

# **Broad-Scale Nonindigenous Species Monitoring along the West Coast in National Marine Sanctuaries and National Estuarine Research Reserves**

## **Report to National Fish & Wildlife Foundation**

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## Summary

Nonindigenous species have caused substantial environmental and economic damage to coastal areas. Moreover, the extent and impacts of nonindigenous species are increasing over time. To develop predictive models and to identify which areas should be targeted for impact mitigation or early detection, we need a basic foundation of knowledge about the spatial and temporal patterns of invasions. This project was developed because we lacked the necessary data to rigorously evaluate the patterns of coastal invasions. This collaborative project, between the Smithsonian Environmental Research Center, the National Estuarine Research Reserve System (NERRS) and the National Marine Sanctuary Program (NMSP), established a rigorous, large-scale monitoring and research program for invasive species in nine protected coastal areas along the US West Coast from San Diego, CA, to Kachemak Bay, AK. Our research included two components, broad-scale and site-specific projects.

The broad-scale component focused on using standardized protocols to collect data on the composition of fouling communities and nearshore fish and crabs. We collected data from 310 settling plates and 140 traps across nine NERRS Reserves and NMSP Sanctuaries. The four most common taxa on the settling plates were Bryozoa, Tunicata, Cirripedia, and Hydrozoa. We identified these four taxa and also Nudibranchia, a mobile molluscan taxa often associated with fouling organisms, to species and noted which were nonindigenous. We found 132 species in the 5 taxa under study. NIS accounted for over one quarter of the diversity in these taxa, with 31 NIS identified. Over half of tunicate species were non-native. The documented NIS included two new US west coast sitings plus 3 other range extensions. We documented two patterns in NIS, a latitudinal pattern and differences between NIS impacts in marinas versus non-marina sites; research on salinity differences is still underway. Both the number and percent of NIS decreased with increasing latitude. Tijuana River had the most, 21, NIS and Monterey Bay had the highest proportion of NIS (57%). The same pattern of decreasing NIS with increasing latitude was observed when we examined Tunicata only and Bryozoa only. Across latitudes, plates in marinas were more impacted by NIS than were plates in more natural areas. All NIS but one were found at marinas, whereas only half the NIS were found at the non-marina sites. In addition, NIS at marinas accounted for almost 80% of the NIS per site. Therefore, we were able to provide information on the relative risk of invasions for different taxonomic groups and geographic regions. The spatial and habitat patterns can be used for future predictions and will be of even more value once they are confirmed with additional taxonomic groups and hypothesis-driven studies that will continue from this initial study. Our broad-scale trapping study illustrated how recently-introduced NIS quickly can become numerically dominant. Although we only found *Carcinus maenas* at Elkhorn Slough NERR, this recently introduced nonindigenous crab was very common at this Reserve and was the most abundant crab in our traps at 3 of 7 Elkhorn sites.

The site-specific projects were conducted at each Reserve plus Olympic Coast and Monterey Bay Sanctuaries. Several are serving as the first important step in longer term research, such as examining whether a change in shipping policy in Kachemak Bay will increase NIS. Others, such as the South Slough project examining the effect of a salinity cline on the number and proportion of NIS, will be expanded to test hypotheses across several protected areas. Many of these site-specific projects still need further analyses, and analysis is underway.

## Introduction

The extent and impacts of nonindigenous species (NIS) invasions in coastal marine ecosystems have become increasingly evident in recent years (Carlton 1989, 1996a; Carlton & Geller 1993; Ruiz et al. 1997, Grosholz 2002). We know of at least 500 NIS that have invaded marine and estuarine habitats of the U.S. (Cohen & Carlton 1998, Ruiz et al. 2000). Moreover, the tempo of invasion appears to be increasing rapidly (Ruiz et al. 2000). NIS have caused substantial environmental and economic damage to coastal areas (Carlton 2001). In marine and estuarine environments, the impacts of biological invasions include dramatic changes to ecological community structure and ecosystem dynamics, parasite and pathogen interactions with native species, commercial fisheries stress, and detrimental alterations to physical habitat structure (Carlton 2001, Grosholz 2002, Levin 2002).

Understanding patterns of NIS invasions among systems is crucial to developing effective management strategies (Ruiz & Carlton 2003). Without this foundation of basic information, we cannot assess the relative risk of invasions for different taxonomic groups, geographic regions, habitat types, or vectors. Moreover, spatial patterns of invasion are key to focusing monitoring, early detection, and vector management efforts to reduce risks of new invasions. Although existing data underscore the increasing ecological and economic consequences of NIS invasions (OTA 1993; Ruiz et al. 1999; GAO 2002, Perrings et. al 2002), there are serious limitations in our present knowledge about marine invasions (Ruiz et al. 2000). Such information gaps affect critical management decisions and hamper development of invasion biology as a predictive science (Carlton 1996b; Kareiva 1996; Vermeij 1996; Ruiz et al. 1997; Ruiz & Hewitt 2002).

