location. As some species show preferential recruitment to different substrates, two settling plate materials were chosen (Marsden and Lansky, 2000; Field, Glassom, and Bythell, 2007; Hoover and Purcell, 2009).

1. **Floating setup:** For the sampling locations with no artificial shoreline structures present and/or available to tie off to, a floating setup was constructed. The floating setup consisted of two settling plates (one wood and one acrylic) mounted to a PVC frame, a sealed PVC tube for buoyancy, a concrete block to weigh the structure down, and a retrieval line subsurface floating weight (Figure 6). Figure 7 shows how it would look once deployed.
2. **Line tie-off set-up:** For those sampling locations where artificial shoreline structures were available to attach the settling plates to, a total of four settling plates (two wood and two acrylic, one each at the surface and the bottom) were attached to a nylon rope with weight attached to each line (Figure 8). The surface deployed plates were within half a meter of the surface during low tide, and bottom deployed plates were within 1 m of the bottom. For this study, “artificial shoreline structures” were predominantly piers. Example of deployment is provided in Figure 9.

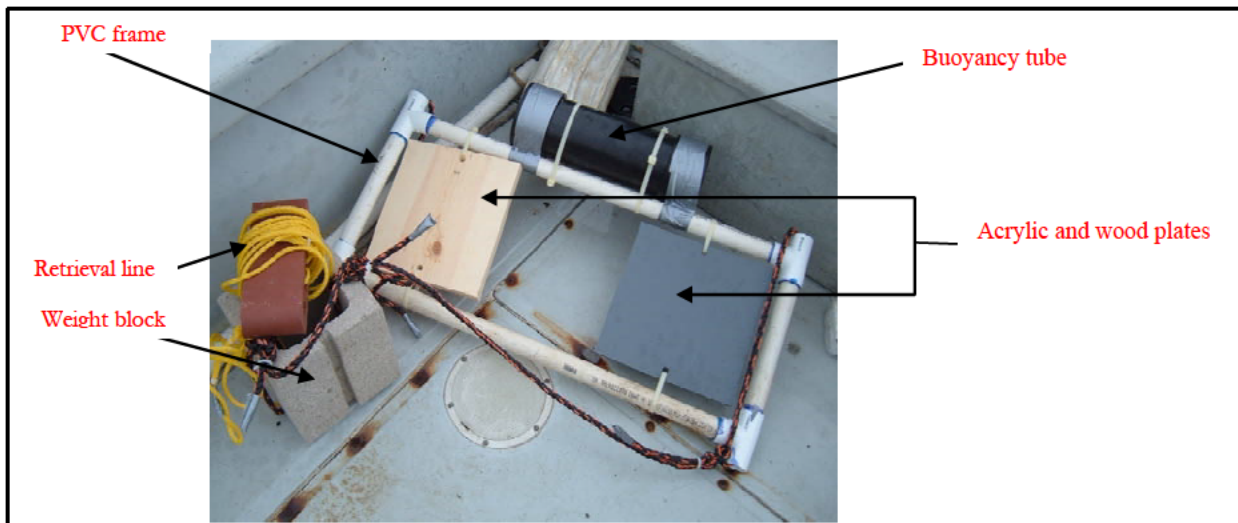


Figure 6. Floating settling plate set-up.

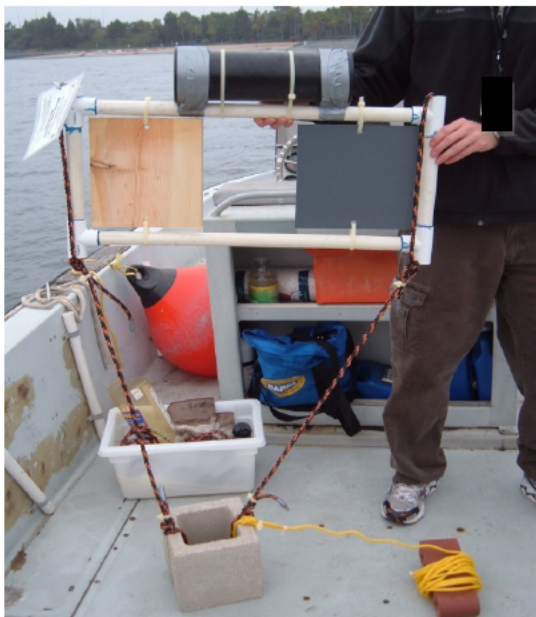


Figure 7. Floating settling plate deployment configuration.

This image is of a floating set-up just prior to deployment. Note that the buoyancy tube keeps the floating set-up vertical in the water-column while the half-cinder block weights it to the sediment. The lighter brick is attached to a 100-foot line that is launched near the settling plate. The secondary line acts as a grab line to allow for more easy retrieval with a grappling hook. Note the identification tag fastened to the PVC frame. Each settling plate setup contained an identification tag including contact information if it was accidentally retrieved by an outside party.

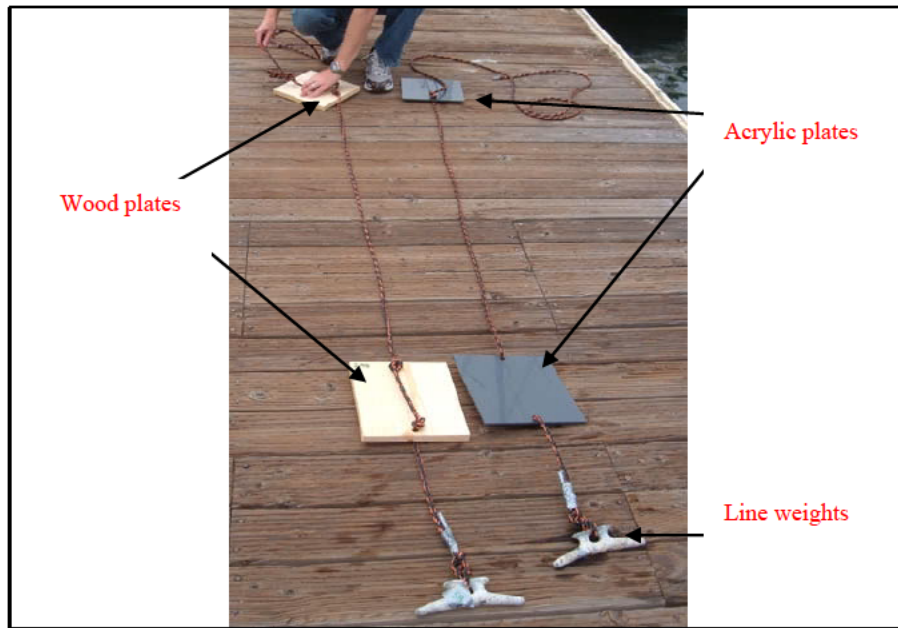


Figure 8. Line tie-off set-up.

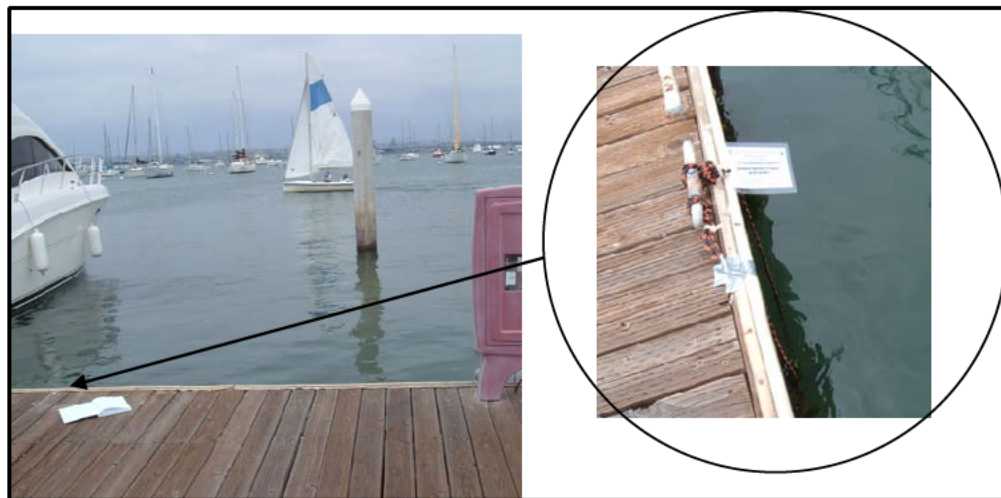


Figure 9. Line tie-off settling plate deployment configuration.

Note: Arrow indicates approximate location of deployment relative to pier. Line is perpendicular to water column indicating sufficient weight loading at bottom of line. Also, note presence of identification placard on line. This particular sampling location was for S14.

3. Deployment of settling plates: Port officials, marinas, or Navy officials granted prior permissions to deploy a set of settling plates at the different sampling locations. A CATEX for settling plate deployment was issued from Navy Region Southwest in June 2010 (Appendix B). In accordance with the CATEX, four or more plates were deployed at each of the study locations that included Naval Submarine Base (SUBASE) beach, SSC Pacific Pier 169, Naval Amphibious Base (NAB) Fishing Pier, Shelter Island Yacht Club, Hilton

Pier (across from Spanish Landing), Sunroad San Diego Marina, Bay View Park (Coronado), San Diego Marriott Hotel and Marina, Tidelands Park (Coronado), Amphibious Base Fuel Pier, Chollas Creek basin, Paletta Creek basin, Fiddler Cove RV campground, Grand Caribe Shoreline Park (Coronado Cays), and Chula Vista harbor pier. Table 1 lists settling plate deployment locations, along with determination of whether they were floating or tie-off set-ups. Note that the settling plates were only deployed at study locations where there was sufficient depth and suitable anchor points. Thus, settling plates for sites S18 were not included because the mud flat habitat at these two locations was too shallow to deploy the plates. Each settling plate was to be deployed a minimum of 6 months prior to the rapid assessment survey.

Table 1. Settling plate deployment locations, including region and type of set-up.

Eco-Region	Site Name	Site ID	Deployment (Hanging/Float)
Marine Region	SUBASE beach	S1	Float
	SSC Pacific Floating Pier	S2	Hanging
	NAB Fishing Pier	S3	Float
	Sun Harbor Marina	S4	Hanging
	Harbor Island West Marina	S5	Hanging
Thermal Region	Sunroad San Diego Marina	S6	Hanging
	Bayview Park Coronado	S7	Float
	San Diego Marriott Hotel and Marina	S8	Hanging
	Tidelands Park Coronado	S9	Float
Seasonally Hypersaline Region	Amphib Base Fuel Pier	S10	Float
	Chollas Creek	S11	Float
	Paleta Creek	S12	Float
	Fiddler's Cove Marina	S14	Hanging
Estuarine Region	Chula Vista Nature Reserve	S15	None deployed
	Grand Caribe Shoreline Park	S16	Float
	Chula Vista Harbor Pier	S17	Hanging
	South Bay Marine Biological Center	S18	None deployed

4. Retrieval of Settling plates: Each set-up of settling plates was collected at a maximum of a 2 to 3 days prior to sorting and identification by the expert taxonomists. For the floating set-up, plates were collected using a grappling hook. Settling plates were retrieved from a boat by dragging a grappling hook attached to a line. Once retrieved, each set-up was placed in a separate cooler filled with seawater and transported back to the laboratory. For the tie-off set-up, they were disconnected from the artificial structure and placed in a cooler of seawater for transportation back to the laboratory. Once back at the laboratory,, each cooler was then placed in a controlled temperature wet-lab with a O₂ line placed in the cooler to maintain oxygenation of the water until such time that each plate was sorted and identified. Voucher samples of invasive and/or novel organisms were collected and stored in 90% ETOH from each plate.

D. Rapid Assessment Survey

The rapid assessment survey consisted of three main parts, benthic sediment sampling, field sampling, and sorting/identification/classification. Study design was based on the Chesapeake Bay and San Francisco Bay estuarine monitoring program (Bowman, Dohner, and Dohner, 1993). Sixteen sites were included in the rapid assessment survey, with sampling and identification taking place over 5 days.. The survey focused primarily on the fouling community and macrofaunal species within the bay.

1. **Benthic Sediment Sampling:** A senior biologist from ABC laboratory and a scientist from SSC Pacific collected benthic sediment samples using a 20-foot Boston whaler and a Van Veen grab. A 0.25-m² Van Veen grab sampler was used to collect sediment samples from 13 sampling sites. Sample sites including location are listed in Table 2. Sample retrieval was unsuccessful at stations S8, S3, S11, and S12. Grab event information was recorded on field data sheets. Three or more grabs were collected from each sampling location to fill the 1-L collection jar. Samples were then sieved through a 1-mm mesh screen. Material retained on the screen was placed for at least 30 min in a relaxant solution of 1-kg MgSO₄ per 20 L of seawater, and then preserved in 90% ETOH and stored at 4 °C until sorting and identification. The near-shore benthic sediment samples were collected July 6–8, 2011.

Table 2. Benthic sediment sampling locations.

REGION	Site Name	Site ID	Latitude	Longitude
Marine Region	SUBASE beach	S1	32.68647	117.235
	SSC Pacific Floating Pier	S2	32.70508	117.23621
	NAB Fishing Pier	S3	32.71003	117.21861
	Sun Harbor Marina	S4	32.72339	117.22503
	Harbor Island West Marina	S5	32.72775	117.2100
Thermal Region	Sunroad San Diego Marina	S6	32.72687	117.1900
	Bayview Park Coronado	S7	32.70216	117.17864
	San Diego Marriott Hotel and Marina	S8	32.70725	117.16557
	Tidelands Park Coronado	S9	32.69064	117.16377
Seasonally Hypersaline Region	Amphib Base Fuel Pier	S10	32.67552	117.16537
	Chollas Creek	S11	32.68736	117.12996
	Paleta Creek	S12	32.67394	117.11649
	Fiddler's Cove Marina	S14	32.65188	117.14472
Estuarine Region	Grand Caribe Shoreline Park	S16	32.62332	117.12831
	Chula Vista Harbor Pier	S17	32.62474	117.10539
	South Bay Marine Biological Center	S18	32.60796	117.11941

2. **Rapid Assessment Survey (RAS):** Field sampling was conducted July 11–15, 2011. Approximately four sites were surveyed per day. Survey time allotted to each location was approximately 1 to 2 h. A determination of sampling location scheduling was developed based on tidal conditions (ideally during low tide) and type of sampling methods to be completed at each location. Figure 10 provides tidal conditions for the week of July 11, 2011. The sampling schedule is provided in Table 3. Two sampling teams of Navy scientists and student support conducted the in-field sampling. Sampling team preparation is described in detail below. The specific sampling methodology for each strategy was based on the habitat(s) found within each sampling site (i.e., intertidal vs. subtidal) within each hydrographic region. A summary of study

sites based on hydrographic region, habitat type, and potential sampling type associated with each site is provided in Table 4. Sampling methodology associated with sampling types is listed in Table 5 and includes line transects/quadrat/sieving, perpendicular beach seine, timed search, and small quadrat/scraping. Detailed descriptions of each sampling protocol is provided in Appendix A. General information about each sampling site including GPS location, habitat description, weather conditions, and tide was collected at each sampling location.

Table 3. Schedule of events for the Rapid Assessment Survey.

M – July 4th <i>No collections</i>	T – July 5th <i>No collections</i>	W – July 6th <i>Sediment Collection:</i> Smugglers Cove S1 SSC S2 NAB Pier S3 Sun Harbor S4 Harbor Island S5 Sun Road S6	R – July 7th <i>Sediment Collection:</i> Bayview Park S7 SD Marriot S8 Tidelands S9 Amphib Fuel Pier S10 Fiddlers Cove S14	F – July 8th <i>Sediment Collection:</i> Grand Caribe S16 Chula Vista Pier S17 SB Marine Biological Center S18
M – July 11th RAS Day 1 Plate collection All staff S1 and S2 Taxonomist (Lab)-Start ID of sediment grabs and two plates	T – July 12th RAS Day 2 Plate collection Team 1: S18 then S17 (plate) Team 2: S16 then S14 (plate) Taxonomist (Lab)-Plates S14-S18 + field collection samples	W – July 13th RAS Day 3 Team 1: S9 and S10 (plate, needs grappling hook) Team 2: S7 and S3 Taxonomist (Lab)-Plates S3, S7-S10 + field	R – July 14th RAS Day 4 Team 1: S5 (plate) Taxonomist (Lab)-Plates S5, S12 + field	F – July 15th RAS Day 5 Final identification.

Table 4. Sampling methodology based on habitat type.

Sampling Methodology	Fixed Fouling	Floating Fouling	Near-shore Benthic	Rip-Rap	Sandy Beach	Mudflat
Line Transect/ Quadrat / Seiving					X	X
Beach Seine					X	X
Van Veen Grab			X			
Timed Search	X	X		X		
Small Quadrat / Scraping	X	X		X		

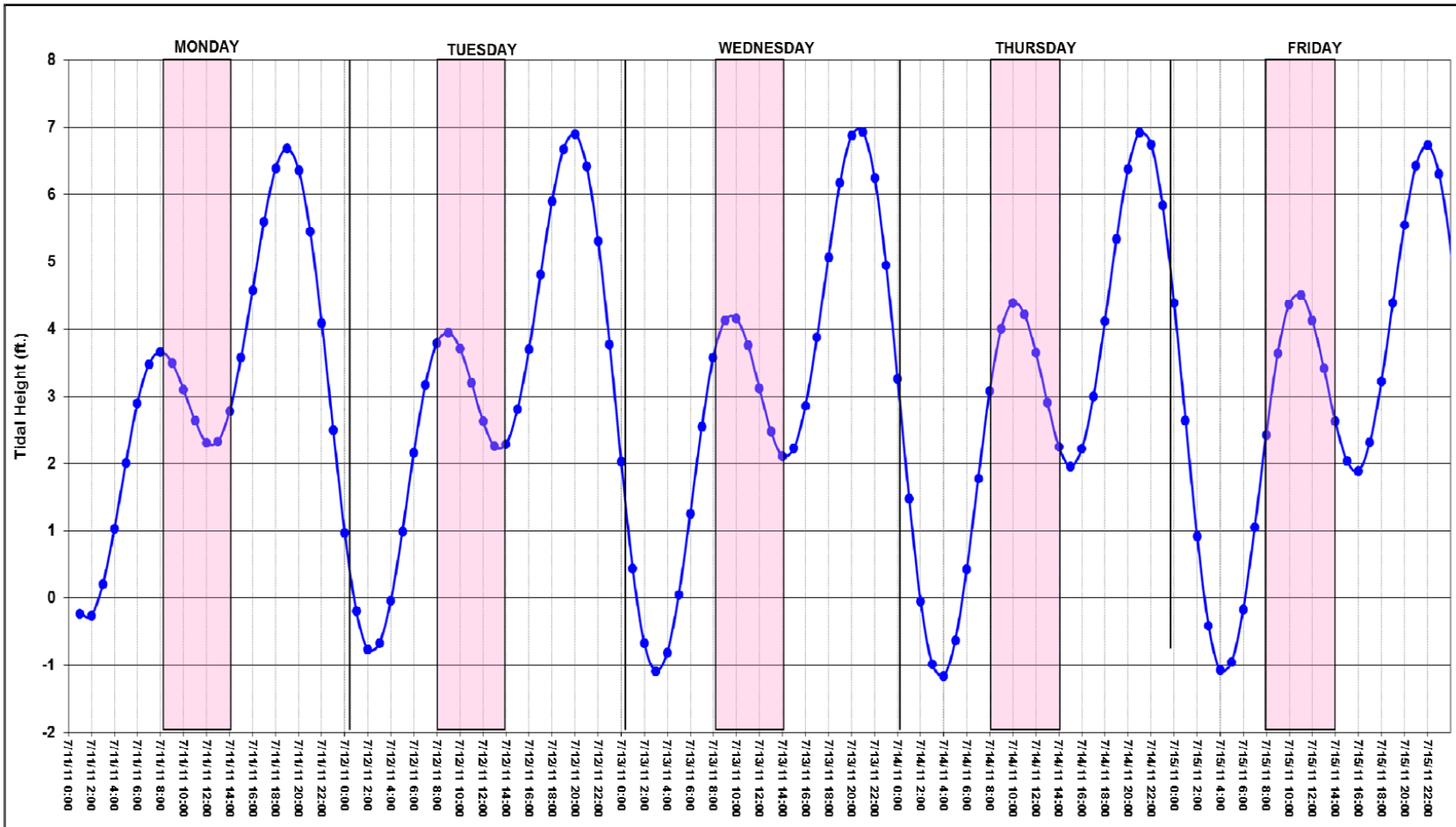


Figure 10. Tidal conditions data obtained from <http://tidesandcurrents.noaa.gov/>.

Table 5. Habitat type for the different sampling sites.