The rates and spatial patterns of marine invasions remain largely unresolved (Ruiz et al. 2000, Ruiz & Hewitt 2002). Although literature-based syntheses have identified many apparent patterns of invasion among coastal systems, these studies lack standardized, quantitative, and contemporary field surveys (Carlton 1979; Cohen & Carlton 1995; Reise et al. 1999, Hewitt et al. 1999, Ruiz et al. 2000). The quality and quantity of information in these reports is very uneven among sites and reflects distinct differences in sampling methods as well as differences in spatial and temporal search effort among taxonomic groups, habitats, bays, and geographic regions (Ruiz et al. 2000). For example, an extensive and on-going sampling program of soft-sediment benthic invertebrates may exist for some sites, but it may have been decades since this biota was sampled at other sites included in the same synthesis. Even where recent faunal surveys exist, the sampling designs and level of taxonomic analyses usually differ among sites and over time. Thus, spatial and temporal patterns of invasion, both within and among sites, are confounded by sampling biases present in the available data (Ruiz et al. 2000). Therefore, the Forum on Ecological Surveys (sponsored by U.S. Fish and Wildlife Service, April 1998) concluded that fundamental data for assessing such risks in coastal marine systems are lacking. The uneven and incomplete nature of existing data remains a critical information gap, limiting guidance and evaluation of management decisions.

The threats posed by current and future marine invasions are of critical concern to our nation's coastal protected areas. The National Estuarine Research Reserve System (NERRS) and the National Marine Sanctuary Program (NMSP) are two systems of protected areas charged with the conservation and stewardship of ecologically sensitive, biologically diverse areas along all

coasts of the United States. Most NERRS Reserves have been affected by biological invaders (Wasson et al. 2002). Similarly, a 2001 assessment of the scientific information needs for the NMSP indicated that, for the majority of sites, the identification of marine invasive species sources was of high concern. Both NERRS and NMSP managers require scientific information to address the persistent and expanding non-indigenous species threat. They concluded substantial increases in research and monitoring activity are necessary to effectively support management (Gittings et al. 2002).

To address the information gaps about invasion patterns and the extent of invasion in these protected areas, the NERRS, NMSP, and Smithsonian Environmental Research Center (SERC) initiated a joint research program studying NIS in the west coast NERRS and NMSP protected areas. Few attempts have been made to monitor marine invasions at large spatial scales, although standardized surveys across large regions are necessary to examine hypotheses of invasion success and habitat resistance at these scales. Therefore, we set out to create a broad-scale, multi-habitat platform to study, compare, and contrast invasion patterns and their associated ecological and economic implications for protected area management nationally.

This research program had two main objectives. First, we planned to initiate and develop capacity for a broad-scale NIS monitoring network that uses standardized protocols. A quantitative baseline of standardized data could be used to identify patterns of established non-indigenous species, detect the arrival and spread of new marine invasions, and test hypotheses about the causes and impacts of invasions in coastal ecosystems. Our second goal was to develop site-specific projects of local interest that could serve as pilots for broader scale future projects. Below we address the broad-scale project followed by the site-specific projects.

## **BROAD-SCALE MONITORING FOR SESSILE INVERTEBRATES AND NEARSHORE CRABS AND FISH**

### **Goals, Broad-Scale Project**

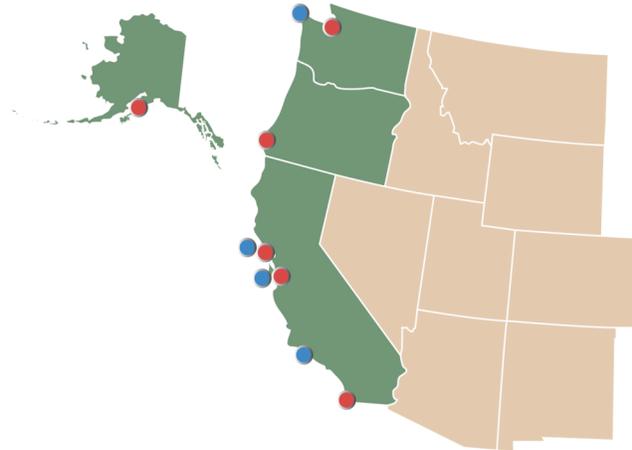
- Provide a baseline against which the impact of future invasions may be compared.
- Contribute to the nationwide effort to track the diversity, range, and abundance of non-indigenous species.
- Delineate the current geographic range of established invaders and track their spread.
- Provide early detection of new invasion events so that rapid, focused responses may be initiated.
- Link data on invasive species in the monitoring network for shared information and large-scale analysis, as well as for inclusion in the growing national database on marine invasions established at SERC.
- Develop capacity to assess the success and performance of non-native species across sites.

- Test several hypotheses about the spatial and temporal patterns of NIS, including:
  1. Across NERRS and NMSP sites, the number of established non-indigenous species decreases with increasing latitude;
  2. Along the marine to estuarine gradient at a given latitude, the number of non-indigenous species will decrease, and the percentage of non-native species will increase, with decreasing salinity.
  3. The number of established non-indigenous species is significantly greater in estuaries and bays (i.e., NERRS sites) than more exposed marine habitats (NMSP sites);
  4. The rate of new invasions will decline with increasingly effective management efforts.
  5. The rate of spread of non-indigenous species among sites is related to life history characteristics and coast-wise transport vectors.

## Methods, Broad-Scale Project

### Reserves and Sanctuaries

Our research program employed standardized field approaches in the protected areas along the length of the U.S. West Coast from Kachemak Bay AK to San Diego CA. We sampled in and near all five west coast NERRS Reserves: Kachemak Bay AK, Padilla Bay WA, South Slough OR, Elkhorn Slough CA, Tijuana River Estuary CA, and four West Coast NMSP Sanctuaries, Olympic Coast WA, Gulf of Farallones CA, Monterey Bay CA, Channel Islands CA (**Fig 1**; see web content for map of Reserves and Sanctuaries along the west coast and of sites within and near these protected areas). The five Reserves and the Olympic Coast and Monterey Bay NMSP Sanctuaries were active participants in the project. After we began sampling, a sixth NERRS Reserve was added to the west coast, but we did not include this Reserve in our sampling regime. At multiple sites at each Reserve and Sanctuary, we sampled for fouling organisms using settling plates and, at the Reserves, for nearshore crabs and fish using traps.



**Figure 1.** Map of the US West Coast National Estuarine Research Reserves (red) and National Marine Sanctuaries (blue) in this study.

Henceforth, we use ‘Reserves’ to refer to the participating NERRS, ‘Sanctuaries’ to refer to the NMSP sanctuaries sampled in the project, and ‘protected areas’ to refer to both.

## Settling Plate Methods

### *Site selection*

We sampled with settling plates at two marinas or piers and at several non-marina sites in or near each of the protected areas (**Table 1, Web content**). We used the two nearest marinas that were willing to participate and were not in the same harbor. There was one exception to this rule: because Elkhorn Slough opens into Monterey Bay, we used the one marina at the mouth of Elkhorn Slough, Moss Landing, midway up Monterey Bay, plus the marinas at the north and south ends of Monterey Bay. Hence, we sampled just three marinas for these two protected areas combined. We sampled both the north and south parts of the Moss Landing marina but have combined the results from the two parts for analysis because there was very little difference between them in fouling community composition.

We sampled at four to eight non-marina sites in each protected area. The non-marina areas for settling plates were selected by the actively participating sites, and were selected based on three different criteria. First, all NERRS sites participate in a system-wide water monitoring program (SWMP) and each Reserve has four to five water monitoring stations. We sampled at each of these SWMP stations to help start collecting biological data to go along with the water quality data. We deployed between one and six plates at each SWMP station, depending on feasibility of deploying multiple plates. Second, we deployed plates at additional areas that would help sample along a gradient or test a hypothesis of local interest, usually related to the SWMP station's gradient. For example, in Elkhorn Slough, the SWMP sites were selected to measure water quality at different distances from the mouth and also at different water flow regimes (high vs. low). We therefore, deployed plates at high and low flow areas at different distances from the mouth of the slough. Similarly, we sampled extra sites in South Slough to better measure the effect of salinity on recruitment.

Third, we took advantage of already-existing structure or buoys used by other sampling programs to sample deep water, and hard bottomed sites in the protected areas. For example, we deployed plates at buoys used by the Partnership for Interdisciplinary Studies of Coastal Oceans (PISCO) in the Channel Islands and Monterey Bay Sanctuaries. Olympic Coast maintains its own offshore buoys, which we used for settling plates. In the Gulf of Farallones area, we attached plates to buoys maintained by a graduate student in Tomales Bay.

We measured several environmental variables at each site. We measured temperature every 4 hrs with a datalogger. We also measured temperature, salinity, and dissolved oxygen at deployment and retrieval with a YSI probe. The NERRS records readings every 30 min of temperature, salinity, dissolved oxygen, and other water measurements at each of their SWMP sites. No other site variables were measured. Sites varied in flow and sediment type, and these were noted based on broad visual assessment. The marina sites tended to have low flow as did several sites in Elkhorn Slough (Azevedo Pond, Hudson Landing West, North Marsh, and Whistle Stop Lagoon) and one of the Tijuana River sites (Tidal Linkage). All other sites seemed to have moderate to high flow. The four southern NERRS, Tijuana River, Elkhorn Slough, South Slough, and Padilla Bay had soft sediment, mostly silt, at all of the sites we used. A few of the sites in these Reserves had riprap in the intertidal zone nearby (e.g., Elkhorn Slough's North Marsh, Vierra, and Whistle

Stop Lagoon). The sites we used at the edge of the Gulf of Farallones NMS, sites in Tomales Bay and Bodega Bay, were also soft bottomed. Kachemak Bay and the non-marina sites in and by the Sanctuaries had sandy or rocky substrates for the most part.

### *Plate deployment*

Following standardized protocols, we sampled the sessile invertebrate community at each site with settling plates, also called fouling panels. These plates were 13.5 cm x 13.5 cm x 0.5 cm polyvinyl chloride (PVC) squares. They served as passive collectors that accumulated organisms over time through larval recruitment and immigration. Plates were sanded on one side to facilitate attachment of recruiting invertebrates. All plates faced sanded side down to maximize invertebrate recruitment, minimize algal growth, and minimize sediment deposition on the target organisms.

Plates were suspended at least 0.25 m above the substrate so they never touched the substrate. They either were hung from a floating structure or suspended to be at least 0.5 m below MLLW; they were in water all or almost all of the time. The plates were deployed in different ways depending on available structure, sediment type, and depth at the different sites. For all deployment methods, we recorded plate depth and elevation above the substrate.