Eco-Region	Site Name	Site ID	Habitat Type					
			Subtidal				Intertidal	
			Float Fouling	Fixed Fouling	Settling Plates	Subtidal Sediment	Hard Substrate	Intertidal Sediment
Marine Region	SUBASE beach	S1			X	X		Sandy beach
	SSC Pacific Floating Pier	S2	X	X	X	X		Sandy beach
	NAB Fishing Pier	S3	X	X	X	X	Rip-rap	Sandy beach
	Sun Harbor Marina	S4	X	X	X	X		
	Harbor Island West Marina	S5			X	X		Sandy beach
Thermal Region	Sunroad San Diego Marina	S6	X	X	X	X		
	Bayview Park Coronado	S7			X	X	Rip-rap	Sandy beach
	San Diego Marriott Hotel and Marina	S8	X	X	X	X		
	Tidelands Park Coronado	S9			X	X	Rip-rap	Sandy beach
Seasonally Hypersaline Region	Amphib Base Fuel Pier	S10	X	X	X	X	Rip-rap	Sandy beach
	Chollas Creek	S11	X	X	X	X		
	Paleta Creek	S12	X	X	X	X		
	Fiddler's Cove Marina	S14			X	X		Sandy beach
Estuarine Region	Grand Caribe Shoreline Park	S16			X	X		Sandy Beach
	Chula Vista Harbor Pier	S17				X	Rip-rap	Sandy Beach
	South Bay Marine Biological Center	S18				X		Mud flat

3. **Sampling Team Preparation, Specimen Collection, and Pre-sorting:** Field sampling efforts were split into two teams to complete the four scheduled locations. Each team received a set of supplies, description of activities for the day, and images of study locations. Each team completed activities on schedule for locations, recorded general information on field datasheets, and collected specimens for further identification and sorting back at the laboratory. Specimens were collected in 1-L pre-cleaned glass jars filled with seawater and transported back to the laboratory within 2 h of collection. Once in the lab collection jars were placed in a temperature-controlled wet-room with O₂ circulated through jars until a relaxant solution of 1 kg MgSO₄ per 20 L of seawater could be added (within 12 h of collection). Organisms from a sample jar were then sorted into taxonomic categories, preserved in 90% ETOH and distributed to experienced taxonomists for species identification and enumeration. A field packet including data sheets, activity schedule/ location, and site images is provided in Appendix C and Appendix A (work plan).
4. **Sorting/Identification/Classification:** A team of Navy scientists and a technician and an experienced senior biologist from ABC Laboratory conducted initial sample separation and sorting by phylogenetic group. Once samples were separated and sorted, expert taxonomists performed species identification, voucher separation, and enumeration. A brief description of sample separation, sorting, and species identification is provided below:
 - a. **Site Sample Separation:** The 1-L jars collected by the two field research teams from each of the sampling methodologies at each location were initially separated into phylogenetic groups by placing the entire sample contents into a large, flat photographic tray. The relaxant solution (MgSO₄) was poured off the sample and a small amount of 90% ETOH was added. Samples were then gently agitated until equally distributed across the tray. The sample was then sorted by standard sorting procedure using with fine forceps into petri dishes for each major phyla.

E. Grain Size Analysis

Sediment samples were collected from each of the four hydrographic regions and a grain size analysis was conducted. Grain size analysis can be broken down into sediment collection methods and analysis methods, which are described below.

1. Sediment collection: Sediment samples were collected at at least two locations in each of the four management regions. Nine sediment samples were collected for grain size analysis using a 0.05-m² Young-modified Van Veen grab. Between sites, the grab was rinsed with seawater. The top 5 cm were subsampled and placed in a clean, labeled zip-top bag for grain size analysis. Sample locations were the same as for specimen collection.
2. Sediment analysis: A sieving/sedimentation technique similar to ASTM D422 (ASTM, 2007) was used to perform grain size analysis of the sediments. After determining the dry=weight percentage for each sample, a wet sieving technique was used to isolate sand fractions according to the Wentworth size classes (Wentworth, 1922). Sieve mesh sizes used were 1.0, 0.50, 0.25, 0.125, and 0.063 mm. Analysis of the fines fraction (< 63 µm) was performed using gravimetric settling according to the Stokes relationship (Stokes, 1847) to identify the particle size diameter and a dedicated hydrometer (152H) to quantify the particle concentration. The Stokes Law relates velocity of settling particles to its effective diameter and the 152H hydrometer determines particle load based on the buoyancy of the measured sample. The particle distribution is reported as percent dry mass sample per size class. For the samples containing low fines content, only sieving was performed, and the less than 63 µm fraction reported as % fines. For samples with < 63 µm, the ASTM 1998 method was used that combined the mechanical sieving and hydrometer method. The sieves were used to quantify particle sizes in the sand range (> 63 µm) and larger, and a sedimentation-hydrometer technique was used to quantify the silt and clay size ranges (< 63 µm).

F. DNA Barcoding Analysis

Twenty samples were submitted for DNA barcode sequencing to the Smithsonian Institute, USA. Samples were selected from voucher samples collected during the Rapid Survey (July 11–15, 2011) and at the recommendation of taxonomists (per communication with Leslie Harris, Tony Phillips and John Ljubenkov, March, 2012).

All samples were put in 90 to 95% ethanol within a few hours of collection on the dates listed in Table 6.

1. Sample extraction and amplification: The Smithsonian Institute is part of the global Consortium for the Barcode of Life (CBOL) which develops and employs standardized protocols for analyzing specimens. These standard processes can be found at <http://barcoding.si.edu/dnabarcoding.htm>. In short, methodology for DNA extraction and amplification is as follows: Genomic DNA was extracted in accordance to the manufacturer's protocol using the DNeasy 96 Blood and Tissue kit (Qiagen). DNA integrity of each specimen was determined by first amplifying the subunit nuclear ribosomal RNA 18S. Universal primers used for amplification were 18S: (5'-AACCTGGTTGATCCTGCCAGT-3') and 18S: (5'-TGATCCTTCTGCAGGTTACCTAC-3') (Medlin, Elwood, and Sogin, 1988). Once DNA integrity was determined primers LCO1490 (5'-GGTCAACAAATCAT AAAGATATTGG-3') and HCO2198 (5'-TAAACTTCAGGGTGA CCAAAAAATCA-3') for the mitochondrial cytochrome oxidase subunit I gene (COI) were used to amplify a 658 base-pair (bp) fragment (Folmer et

al., 1994). A standard polymerase chain reaction (PCR) included 2° µl of DNA extract, 10 pM of each PCR primer and Ready-To-Go PCR beads (Amersham Pharmacia Biotech). Standard PCR conditions consisted of 1 min at 94 °C followed by 5 cycles of 40 s at 94 °C, 40 s at 45 °C, 60 s at 72 °C, followed by 35 cycles of 40 s at 94 °C, 40 s at 51 °C, 60 s at 72 °C, followed by 5 min at 72 °C. Successful amplification product was purified using QIAquick PCR Purification Kit (Qiagen). The PCR product was run on an agarose gel (2%) containing EtBr and the target fragment was excised from the gel and purified using QIAquick Gel Extraction Kit (Qiagen). Automated sequencing was performed directly on purified PCR products using ABI BigDye terminator V3.1. Sequence reactions were purified using Millipore 96-well plates loaded with Sephadex G-50 and run on an ABI 3130xl genetic analyzer (Applied Biosystems). Products were sequenced in both directions using LCO1490 and HCO2198. Sequence results were then analyzed using NCBI Blast® Search, Barcode of Life Data Systems (BOLD) database search, and ClustalW Multiple Sequence Alignment Software to identify nearest sequence matches.

Table 6. Voucher samples submitted for DNA barcode sequencing,

DNA SELECTIONS					
Sample ID	Phylum	Species	Date of Collection	Region	Notes
EIS-GL-0027	Chordata	Ascidia ceratodes	13-Jul-2011	Marine	native
EIS-JL-0185	Gastropoda	Diaulula sandiegensis	13-Jul-2011	Marine	
EIS-TP-0025	Arth	Amphideutopus oculatus	15-Jul-2011	Marine	4 found
EIS-TP-0065	Arth	Jassa slatteryi	15-Jul-2011	Marine	2 found
EIS-GL-0021	Chordata	Aplidium californicum (?)	13-Jul-2011	Thermal	native
EIS-JL-0014	Pelecypoda	Lyonsia californica	16-Jul-2011	Thermal	
EIS-LH-0137	ANN	Syllis nipponica	13-Jul-2011	Thermal	
EIS-LH-0141	ANN	Platynereis bicanaliculata	13-Jul-2011	Thermal	juvenile
EIS-TP-0012	Arth	Neotrypaea californiensis	16-Jul-2011	Thermal	1 found
EIS-GL-0014	Chordata	Botrylloides diegensis	12-Jul-2011	Seasonally Hypersaline	nonindigenous
EIS-JL-0040	Bryozoa	Smittoidea prolifica	12-Jul-2011	Seasonally Hypersaline	
EIS-JL-0138	Cephalopoda	Octopus bimaculatus/bimaculooides	12-Jul-2011	Seasonally Hypersaline	impossible to tell species this young
EIS-JL-0230	Pelecypoda	Chione undatella	14-Jul-2011	Seasonally Hypersaline	
EIS-LH-0061	ANN	Nicolea sp A Harris	12-Jul-2011	Seasonally Hypersaline	typically the dominant terebellid in southern California harbor fouling; apparently undescribed & introduced
EIS-TP-0016	Pyc	Rhynchothorax philosammum	12-Jul-2011	Seasonally Hypersaline	1 found
EIS-GL-0001	Chordata	Botryllus schlosseri	11-Jul-2011	Estuarine	nonindigenous; on Styela clava
EIS-GL-0008	Chordata	Molgula ficus	11-Jul-2011	Estuarine	nonindigenous
EIS-JL-0038	Bryozoa	Scrupocellaria sp	11-Jul-2011	Estuarine	
EIS-LH-0002	ANN	Harmothoe imbricata complex	11-Jul-2011	Estuarine	
EIS-TP-0043	Arth	Ampithoe velida	16-Jul-2011	Estuarine	1 found

G. Community Metrics

Species richness, the Shannon Weiner Diversity Index, and similarity indices were calculated for each station. The Shannon Weiner Diversity Index was calculated using Shannon Weiner Diversity Index (H'):

$$H' = - \sum_{i=1}^s p_i \ln p_i$$

where H' is the species diversity index, s is the number of species, and p_i is the proportion of individuals of each species belonging to the i th species of the total number of individuals. The diversity index was calculated individually for each sampling method/habitat used (e.g., infauna, intertidal, beach seine, fouling community).

Additionally, to compare the various stations, similarity indices were calculated in pairs for all the stations within the study. Similarity indices were calculated using the following equation:

$$SI = \frac{2 \sum_{i=1}^{n_i} \min(p_{ia}, p_{ib})}{\sum_{i=1}^{n_i} p_{ia} + p_{ib}}$$

where SI = The calculated similarity index, n = the total number of species, p_i = the proportion of a particular species abundance to the total species abundance, p_{ia} = the proportion of a particular species abundance to the total species abundance for set (A) of data sets (A) and (B), and p_{ib} = the proportion of a particular species abundance to the total species abundance for set (B) of data sets (A) and (B), where data sets (A) and (B) would represent the two stations being compared. Similarity indices were calculated with the total species list for each station.

BACKGROUND HISTORICAL DATA SURVEY

The objective of this historical data survey is to review the literature, collection of records, and unpublished benthic infaunal data from San Diego Bay to assess the status of benthic species in San Diego Bay. Additionally, these historical data were compared to the results from the current San Diego Bay Marine Ecological Index Study within the four hydrographic regions discussed in this report.

For nearly two decades, various environmental studies were conducted in San Diego Bay to evaluate the impairment to health of benthic organisms—a primary beneficial use concern—using multiple measures of sediment quality, including chemistry, toxicity, benthic community composition, and bioaccumulation. While a large number of studies were performed over the past 20 years, only those studies from which benthic-related data could be readily accessed have been included in this historical review.

Summary of Historical Surveys

A. Southern California Coastal Water Research Project (SCCWRP)

Perhaps the most extensive of all the environmental monitoring programs in southern California is the Southern California Coastal Water Research Project (SCCWRP). This program is an environmental monitoring program whose aim is to assess the potential effects of human activities on southern California's coastal ocean. The Coastal Ecology component of the Southern California Bight (Bight¹) regional monitoring effort seeks to determine the spatial extent of contaminant accumulation in marine sediments and assess the effects of this contamination on living marine resources. Coastal Ecology was the original component of the Bight program, initially implemented as the 1994 Regional Monitoring Pilot Project. Coastal Ecology regional monitoring

¹ A bight is defined as a bend in the coastline, and the Southern California Bight (SCB) is the 700 km (400 miles) of recessed coastline from Point Conception, in Santa Barbara County, California, to Cabo Colnett, just south of Ensenada, Mexico. Here, subtropical waters flow north close to the shore, while subarctic waters flow south offshore. This unique ocean circulation pattern creates a biological transition zone that supports approximately 500 marine fish species and more than 5,000 invertebrate species (SCCWRP, 2013).

is now conducted every 5 years. The most recent completed effort was initiated in the summer of 2008. More than 60 organizations have participated as partners in the Coastal Ecology portion of SCCWRP's bight-wide regional monitoring efforts (SCCWRP, 2013). A large amount of data is publicly available from these studies (Table 7).

Benthic infaunal data from the Bight '98 and Bight '03 studies were compiled for this historical review. While data from the 1994 pilot study has been published, it is not easily accessible and the data from the 2008 study has yet to be released. These data are presented along with data from other San Diego Bay studies, including the SSC Pacific EIS Study, in subsequent sections.

B. SSC Pacific Environmental Studies

Various marine environmental studies were conducted in San Diego Bay by scientists at SSC Pacific, with a focus primarily on Navy areas of interest. The three studies described below all had benthic assessment components.

1. Sediment Assessment Study for the Mouths of Chollas and Paleta Creek, San Diego (2001): This report describes results of an investigation into the potential impairment of beneficial uses at the mouths of Chollas Creek and Paleta Creek (also known as Seventh Street Channel) where they enter San Diego Bay. The goal of the investigation was to develop a comprehensive weight of evidence (WOE) evaluation of the impairment of aquatic life beneficial uses as well as a screening level evaluation of wildlife and human health beneficial uses at both sites. The investigation was prompted by the designation of these two sites by the Regional Water Quality Control Board, San Diego Region (SDRWQCB) as a Toxic Hot Spot based on chemical contamination of sediments and aquatic life impacts. Additionally, the SDRWQCB also initiated development of a Total Maximum Daily Load (TMDL) assessment to address potential source reduction requirements at these two sites because of benthic community degradation and sediment toxicity. This investigation was a joint effort by the SDRWQCB, NRSW, and the City of San Diego (SCCWRP and SSC San Diego, 2005).
2. Sediment Site Assessment Study, Submarine Base San Diego (2004): This report details an investigation of the nature and extent of impaired San Diego Bay sediments adjacent to Submarine Base San Diego. The investigation was prompted by the designation of the site by the San Diego Regional Water Quality Control Board as having contaminated sediments and aquatic life impacts. The study was conducted by personnel from the Space and Naval Warfare Systems Center San Diego for Navy Region Southwest. The primary beneficial use concern is impairment to health of benthic organisms (Aquatic Life Beneficial Use), focusing on invertebrates such as crustaceans, polychaetes and molluscs that live in and on the sediment. There is also concern for potential exposure and impact to fish and birds that prey on these benthic organisms (Aquatic Dependent Wildlife Beneficial Use) as well as potential exposure to humans that may occur through fishing activities (Human Health Beneficial Use). The conceptual approach taken in this study was to use multiple measures of sediment quality, including chemistry, toxicity, benthic community composition, and bioaccumulation to assess the potential for impairment to each of these three beneficial uses (SSC San Diego [now, SSC Pacific], 2007).

Table 7. Southern California Coastal Water Research Project (SCCWRP) Bight Studies: Available Data.

Southern California Coastal Water Research Project (SCCWRP) Bight Studies			Data Available (http://www.sccwrp.org/Data/SearchAndMapData/DataCatalog.aspx)				
Study	Year	Purpose	Category	Metadata	Map	Zip	Excel
1994 Pilot Project Survey (Bight)	1994	A 261 site survey conducted on the continental shelf of the the Southern California Bight between Point Conception and the Mexican border. The data include, metadata, sediment chemistry, infaunal biomass and abundance, and trawl fish and invertebrate data collected for each time period	Fish Abundance	X	X	N/A	X
			Fish Biomass	X	X	N/A	X
			Infaunal Abundance	X	X	N/A	X
			Invertebrate Abundance	X	X	N/A	X
			Invertebrate Biomass	X	X	N/A	X
			Trawl Data	N/A	N/A	X	N/A
			Sediment Chemistry	X	X	N/A	X
			Toxicity	N/A	N/A	X	N/A
			Benthic Data	N/A	N/A	X	N/A
			Chemistry Data	N/A	N/A	X	N/A
			CTD Data	N/A	N/A	X	N/A
Fish Tissue Chemistry Data	N/A	N/A	X	N/A			
Bight 1998 Survey	1998	A survey conducted on the continental shelf of the the Southern California Bight during 1998. The data include metadata, sediment chemistry, sediment toxicity, biomarker, infaunal biomass and abundance, and trawl fish and invertebrate data.	Fish Abundance	X	X	N/A	X
			Fish Biomass	X	X	N/A	X
			Infaunal Abundance	X	X	N/A	X
			Invertebrate Abundance	X	X	N/A	X
			Invertebrate Biomass	X	X	N/A	X
			Trawl Data	N/A	N/A	X	N/A
			Sediment Chemistry	X	X	N/A	X
			Toxicity	N/A	N/A	X	N/A
			Storm Event Shoreline Microbiology Project	N/A	N/A	X	N/A
			Shoreline Microbiology Survey Winter 1999	N/A	N/A	X	N/A
			Shoreline Microbiology Survey Summer 1998	N/A	N/A	X	N/A
			Chemistry Data	N/A	N/A	X	N/A
			Benthic Data	N/A	N/A	X	N/A
Biomarker Data	N/A	N/A	X	N/A			
Bight 2003 Survey	2003	This is the third regional survey of the continental shelf of the Southern California Bight since 1994. The data include metadata, sediment chemistry, sediment toxicity, infaunal biomass and abundance, and trawl fish and invertebrate data.	Fish Abundance	X	X	X	X
			Fish Biomass	X	X	X	X
			Infaunal Abundance	X	X	X	X
			Invertebrate Abundance	X	X	X	X
			Invertebrate Biomass	X	X	X	X
			Trawl Debris	X	X	X	X
			Sediment Chemistry	X	X	X	X
Toxicity	N/A	N/A	X	N/A			
Bight 2008 Survey	2008	This is the fourth regional survey of the continental shelf of the Southern California Bight since 2003. The data include metadata, sediment chemistry, sediment toxicity, infaunal biomass and abundance, and trawl fish and invertebrate data.	Data not available for download. Reports are available.				

3. Sediment Site Assessment Study, Naval Station (NAVSTA) San Diego (2008): This report describes results of an investigation into the potential impairment of beneficial uses to San Diego Bay sediments in the middle pier area of Naval Base San Diego (NBSD). The investigation was a Phase I Total Maximum Daily Load (TMDL) evaluation of the magnitude and spatial extent of sediment impairments to sensitive beneficial uses. The goal of the investigation was to develop a comprehensive weight of evidence (WOE) evaluation of impairment to aquatic life beneficial uses as well as a screening level evaluation of wildlife and human health beneficial uses. The investigation was conducted in response to a request from the San Diego Regional Water Quality Control Board (SDRWQCB) to evaluate the site because of its inclusion under the California State Water Resources Control Board's (SWRCB) Clean Water Act Section 303(d) list. Specifically, Piers 2 through 7, an area of approximately 103 acres, was listed as a medium priority TMDL site for benthic community effects and sediment toxicity. Two sites, Piers 3 and 4 and Piers 5 and 6, were originally listed as a moderate priority Sites of Concern under the State of California's Bay Protection Toxic Cleanup Program (BPTCP) for aquatic life impacts (SSC Pacific, 2009).

C. Data Summary and Analysis Overview of Benthic Community Data

Biological community metric data (e.g., abundance, total numbers of taxa, Shannon-Wiener Diversity, and the BRI) were queried from each of the data sets associated with the studies discussed above. To maintain consistency in comparing data sets, only data collected from Van Veen sediment grab samples were included. The principal goals of this historical review are to (1) provide a synopsis of the total species abundance and types (species) of benthic organisms that have been found in San Diego Bay over the past 15 years, and (2) evaluate the EIS Study results in context with the historical data. While the primary goal of this historical data summary is to provide an overview of the relative abundance and diversity of species found in San Diego Bay, other biological community metrics such as Shannon-Wiener Diversity and the BRI are reported as well.

D. Sample Locations

In all, 170 sample locations were identified from the six studies, including the samples collected as part of the EIS study. As can be seen in Figure 11–15, benthic community data were collected at locations throughout most of San Diego Bay.

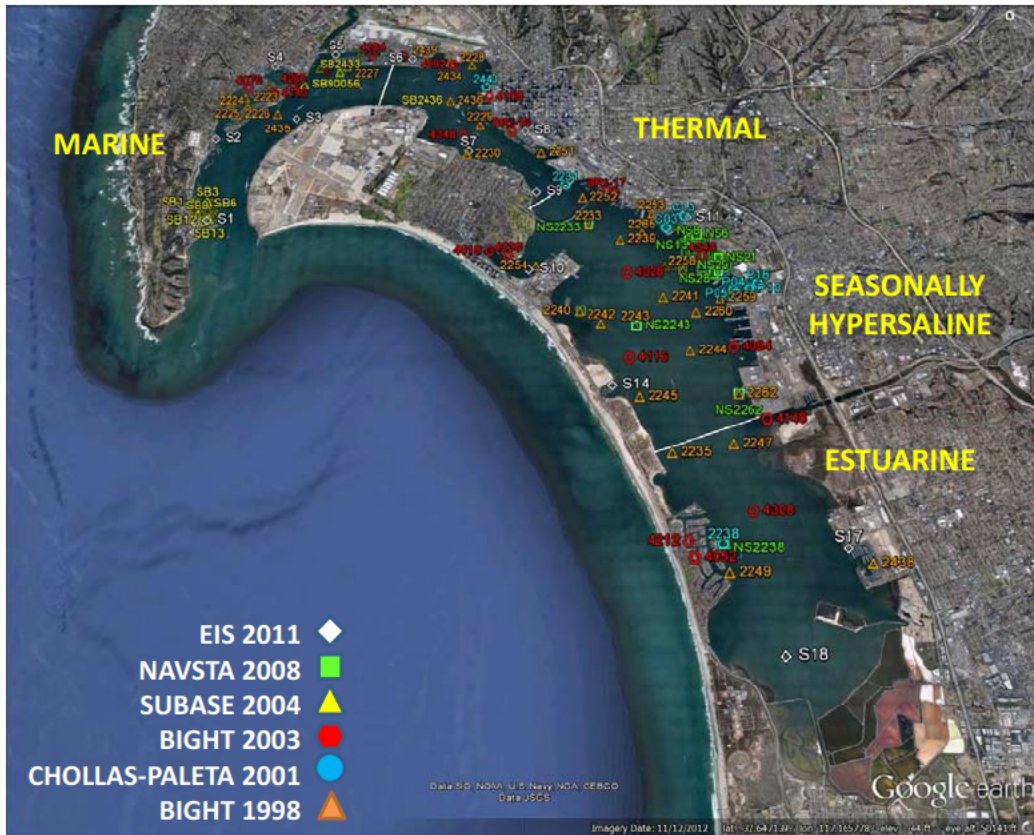


Figure 11. Sampling locations from six studies conducted 1998–2011 in San Diego Bay showing four hydrographic regions.