We divided each marina and pier into six equally sized parts and deployed one plate at a randomly selected spot in each part. Marina and pier plates were weighted with a brick. At marinas, each plate and brick unit was hung from a floating dock typically at  $1.0 \pm 0.2$  m below the water surface, depth allowing. Plates suspended from shallow water floats were closer to the surface, as shallow as 0.4 m deep, so the plates would not touch the bottom when the tide was out. At piers, plates were suspended to an estimated height of 1 m below mean lower low water (MLLW).

The SWMP and in-estuary plates were attached to pilings or temporary stands in the four southern NERRS and to buoyed lines in Kachemak Bay and the deep water Padilla buoy SWMPs (see below). Plates were deployed from pilings when these were available at a site, as was the case with the South Slough SWMP sites and some Kachemak Bay plates (as indicated in **Table 1**). Piling plates were suspended 0.5 to 1.0 m below MLLW, were attached to a PVC pole, and were held at least 0.4 m away from the piling so they would be affected only minimally by anti-fouling substances on the pilings. All other Reserve sites lacked pre-existing structure. Therefore, in soft-sediment areas in Reserves (Tijuana River, Elkhorn Slough, South Slough, and most Padilla Bay sites), plates were attached to PVC pipe stands. The plates hung 0.25 to 0.5 m above the substrate and 0.5 m below MLLW. Each plate was at least 10 m from all other plates. In 2004, when we did additional sampling of South Slough for the site-specific project, we put two plates on each frame; these plates were only 7 cm from their pairs. Frames were still located at least 10 m from one another.

At the non-marina sites of the Sanctuaries, Kachemak Bay NERR, and the 'Gong' site of Padilla Bay NERR, plates were deployed from anchored buoys. These plates were suspended between 1 and 2.5 m below the water's surface. We also deployed a second plate from the Reserve buoys 1

m above the substrate. The plates on buoys were held away from the line and, like the other plates, were oriented horizontally with the sanded side down.

Most plates were deployed in June and July in 2003 and 2004, and retrieved approximately 3 months (86-113 days) later. The exceptions were plates deployed from buoys in Sanctuaries in 2004. These plates were deployed in May and retrieved 128-131 days (Olympic Coast, Monterey Bay) or 182 days later (Channel Islands).

### ***Retrieval, point counts, and vouchers***

We retrieved the settling plates once they had soaked for approximately 3 months. We held retrieved plates in water from each site as we processed them. We scraped away organisms growing on the top and sides of each plate, isolating the ones on the bottom side. We photographed the plates to provide an archival record and to provide pictures for web-based taxonomic guides, and we measured the amount of water they displaced to have a measure of biovolume (biomass).

We preserved plates so a small team could process all of them at once each year, for consistency. We put each plate into a nylon stocking to keep together all the mobile and sessile organisms associated with the plate, then soaked the plate for 15 to 30 min in a Magnesium Chloride (MgCl) or Magnesium Sulfate (MgSO<sub>4</sub>) bath to relax the organisms for easier identification. We used 64 g MgCl or MgSO<sub>4</sub> per liter of water, and used a mixture of seawater and tap water so that the final mixture including the magnesium salt was a similar salinity to the site. The plates were then placed in a 10% Formalin bath, made with seawater, for 24 to 48 hrs, and finally into a 70% ethanol bath for 24 hrs or more. After all plates had soaked in alcohol for at least 24 hrs, they were drained and shipped overnight to SERC for further processing.

A team of four conducted point counts on the plates and collected multiple specimens of all species. We classified organisms by higher taxonomic group, numbering each morphologically different organism on a plate as a new morphotype for that group (e.g. Tunicata1, Tunicata 2, Bryozoa 1... for each plate), and noted genus or species identification when known. We identified to morphotype the organism directly attached to the plate at 50 point counts on each plate, 49 points along a 7x7 grid plus a 50<sup>th</sup> randomly selected point. The point counts provided a measure of community structure and dominance of sessile invertebrates.

We collected and archived multiple specimens of each morphotype on each plate. Taxonomic experts verified species identification of all collected hydroids, nudibranchs, cirripedia, bryozoa, and tunicates. These specimens provided a measure of species richness and the number and percent of NIS on each plate, at each site, and at each protected area. All vouchered specimens were stored with a unique barcode in an archive at SERC; their identifications and site-specific information were maintained in a database.

### ***Data analysis***

We counted number of species from the five identified taxa in three ways. First we counted the maximum number of species in these five groups that may have been at a site and protected area.

This count included all the fully identified species plus those that have not yet been given names and had published descriptions, plus samples that were identified to a higher-level taxonomic grouping but not all the way to species (due to damaged and partially eaten specimens...). Multiple samples identified to family of that family were counted as one entry. This maximum number of species is given in Table 2 but not used in analyses. Second, we tallied a minimum number of species for each plate, site, and protected area. This included the named and not-yet-named fully identified species plus any higher-level identifications that were not represented by species in that same group. For example, a site that had a sample identified as the thecate Hydrozoa Family Campanulinidae and had no samples in this family identified to genus or species (no Campanulina, Cuspidella, and Opercularella), then we would count the family once in the minimum number of species count. Third we tallied the number of named species. This excluded the higher-level identifications and the identifications of not-yet-named species. The exception is for the tunicates; we included not-yet-named tunicate species in this count because these were well documented, well described species whose non-native status was documented in some cases. We used the minimum number of species in analyses about species diversity.

To determine the percent of the species that were non-indigenous, we divided the number of species that are documented as NIS by the number of named species. We used the number of named species as the denominator rather than the minimum number of species found because a species must be identified and named to be documented as an NIS. The exception is for the tunicates. This taxon includes NIS for identified but only partly-named species (e.g. *Didemnum* sp. A). Therefore, for the Tunicata, all organisms identified to species were included for the denominator. We excluded cryptogenic species from the list of NIS.

To allow comparison of sites with different sampling intensities, we counted the number of species on 5 randomly-selected plates at each site. Analysis on plates from previous west coast sampling showed that species accumulation tailed off after five plates at most sites (GM Ruiz, unpublished data). When a site was visited on two years, we selected all the plates from just one year (the first year with five plates). Because not all sites had five plates, we either excluded sites with fewer plates from analysis or, when possible, combined sites. We only combined sites when they were near to each other, in similar habitat, and deployed with the same method (buoys with buoys, or frames with frames). We combined the Channel Island area plates deployed from buoys into two groups, the southern three sites (Ellwood, Alegria, and Jalama) and the northern three sites (Arguello, Purisima Point, and Point Sal). Similarly, the Monterey Bay buoy-deployed plates were combined as were the Gulf of Farallones area Tomales Bay buoy plates and also the Olympic Coast buoy plates. Some Elkhorn Slough sites were combined: we grouped the two high flow sites near the mouth of the slough (Vierra and South Marsh Restoration), we grouped the low flow sites mid estuary (Whistle Stop Landing with North Marsh), and we grouped the low flow sites higher in the estuary (Azevedo Pond with Hudson Landing West). All other sites with fewer than five plates were excluded except for Cap Sante Boat Haven in Padilla Bay, which had four species-rich plates.

The point count data provided information on plate species diversity beyond the five taxa that were sent out for taxonomic identification. These point count data use morphotypes, what looks like a species rather than a verified named species, so cannot be used for site-level species accumulation curves (the identifier given to an organism on one plate was not necessarily the

same identifier given to the same species on another plate). Therefore, we calculated an index of diversity, the Shannon-Weiner Index, for each plate. This index accounts for both species richness (number of species per plate) and how the individuals are apportioned across the component species (distribution of counts across morphotypes). The Shannon Index calculates the amount of information that is needed to describe (to morphotype) every member of the community. To calculate this index, the relative abundance of each morphotype (the number of primary point counts of an invertebrate morphotype divided by the number of primary point counts of all invertebrates on the plate) was multiplied by the natural log of the relative abundance and summed across all morphotypes on a plate then multiplied by -1. Thus a plate with just one morphotype had a value of zero and the value increased with number of morphotypes and evenness of the counts across morphotypes. These data include plates from sites that do not yet have taxonomic identifications, so the sample size is somewhat larger than that used for analyses requiring species identifications.

The point count data also provided information on the proportion of each plate that was covered by invertebrates. These data were also used to calculate the proportion of points that each taxon covered.

We categorized sites as ‘marina sites’, plates hung from floating docks or from piers, and ‘other sites’, plates deployed from buoys, from frames stuck into the substrate, or from ells attached to individual pilings. **Table 2** includes small piers inside protected areas as non-marina plates, but all analyses use the aforementioned categories. The NERRS SWMP sites were all counted as in-estuary, non-marina plates.

All variables used in statistical analyses were transformed to meet the assumption of a normal distribution. Means are given with the raw data, and figures are presented with the raw data or backtransformed axis labels for ease of use.

## **Crab Trapping**

We trapped for nearshore fish and crabs at the five Reserves. We selected one to seven sites at each Reserve for trapping, typically at settling plate sites (see **Table 1**). When possible, we used sites with structure, including eel grass (*Zostera marina*), cobble, and rip-rap. We trapped more than our target of three sites at Elkhorn Slough because the nonindigenous crab *Carcinus maenas* are abundant there and are under study at this Reserve. We trapped fewer than our target amount in Kachemak Bay due to a lack of accessible trapping sites and similar logistical problems.

Traps were deployed during fall low tides in 2003 and 2004, around the plate retrievals. We deployed 8 traps at each site at approximately MLLW. Some traps were not set properly or were lost; the number of traps per site we retrieved is given in **Table 1** and **Appendix B**. Two types of traps were set out at each site. We used 4 collapsible, box traps (61 x 46 x 20 cm, with two 46 cm openings and 13 mm mesh; made by Fukui) and 4 minnow traps (vinyl-coated steel tapered cylinders, 42 cm long, 23 cm diameter, with  $5.0 \pm 0.5$  cm diameter openings on each side and ~ 1 cm mesh) interspersed between them. All traps were 20 to 30 m from their neighbors. Traps were tethered into place (tied to structure or to a temporary pole) and were weighted where currents

were strong. Traps were baited with frozen fish, when available, or cat food (Kachemak Bay in 2003, Padilla Bay).

We retrieved traps after 24 hrs and identified all fish and crabs within to species. We also noted gender and injuries, and measured length (fish) or carapace width (crabs). We report traps from 2003, and we also report Kachemak Bay trap results from 2004.