Figure 12. Sampling locations from six studies conducted 1998–2011 in San Diego Bay within the Marine Hydrographic Region.



Figure 13. Sampling locations from six studies conducted 1998–2011 in San Diego Bay within the Thermal Hydrographic Region.

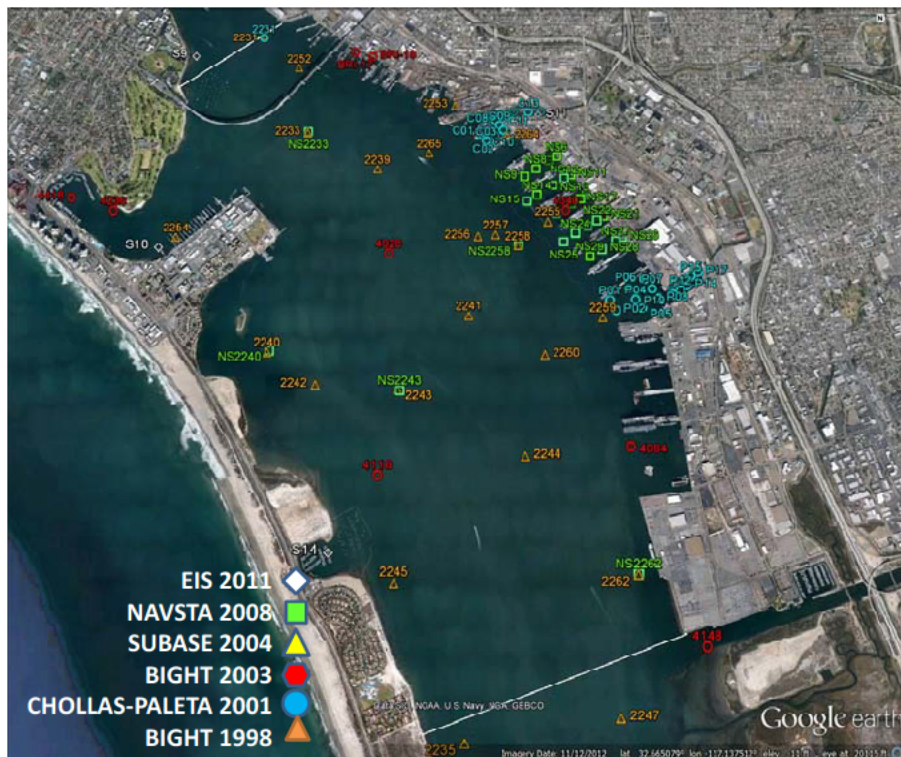


Figure 14 Sampling locations from six studies conducted 1998–2011 in San Diego Bay within the seasonally Hypersaline Hydrographic Region.



Figure 15. Sampling locations from six studies conducted 1998–2011 in San Diego Bay within the Estuarine Hydrographic Region.

E. Benthic Community Data

The benthic abundance² for each location sampled from each study was tabulated and segregated within each of the four hydrographic regions (Tables 9–13). Benthic abundance ranged from less than 10 organisms counted per sample to over 5,000 organisms per sample at two different locations (Figure 16).

More than 43% of the sampling locations (73 out of 170) yielded abundance counts in the range of 0–500 organisms/sample, followed by 29% in the range of 500–1,000 organisms/sample, 21% in the 1,000–2,000 organisms/sample range, and the remaining 7% falling within 2,000 to greater than 5,000 organisms/sample. Two samples yielded very high organism counts (Sediment Site Assessment Study, Submarine Base San Diego (2004), Subbase14 = 5,895 and Sediment Assessment Study for the Mouths of Chollas and Paleta Creek, San Diego (2001), Chollas-Paleta2231 = 6,343, a reference location). Abundance data from the samples collected as part of the EIS Study ranged from 3–182 organisms/sample (Table). Because the EIS Study locations were not co-located specifically to past sampling locations, a direct comparison of abundance data over time cannot be made. However, in comparing data from EIS study locations that are near sample locations from past studies, it appears that the abundance data from the EIS study samples is generally lower overall when compared to the historic data (Table 14). For example, EIS station S1 had an abundance count of 27 organisms, whereas Subbase Station SB12 had a count of 532

² Benthic Abundance - The simplest measure of population composition is the total numbers of organisms (abundance) collected per sampling effort.

organisms. Similar results are observed for all of the samples where data are available. However, because of the limited amount of data available, the length of time between which samples were collected, seasonal variations, lack of co-located sampling locations, and potential differences in sample collection and handling, it is difficult to draw any concrete conclusions between this study and previous studies. The EIS abundance data appear to trend lower.

Additional Benthic Community metrics such as taxa, Shannon-Wiener Diversity, and Benthic Response Index are also shown in Tables 8–13. While it is beyond the scope of this historical review to provide a discussion of all of the results from each study, the data are presented to provide the reader with an overview of benthic community health throughout San Diego Bay over the past 15 years.

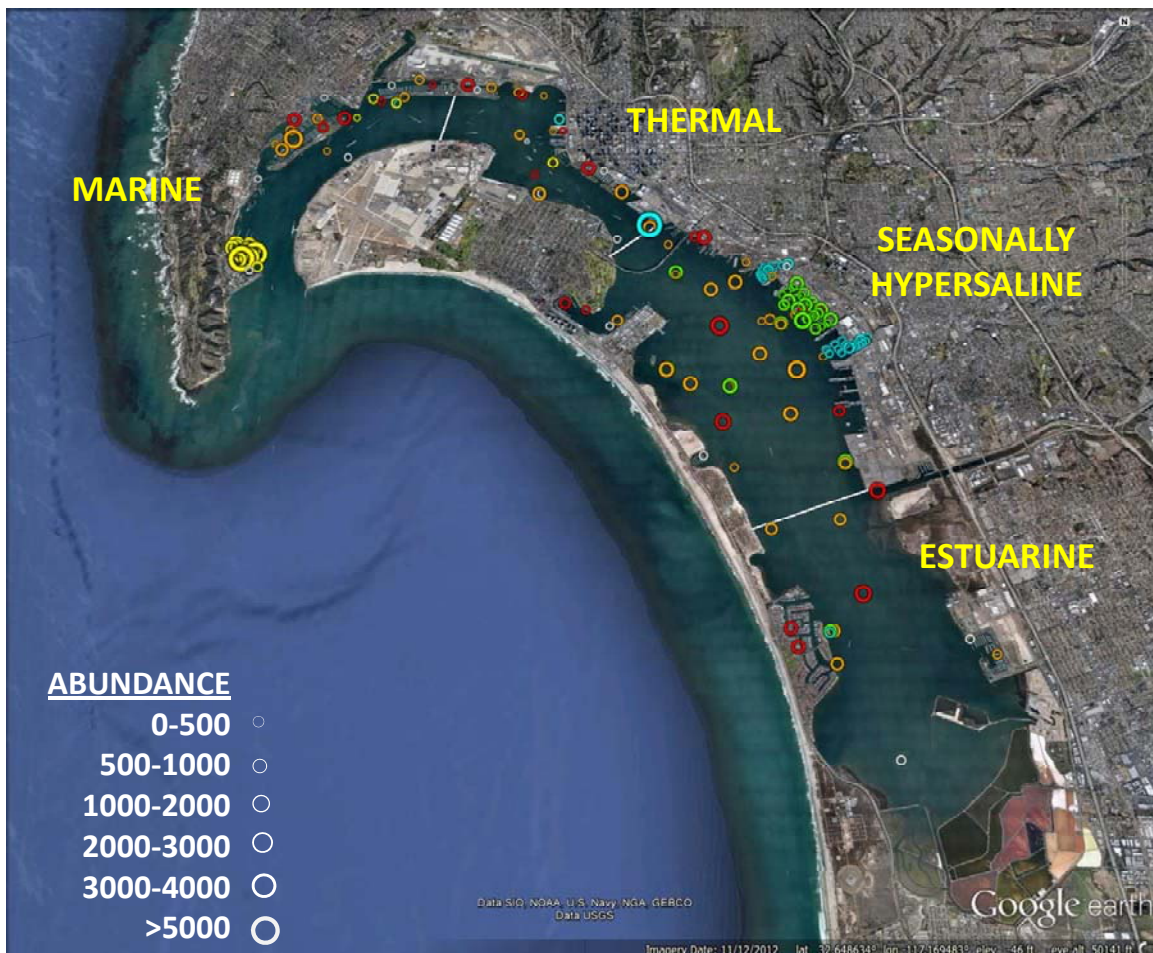


Figure 16. Benthic Abundance data for each sample collected from six studies conducted 1998–2011 in San Diego Bay showing four hydrographic regions. Abundance is based on circle diameter with the colors representing different studies; reference color-coding is shown in Figure 11.

Table 8. EIS Study 2010–1011: benthic community data.

EIS Study 2010-2011							
Zone	Station	Abundance	# Taxa	S-W Diversity	BRI	Longitude	Latitude
Marine	S1	27	8	1.64	NA	-117.23500	32.68647
Marine	S2	54	16	2.36	NA	-117.23617	32.70538
Marine	S3	9	7	0.61	NA	-117.21838	32.71026
Marine	S4	9	6	1.34	NA	-117.22494	32.72364
Marine	S5	3	2	0.64	NA	-117.21085	32.72677
Thermal	S6	15	8	1.71	NA	-117.19249	32.72566
Thermal	S7	126	32	3.14	NA	-117.17832	32.70238
Thermal	S8	39	10	1.73	NA	-117.16557	32.70725
Thermal	S9	48	15	2.21	NA	-117.16306	32.69281
Seasonally Hypersaline	S10	13	6	1.48	NA	-117.16475	32.67581
Seasonally Hypersaline	S11	47	20	2.67	NA	-117.12996	32.68736
Seasonally Hypersaline	S12	----	----	----	NA	-117.11649	32.67394
Seasonally Hypersaline	S13	----	----	----	NA	NA	NA
Seasonally Hypersaline	S14	84	19	2.37	NA	-117.14819	32.65284
Estuarine	S15	----	----	----	NA	NA	NA
Estuarine	S16	----	----	----	NA	-117.12783	32.62664
Estuarine	S17	5	3	1.05	NA	-117.10552	32.62449
Estuarine	S18	182	30	2.81	NA	-117.11941	32.60796

Empty Cells: S12, S13, S15, S16—not sampled and/or sorted.

Table 9. Naval Station San Diego (NAVSTA) TMDL Study 2008: benthic community data.

NAVSTA TMDL 2008							
Zone	Station	Abundance	# Taxa	S-W Diversity	BRI	Longitude	Latitude
Seasonally Hypersaline	NS2240*	1383	39	2.22	50	-117.15410	32.66754
Seasonally Hypersaline	NS2243*	1391	50	2.17	35	-117.14263	32.66450
Seasonally Hypersaline	NS2233*	1055	50	2.60	34	-117.15174	32.68581
Seasonally Hypersaline	NS2258*	1285	47	2.23	19	-117.13212	32.67601
Seasonally Hypersaline	NS2262*	1619	35	2.05	25	-117.12293	32.65144
Seasonally Hypersaline	NS12	265	33	2.65	30	-117.12771	32.68165
Seasonally Hypersaline	NS15	807	40	1.94	42	-117.13122	32.67967
Seasonally Hypersaline	NS19	1265	42	1.77	20	-117.12857	32.67865
Seasonally Hypersaline	NS22	560	38	2.66	23	-117.12487	32.67811
Seasonally Hypersaline	NS23	802	43	2.45	33	-117.12571	32.67761
Seasonally Hypersaline	NS24	696	46	2.50	29	-117.12689	32.67705
Seasonally Hypersaline	NS25	2496	49	1.53	15	-117.12798	32.67632
Seasonally Hypersaline	NS26	1799	56	2.44	36	-117.12261	32.67669
Seasonally Hypersaline	NS27	409	34	2.54	25	-117.12326	32.67639
Seasonally Hypersaline	NS28	278	28	2.62	30	-117.12454	32.67569
Seasonally Hypersaline	NS29	581	40	2.05	37	-117.12566	32.67511
Seasonally Hypersaline	NS6	1383	52	2.38	32	-117.12836	32.68356
Seasonally Hypersaline	NS11	962	44	1.24	36	-117.12710	32.68197
Seasonally Hypersaline	NS14	1251	36	2.53	31	-117.13030	32.68021
Seasonally Hypersaline	NS16	784	38	2.52	50	-117.12577	32.68018
Seasonally Hypersaline	NS17	414	37	2.18	36	-117.12631	32.67986
Seasonally Hypersaline	NS18	1143	44	2.32	44	-117.12715	32.67947
Seasonally Hypersaline	NS21	1164	40	2.33	27	-117.12426	32.67845
Seasonally Hypersaline	NS7	187	26	2.33	38	-117.12912	32.68321
Seasonally Hypersaline	NS8	468	24	1.62	28	-117.13032	32.68249
Seasonally Hypersaline	NS9	377	30	1.91	36	-117.13138	32.68181
Seasonally Hypersaline	NS13	450	29	1.68	30	-117.12879	32.68104
Estuarine	NS2238*	668	34	1.85	43	-117.12861	32.62542

*Includes San Diego Bay Reference Stations used for this study.

Table 10. Subase Sediment Assessment Study 2004: benthic community data.

Subase Sediment Assessment Study 2004							
Zone	Station	Abundance	# Taxa	S-W Diversity	BRI	Longitude	Latitude
Marine	SB2433*	906	63	2.86	19	-117.20949	32.72237
Marine	SB2441*	994	67	2.78	28	-117.23781	32.69144
Marine	SB90056*	495	66	3.34	11	-117.21748	32.71916
Marine	SBC001SS31*	611	55	2.85	19	-117.21446	32.72334
Marine	SB1	1018	103	3.22	22	-117.23894	32.69056
Marine	SB2	1111	71	2.72	31	-117.23807	32.69077
Marine	SB3	2086	129	2.85	18	-117.23571	32.69102
Marine	SB4	1650	89	2.95	24	-117.23825	32.68918
Marine	SB5	2386	77	2.65	27	-117.23701	32.68988
Marine	SB6	1547	104	2.82	23	-117.23593	32.69032
Marine	SB7	2549	112	2.48	20	-117.23473	32.69079
Marine	SB8	2227	96	2.86	24	-117.23740	32.68798
Marine	SB9	3189	85	2.72	26	-117.23661	32.68841
Marine	SB10	4569	129	2.61	23	-117.23528	32.68895
Marine	SB11	1032	130	3.44	15	-117.23412	32.68950
Marine	SB12	532	62	2.78	26	-117.23563	32.68658
Marine	SB13	636	82	3.15	22	-117.23465	32.68707
Marine	SB14	5895	108	1.87	34	-117.23350	32.68762
Thermal	SB2229*	510	62	3.10	16	-117.17604	32.70893
Thermal	SB2436*	682	57	2.69	22	-117.18310	32.71505

*Includes San Diego Bay Reference Stations used for this study.

Table 11. Bight 2003 Study: benthic community data.

Bight 2003							
Zone	Station	Abundance	# Taxa	S-W Diversity	BRI	Longitude	Latitude
Marine	4284	168	18	2.13	NA	-117.20206	32.72692
Marine	4268	1612	49	1.92	NA	-117.22003	32.71868
Marine	4076	1129	33	2.12	NA	-117.23046	32.71810
Marine	4156	6	6	1.79	NA	-117.21263	32.72288
Marine	4140	566	29	2.04	NA	-117.22429	32.71683
Thermal	4092	834	53	2.54	NA	-117.18276	32.72427
Thermal	4108	411	30	2.52	NA	-117.17404	32.71598
Thermal	4348	420	31	2.43	NA	-117.17970	32.70647
Thermal	BRI-15	1468	25	1.70	NA	-117.19450	32.72650
Thermal	BRI-16	1073	24	1.99	NA	-117.16867	32.70750
Seasonally Hypersaline	4028	2329	49	2.48	NA	-117.14385	32.67537
Seasonally Hypersaline	4116	1285	45	2.50	NA	-117.14434	32.65828
Seasonally Hypersaline	4084	680	35	2.55	NA	-117.12292	32.66033
Seasonally Hypersaline	4340	245	29	2.28	NA	-117.12767	32.67891
Seasonally Hypersaline	4236	466	31	2.54	NA	-117.16925	32.67889
Seasonally Hypersaline	BRI-20	179	26	2.76	NA	-117.11617	32.67317
Seasonally Hypersaline	BRI-19	507	36	2.41	NA	-117.12438	32.67853
Seasonally Hypersaline	BRI-18	1035	48	2.61	NA	-117.14583	32.69283
Seasonally Hypersaline	BRI-17	772	37	2.23	NA	-117.14757	32.69323
Seasonally Hypersaline	4418	842	28	2.00	NA	-117.17326	32.67998
Estuarine	4148	1049	42	2.50	NA	-117.11780	32.64680
Estuarine	4308	1209	34	2.21	NA	-117.12242	32.63080
Estuarine	4212	813	27	2.60	NA	-117.13514	32.62586
Estuarine	4052	991	26	1.45	NA	-117.13415	32.62322

Table 12. Chollas Paleta Sediment Assessment Study 2001: benthic community data.

Chollas Paleta Study 2001							
Zone	Station	Abundance	# Taxa	S-W Diversity	BRI	Longitude	Latitude
Marine	2433	421	57	2.88	30	-117.20919	32.72238
Marine	2441	476	66	2.56	60	-117.23803	32.69129
Thermal	2440	918	66	2.93	30	-117.17485	32.71846
Seasonally Hypersaline	2231	6343	88	1.09	39	-117.15658	32.69463
Seasonally Hypersaline	C01	375	34	2.49	50	-117.13539	32.68573
Seasonally Hypersaline	C02	154	32	2.47	48	-117.13520	32.68540
Seasonally Hypersaline	C03	163	22	2.05	54	-117.13493	32.68500
Seasonally Hypersaline	C04	471	29	2.44	55	-117.13495	32.68646
Seasonally Hypersaline	C05	206	21	1.89	56	-117.13456	32.68594
Seasonally Hypersaline	C06	301	32	2.63	50	-117.13407	32.68545
Seasonally Hypersaline	C07	431	40	2.40	45	-117.13439	32.68723
Seasonally Hypersaline	C08	20	6	1.16	65	-117.13403	32.68686
Seasonally Hypersaline	C09	642	43	2.67	53	-117.13364	32.68641
Seasonally Hypersaline	C10	314	30	2.46	53	-117.13330	32.68595
Seasonally Hypersaline	C11	7	7	1.95	30	-117.13353	32.68726
Seasonally Hypersaline	C12	34	14	2.27	55	-117.13229	32.68760
Seasonally Hypersaline	C13	190	26	2.15	72	-117.13088	32.68758
Seasonally Hypersaline	C14	553	10	0.44	83	-117.12971	32.68763
Seasonally Hypersaline	P01	155	31	2.76	32	-117.12407	32.67153
Seasonally Hypersaline	P02	125	22	2.47	41	-117.12357	32.67069
Seasonally Hypersaline	P03	254	31	2.42	54	-117.12234	32.67247
Seasonally Hypersaline	P04	210	24	2.26	50	-117.12177	32.67158
Seasonally Hypersaline	P05	127	16	1.81	51	-117.12123	32.67089
Seasonally Hypersaline	P06	70	15	2.09	56	-117.12091	32.67321
Seasonally Hypersaline	P07	196	22	2.25	53	-117.12023	32.67243
Seasonally Hypersaline	P08	773	33	2.21	44	-117.11969	32.67164
Seasonally Hypersaline	P09	39	18	2.67	48	-117.11840	32.67236
Seasonally Hypersaline	P10	255	26	2.50	54	-117.11840	32.67197
Seasonally Hypersaline	P11	88	24	2.82	55	-117.11822	32.67265
Seasonally Hypersaline	P12	304	36	2.69	43	-117.11770	32.67232
Seasonally Hypersaline	P13	768	35	2.35	51	-117.11733	32.67306
Seasonally Hypersaline	P14	487	36	2.42	57	-117.11709	32.67268
Seasonally Hypersaline	P15	114	21	2.24	59	-117.11669	32.67342
Seasonally Hypersaline	P16	153	19	2.06	69	-117.11642	32.67305
Seasonally Hypersaline	P17	151	20	2.63	65	-117.11601	32.67376
Estuarine	2238	419	32	2.82	23	-117.12869	32.62537

Table 13. Bight 1998 Study: benthic community data.