## Results, Broad-Scale Project

### Settling Plates

#### *Species diversity and composition in and near the West Coast Reserves and Sanctuaries*

We found at least 132 species out of 5 taxa in our fouling plate survey, 20 hydroids, 19 nudibranchs, 11 barnacles, 26 tunicates, and 56 bryozoans (**Table 2, Fig 2a**). We found the most species in bays near Tijuana River NERR, and there was a general decrease in number of species from the southernmost to northernmost protected areas. Tijuana River NERR, however, had the most settling plates in marinas. The high number of plates as well as their habitat location could bias the trend.

Analysis of the number of species on 5 randomly-selected plates per site, which provides an estimate of alpha diversity (species richness per site) for the 5 taxa under study and also allows comparison of sites that were sampled with different intensities, yielded a coastwide average of  $14.8 \pm 9.2$  species per 5 plates (mean  $\pm$  SD, N = 39 sites). Alpha diversity ranged from 2 species on 5 Hummingbird Island plates, Elkhorn Slough NERR, and on Seldovia piling plates, Kachemak Bay NERR, to 36 species at Dana Marina, Mission Bay, near Tijuana River NERR (**Appendix C**). On the protected area level, this measure also showed a decrease in richness from southern protected areas to more polar ones (**Fig 2b**). However, the Gulf of Farallones had the highest number of species, with  $25.25 \pm 5.41$  species per 5 plates (mean  $\pm$  SE, N = 4 sites) and Elkhorn Slough had the lowest number, with  $9.20 \pm 1.93$  species (N = 5 sites; **Fig 2b**). Tijuana River had the second highest species richness per five plates (**Fig 2b**).

Species diversity (accounting for both richness and evenness of distribution among species) on individual plates in the 76 sites also decreased from southern to northern sites but was highest at the Gulf of Farallones area plates (Shannon Index,  $H = 1.04 \pm 0.11$ , mean  $\pm$  SE, N = 26) and was lowest on the Padilla Bay plates ( $0.55 \pm 0.09$ , N = 25; **Fig 2c**).

Invertebrates covered  $64.1 \pm 31.1\%$  (mean  $\pm$  SD, N = 310 plates) of the observed points on plates. Percent cover was highest on plates in South Slough ( $70.8 \pm 3.8\%$  cover, mean  $\pm$  SE, N = 76 plates) and Tijuana River ( $70.7 \pm 4.7\%$ , N = 33) and lowest in Padilla Bay ( $50.6 \pm 6.3\%$ , N = 25; **Fig 2d**). Our second measure of abundance and growth of organisms on plates, biovolume, yielded an average of  $167 \pm 87$  ml displaced (N = 295 plates). Gulf of Farallones area plates had the highest biovolume ( $256 \pm 31$  ml, mean  $\pm$  SD N = 26), while Padilla Bay, again, had the lowest ( $111 \pm 7$  ml, mean  $\pm$  SEN = 25; **Fig 2e**).

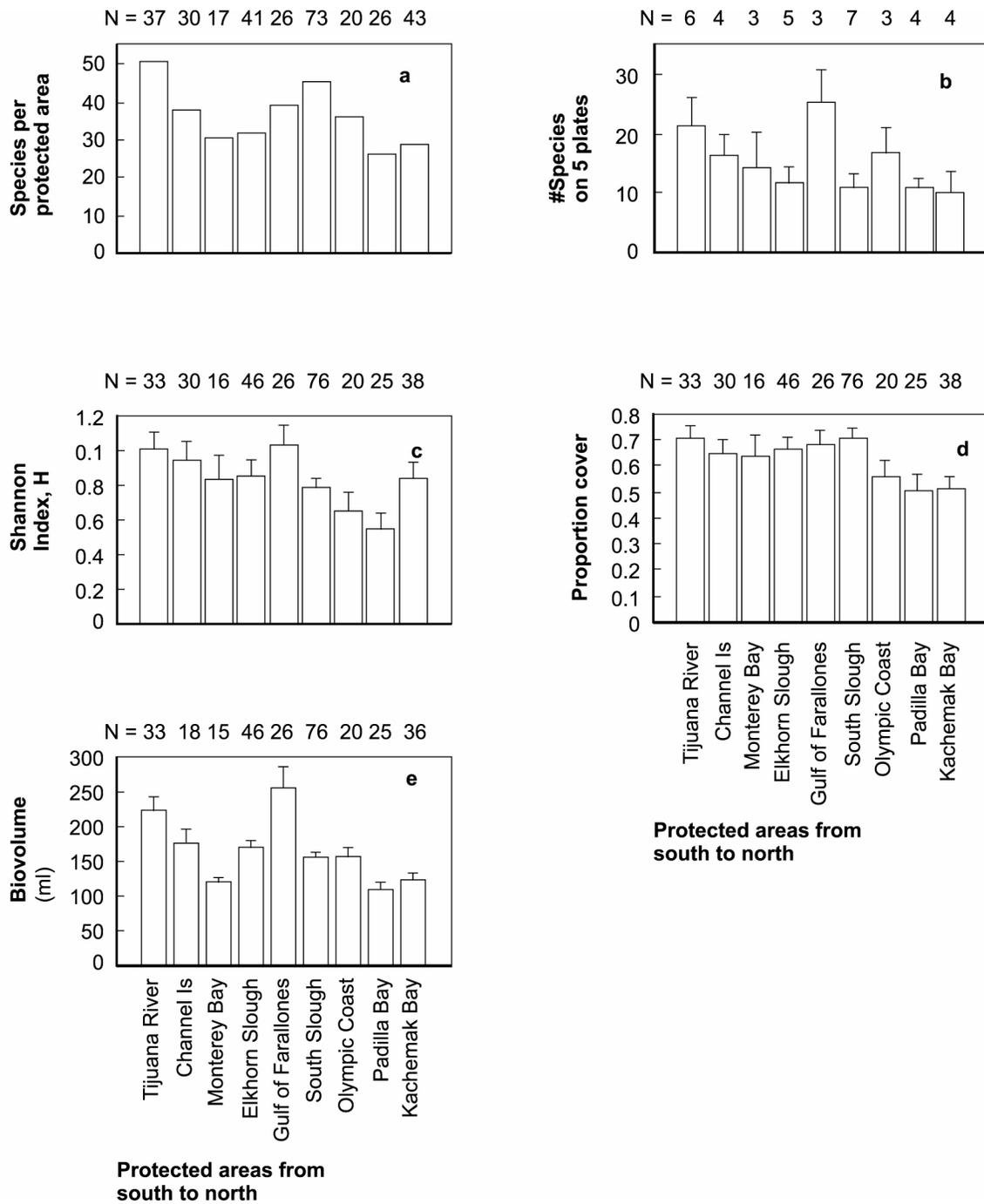


Figure 2. Species diversity and abundance on settling plates across protected areas, as measured by: a) Species richness from all plates, b) Species richness on 5 plates, c) Species diversity (Shannon diversity index) per plate, d) Proportion cover per plate, and e) Biovolume (water displaced) per plate. Bars in b-e show mean  $\pm 1$  SE; N, number of sites in b or number of plates in a, c, d, and e, is given along the top.

There were more Bryozoa species than other taxa overall and at each protected area (**Table 2**). Bryozoa also were on more plates than any other taxa: they were on 250 of the 310 plates in the study, while the second most common taxa, Tunicata, were on just 128 of the plates, Cnidaria were on 117 (Hydrozoa were on 107 of those), and Cirripedia were found on 111 plates. Other taxa were much less common. Of the plates with Bryozoa, Bryozoa also occupied more of the observed points than any other taxa ( $48.7 \pm 1.2\%$  of the points, mean  $\pm$  SE, N = 250 plates **Fig 3**). Tunicates, hydrozoa, and barnacles also accounted for much of the percent cover on plates with these taxa, whereas other taxa were much less abundant (**Fig 3**).

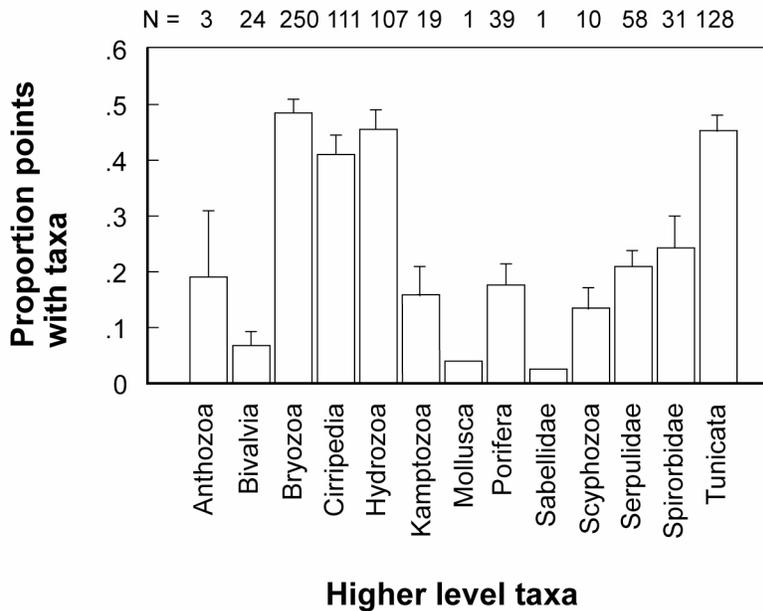


Figure 3. Species dominance on settling plates as measured by proportion cover on plates. Bars show mean  $\pm$  SE; N, number of plates with each taxon, given along the top.

### *NIS diversity and abundance*

Nonindigenous species (NIS) accounted for over one fourth of the diversity of the more common taxonomic groups in protected areas along the US west coast (**Table 2**). We found 31 NIS on the plates, one each of Hydrozoa, Serpulidae, and Nudibranchia, plus 3 Cirripedia, 11 Bryozoa, and 14 Tunicata NIS (**Tables 2, 3**). NIS represent 27% of the species richness of these taxa excluding the Serpulidae, which are not yet fully documented in our samples: 7.7% of the fully-identified, described hydroid species were NIS, as were 5.9% of the nudibranchs, 27.3% of the barnacles, 27.0% of the bryozoans, and 53.8% of the tunicate species (**Table 2**).