Bight 1998							
Zone	Station	Abundance	# Taxa	S-W Diversity	BRI	Longitude	Latitude
Marine	2221	824	35	2.617	39	-117.20500	32.7279
Marine	2222	693	35	1.785	45	-117.22571	32.71867
Marine	2223	816	37	2.659	43	-117.23093	32.71576
Marine	2224	383	41	2.896	29	-117.23367	32.71292
Marine	2225	3149	70	2.302	38	-117.23004	32.71338
Marine	2226	1012	57	2.594	38	-117.23213	32.71138
Marine	2227	933	52	2.849	25	-117.20790	32.72376
Marine	2433	709	59	3.076	21	-117.20950	32.72239
Marine	2435	466	60	3.409	-1	-117.22289	32.71159
Marine	2441	1672	86	3.234	17	-117.23803	32.69111
Marine	2442	388	52	2.874	21	-117.23707	32.68926
Thermal	2228	251	41	3.134	33	-117.17820	32.72413
Thermal	2229	705	63	3.124	16	-117.17600	32.70894
Thermal	2230	1372	72	2.701	18	-117.17870	32.70192
Thermal	2240	1201	40	2.183	29	-117.15428	32.66743
Thermal	2251	1194	34	1.852	43	-117.16198	32.70226
Thermal	2263	343	44	3.243	26	-117.17615	32.71608
Thermal	2434	576	50	3.305	24	-117.18359	32.72487
Thermal	2436	599	48	3.064	19	-117.18308	32.71504
Thermal	2439	536	33	2.368	38	-117.18946	32.72603
Thermal	2440	651	59	3.158	32	-117.17484	32.71851
Seasonally Hypersaline	2231	1502	70	2.753	16	-117.15653	32.69485
Seasonally Hypersaline	2233	395	39	2.727	29	-117.15179	32.68573
Seasonally Hypersaline	2239	1030	25	1.663	38	-117.14509	32.68248
Seasonally Hypersaline	2241	1526	44	2.305	35	-117.13662	32.67029
Seasonally Hypersaline	2242	1117	28	1.795	37	-117.14993	32.66493
Seasonally Hypersaline	2243	966	47	2.741	36	-117.14282	32.66459
Seasonally Hypersaline	2244	1376	48	2.685	31	-117.13189	32.65966
Seasonally Hypersaline	2245	487	25	2.161	43	-117.14279	32.65091
Seasonally Hypersaline	2252	327	37	2.086	4	-117.15298	32.69177
Seasonally Hypersaline	2253	465	33	2.267	45	-117.13786	32.68821
Seasonally Hypersaline	2254	684	33	2.172	47	-117.16326	32.67672
Seasonally Hypersaline	2255	391	30	2.126	37	-117.12937	32.67801
Seasonally Hypersaline	2256	237	28	2.658	38	-117.13577	32.67678
Seasonally Hypersaline	2257	503	37	2.31	38	-117.13415	32.67693
Seasonally Hypersaline	2258	826	36	2.292	43	-117.13215	32.67600
Seasonally Hypersaline	2259	102	22	2.616	38	-117.12477	32.67015
Seasonally Hypersaline	2260	2263	49	1.828	39	-117.12999	32.66723
Seasonally Hypersaline	2262	542	29	2.101	41	-117.12293	32.65143
Seasonally Hypersaline	2264	237	28	2.719	44	-117.13293	32.68540
Seasonally Hypersaline	2265	1543	48	2.388	27	-117.14028	32.68390
Estuarine	2235	551	29	2.074	42	-117.13707	32.64081
Estuarine	2238	760	41	2.469	38	-117.12802	32.62548
Estuarine	2247	900	33	2.087	34	-117.12493	32.64235
Estuarine	2249	600	37	2.265	45	-117.12800	32.62083
Estuarine	2438	384	34	2.639	48	-117.10144	32.62229

Table 14. Comparison of benthic abundance data between EIS study data and historical study data (stations near each other).

EIS Study/Station	Abundance	Other Study/Station	Abundance
S1	27	Subase/SB12	532
S2	54	none nearby	NA
S3	9	Bight 98/2435	466
S4	9	none nearby	NA
S5	3	Bight 98/2221	824
S6	15	Bight 98/2439	536
S7	126	Bight 98/2230	1372
S8	39	none nearby	NA
S9	48	none nearby	NA
S10	13	Bight 98/2254	684
S11	47	Chollas-Paletta/CP13	190
S12	NA	NA	NA
S13	NA	NA	NA
S14	84	Bight 98/2245	487
S15	NA	NA	NA
S16	NA	NA	NA
S17	5	Bight98/2438	384
S18	182	none nearby	NA

RESULTS

The results section is broken down into six main subsections, (A) Sampling Locations, (B) Settling Plate Study, (C) Rapid Assessment Survey, (D) Grain Size Analysis, (E) DNA Barcoding, and (F) Statistical Analysis–Community Metrics. General results for each subsection are provided below.

A. Sampling Locations

Eighteen site locations were initially identified for sampling in this study, five in the Marine Region, four in the Thermal Region, five in the Hypersaline Region, and four in the Estuarine Region. However, two sampling sites, S13 and S15, were dropped from the study due to lack of availability at the time of the survey. A summary of sampling methods by locations is provided in Table 15. Note that for the designation of (SP), organisms were collected primarily from the settling plates but there were some instances when organisms of interest were found on support structure, including rope, frame, float, and ballast.

Table 15. Summary of sampling methods/substrate collected per sampling station.

Sampling Methods					
Station ID	SP	Seine	SED	DO	Sieve
S1	x	x	x		x
S2	X	x	x		x
S3	x		x	x	x
S4	x		x	x	
S5	x		x		x
S6	x		x	x	
S7	x		x		x
S8	x		x	x	
S9	x		x	x	x
S10	x		x	x	x
S11	X		x		
S12	x				
S14	x		x	x	x
S16	x		x		x
S17	x	x	x		x
S18			x		x

(x) in red indicates sampling plate deployed but not retrieved. (sp) Settling plate. (Seine) beach seine. (SED) sediment grab. (DO) Dock time search. (Sieve) Beach sieve

B. Settling Plate Study

Of the 16 settling plates deployed, 10 were recovered and 9 were in good condition. The settling plate for S11 was compromised due to plates dropping onto the sediment floor. For S17, while this was a hanging set-up, the ropes were cut and the plates were missing. For S18, as mentioned previously, no settling plate was deployed due to the shallowness of the water. Collection of floating settling plates posed more of a challenge as it required returning to the exact location and grappling the float to locate the plates; as a result, some of them were not retrievable. A summary of settling plate locations and retrieval information is provided in Table 16.

Table 16. Settling plate deployment locations and retrieval history

Eco-Region	Site Name	Site ID	Deployment (Hanging/Float)	Deployment Date	Deployment Time	Latitude	Longitude	Depth (ft)	Retrieval Date	Retrieval Time	Comments
Marine Region	SUBASE beach	S1	Float	7/19/2010		32.68530	117.2345	20	Attempted, 7/14/2011		no retrieval
	SSC Pacific Floating Pier	S2	Hanging	7/13/2010	14:35	32.70538	117.23617	14.7	7/15/2011	9:00	all in good condition
	NAB Fishing Pier	S3	Float	7/8/2010	9:15	32.71026	117.21838	44.9	Attempted, 7/14/2011		no retrieval
	Sun Harbor Marina	S4	Hanging	7/13/2010	10:40	32.72364	117.22494	18.2	7/13/2011	9:35	all in good condition
	Harbor Island West Marina	S5	Hanging	7/6/2010	14:00	32.72677	117.21085	10.8	7/13/2011	13:45	all in good condition
Thermal Region	Sunroad San Diego Marina	S6	Hanging	7/9/2010	11:42	32.72566	117.19249	12.1	7/13/2011	10:50	all in good condition
	Bayview Park Coronado	S7	Float	7/8/2010	9:40	32.70238	117.17832	6.5	Attempted, 7/14/2011		no retrieval
	San Diego Marriott Hotel and Marina	S8	Hanging	7/13/2010	9:50	32.70725	117.16557	12.4	7/13/2011	10:15	Wood plates deteriorated
	Tidelands Park Coronado	S9	Float	7/8/2010	10:10	32.69281	117.16306	5.6	7/7/2011	12:00	No Float, plates on sediment. Plates ruined due to poor SW flow stoppage
Seasonally Hypersaline Region	Amphib Base Fuel Pier	S10	Float	7/13/2010	13:20	32.67581	117.16475	16.8	7/14/2011	11:30	all in good condition
	Chollas Creek	S11	Float	7/9/2010	9:50	32.68736	117.12996	13.5	Attempted 7/11/2011		no retrieval
	Paleta Creek	S12	Float	7/9/2010	10:15	32.67394	117.11649	18	7/11/2011	13:15	all in good condition
	Fiddler's Cove Marina	S14	Hanging	7/6/2010	14:40	32.65284	117.14819	13.1	7/12/2011	12:50	all in good condition
Estuarine Region	Grand Caribe Shoreline Park	S16	Float	7/8/2010	10:37	32.62664	117.12783	8.6	7/7/2011	10:55	octopus and eggs in cinder block
	Chula Vista Harbor Pier	S17	Hanging	7/9/2010	10:50	32.62449	117.10552	11.8	Attempted 7/12/11		no retrieval-rope was cut

The 12-month deployment of plates resulted in a rich diversity of organisms. Not only were the settling plates completely fouled but most of the support structure as well (Figure 17). In total, there were 180 different species of organisms found on the 10 settling plates within the four different management regions. Five of these were found in all four regions (e.g., *Harmothoe imbricata* complex, *Zoobotryon verticillatum*, *Leucothoe alata*, *Paracerceis sculpta*, and *Paranthura elegans*). At the phylum level, this equates to one annelid, one bryozoan, and three arthropoda). Twenty-one of the species were found in three of the four hydrographic regions (Table 17), and 45 species were found in at least two of the four hydrographic regions (Table 18).

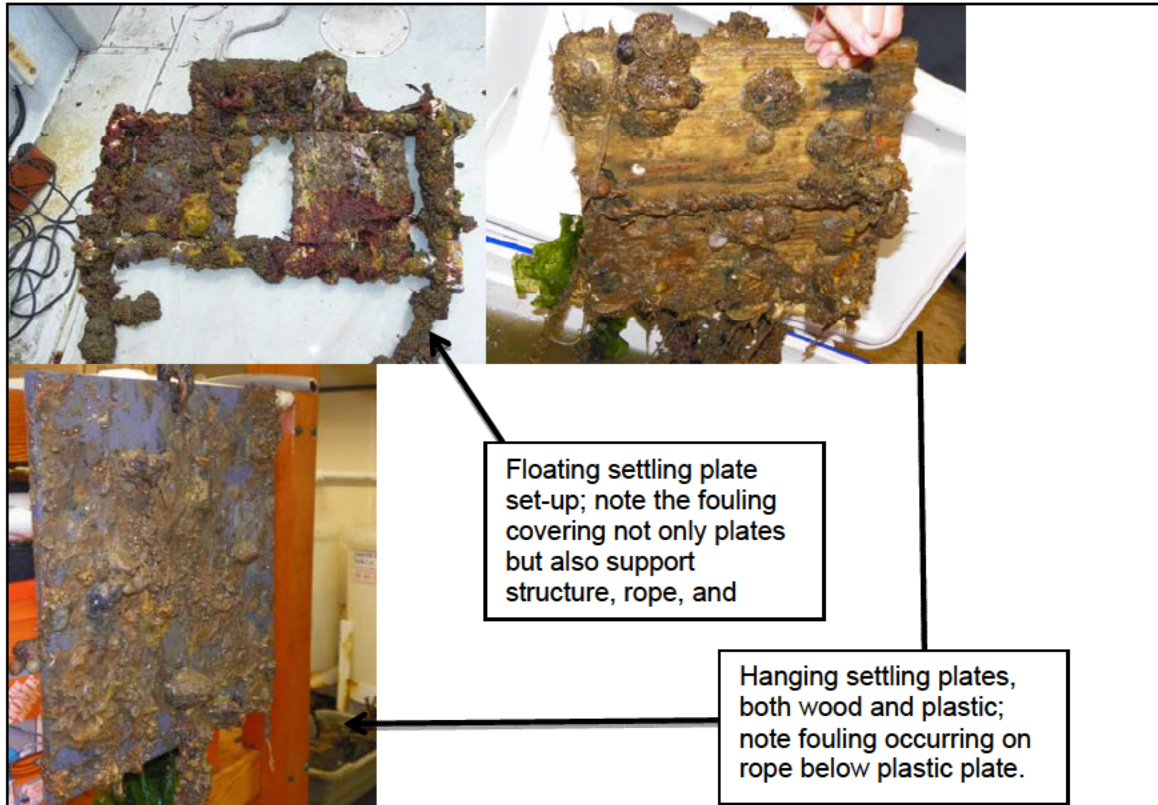


Figure 17. Samples images of recently recovered settling plates. Images are representative of both the floating and hanging set-up. Settling plates from all locations essentially looked the same.

Table 17. Settling plate species found on three of the four hydrographic regions.

Sample Type	Region	Phylum	Species
SP	Seasonally Hypersaline, Marine, Thermal	Annelida	Brania complex
SP	Seasonally Hypersaline, Marine, Thermal	Annelida	Cirratulus cf cingulatus
SP	Estuarine, Thermal, Marine	Annelida	Parasabella pallidus ?
SP	Seasonally Hypersaline, Estuarine, Thermal	Annelida	Exogone lourei
SP	Seasonally Hypersaline, Marine, Thermal	Annelida	Halosydna johnsoni
SP	Seasonally Hypersaline, Estuarine, Thermal	Annelida	Hydroides diramphus
SP	Seasonally Hypersaline, Marine, Thermal	Annelida	Myxicola
SP	Seasonally Hypersaline, Marine, Thermal	Annelida	Nicolea sp A Harris
SP	Seasonally Hypersaline, Estuarine, Thermal	Annelida	Odontosyllis phosphorea
SP	Seasonally Hypersaline, Estuarine, Thermal	Annelida	Polycirrus
SP	Seasonally Hypersaline, Estuarine, Thermal	Annelida	Scoletoma perkinsi ?
SP	Seasonally Hypersaline, Marine, Thermal	Annelida	Syllis nipponica
SP	Seasonally Hypersaline, Marine, Thermal	Platyhelminthes	Eurylepta aurantiaca
SP	Seasonally Hypersaline, Marine, Thermal	Bryozoa	Watersipora arcuata
SP	Seasonally Hypersaline, Estuarine, Marine	Mollusca	Musculista senhousia
SP	Seasonally Hypersaline, Estuarine, Marine,	Mollusca	Mytilus galloprovincialis
SP	Seasonally Hypersaline, Marine, Thermal	Arthropoda	Dissiminassa dissimilis
SP	Seasonally Hypersaline, Marine, Thermal	Arthropoda	Lophopanopeus bellus
SP	Seasonally Hypersaline, Estuarine, Thermal	Arthropoda	Pyromaia tuberculata
SP	Seasonally Hypersaline, Marine, Thermal	Arthropoda	Zeuxo
SP	Seasonally Hypersaline, Marine, Thermal	Arthropoda	Monocorophium acherusicum

Note taxonomist labeled a sample identification with (?) if there was a potential unsurety in identification. Most likely cause of unsurety was due to sample degradation.

Table 18. Settling plate species list present in two different hydrographic regions.

Sample Type	Region	Phylum	Species
SP	Seasonally Hypersaline, Estuarine	Chordata	Botryllus schlosseri
SP	Seasonally Hypersaline, Estuarine	Chordata	Ciona intestinalis
SP	Seasonally Hypersaline, Estuarine	Chordata	Ciona savignyi
SP	Seasonally Hypersaline, Estuarine	Chordata	Symplegma reptans
SP	Estuarine, Thermal	Chordata	Microcosmus squamiger
SP	Estuarine, Thermal	Chordata	Molgula ficus
SP	Estuarine, Marine	Chordata	Polyandrocarpa zorritensis
SP	Estuarine, Marine	Chordata	Styela plicata
SP	Seasonally Hypersaline, Thermal	Annelida	Amblyosyllis speciosa
SP	Seasonally Hypersaline, Thermal	Annelida	Branchiomma
SP	Seasonally Hypersaline, Thermal	Annelida	Capitella capitata complex
SP	Seasonally Hypersaline, Marine	Annelida	Cirratulidae juveniles
SP	Seasonally Hypersaline, Estuarine	Annelida	Dorvillea (Schistomeringos) annulata
SP	Seasonally Hypersaline, Thermal	Annelida	Eulalia quadrioculata
SP	Seasonally Hypersaline, Thermal	Annelida	Naineris dendritica
SP	Seasonally Hypersaline, Estuarine	Annelida	Neanthes acuminata complex
SP	Seasonally Hypersaline, Estuarine	Arthropoda	Paracerceis
SP	Marine, thermal	Annelida	Pileolaria marginata
SP	Marine, thermal	Annelida	Platynereis bicanaliculata
SP	Seasonally Hypersaline, Thermal	Annelida	Polydora
SP	Seasonally Hypersaline, Thermal	Annelida	Polydora narica
SP	Marine, thermal	Annelida	Salmacina tribranchiata
SP	Seasonally Hypersaline, Thermal	Annelida	Syllis gracilis complex
SP	Seasonally Hypersaline, Thermal	Porifera	Porifera ?
SP	Seasonally Hypersaline, Estuarine	Cnidaria	Hydrozoa ?
SP	Seasonally Hypersaline, Estuarine	Mollusca	Ostrea edulis
SP	Seasonally Hypersaline, Thermal	Bryozoa	Diaperoforma californica
SP	Seasonally Hypersaline, Marine	Mollusca	Octopus bimaculatus/bimaculoides ?
SP	Estuarine, Thermal	Cnidaria	Diadumene
SP	Estuarine, Marine	Bryozoa	Scrupocellaria
SP	Estuarine, Marine	Bryozoa	Thalamoporella
SP	Marine, thermal	Mollusca	Hiatella arctica
SP	Marine, thermal	Platyhelminthes	Pseudoceros canadensis ?
SP	Seasonally Hypersaline, Thermal	Arthropoda	Eualus lineatus
SP	Seasonally Hypersaline, Thermal	Arthropoda	Bemlos concavus
SP	Seasonally Hypersaline, Estuarine	Arthropoda	Erichthonius brasiliensis
SP	Seasonally Hypersaline, Marine	Arthropoda	Nymphon heterodenticulatum
SP	Seasonally Hypersaline, Thermal	Arthropoda	Podocerus fulanus
SP	Seasonally Hypersaline, Estuarine	Echinodermata	Amphipholis squamata
SP	Seasonally Hypersaline, Thermal	Arthropoda	Ampithoe plumulosa
SP	Seasonally Hypersaline, Thermal	Arthropoda	Elasmopus serricatus
SP	Seasonally Hypersaline, Marine	Arthropoda	Leptochelia dubia
SP	Marine, thermal	Arthropoda	Apolochus picadurus
SP	Marine, thermal	Arthropoda	Balanus trigonus
SP	Marine, thermal	Arthropoda	Protomedeia articulata cmlpx

Additionally, 15 species were found only in the Estuarine Region, 23 species were found only in the Marine Region, 29 were found only in the Thermal Region, and 41 were found only in the seasonally Hypersaline Region. Tables 19 through 22 provide a summary of those species.

Table 19. Settling plate species only in the Estuarine Region.

Sample Type	Region	Phylum	Species
SP	Estuarine	Chordata	Diplosoma listerianum
SP	Estuarine	Chordata	Perophora annectens
SP	Estuarine	Chordata	Styela clava
SP	Estuarine	Annelida	Armandia brevis
SP	Estuarine	Annelida	Leitoscoloplos pugettensis
SP	Estuarine	Annelida	Mediomastus
SP	Estuarine	Annelida	Pista cf brevibranchiata
SP	Estuarine	Annelida	Scoletoma sp C
SP	Estuarine	Annelida	Syllis
SP	Estuarine	Bryozoa	Bryozoa ?
SP	Estuarine	Platyhelminthes	Armatoplana reishi
SP	Estuarine	Mollusca	Barleeia
SP	Estuarine	Bryozoa	Disporella
SP	Estuarine	Mollusca	Mitrella aurantiaca
SP	Estuarine	Nemertea	Nemertea

Table 20. Settling plate species only in the Hypersaline Region.

Sample Type	Region	Phylum	Species
SP	Seasonally Hypersaline	Chordata	Distaplia occidentalis
SP	Seasonally Hypersaline	Chordata	Botrylloides perspicuum
SP	Seasonally Hypersaline	Chordata	Aplidium californicum ?
SP	Seasonally Hypersaline	Chordata	Botrylloides diegensis
SP	Seasonally Hypersaline	Annelida	autolytid epitokes
SP	Seasonally Hypersaline	Annelida	Branchiosyllis exilis ?
SP	Seasonally Hypersaline	Annelida	Cirriformia ?
SP	Seasonally Hypersaline	Annelida	Eupolymnia heterobranchiata ?
SP	Seasonally Hypersaline	Annelida	Ophryotrocha
SP	Seasonally Hypersaline	Annelida	Phyllodoce medipapillata
SP	Seasonally Hypersaline	Annelida	Polydorin
SP	Seasonally Hypersaline	Annelida	Protocirrineris
SP	Seasonally Hypersaline	Annelida	Scoletoma erecta
SP	Seasonally Hypersaline	Annelida	Timarete luxuriosa ?
SP	Seasonally Hypersaline	Annelida	Trypanosyllis
SP	Seasonally Hypersaline	Bryozoa	Crisia
SP	Seasonally Hypersaline	Mollusca	Ostrea conchaphila
SP	Seasonally Hypersaline	Platyhelminthes	Stylochoplana
SP	Seasonally Hypersaline	Cnidaria	Diadumene lineata ?
SP	Seasonally Hypersaline	Nemertea	Baseodiscus delineatus
SP	Seasonally Hypersaline	Mollusca	Crepidula onyx
SP	Seasonally Hypersaline	Mollusca	Crucibulum spinosum
SP	Seasonally Hypersaline	Porifera	Haliclona
SP	Seasonally Hypersaline	Bryozoa	Smittoidea prolifica
SP	Seasonally Hypersaline	Platyhelminthes	Stylostomum lentum
SP	Seasonally Hypersaline	Nemertea	Tetrastemma aberrans
SP	Seasonally Hypersaline	Mollusca	Vayssierea felis
SP	Seasonally Hypersaline	Mollusca	Volvarina taeniolata
SP	Seasonally Hypersaline	Cnidaria	Bimeria/Garveia ?
SP	Seasonally Hypersaline	Porifera	Leucilla nuttingii
SP	Seasonally Hypersaline	Mollusca	Mitrella aurantiaca
SP	Seasonally Hypersaline	Mollusca	Odostomia
SP	Seasonally Hypersaline	Mollusca	Polycera hedgpethi
SP	Seasonally Hypersaline	Bryozoa	Watersipora subtorquata
SP	Seasonally Hypersaline	Arthropoda	Pelia tumida
SP	Seasonally Hypersaline	Arthropoda	Caprella
SP	Seasonally Hypersaline	Arthropoda	Colomastix
SP	Seasonally Hypersaline	Arthropoda	Liljborgia geminata
SP	Seasonally Hypersaline	Arthropoda	Postasterope barnesi
SP	Seasonally Hypersaline	Arthropoda	Rhynchothorax philopsammum
SP	Seasonally Hypersaline	Arthropoda	Quadrimaera vigota

Table 21. Settling plate species only in the Marine Region.

Sample Type	Region	Phylum	Species
SP	Marine	Chordata	Ascidia ceratodes
SP	Marine	Chordata	Styela clava
SP	Marine	Annelida	Harmothoe cf. hirsuta
SP	Marine	Annelida	Nereis
SP	Marine	Cnidaria	Aglaophenia
SP	Marine	Nemertea	Amphiporus
SP	Marine	Mollusca	Hermisenda crassicornis
SP	Marine	Mollusca	Lamellaria diegoensis
SP	Marine	Cnidaria	Plumularia
SP	Marine	Mollusca	Teredo navalis
SP	Marine	Bryozoa	Celleporaria brunnea
SP	Marine	Bryozoa	Bugula neretina
SP	Marine	Mollusca	Crepidula
SP	Marine	Mollusca	Diaulula sandiegensis
SP	Marine	Mollusca	Leptopecten latiauratus
SP	Marine	Mollusca	Navanax inermis
SP	Marine	Arthropoda	Cancer branneri
SP	Marine	Arthropoda	Caprella californica
SP	Marine	Arthropoda	Caprella mendax
SP	Marine	Arthropoda	Caprella mutica
SP	Marine	Arthropoda	Jassa slatteryi
SP	Marine	Arthropoda	Pugettia producta
SP	Marine	Arthropoda	Tanystylum intermedium

Table 22. Settling plate species only in the Thermal Region.

Sample Type	Region	Phylum	Species
SP	Thermal	Annelida	Dodecaceria
SP	Thermal	Annelida	Epigamia noroi
SP	Thermal	Annelida	Eulalia californiensis
SP	Thermal	Annelida	Eumida
SP	Thermal	Annelida	Exogone
SP	Thermal	Annelida	Hydroides elegans
SP	Thermal	Annelida	Myrianida pachycera
SP	Thermal	Annelida	Dipolydora sp A Harris
SP	Thermal	Annelida	Boccardiella hamata
SP	Thermal	Arthropoda	Pontonia
SP	Thermal	Platyhelminthes	Acerotisa californica
SP	Thermal	Platyhelminthes	Euryleptodes insularis
SP	Thermal	Cnidaria	Hydrozoa ?
SP	Thermal	Bryozoa	Porellidae
SP	Thermal	Platyhelminthes	Prostheceraeus bellastratus
SP	Thermal	Mollusca	Alia carinata
SP	Thermal	Cnidaria	Aurelia aurita
SP	Thermal	Mollusca	Cerithiopsis cosmia
SP	Thermal	Bryozoa	Crisia
SP	Thermal	Mollusca	Janolus barborensis
SP	Thermal	Platyhelminthes	Prosthlostomum latocelis
SP	Thermal	Arthropoda	Limnoria quadripunctata
SP	Thermal	Arthropoda	Pachygrapsus crassipes
SP	Thermal	Arthropoda	Protohyale frequens
SP	Thermal	Arthropoda	Amphibalanus amphitrite
SP	Thermal	Arthropoda	Bemlos macromanus
SP	Thermal	Arthropoda	Cancer amphioetus
SP	Thermal	Arthropoda	Paradexamine
SP	Thermal	Arthropoda	Synalpheus lockingtoni

C. Rapid Assessment Survey

The rapid assessment survey was conducted July 11–15. While the bulk of sample collection (e.g., retrieval of settling plates, line transects, beach seine, and time searches, sorting, and identification) occurred during this week, the near off-shore Van Veen sediment grabs, described in the Methods section, were collected July 6–8. Sorting and identification of these samples was completed the week of the Rapid Assessment Survey. A total of 6,477 organisms, with 299 species represented from 13 phyla, were collected and counted. This section is broken into two main parts, a summary of species identified by sampling method and a summary of the species assemblages and community metrics from each of the 16 study stations. For a complete listing of identified species by both sampling station and by alphabetical order please refer to Appendix E and F.

1. Sampling Method Species Assemblages: As settling plates have already been summarized above in Tables 19–22, the following is a brief summary of results for the other collection methodologies (refer to Results, Section A,). Twenty-four species identified from sieving samples with potentially an additional two to five identified to genus level. Of those 24 species, two were found in all four regions, *Gemma gemma* and *Solen sicarius* and three species were found in three out of the four hydrographic regions (i.e., *Donax gouldii*, *Musculista senhousia*, *Acanthina spirata*). An additional six species were found in at least two of the four regions (i.e., *Golfingia margaritacea californiensis*, *Cylichnella inculta*, *Neotrypaea californiensis*, *Tagelus californianus*, *Macron lividus*, and *Chione undatella*). The remaining species identified were only found in one of the four regions (Table 23).

Table 23. Summary of single region species found by Sieve Method.

Region	Type Sample	Phylum	Species
Marine	Sieve	Arthropoda	Caprella californica
Marine	Sieve	Mollusca	Nassarius fossatus
Marine	Sieve	Arthropoda	Taliepis nuttallii
Marine	Sieve	Arthropoda	Pilumnus spinohirsutus
Marine	Sieve	Echinodermata	Amphipholis squamata
Thermal	Sieve	Mollusca	Notoucmea insessa
Seasonally Hypersaline	Sieve	Mollusca	Crucibulum spinosum
Estuarine	Sieve	Mollusca	Tellina meropsis
Estuarine	Sieve	Mollusca	Cerithidia californica
Marine	Sieve	Arthropoda	Neotrypaea affinis
Marine	Sieve	Arthropoda	Cirolana harfordi
Marine	Sieve	Arthropoda	Paranthurus elegans

For the beach seine method of collection, 14 species were identified, with an additional 6–10 identified to genus level. All species identified came from either the Marine or Estuarine Hydrographic Region, with none of the 14 occurring in more than one region. A summary is provided in Table 26. Note that the beach seine method of collection was only conducted in the Marine and Estuarine regions (refer to Table 24).

Table 24. Summary of species found in Seine Method by region.

Region	Type Sample	Phylum	Species
Estuarine	Seine	Echinodermata	Amphipholis squamata
Estuarine	Seine	Chordata	Leuresthes tenuis
Estuarine	Seine	Mollusca	Musculista senhousia
Estuarine	Seine	Arthropoda	Pachygrapsus crassipes
Estuarine	Seine	Cnidaria	Anthopleura
Estuarine	Seine	Mollusca	Mitrella aurantiaca
Marine	Seine	Mollusca	Olivella biplicata
Marine	Seine	Mollusca	Arcularia tiarula
Marine	Seine	Arthropoda	Rocinela angustata
Marine	Seine	Arthropoda	Caprella californica
Marine	Seine	Arthropoda	Cirolana harfordi
Marine	Seine	Arthropoda	Idotea fewkesi
Marine	Seine	Arthropoda	Leptochelia dubia
Marine	Seine	Arthropoda	Exosphaeroma inornata

For the near off-shore sediment collection method (refer to Methods Section), 161 species were identified, with an additional 20 to 21 identified to genus level. Of those 161 species, four were found in all four hydrographic regions (e.g., *Musculista senhousia*, *Caprella californica*, *Heterophoxus ellisi*, and *Protohyale frequens*), five in at least three of the regions (e.g., *Gemma gemma*, *Amphipholis squamata*, *Tellina meropsis*, *Notoacmaea depicta*, *Paranthurus elegans*) and 17 species in at least two of the regions (Table 25). The remaining species were found in only one region (Table 26).

Table 25. Species identified in SED Method Collection of three management regions.

Region	Type Sample	Phylum	Species
Estuarine, Thermal	SED	Arthropoda	Ampithoe valida
Marine, Thermal	SED	Mollusca	Donax gouldii
Marine, Thermal	SED	Mollusca	Solen sicarius
Marine, Thermal	SED	Arthropoda	Neotrypaea californiensis
Marine, Thermal	SED	Arthropoda	Ampelisca cristata microdentata
Marine, Thermal	SED	Arthropoda	Amphideutopus oculus
Marine, Thermal	SED	Arthropoda	Foxiphalus golfensis
Marine, Thermal	SED	Arthropoda	Leptochelia dubia
Marine, Thermal	SED	Arthropoda	Monocorophium acherusicum
Seasonally Hypersaline, Estuarine	SED	Annelida	Harmothoe imbricata complex
Seasonally Hypersaline, Estuarine	SED	Annelida	Naineris dendritica
Seasonally Hypersaline, Estuarine	SED	Bryozoa	Disporella
Seasonally Hypersaline, Marine	SED	Mollusca	Chione undatella
Seasonally Hypersaline, Thermal	SED	Other Phyla	Golfingia margaritacea californiensis ?
Seasonally Hypersaline, Thermal	SED	Mollusca	Arctularia tiarula
Seasonally Hypersaline, Thermal	SED	Mollusca	Acteocina inculta
Seasonally Hypersaline, Thermal	SED	Mollusca	Lyonsia californica
Seasonally Hypersaline, Thermal	SED	Arthropoda	Pyromaia tuberculata

Finally, the last sampling method was that of time searches (refer to Methods section, Appendix A). The majority of species identified were found on docks/floating structures. Due to the extensive volume of material needing to be sorted from the SP, Seine, SED, and Sieve sample collection, the methods sample collection from the time searches focused more on species of interest by the taxonomists. This data set should not be considered comprehensive but rather more opportunistic. Thus, this list is provided for information. Sixteen species were identified, with an additional six identified down to genus level. Samples were found from locations within the Marine, Thermal, and Seasonally Hypersaline regions. Note that no time searches were conducted in the Estuarine Region (refer to Results section, Table 15), and thus, no samples collected. None of the 16 species identified were found in more than one hydrographic region. Table 27 provides a summary of species identified.

2. Summary of the Species Assemblages and Community Metrics: The following presents a summary of the species assemblages and community metrics from each of the study stations. This information is a compilation of all sampling methodologies at each location. A complete species listing by site location is in Appendix F.
 - a. Individual Stations in the Marine Region
 - i. Station S1 – Smugglers Cove: Twenty-seven species were observed at this station. Four phyla were present, with arthropods making up 58%, mollusks making up 28%, nemertean making up 12%, and one species of cnidaria at 4%. The arthropod *Cirolana harfordi* was the most abundance species with 373 collected. *Cirolana harfordi* is an isopod found in the intertidal zone. The average species diversity calculated for this site was 1.31, with 505 collected specimens.

Table 26. Summary of single management region species identified in SED Method Collection.

Region	Type	Phylum	Species
Estuarine	SED	Annelida	Armandia brevis
Estuarine	SED	Annelida	Euclymeninae
Estuarine	SED	Annelida	Glycera americana
Estuarine	SED	Annelida	Harmothoe cf. hirsuta
Estuarine	SED	Annelida	Leitoscoloplos pugettensis
Estuarine	SED	Annelida	Mediomastus
Estuarine	SED	Annelida	Neanthes acuminata complex
Estuarine	SED	Annelida	Pista cf. brevirbranchiata
Estuarine	SED	Annelida	Scoloplos armiger complex
Estuarine	SED	Annelida	Scyphoproctus oculatus
Estuarine	SED	Arthropoda	Alpheus californiensis
Estuarine	SED	Arthropoda	Bemlos macromanus
Estuarine	SED	Arthropoda	Leucothoe alata ?
Estuarine	SED	Arthropoda	Paracerceis sculpta
Marine	SED	Mollusca	Mytilus californianus
Marine	SED	Arthropoda	Incisocallope newportensis
Marine	SED	Arthropoda	Hartmanodes hartmanae
Marine	SED	Arthropoda	Bemlos concavus
Marine	SED	Arthropoda	Erichthonius brasiliensis
Marine	SED	Arthropoda	Photis parvidons
Marine	SED	Arthropoda	Protomeдея articulata cmlpx
Marine	SED	Arthropoda	Stenothoe estacola
Marine	SED	Arthropoda	Elasmopus bampo
Marine	SED	Cnidaria	Edwardsia californica
Marine	SED	Nemertea	Carinoma mutabilis
Marine	SED	Nemertea	Paranemertes californica
Marine	SED	Nemertea	Tubulanus polymorphus
Seasonally Hypersaline	SED	Chordata	Coryphopterus nicholsi
Seasonally Hypersaline	SED	Annelida	Boccardiella hamata
Seasonally Hypersaline	SED	Annelida	Branchiomma
Seasonally Hypersaline	SED	Annelida	Capitella capitata complex 1
Seasonally Hypersaline	SED	Annelida	Dorvillea (Schistomeringos) annulata
Seasonally Hypersaline	SED	Annelida	Epigamia noroi
Seasonally Hypersaline	SED	Annelida	Eulalia californiensis
Seasonally Hypersaline	SED	Annelida	Halosydna johnsoni
Seasonally Hypersaline	SED	Annelida	Ophiodromus pugettensis
Seasonally Hypersaline	SED	Annelida	Platynereis bicanaliculata
Seasonally Hypersaline	SED	Annelida	Polydora narica
Seasonally Hypersaline	SED	Annelida	Proceraea nigropunctata
Seasonally Hypersaline	SED	Annelida	Syllis nipponica
Seasonally Hypersaline	SED	Mollusca	Solen rostriformis
Seasonally Hypersaline	SED	Cnidaria	Scolathus scamiti
Seasonally Hypersaline	SED	Arthropoda	Lophopanopeus leucomanus
Thermal	SED	Mollusca	Tagelus californianus
Thermal	SED	Arthropoda	Taliepis nuttallii
Thermal	SED	Annelida	Neanthes succinea
Thermal	SED	Mollusca	Acanthina spirata
Thermal	SED	Mollusca	Asthenothaerus diegensis
Thermal	SED	Mollusca	Bulla gouldiana
Thermal	SED	Mollusca	Olivella biplicata
Thermal	SED	Arthropoda	Heteroserolis carinata
Thermal	SED	Arthropoda	Hippolyte californiensis
Thermal	SED	Arthropoda	Aoroides secundus
Thermal	SED	Arthropoda	Dulichia rhabdoplastis
Thermal	SED	Arthropoda	Hemigrapsus nudus

Table 27. Summary of species identified by region from the Time Search Method.

Region	Type Sample	Phylum	Species
Marine	DO	Chordata	Symplegma reptans
Thermal	DO	Chordata	Aplidium californicum
Thermal	DO	Chordata	Ascidia zara
Thermal	DO	Chordata	Botrylloides violaceus
Thermal	DO	Chordata	Diplosoma listerianum
Thermal	DO	Annelida	Brania complex
Thermal	DO	Annelida	Dorvillea moniloceros
Thermal	DO	Annelida	Odontosyllis phosphorea
Thermal	DO	Annelida	Platynereis bicanaliculata
Thermal	DO	Annelida	Syllis nipponica
Thermal	DO	Platyhelm	Acerotisa californica
Thermal	DO	Bryozoa	Diaperoforma californica
Thermal	DO	Platyhelm	Eurylepta aurantiaca
Thermal	DO	Bryozoa	Zoobotryon verticillatum
Thermal	DO	Arthropoda	Aoroides secundus
Thermal	DO	Arthropoda	Podocerus fulanus

- ii. Station S2 – SSC-Pacific Pier 169: Fifty-one species were observed at this station. Four phyla were present, with arthropods making up 71% of the species assemblage, mollusks making up 22%, cnidarians making up 4%, and one species of nemertean at 4%. The arthropod *Cirolana harfordi* was the most abundance species with 142 collected. *Cirolana harfordi* is an isopod found in the intertidal zone. The average species diversity calculated for this site was 1.74, with 313 collected specimens.
- iii. Station S3 – NAB Fishing Pier: Twenty species were observed at this station. Six phyla were present, with arthropods making up 59% of the species assemblage, mollusks and nemerteans each making up 13%, cnidarians and sipunculas each making up 6%, and one species of echinoderm at 3%. The arthropods *Cirolana harfordi* and *Exosphaeroma inornata* were the most abundance species, each with 167 collected. *Cirolana harfordi* and *Exosphaeroma inornata* are both isopods found in the intertidal zone, with *E. inornata* primarily found in California. The average species diversity calculated for this site was 0.85, with 377 collected specimens.
- iv. Station S4 – Sun Harbor Marina: Twenty-five species were observed at this station. Six phyla were present, with arthropods making up 35% of the species assemblage, annelids making up 32%, mollusks and chordates each making up 12%, bryozoans making up 6%, and one species of platyhelminthes at 3%. No single species had an overly dominant abundance, with all counts less than 20 per species. The arthropod *Heterophoxus ellisi*, an amphipod, was the most abundant species with 17 collected, while three species of polychaetes were the second most abundant, each with 10 specimens of *Cirratulus cf cingulatus*, *Pileolaria marginata*, and *Salmacina tribranchiata*. The average species diversity calculated for this site was 1.78, with 94 collected specimens.

- v. Station S5 – Harbor Island West Marina: Thirty-one species were observed at this station. Six phyla were present, with arthropods making up 49% of the species assemblage, annelids making up 22%, mollusks making up 19%, with chordates, nemerteans, phoronids, and platyhelminthes each making up 6% with a single species present. The arthropod *Monocorophium acherusicum* was the most abundant species with 216 collected. *Monocorophium acherusicum* is a tube-building amphipod associated with float fouling communities and soft substrates. The average species diversity calculated for this site was 1.20, with 357 collected specimens.
- b. Individual Stations in the Thermal Region
- i. Station S6 – Sunroad San Diego Marina: Sixty-three species were observed at this station. Five Phyla were present, with arthropods making up 42% of the species assemblage, annelids making up 41%, mollusks making up 8%, platyhelminthes making up 7%, and 2% of the species found were from the phylum aceolomorpha. No single species had an overly dominant abundance, with all counts 20 or less per species. Two species of polychaetes, *Pileolaria marginata* and *Syllis nipponica*, were the most abundant, each with 20 specimens. The average species diversity calculated for this site was 2.57, with 254 collected specimens.
- ii. Station S7 – Bayview Park Coronado: Thirty six species were observed at this station. Six phyla were present, with arthropods making up 63% of the species assemblage, mollusks making up 26%, platyhelminthes making up 5%, with chordates, cnidarians, and echinoderms each making up 2% of the assemblage with one species. The arthropods *Leptocheilia dubia* and *Foxiphalus cognatus* had the highest abundance with 50 and 46 individuals, respectively. *L. dubia* is a benthic tube builder found in soft sediments, and *F. cognatus* is a benthic amphipod. The average species diversity calculated for this site was 1.24, with 227 collected specimens.
- iii. Station S8 – San Diego Marriott Hotel and Marina: Sixty-three species were observed at this station. Eight phyla were present, with arthropods making up 42% of the species assemblage, annelids making up 30%, mollusks and chordates each making up 9%, platyhelminthes making up 4%, cnidarians making up 3%, and bryozoans and echinoderms each making up 1% of the assemblage with one species. The annelid *Salmacina tribranchiata* had the highest abundance with 1,002 individuals. *S. tribranchiata* is a tub-building polychaete. The average species diversity calculated for this site was 1.87, with 1,373 collected specimens.
- iv. Station S9 – Tidelands Park Coronado: Twenty-six species were observed at this station. Three phyla were present, with arthropods making up 70% of the species assemblage, mollusks making up 26%, and echinoderms making up 4%. The arthropods *Exosphaeroma inornata* and *Allorchestes angusta* had the highest abundance with 490 and 303 individuals, respectively. *E. inornata* is an isopod found primarily in California, and *A. angusta* is an intertidal amphipod. The average species diversity calculated for this site was 1.17, with 908 collected specimens.

- c. Individual Stations in the Seasonally Hypersaline Region
- i. Station S10 – Amphibious Base Fuel Piers: Seventeen species were observed at this station. Six phyla were present, with annelids and mollusks each making up 31% of the species assemblage, arthropods making up 28%, and chordates, nemerteans, and platyhelminthes each at 3%. The arthropod *Cirolana harfordi* was the most abundance species with 88 collected. *C. harfordi* is an isopod found in the intertidal zone. The average species diversity calculated for this site was 1.44, with 151 collected specimens.
 - ii. Station S11 – Chollas Creek: Twenty species were observed at this station. Two phyla were present, with annelids making up 95%, and mollusks making up 5% of the species assemblage. Abundance at this station was generally low, with *Platynereis bicanaliculata* having the highest count with 12 individuals. *P. bicanaliculata* is a tube-building polychaete commonly associated with eelgrass beds or algal holdfasts. The species diversity calculated for this site was 2.67, with 47 collected specimens.
 - iii. Station S12 – Paleta Creek: Sixty-eight species were observed at this station. Ten phyla were present, with annelids making up 40% of the species assemblage, arthropods making up 27%, mollusks making up 12% of the species assemblage, with platyhelminthes (4%), bryozoa (3%), chordata (3%), cnidaria (3%), nemertea (3%), porifera (3%), and echinoderms (1%) making up the remaining phyla. No single species had an overly dominant abundance, with all counts 17 or less per species. Two species of polychaetes, *Exogone lourei* and *Cirratulidae juveniles*, were the most abundant, each with 17 specimens. The average species diversity calculated for this site was 3.80, with 248 collected specimens.
 - iv. Station S14 – Fiddler’s Cove Marina: Eighty-three species were observed at this station. Ten phyla were present, with annelids making up 22% of the species assemblage, arthropods making up 33%, mollusks making up 26%, chordates making up 9%, nemerteans making up 3%, with byrozoa, cnidaria, echinoderms, phoronida, and porifera each making up 1%. The mollusk *Musculista senhousia* was the most abundant species with 501 individuals. *M. senhousia*, also known as the “Asian Mussel,” is a small greenish-brown exotic mussel. It was first reported in San Diego Bay in 1976 (Carlton, 1979). The average species diversity calculated for this site was 1.58, with 1,004 collected specimens.
- d. Individual Stations in the Estuarine Region
- i. Station S16 – Grand Caribe Shoreline Park: Fifty-one species were observed at this station. Ten Phyla were present, with annelids making up 32% of the species assemblage, arthropods making up 21%, chordates making up 21%, mollusks making up 15%, and each with 2% were bryozoa , cnidaria, echinoderms, nemerteans, phoronidans, and platyhelminthes. No single species had an overly dominant abundance. The tunicate *Ciona intestinalis* and mollusk *Musculista senhousia* were the most abundant with 20 and 15 counts, respectively. *C. intestinalis* is a solitary tunicate that may form dense aggregates on floating/ submerged substrates. *C. intestinalis* is an exotic species first introduced to the West Coast of the United States via San Diego Bay in 1897 (Fofonoff et al., 2003). *M. senhousia* is a small greenish-brown exotic mussel, also known as the “Asian mussel.” It was first reported in San Diego Bay in 1976 (Carlton, 1979).

The average species diversity calculated for this site was 2.56, with 222 collected specimens.

- ii. Station S17 – Chula Vista Harbor Pier: Twenty-two species were observed at this station. Six phyla were present, with arthropods making up 40% of the species assemblage, mollusks making up 37%, cnidarians and nemerteans making up 9%, and chordates and echinoderms each making up 3%. No single species had an overly dominant abundance. The mollusk *Musculista senhousia* had 41 counts, the fish *Leuresthes tenuis* had 25 individuals, and the arthropod *Nebalia pugettensis complex* had 23. *M. senhousia* is a small greenish-brown exotic mussel, also known as the “Asian mussel.” It was first reported in San Diego Bay in 1976 (Carlton, 1979). *L. tenuis* is the California grunion, a small fish with a bluish-green back and silvery sides and bellies. *N. pugettensis complex* is a small crustacean (Leptostraca) often associated with highly organic sediments. The average species diversity calculated for this site was 1.42, with 128 collected specimens.
- iii. Station S18 – South Bay Marine Biological Center: Thirty-eight species were observed at this station. Five phyla were present, with annelids making up 41% of the species assemblage, arthropods making up 35%, mollusks making up 19%, and bryozoans and echinoderms each making up 3%. The mollusk *Musculista senhousia* had the greatest abundance with 64 individuals. *M. senhousia* is a small greenish-brown exotic mussel, also known as the “Asian mussel.” It was first reported in San Diego Bay in 1976 (Carlton, 1979). The average species diversity calculated for this site was 1.54, with 240 collected specimens.

3. Among Site Comparisons

The community metrics varied across the different study stations. In general, there was a lot of inter-station variability with no general patterns across various regions of the bay. The level of effort may have varied slightly at the different stations and should be considered when drawing comparisons between stations using these community metrics.

Summaries of species richness, abundance, and Shannon diversity are shown in Table 28. Metrics are separated by sampling methodology/habitat.

- a. Species Richness: Species richness varied among the different study stations and regions. In general, richness was highest from the infaunal and fouling communities, compared to samples collected using the beach seine and from within the intertidal zone (Figure 18). Richness also varied at stations within the regions (Figure 19). The greatest number of species was observed at stations S14, with 83 species, the majority of which come from the fouling community. Stations S6 and S8, both with 63 species, and S14, with 62 species, primarily from the fouling community, were the stations with the next highest species richness.

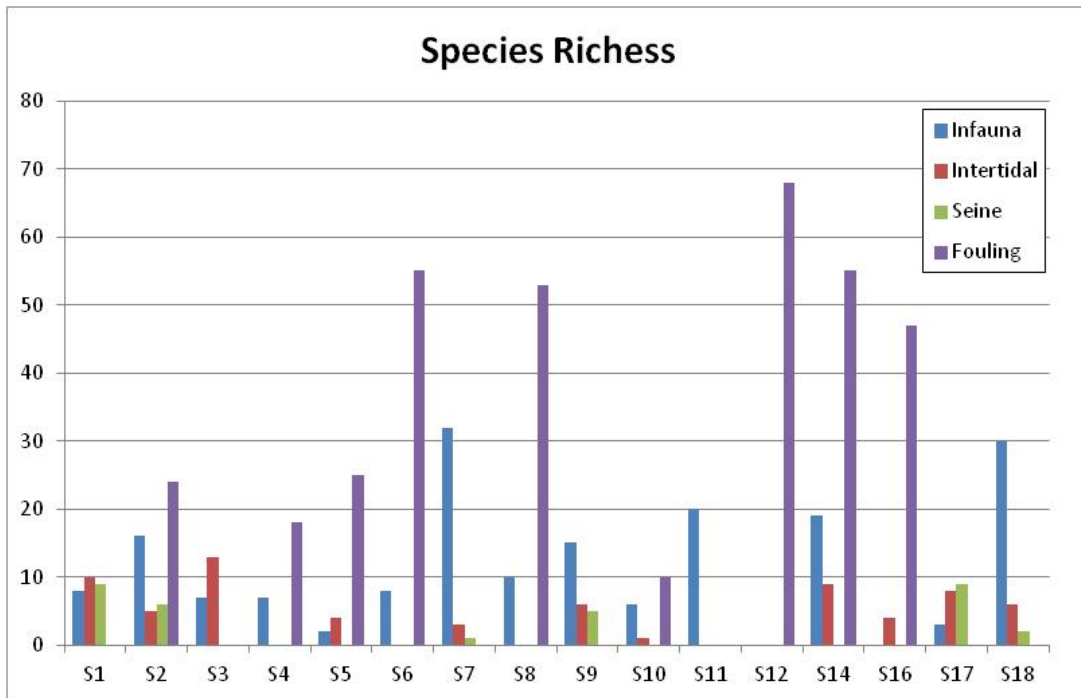


Figure 18. Species richness from the different study stations per different habitat/sampling methods.

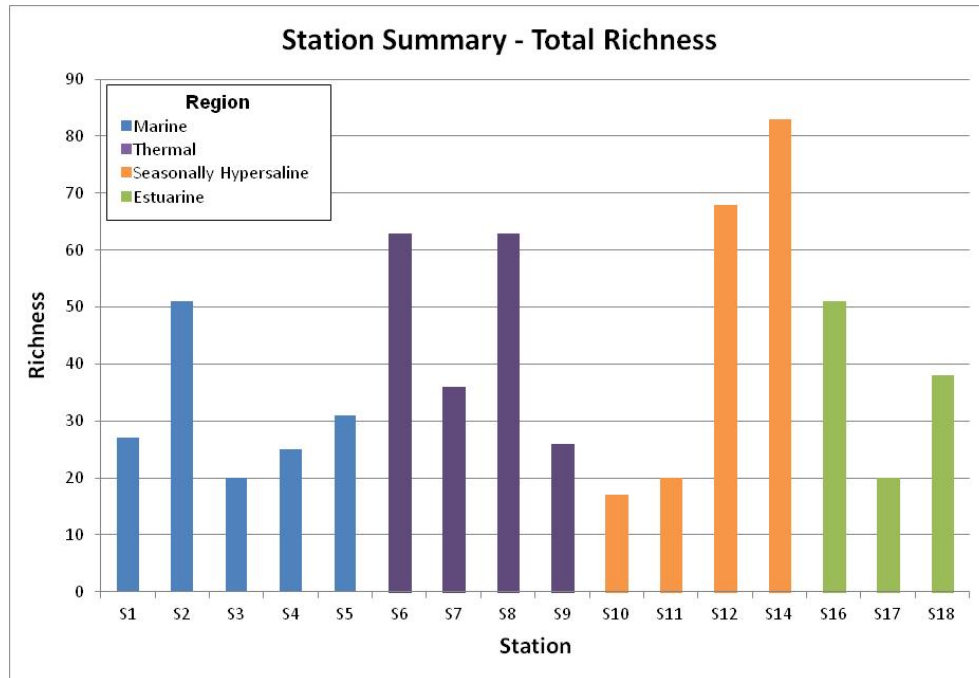


Figure 19. Summary of total species richness found at each site by region.

- b. Species Abundance: Figure 20 shows the species abundance at all study stations by habitat/ sampling methodology and Figure 21 show totals for each station by region. Abundance, in terms of individual organisms, was between approximately 50–500, with the exception of three stations. Stations S8 (Marriott Marina), S9 (Tidelands Park Coronado) and S14 (Fiddlers Cove Marina) had greater abundances with 1,373, 1,004, and 908 individuals, respectively. The increased station abundances were due to one or two individual species: a polychaete (*Salmacina tribranchiata*) at station S8, two arthropods (*Exosphaeroma inornata* and *Allorchestes angusta*) at station S9, and the mollusk *Musculista senhousia* at station S14. High organism abundance at S8 was almost exclusively from the infaunal community, while S9 and S14 where more evenly spread among the various habitat/sampling methods.

Table 28. Community metrics summary for each station

	Station	Infauna			Intertidal			Beach Seine			Fouling			Station Summary		
		Richness	Abundance	Diversity	Richness	Abundance	Diversity	Richness	Abundance	Diversity	Richness	Abundance	Diversity	Total Richness	Total Abundance	Average Diversity
Marine Region	S1	8	27	1.64	10	67	1.86	9	411	0.44	----	----	----	27	505	1.31
	S2	16	54	2.36	5	151	0.29	6	14	1.47	24	94	2.86	51	313	1.74
	S3	7	9	0.61	13	368	1.09	----	----	----	----	----	----	20	377	0.85
	S4	7	26	1.03	----	----	----	----	----	----	18	68	2.53	25	94	1.78
	S5	2	3	0.64	4	222	0.15	----	----	----	25	132	2.81	31	357	1.20
Thermal Region	S6	8	15	1.71	----	----	----	----	----	----	55	239	3.43	63	254	2.57
	S7	32	200	2.64	3	3	1.10	1	24	0.00	----	----	----	36	227	1.24
	S8	10	39	1.73	----	----	----	----	----	----	53	1334	2.00	63	1373	1.87
	S9	15	48	2.21	6	356	0.45	5	504	0.83	----	----	----	26	908	1.17
Seasonal Hypersaline	S10	6	13	1.48	1	117	0.77	----	----	----	10	21	2.06	17	151	1.44
	S11	20	47	2.67	----	----	----	----	----	----	----	----	----	20	47	2.67
	S12	----	----	----	----	----	----	----	----	----	68	248	3.80	68	248	3.80
	S14	19	84	2.37	9	511	0.14	----	----	----	55	409	2.25	83	1004	1.58
Estuarine	S16	----	----	----	4	48	0.37	----	----	----	47	203	3.45	51	251	1.91
	S17	3	5	1.05	8	44	1.36	9	79	1.68	----	----	----	20	128	1.37
	S18	30	182	2.81	6	39	1.28	2	19	0.51	----	----	----	38	240	1.54

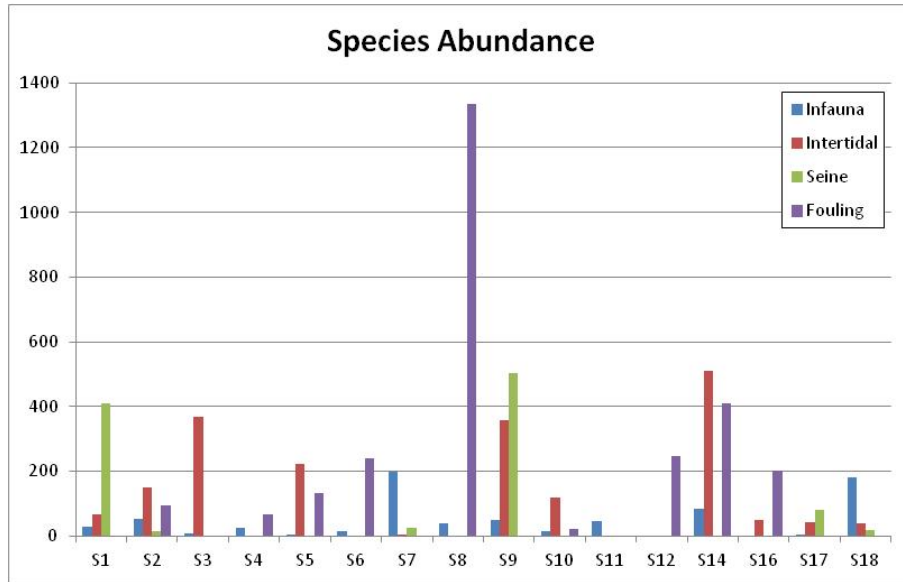


Figure 20. Species abundance at stations by habitat/sampling method.

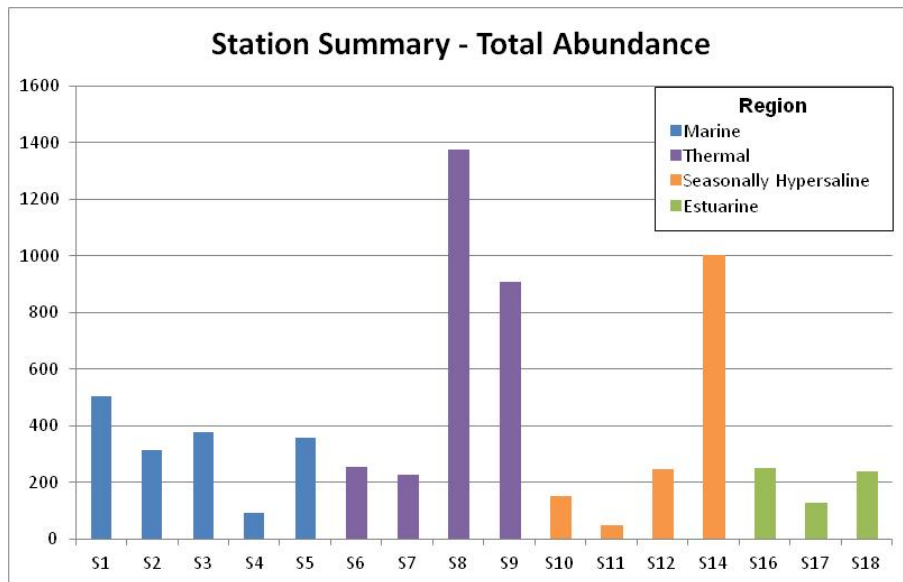


Figure 21. Total species abundance at each station by region.

- c. **Species Diversity:** Species diversity (Figure 22 and Figure 23) also varied between the different stations, and by habitat/sampling methodologies. As with the other metrics, there were no clear spatial patterns of diversity across the stations or regions of the bay. Of the various habitats sampled, the fouling community tended to be the most diverse with the infaunal community having the second highest diversity (Figure 22). The high average diversity at station S12 (Figure 23) is slightly biased because only habitat/sampling method (fouling community) is represented.

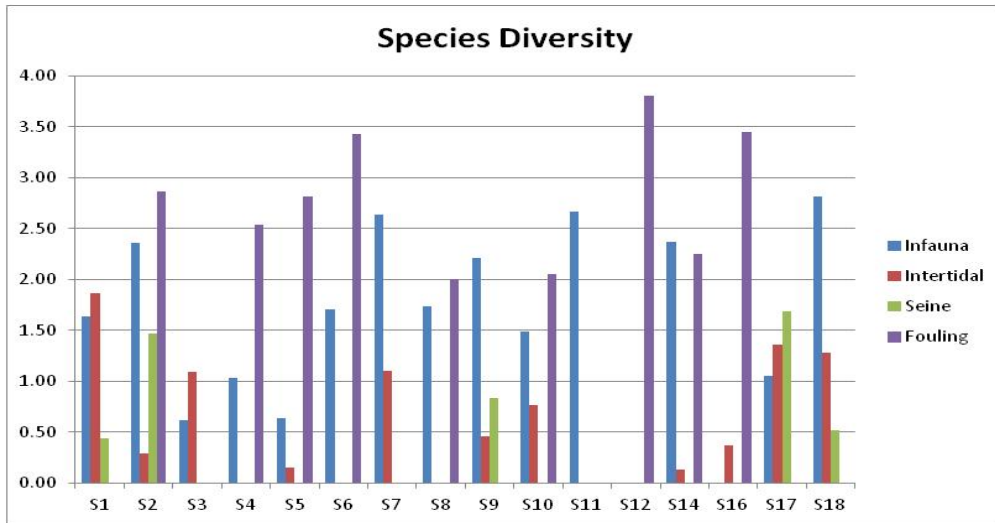


Figure 22. Shannon Weiner Diversity at each station by sampling method.

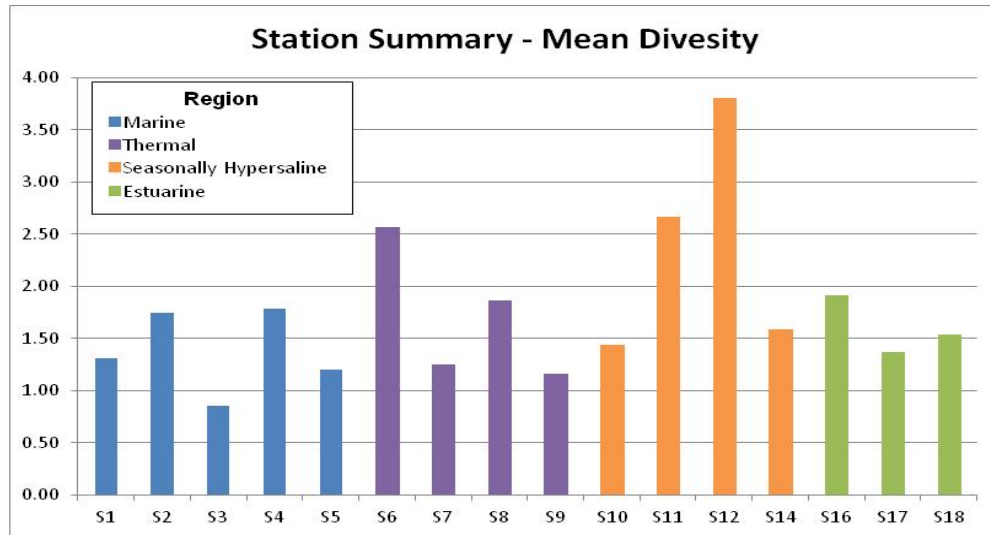


Figure 23. Average Shannon-Weiner Diversity at each station by region.

- d. **Species Assemblage:** The species assemblage in terms of representative phyla did show some spatial variability throughout the bay (Figure 24). The most representative phyla were arthropoda, annelida, and mollusca. Arthropods and mollusks were the most predominant phyla at the three stations closest to the mouth of the bay, S1, S2, and S3, where no annelids were observed. Annelids made up a greater proportion of the species assemblage at sites towards the back of the bay. Annelids made up 70% and 90% of the species at stations S9 and S11, respectively, although they were absent entirely from station S17. Mollusks were the only phylum found at all of the stations, although with a more modest percentage than arthropods or annelids, ranging from 5–37% with an average of 19%.

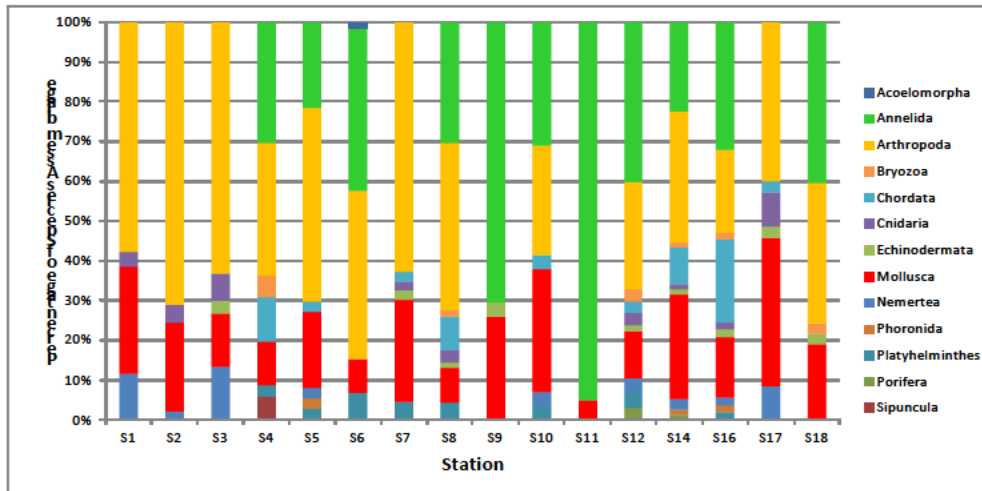


Figure 24. Cumulative percentage of species assemblage by major phyla.

One notable spatial trend for species assemblage was the abundance of the exotic Asian Mussel, *Musculista senhousia*. This species was observed at 8 of the 16 stations, with a greater abundance in the back bay. *M. senhousia* made up less than 5% of the total abundance at stations S2, S4, S5, S6, S7, S8, and S10 and was generally the most abundant species at Stations S14 (50%, n = 501), S16 (7%, n = 15), S17 (32%, n = 41), and S18 (27%, n = 64).

Similarity indices comparing two stations to each other ranged from 0.00–0.65 (Table 29). Similarity indices found within the 90th percentile of the entire similarity data set were between 0.34 and 0.65. Evaluating the 90th percentile values, there appears to be two clusters of values (Table 28) that display a strong spatial trend. Stations at the extreme front and back bay were most similar to each other, with sites S1, S2, and S3 as one group in the front bay and S14, S17, and S18 in the back bay. S16 compared to the surrounding S14, S17, and S18 was lower with indices ranging from 0.22–0.27. Additional stations exhibited a high degree of similarity with a strong spatial relationship. Site S10 was more similar to stations in the front bay (S1, S2, and S3) than stations found in closer proximity. Station S9 was similar to S3, but not S2, S1, or S10. The pairs S11/S5 and S6/S12 were more similar to each other than adjacent site, respectively.

Table 29. Similarity indices for pairs of stations throughout San Diego Bay. Cells highlighted red are from the 90th percentile.

	Marine					Thermal				S. HyperS			Estuarine			
	S1	S2	S3	S4	S5	S6	S7	S8	S9	S10	S11	S12	S14	S16	S17	S18
S1	----															
S2	0.62	----														
S3	0.54	0.49	----													
S4	0.07	0.08	0.05	----												
S5	0.03	0.14	0.05	0.30	----											
S6	0.06	0.15	0.04	0.30	0.28	----										
S7	0.15	0.34	0.22	0.12	0.19	0.08	----									
S8	0.02	0.10	0.02	0.21	0.23	0.24	0.10	----								
S9	0.11	0.09	0.56	0.04	0.02	0.04	0.18	0.02	----							
S10	0.65	0.54	0.58	0.10	0.06	0.15	0.16	0.06	0.16	----						
S11	0.03	0.00	0.00	0.00	0.60	0.20	0.01	0.05	0.01	0.06	----					
S12	0.00	0.10	0.00	0.22	0.27	0.40	0.07	0.17	0.00	0.06	0.27	----				
S14	0.04	0.09	0.05	0.14	0.12	0.12	0.14	0.10	0.09	0.06	0.03	0.23	----			
S16	0.09	0.11	0.10	0.20	0.17	0.22	0.16	0.14	0.10	0.17	0.14	0.28	0.22	----		
S17	0.07	0.07	0.07	0.09	0.09	0.09	0.13	0.07	0.05	0.10	0.00	0.05	0.38	0.22	----	
S18	0.04	0.05	0.05	0.18	0.09	0.11	0.21	0.07	0.07	0.08	0.03	0.11	0.46	0.27	0.42	----

D. Grain Size Analysis

Excluding the marine management region, two sediment samples were collected and analyzed for grain size in each of the four management regions. A summary of samples, station location, and percent dry mass sample per size class is provided in Table 30. In general, a unique distribution of size class was observed for each of the four hydrographic regions (Figure 25), along with differences in median grain size (Figure 26 and Figure 27). Distributions of size class by region were as follows: the median grain size (M_d) for the marine region was 59 μm with a fairly even distribution of sand, silts, and fines for two of the three samples analyzed. The sample analyzed from the S5 sampling station-Harbor Island West Marina had a distribution signature more similar to stations S7 and S9 in the Thermal region in that it consisted predominantly of sand with some clay and silt. The Thermal region had an M_d that ranged from 125–179 μm with predominate sand size fraction. The Hypersaline region had an M_d that ranged from 89–140 μm with only sands and fines size fractions present. The Estuarine region had an M_d that ranged from 37–69 μm with a fairly even distribution of sand, silt, and fines in the first station sample (S16) and predominantly fines in the second station sample (S17).

Table 30. Summary of Grain Size Analysis by Station and Region.

Size Class	Size Range	Sample Station								
		Marine Region			Thermal Region		HyperSaline Region		Eustaurine Region	
		S2	S-4	S5	S-7	S-9	S-10	S-14	S-16	S-17
V. Coarse Sand	2 - 1 mm	3.3%	0.0%	46.9%	1.5%	0.0%	0.1%	0.3%	0.6%	0.1%
Coarse Sand	1000 - 500 μm	3.6%	0.1%	18.5%	2.8%	0.1%	0.2%	0.5%	0.6%	0.0%
Medium Sand	500 - 250 μm	13.9%	0.7%	12.5%	14.3%	2.6%	2.0%	11.4%	3.4%	0.3%
Fine sand	250 - 125 μm	25.7%	10.7%	13.9%	66.1%	47.5%	36.1%	45.3%	18.2%	0.4%
V. Fine Sand	125 - 63 μm	14.3%	30.0%	5.4%	9.5%	30.1%	24.3%	15.7%	32.2%	3.3%
Coarse Silt	63 - 33 μm	39.2%	46.8%	2.6%	0.2%	na	na	na	29.1%	60.8%
Medium Silt	33 - 15.6 μm	na	1.3%	na	0.6%	na	na	na	3.0%	3.9%
Fine Silt	15.6 - 7.8 μm	na	1.6%	na	1.9%	na	na	na	1.8%	4.9%
V. Fine Silt	7.8 - 3.9 μm	na	0.6%	na	0.0%	na	na	na	1.8%	1.9%
Clay	< 3.9 μm	na	8.1%	na	3.1%	na	na	na	9.5%	24.4%
Sand	2 - .063 mm	60.8%	41.5%	97.2%	94.1%	80.3%	62.6%	73.3%	55.0%	4.1%
Silt	63 - 3.9 μm	39.2%	50.4%	2.6%	2.7%	na	na	na	35.6%	71.5%
Clay	< 3.9 μm	na	8.1%	na	3.1%	na	na	na	9.5%	24.4%
Fines (< 63 μm)	< 63 μm	39.2%	58.5%	2.6%	5.9%	19.7%	37.4%	26.7%	45.0%	95.9%
Graphical Mean (M_z), μm			59.1		185.7	112.4	89.8	116.2	70.6	14.5
Median Size (M_d), μm			55.1		179.8	125.4	89.6	140.3	69.6	37.0

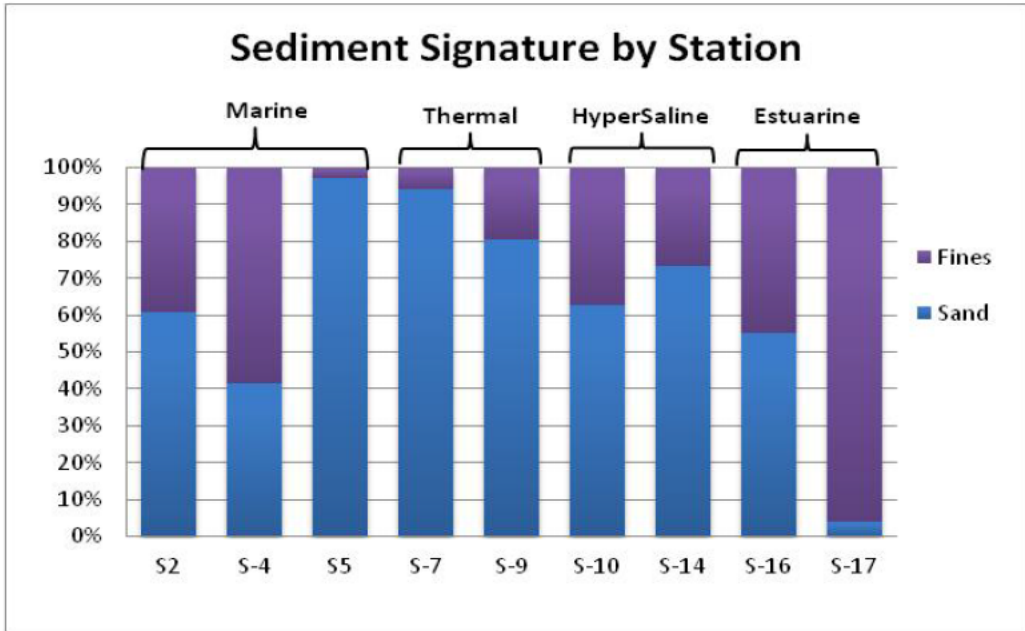


Figure 25. Sediment signature by station.

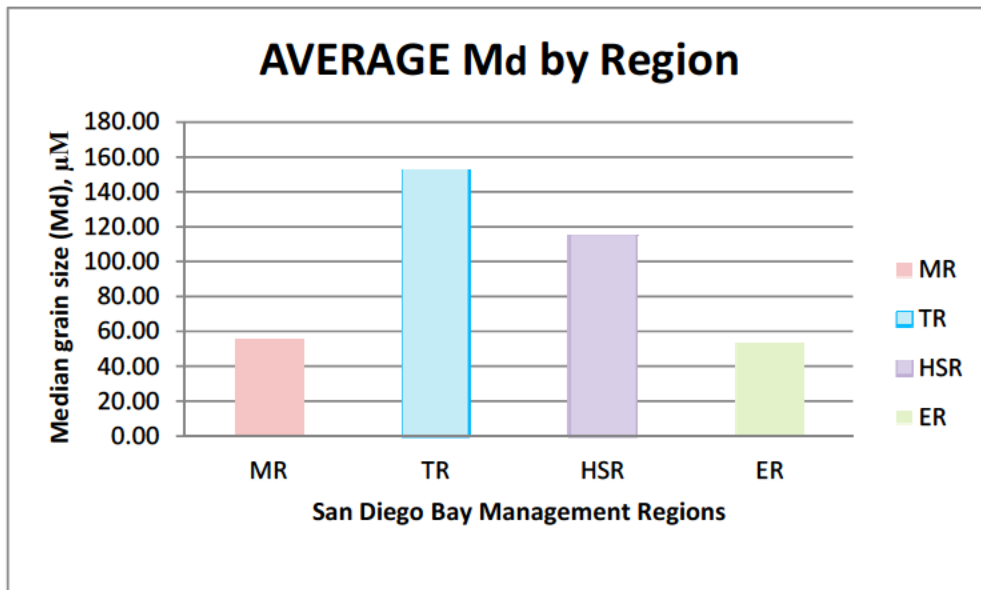


Figure 26. Average M_d by region.

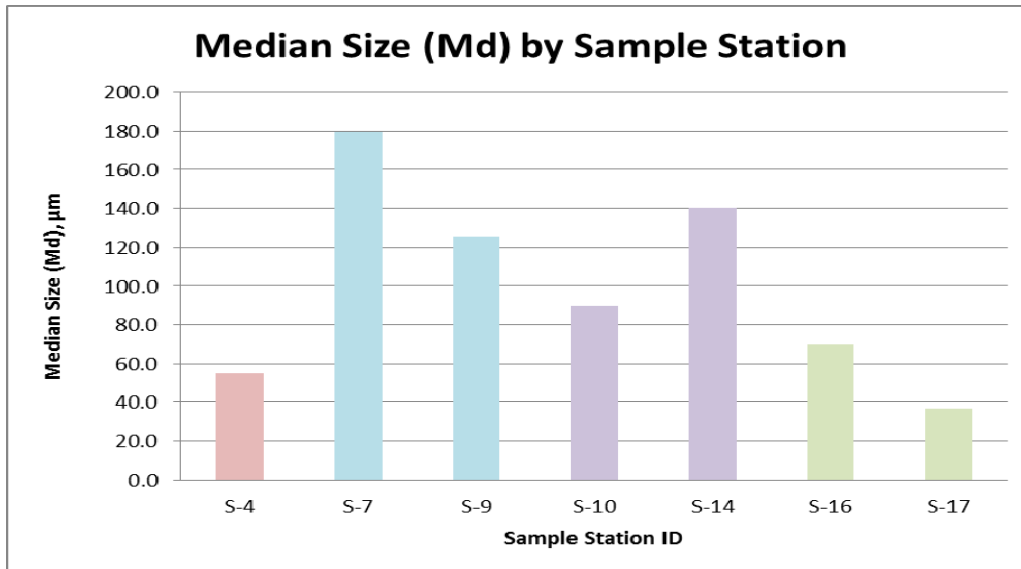


Figure 27. Average M_d by sampling station.

E. DNA Bar Code Analysis

All vouchers were initially amplified using universal 18S primers to determine quality of DNA. Of the 20 samples submitted for analysis, 11 were successfully amplified via 18S. Thus, nine vouchers failed to amplify with the universal 18S primers. This equates to a 55% sequencing success. The remaining 11 samples were then amplified to produce a ~ 650-bp band using universal CO1 primers. DNA Barcoding sequencing results are provided in Appendix G. The 11 658-bp sequences were then analyzed using NCBI Blast[®] Search, Barcode of Life Data Systems (BOLD) database search, and ClustalW Multiple Sequence Alignment Software to identify nearest sequence matches. Blast and BOLD analysis failed to identify one of the eleven 650-bp sequences (Sample EIS-GL-0021); however, (BOLD) database analysis did indicate a 74% similarity to specimens in the *Chordata* phylum. ClustalW comparisons to the remaining 10 nearest sequence matches are provided in Appendix H. A summary of voucher identification based on DNA barcoding results are provided in Table 31. Of the 11 650-bp sequences, 10 (91%) were successfully identified up to a similar Order classification as that classified by the expert Taxonomist panel. Further, 73% were identified \geq to a genus classification, with 64% (7) matching similar species classification as that identified by the expert taxonomist panel. Blast[®] and ClustalW analysis results as they compare to expert panel classification is also provided in Table 31.

Table 31. Summary of DNA Bar Code Analysis.

SAMPLE ID	Taxonomist Identification		18s Amp	Blast® and ClustalW® Analysis			BOLD®	
	Phylum	Species		BLAST® ID	bp	Similarity		Match to Taxonomist ID
EIS-GL-0027	Chordata	Ascidia ceratodes						
EIS-JL-0185	Gastropoda	Diaulula sandiegensis	X	Diaulula sandiegensis	658	93%	Diaulula sandiegensis	to species
EIS-TP-0025	Arthropod	Amphideutopus oculus	X	Amphipoda	658	82%	Amphideutopus oculus (99%)	to species
EIS-TP-0065	Arthropod	Jassa slatteryi	X	Jassa slatteryi	658	98-99%	Jassa slatteryi	to species
EIS-GL-0021	Chordata	Aplidium californicum	X	no match*	658		Chordata, Ascidiacea (74%)	to class
EIS-JL-0014	Pelecypoda	Lyonsia californica						
EIS-LH-0137	Annelida	Syllis nipponica						
EIS-LH-0141	Annelida	Platynereis bicanaliculata						
EIS-TP-0012	Arthropod	Neotrypaea californiensis	X	Neotrypaea californiensis	658	99-100%	Malacostraca, Decapoda (88%)	to species
EIS-GL-0014	Chordata	Botrylloides diegensis	X	Botrylloides diegensis	658	84-100%	no match	to species
EIS-JL-0040	Bryozoa	Smittoidea prolifica						
EIS-JL-0138	Cephalopoda	Octopus bimaculatus/bimaculoides	X	Octopus bimaculoides	658	90-100%	Octopus bimaculoides	to species
EIS-JL-0230	Pelecypoda	Chione undatella	X	Chione subimbricata	658	93%	Chione subimbricata	to genus
EIS-LH-0061	Annelida	Nicolea sp A Hamis						
EIS-TP-0016	Pycnogonida	Rhynchothorax philopsammum	X	Ammonothea hilgendorffi	658	87-97%	Pantopoda	to order
EIS-GL-0001	Chordata	Botryllus schlosseri						
EIS-GL-0008	Chordata	Molgula ficus						
EIS-JL-0038	Bryozoa	Scrupocellaria sp						
EIS-LH-0002	Annelida	Harmothoe imbricata complex	X	Harmothoe imbricata	658	87%	Harmothoe imbricata	to species
EIS-TP-0043	Arthropod	Ampithoe valida	X	Ampithoe longimana	658	89%	Ampithoe longimana	to genus

(*) while no match was found a search in Barcode of Life Data Systems (BOLD) database indicated 74% similarity to specimens in the *Chordata* phylum

F. Voucher Collection and Archiving

As part of this effort, representative examples of indigenous, non-indigenous, cryptogenic, and invasive species were collected from the various sampling locations within the four hydrographic regions. Two hundred and eighty-four voucher samples have been collected and are being stored at -80 °C at SSC Pacific. A subset of vouchers (20) were sent to the Smithsonian Institution, USA for DNA barcoding sequencing (refer to DNA Bar Coding section under Methods). In addition, an additional 150 archiving samples were collected from taxa of interest and were donated to the natural history museums, Los Angeles (Appendix I).

DISCUSSION

This report has summarized an ecological index, early detection survey. The purpose of this study was to identify and catalog native and non-indigenous species near naval facilities within the four hydrographic regions of the San Diego Bay. The work that was conducted and described herein was similar to a Rapid Assessment Survey (RAS) in that a team of taxonomists was assembled for identification of live specimens over a five days. In addition, post-analysis of preserved benthic samples was also conducted. The main focus of this study was to identify native, introduced, and cryptogenic species present on multiple natural and artificial habitats within these regions. Tasking completed under this effort was broken down into four key objectives: (1) to understand and summarize historic data on species distribution, including presence of exotic species, within the four regions of the Bay; (2) to plan and execute a modified Rapid Assessment (5-day) Survey using a random sampling strategy for identification of species present within the four regions (marine, thermal, seasonally hypersaline, and estuarine); (3) to assess feasibility of the use of DNA barcoding as a tool for augmenting species identification in a rapid assessment platform; and (4) to provide a summary of invertebrate species distribution relative to regions and Navy facilities. The purpose of this section is to briefly discuss some of the key findings under the four major tasking objectives of this effort.

HISTORICAL BACKGROUND DATA SURVEY

The principal goals of the historical review was to provide a synopsis of the total species abundance and types (species) of benthic organisms that were found in San Diego Bay over the past 15 years and to evaluate the EIS Study results in context with the historical data. Five different benthic-related studies performed over the past 15 years in San Diego Bay were queried for benthic infaunal data. Sampling locations and abundance data from each of these studies were mapped and plotted along with the results from the current EIS study. As seen in the previous section, San Diego Bay has been widely sampled over the past 15 years with locations spread throughout the bay, although the back part of the bay has been sampled the least. Benthic abundance ranges from greater than five organisms/sample to greater than 5,000 organisms/sample. Additional benthic community metrics were reported as well, but are not discussed in detail because it is beyond the scope of this study. The benthic abundance data from the EIS study ranged from 3–182 organisms/sample, which in general trends lower than results observed from the historical studies. However, due to differences in which this study was performed (i.e., rapid survey) out versus traditional benthic community studies, it is not possible to draw any detailed conclusions about overall changes in benthic abundance in the bay. A more detailed analysis must be performed along with the collection of more data over time.

Rapid Assessment Survey/Species Identification

Results of this study have yielded 6,477 organisms, with 299 species represented from 13 phyla collected and identified at least to the genus level. Organisms were identified using a various collection methods (seine, time searches on rip-rap and piers, beach sieves, settling plates, and sediment grabs), and vouchers were collected from various natural and artificial habitats. Overall species identified in this study were similar to those reported in previous studies; however, there were some differences in spatial distribution throughout the bay. These differences are assumed to be related to differences in sampling methodologies and shifts in substrate types. Differences in substrate type are a known variable in shaping the associated invertebrate community (INRMP 2013). In addition, variances in types of assemblages of species also differ with respect to type of sampling location. Specifically, those species found on man-made substrates can be quite different

from those found on more natural habitat (Lambert and Lambert, 2003; Maloney, 2007; Wasson, Fenm and Pearse, 2005; Glasby, Connell, Holloway, and Hewitt, 2006). A comprehensive discussion of differences is beyond the scope of this effort; however, just a few factors, such as grain size and species status, will be described here.

A. Grain Size Analysis

Grain size plays an important role in infaunal abundance and diversity. The majority of invertebrates that comprise the 650-plus species known to exist in the bay (INRMP, 2013) live in the soft bottom intertidal and subtidal habitats of the bay. The subtidal bottom of the bay consists primarily of unconsolidated sediments including a mixture of sand, silt, and clay. Several studies have indicated a direct relationship between distribution of sediment mixtures and diversity of infaunal species present (Snelgrove and Butman, 1994; Heino, 2009; Whittiker et al., 2003). In this study, sediment samples for grain-size analysis were collected from a minimum of two locations within each of the four hydrographic regions of the bay. Grain size distributions varied widely among the various regions and also at stations within regions. Within the Marine Region stations, S2 and S4, had more similar grain-size distributions, while S5 was almost entirely composed of sand. Species diversity was higher at sights S2 and more similar at S4 and S5. Annelids were only present at station S5, comprising approximately 20% of the species assemblage. Although, looking at Thermal region station S7 had a similar grain-size distribution to station S5, but the species assemblage and diversity at S7 were much higher. Additionally, stations S7 and S9 had similar grainsize distributions but very different species assemblages. Station S9 was mostly comprised of annelids and was the only station that did not have arthropods present. Stations S10 and S14 had similar grain-size distributions and species assemblages, but differed in abundance and primarily due to high numbers of the exotic mussel *Musculista senhousia* (Asian mussel), which was not present at Station S10. Within the Estuarine hydrographic area, grain-size distributions varied widely, with S16 having a relatively even mix of sand and fines and station S17 composed mostly of fines. Both grain-size distribution and community metrics varied between the different stations and hydrographic regions and grain size does not appear to be a strong controlling factor influencing the overall biological community structure. However, due to the limited sample size and sample composition, a more extensive comparison and analysis was not feasible.

B. Native, Introduced, Cryptogenic Species of Interest

Many researchers have indicated that the introduction and spread of non-indigenous marine organisms may be one of the greatest threats to the sustainability of complex marine ecosystems such as San Diego Bay (Zedler, 1992; Maloney et al., 2007; Crooks, 1996; INRMP, 2013). As part of the cataloging and identification process described under this study, some species were further identified as native, introduced, and cryptogenic. Definitions of native, introduced, and cryptogenic were known to differ between researchers. Thus, for this study, definitions are similar to those defined by Maloney et al. (2007). For example, introduced is defined as either a species not previously identified as native to California, it colonizes a new area that it was not previously in, the extension of its range can be linked either directly and/or indirectly to human activity, and/or there is no geographic link between native area and new colonization. Cryptogenic is defined as a species that is neither demonstrably native nor introduced. In general, this is considered a catchall for any species with insufficient life history documentation (Carlton, 1996 and Maloney et al., 2007). Unresolved or unknown is defined as a species that could not be identified beyond family, class, order, or genus level or confidently classified as native, introduced, or cryptogenic, and/or status of species was not provided by taxonomist or is not currently available. Lastly the term invasive typically refers to an introduced species that has caused either ecologic or economic damage to an ecosystem. However, not all introduced species are

considered invasive. In fact, it has been suggested that the term “invasive” tends to be subjective (Maloney et al., 2007); thus, for this report this term will not be used.

Of the samples collected and identified under this study, 34 were classified as introduced, 23 were classified as cryptogenic, and 74 as native to the San Diego Bay. In addition, 39 samples were not identified to the species level and were thus identified as unresolved or unknown. A summary table is provided in Appendix J. A brief discussion of interesting findings of major phyla and the identifying taxonomist is provided herein.

1. **Ascidians:** The fauna identified and recorded by Gretchen Lambert were mainly from the 16 settling plates. Those ascidian species identified were predominantly what was previously reported in the San Diego Bay (Lambert and Lambert, 1998, 2003 and Maloney et al., 2007). Table 32 provides a summary of introduced species previously reported by Lambert and Lambert (1998, 2003). All but two of the introduced species (*Ascidia* sp. and *Styela canopus*) reported by Lambert were identified in this study (Table 33). This may be due in part to sampling time. *Styela canopus* and *Ascidia* sp. are typically found more localized to the seasonally hypersaline region of the bay during the fall and spring (Lambert & Lambert, 2003). Two additional species, *Diplosoma listerianum* and *Molgula fiscus*, not reported by Lambert, were identified in this study. Both of these species were previously reported by Maloney et al. (2007) and are classified as cryptogenic and introduced respectively. While no new introduced ascidian species were identified, there did appear to be some differences in species distribution across the four regions of the bay. In addition, of the 18 species identified, 14 were classified as introduced. A summary of species identified in this study as compared to previous studies is provided Table 33. These differences could indicate movement within the bay or related to variations in seasonal populations. Lambert and Lambert (2003) indicated that some species are more abundant in the fall versus the spring. Further analysis would be required to understand the source of these distribution differences.
2. **Crustaceans:** The list of crustacean species identified by Tony Philips in this survey are those that are commonly found in the shallow subtidal and intertidal rip-rap samples collected from the bays and harbors in the Southern California Bight (SCB). One species, *Leucothoe nagatai*, Ishimaru 1985 (Figure 28), is an invasive species that has only recently been identified in the SCB. It has been confused with a native species, *Leucothoe alata*, Barnard 1959, in collections preserved in formalin and then 70% ethanol. This method of preservation bleaches out the normal reddish-brown polka dot pigmentation seen in live material of *L. nagatai*, not seen in *L. alata*. If a taxonomist is unaware of the morphological differences between the two species, it would commonly be called *L. alata*. Two other species, *Colomastix* sp 1 and *Paradexamine* sp SD1, are provisional species erected by SCAMIT taxonomists. These have been recognized as new species and until they are described as such, a provisional listing has been created to identify these animals in taxonomic listings.

Table 32. Introduced Species Identified in the San Diego Bay between 1994–2000 (Lambert & Lambert, 1998, 2003).

Sampling Location	24th street (National City)	Fiddlers cove, Navy Yacht club (Coronado)	J St (Chula vista)	Harbor Island	Shelter Island
Introduced Species	Seasonally Hypersaline	Estuarine	Marine		
<i>Ciona intestinalis</i>	x	x	x	x	x
<i>Ciona savignyi</i>	x	x	x	x	x
<i>Ascidia Zara</i>		x?	x?	x	x
<i>Ascidia sp</i>	x?	x	x	x	x
<i>Styela canopus</i>	x	x	x	x	x
<i>Styela clava</i>	x	x	x	x	x
<i>Styela plicata</i>	x	x	x	x	x
<i>Polyandrocarpa zorritensis</i>	x	x	x	x	x
<i>Botryllus schlosseri</i>	x?	x	x	x	x
<i>Botrylloides perspicuum</i>	x				x
<i>Botrylloides violaceus</i>		x	x	x	x
<i>Symplegma reptans</i>		x			x
<i>Microcosmus squamiger</i>	x	x	x	x	x
<i>Molgula manhattensis</i>					

Data is compiled from Lambert and Lambert 1998 and 2003. Covers surveys conducted between 1994-2000. (?) indicates that species was identified at this location at least once between 1994-2000 surveys

Table 33. All ascidian species identified in EIS 2011 Study, along with comparison of regions identified in between this study and Lambert Study (1998, 2003).

Identified Species	San Diego Bay Hydrographic Regions					Pre ID Region(a)
	Estuarine	Marine	Seasonally Hypersaline	Thermal	Native	
<i>Aplidium californicum</i>			x	x	x	
<i>Ascidia ceratodes</i>		x			x	
<i>Ascidia zara</i>				x		Estuarine [^] , Marine, Seasonally Hypersaline [^]
<i>Botrylloides diegensis</i>			x		x	
<i>Botrylloides perspicuum</i>			x			Seasonally Hypersaline
<i>Botrylloides violaceus</i>				x		Seasonally Hypersaline, Estuarine, Marine
<i>Botryllus schlosseri</i>	x		x			Estuarine, Marine, Seasonally Hypersaline [^]
<i>Ciona intestinalis</i>	x		x			Estuarine, Marine, Seasonally Hypersaline
<i>Ciona savignyi</i>	x		x			Estuarine, Marine, Seasonally Hypersaline
<i>Distaplia occidentalis</i>			x		x	
<i>Diplosoma listerianum*</i>	x			x		
<i>Microcosmus squamiger</i>	x			x		Estuarine, Marine, Seasonally Hypersaline
<i>Molgula ficus*</i>	x			x		
<i>Perophora annectens</i>	x				x	
<i>Polyandrocarpa zorritensis</i>	x	x				Estuarine, Marine, Seasonally Hypersaline
<i>Styela clava</i>	x	x				Estuarine, Marine, Seasonally Hypersaline
<i>Styela plicata</i>	x	x	x			Estuarine, Marine, Seasonally Hypersaline
<i>Symplegma reptans</i>	x	x	x			Seasonally Hypersaline

(Pre ID)= Previously Identified under Lambert studies. Introduced species previously identified by Lambert and Lambert but not found in this study were *Ascidia sp* and *Styela canopus*; (*) While both *Diplosoma listerianum* and *Molgula ficus* were not reported in Lambert they were both reported in Maloney et al 2007. *Diplosoma listerianum* was classified as a cryptogenic species and *Molgula ficus* as an introduced species. *Molgula ficus* previously reported in estuarine region by Maloney et al (2007). (a) Note, the Lambert studies did not sample in the Thermal region of the Bay. (^) Lambert reported species present in this region only once between 1994-2000.



Figure 28. Live capture image of *Leucothoe nagatai*. Photograph provided by Tony Phillips.

3. **Polychaeta:** The fauna recorded by Leslie Harris were identified from various substrate, including settling plates, beach seine, sieves, and sediment grabs. Most of the species identified are similar to those previously reported and observed in bays and harbors of the Southern California Bight (SCB), and specifically within the San Diego Bay (Maloney et al., 2007; INRMP, 2013). However, there appears to be two new (probably introduced) polychaetes identified in this species. A *Lumbrineris perkinsi* Carrera-Parra, 2001 (Figure 29) and a *Branchiomma* sp. (Figure 30). In addition, two other samples were identified as probable new introductions, a *Nereis* and the new genus of *Spionid* (polydorine group). The new genus of *Spionid* is most likely native as further investigation has indicated that it occurs in other SCB bays and harbors (personal communication with Leslie Harris, June 2013). The *Branchiomma* sp. is a relatively new species believed to be from the Bahamas; however, it has been showing up in several surveys along the bight within the last few years (personal communication with Leslie Harris, June 2013). *Lumbrineris perkinsi* appears to be from Guana Island, British Virgin Islands (personal communication with Leslie Harris, June 2013).
4. **Mollusca and Miscellaneous Phyla-** The fauna recorded by John Ljubenkov for the SSC Pacific stations sampled in July 2011 were what is normally seen in bays and harbors of the Southern California Bight (SCB), whether infauna or fouling. Of potential interest were the non-native species recorded from the fouling samples. The non-native species *Diadumene ?lineata* (Cnidaria), *Musculista senhousia* (Mollusca-Bivalvia), *Mytilus galloprovincialis* (Mollusca-Bivalvia), *Ostrea edulis* (Mollusca-Bivalvia), *Watersipora arcuata* (Ectoprocta) and *Zoobotryon verticillatum* (Ectoprocta) are all common inhabitants of bays and harbors fouling communities within the SCB. These species collected from San Diego Bay confirms the presence of these non-native species among all major bays and harbors of California. Of major interest is the potential first record of a single specimen of the nudibranch *Vayssierea felis* (Collingwood, 1881), found at S12. This is a potential first record of the species from the west coast of the United States; the voucher specimen will need to be confirmed. It is a common species of the Indo-West Pacific, with records from Japan, Australia, Palmyra Atoll, and Tahiti.



Figure 29. Live capture image of *Lumbrineris perkinsi*. Photograph provided by Leslie Harris.



Figure 30. Live capture image of *Branchiomma* sp. Photograph provided by Leslie Harris.

C. DNA Bar Code Analysis

The concept of DNA barcoding was first broadly introduced in 2003 by Herbert et al., who was looking for a means of assisting the taxonomist community in getting a handle on identifying the millions of species that underpin assessment biodiversity challenges. He and other researchers recognized a rate limiting capacity of the scientific community to identify the 1015 million species estimated to exist on the planet (Herbert et al., 2003; Hammond, 1992; Hawksworth and Kalin-Arroyo, 1995). The concept of DNA barcoding is simple, in that it utilizes short sections of DNA from a universal region of a genome to identify species. That DNA sequence could then be used, similar to a supermarket barcode scanner, for identifying different species in a given population. Mitochondrial DNA was selected as the target DNA due to its lack of introns, haploid mode of inheritance, and limited exposure to recombination (Saccone et al., 1999). Robust primers were

developed enabling researchers to routinely retrieve a specific region of the mitochondrial genome (Folmer et al., 1994). Cytochrome c oxidase 1 gene (CO1) primers were designed by Folmer et al. (1994) and were found to work universally in most, if not all animal phyla (Zhang and Hewitt, 1997). The CO1 gene is currently being used as the target gene in a growing field and global Consortium for the Barcode of Life (CBOL) with the primary goal of constructing a comprehensive DNA barcode library for eukaryotic life.

The primary focus of barcoding in this project was to assess the potential application of this tool, including sensitivity and accuracy, as it compares to expert taxonomist identification. This would help determine potential usability when conducting a rapid ecologic index survey. This section of the discussion describes both sequencing and identification success along with potential benefits/utility in a RAS application.

1. Sequencing success: In this study, of the 20 vouchers submitted for analysis 11 (55%) successfully amplified to produce the characteristic 658bp sequence. The universal 18s primers were used to initially amplify all specimens, as they typically work for nearly every phylum of animal (Hillis and Dixon, 1991; Machida and Knowlton, 2012). This was used as a means to test quality of submitted voucher DNA (and therefore the quality of the specimens). One of the primary causes of PCR amplification failure is due to primer mismatch with degraded DNA. Since other specimens of the same Order did successfully amplify, the failure of 9 out of 20 samples to amplify with the universal 18S primers most likely suggests degraded DNA (per communications Smithsonian Institute, CA, July 2012). For marine samples, studies have shown that several factors affect the quality of DNA including chemical and physical environments of storage, type of tissue, and the duration of storage (Dawson et al., 1998). Presence of endogenous nucleases is another common cause of DNA degradation (Dessauer, cole, and Haiher, 1995).

The majority of samples from this study were collected and placed in seawater for transportation back to the laboratory where they were then initially sorted into phyla for eventual species identification by the expert taxonomist panel. While the field collection sites were within a 10 to 30 minute drive to the sorting and identification laboratory and the majority of voucher samples were placed in 90-95% ethanol within 2-4 hours post collection, special consideration for those more sensitive species (i.e. to processing methodology and holding time) was not taken. For example, the genera *Didemnum* and *Bryozoa* are known to be more susceptible to environmental contamination and are thus more sensitive to potential DNA degradation (per communication with Smithsonian DNA barcoding laboratory, July, 2012). Dawson et al. (1998), reported that vouchers from the genera *Aurelia* and *Phragmatopoma* were also more susceptible to DNA degradation. Thus, failure of amplification for samples EIS-JL-0038 and EIS-JL-0040 may be due to sensitivity of these particular species to rapid degradation. The failure of the remaining seven samples to amplify was most likely due to amount of time from field collection to eventual storage in ethanol and or damage/breaking of DNA as a result of storage in ethanol. Flournoy, Adams, and Pandey (1996) found that sample storage in ethanol can cause dehydration resulting in denaturation of protein. Alteration in chemical and physical storage of specimens may be another factor. The primary focus of this study was to conduct a rapid bio-assessment survey with taxonomist identification being the primary goal. While great care was taken in the field to keep samples in seawater and transport them to the laboratory as quickly as possible for initial sorting and identification, the transplantation of the various organism to temporary storage containers would ultimately

result in changes in environmental pH, salt concentration and temperature. Alteration in these key environmental variables is known to impact enzyme activity and consequently DNA degradation (Dixon and Webb, 1979). Finally, length/duration time of storage could be another cause of sequencing failure. Samples stored in ethanol for a long (> 1 year) period of time can result in acidification. The consortium for the barcode of life recommends that barcode analysis should follow collection as quickly as possible (within a couple weeks) to avoid this issue. Barcode analysis was conducted about 10 months following collection thus acidification from storage could be a possibility of sequencing failure for some samples.

2. Identification success: Overall DNA barcoding identification was successful in that 10 of the 11 (91%) successfully amplified vouchers matched taxonomist identification up to a minimum order classification with 73% being identified at greater than genus level. However, species level identification was only 64% (7 of 11). Sequence similarity (i.e. accuracy) analysis with matching sequences ranged from 87-100%. While there could potentially be a discrepancy in classification between the expert panel and sequencing results (e.g. Sample EIS-TP-0043 classified as *Ampithoe valida* but sequencing results indicate could be *Ampithoe Longimana*) the most likely rate limiting factor for species level identification was that of no current sequence information in Genbank[®] and/or BOLD to compare to (e.g. Sample EIS-GL-0021 classified as *Aplidium californicum* by taxonomist but no match in database query). Currently there is about 38,000 marine specific barcodes on record (MarBOL website, April 2012) which equates to about 10% identification for most marine taxa (Radulovici, Archambault, and Dufressne, 2010). Total validated species numbers ranges from 168,000 to 230,000 (WoRMS: <http://www.marinespecies.org>, and Bouchet, 2006), but are estimated to exceed 10 million species (Radulovici et al., 2010).

While species level identification was only 64% when you compare that to the sheer magnitude of number of potential species to identify in a bioinventory/biomonitoring capacity its overall utility as an additional identification tool is quite high. Several authors have indicated that cost-effectiveness for species identification, especially with amphibious/marine type monitoring programs can be quite significant (Smith, Fisher, and Herbert, 2005; Herbert and Gregory, 2005; Smith, 2005). Herbert and Gregory (2005), estimated that cost per sample identification by a team of taxonomic specialists ranges from \$50-\$100 per sample whereas DNA barcode analysis cost range from \$2-\$5 per specimen. Cost is only one aspect when evaluating benefit. Speed, reliability, and accessibility are additional factors to consider. Most biomonitoring programs rely on classic Linnaean based taxonomy identification. This involves careful collection and handling of specimens to preserve distinguishing features. Identification requires careful examination by a highly trained specialist to differentiate subtle differences. This process is time consuming and typically involves a panel of experts to cover the wide array of taxa encountered in a biomonitoring effort. This process, is generally viewed as a rate limiting (i.e. time and money) step for many ecologic and biodiversity monitoring efforts (Herbert and Gregory, 2005). Barcoding offers a fairly rapid, high throughput potential which could be used as a presorting, routine species identification tool (Schwartz, 2007; Herbert et al., 2003; Radulovici et al., 2010). While barcoding cannot completely replace the need for highly trained taxonomists in biomonitoring programs, it could potentially decrease the burden of routine identifications and allow the expert panels to focus on identification of potential invasive and new species along with their associated delineation and how it

relates to higher taxonomy. In short, the addition of barcode applications to biomonitoring programs could provide valuable information for resources managers although considerations would need to be made with respect to sample handling and preservation protocols.

Discussion of Species Abundance and Distribution for Regions and Naval Facilities

Overall species abundance and diversity varied widely throughout the different stations and study regions with no clearly defined spatial patterns. Additionally, t-tests were performed evaluating stations on or in proximity to Navy Facilities (S1, S2, S3, S11, and S12) with non-Navy locations (S4, S5, S6, S7, S8, S9, S10, S14, S16, S17, and S18) with respect to species richness, abundance and diversity. The t-tests showed that there was no significant difference (at the $p = 0.05$ threshold) for the community metrics between the Navy and Non-Navy stations. Although there was no significant differences found between the Navy and Non-Navy stations the stations with the top three greatest abundances were Non-Navy (S8, S9, S14) while the Navy location S12 (Paletta Creek) had the greatest richness and diversity. One notable spatial trend in terms of species assemblage was the abundance of the exotic Asian Mussel, *Musculista senhousia*. This species was observed at 8 of the 16 stations, with a greater abundance in the back bay. *M. senhousia* made up less than 5% of the total abundance at stations S2, S4, S5, S6, S7, S8, and S10 and was generally the most abundant species at Stations S14 (50%, $n = 501$), S16 (7%, $n = 15$), S17 (32%, $n = 41$), and S18 (27%, $n = 64$). Additionally, based on similarity indices the stations within the back bay region (S14, S16, S17, and S18) were more similar to each other than stations found towards the mouth of the bay (S1, S2, S3).

Recommendations

The level of effort undertaken during the Rapid Assessment Survey was ambitious. It is recommended that during future surveys the scope of work be scaled back somewhat to a level more feasibly accomplished during a one week period. There seems to be a natural separation between the infaunal sediment sampling and the fouling community sampling. There is a lot of historic and ongoing work being done in the sediment realm as part of the Bight survey's lead by SCCWRP. It would seem most time and cost effective to leverage with the work that is part of the Bight Program and supplement with biological surveys of the fouling and intertidal communities. Several researchers have inferred that communities present on artificial structures are different from natural habitats. As a large percentage of naval facilities contain man-made structures, it recommended that future studies focus in particular on fouling communities. Overall the recommended frequency of sampling the fouling and intertidal communities depends on the questions to be answered, or goals of the study. If one is interested in the dynamics of the communities then sampling at a higher frequency across various seasons or temporal scales would be recommended. If the study goal is to better understand the presence and distribution of invasive species sampling on a less frequent scale would be appropriate, biannual or longer. However, of interest is the diversity of species present on the 12 month deployed settling plates. A study deploying settling plates and sampling monthly would be beneficial in assessing the progression and changes overtime. Additionally, if as part of the Bight survey there are locations which are not sampled adding locations of interest and performing the same series of analyses would be most useful and provide future possible direct collaboration.

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14. ABSTRACT The purpose of this study was to conduct an ecological index, early detection survey to identify and catalog native and non-indigenous species near naval facilities within the four hydrographic regions in the bay. Work was similar to a Rapid Assessment Survey (RAS) methodology and a team of taxonomists identified live specimens for 5 days. The focus of this study was to identify native, introduced, and cryptogenic species present on multiple natural and artificial habitats within the four hydrographic regions. The team collected and identified 6,477 organisms, with 299 species represented from 13 phyla. Species identified in this study were similar to those reported in previous studies; however, there were some differences in distribution within the bay. In addition, two previously unreported species were identified. Results presented will include the distribution of native and non-indigenous species identified from natural and artificial habitats within the four hydrographic regions.					
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