The most common nonindigenous taxa were Tunicata (on 346 plates at 114 sites in 8 protected areas, all but Kachemak Bay) followed by Bryozoa (on 303 plates at 86 sites in 7 protected areas, all but Kachemak Bay and Olympic Coast). NIS Cirripedia were on only 42 plates at 20 sites in 5 protected areas, Hydrozoa were on 4 plates at 2 sites in 2 protected areas (Tijuana River, South Slough), and a Nudibranchia was on just one plate (Elkhorn Slough). The most common nonindigenous species included three Tunicata, *Botryllus schlosseri* (on 56 plates at 22 sites in 8

protected areas), its congener *Boltrylloides violaceus* (on 67 plates at 20 sites in 8 protected areas), and another colonial tunicate *Diplosoma listerianum* (on 65 plates at 16 sites in 5 protected areas), plus 3 Bryozoa, the encrusting *Watersipora subtorquata* (on 66 plates at 14 sites in 6 protected areas) and *Cryptosula pallasiana* (50 plates at 14 sites in 7 protected areas), and the erect *Bugula neritina* (58 plates at 14 sites in 5 protected areas).

### *NIS ranges and range extensions*

The nonindigenous fouling species on our plates came to the west coast from both sides of the Atlantic, the Indo-Pacific, and the NW Pacific through shipping vectors (hull fouling and ballast water) and, for 10 of the species, also along with oysters brought for mariculture (**Table 3**).

**Appendix D** lists the sites in which each NIS was found. The ranges of NIS found in our survey are given in the web-accessible database and the associated fact sheets. The most widespread species we found was *Obelia longissima*, which was found at all protected areas. The most widespread NIS were *Botryllus schlosseri* and its congener *Boltrylloides violaceus*, both of which were found at all 8 protected areas from Tijuana River, CA, to Padilla Bay, WA. Also extending from Tijuana River to Padilla Bay but at fewer of the protected areas were *Cryptosula pallasiana*, which was missing from Olympic Coast, and *Balanus improvisus*, which was missing from the Channel Islands up through the Gulf of Farallones area sites.

This study documents range extensions for five of the 31 NIS we found on plates, including new records for the US west coast. Our documentation of *Balanus reticulatus* in a Tijuana River NERR area marina, Sunroad Marina in San Diego Bay, is a new record for the US West Coast. It was previously documented much further south in Mazatlan Mexico (Laguna 1985). The Atlantic bryozoan *Bugula fulva*, which we found in Monterey Marina, seems to be a first west coast record as well, although it may be the same species as Cohen et al.'s (1998) *Bugula sp. 2* in Puget Sound. The marinas we sampled near Channel Islands (Oxnard's Vintage Marina) and the Gulf of Farallones (Bodega Bay's Mason's Marina) also had a bryozoan that was either *B. fulva* or *B. flabellate*, a similar-looking NIS (**Appendix D**). Other species had more minor range extensions. *Balanus improvisus* was found in many of our plate sampling sites. It was previously known from San Francisco Bay, Elkhorn Slough, Coos Bay, the Columbia River, and Vancouver Island, mostly in low or variable-salinity sites, but there have been scattered single records from harbors in San Diego and Los Angeles (Carlton 1979, Wasson et al. 2002). We found *B. improvisus* in Tijuana River's Oneonta Slough; in South Slough's Charleston Boat Basin, Empire Pier, Coos Citrus Docks, Sengstacken Arm and YSI, Valino Island, and Winchester YSI sites; in Olympic Coast's La Push Marina, and in Padilla Bay's Bayview Channel. The tunicate *Styela plicata* had a documented range extending from Mexico north to Santa Barbara (Lambert & Lambert 1998, 2003, Cohen et al. 2002). We found it in the two marinas in Mission Bay (near Tijuana River), and both marinas that we sampled near the Channel Islands, but also in the marina at the mouth of Elkhorn Slough (Moss Landing), a northward range extension. This tunicate also has recently colonized San Francisco Bay (1<sup>st</sup> record 2000, Ruiz et al. unpublished). Finally, the bryozoan *Victorella pavidata* was found at multiple sites in two protected areas, in Elkhorn Slough's Hudson Landing, Kirby Park, and Moss Landing marina, and in Tijuana River's Model Marsh and Main Channel (**Appendix D**). This species, which prefers brackish

water, formerly had only been known on the west coast in Merritt Lagoon, San Francisco Bay (Carlton 1979, Cohen & Carlton 1995).

### *NIS across protected areas and latitudes*

On the protected area level, Monterey Bay had the greatest proportion of NIS; well over half (57%) of the identified species in Monterey Bay NMS were introduced (**Table 2, Appendix D**). Elkhorn Slough (53.6% NIS) and Tijuana River (51.2%) also had many NIS relative to native and cryptogenic species richness (Table 2). Kachemak Bay, of course, had the lowest proportion of NIS (0%), and Olympic Coast had the second lowest proportion (0.14, **Table 2, Appendix D**).

The number of documented NIS per protected area (**Fig 4a**) and per site (**Fig 4b**) decreased with increasing latitude (per site:  $Y = 11.02 - 2.83 X$ ,  $r^2 = 0.18$ ;  $F = 15.89$ ,  $N = 73$  sites within the 9 protected areas,  $P = 0.0002$ ). Tijuana River had the most NIS, 21 known nonindigenous species, in the five identified taxa on the settling plates, and Kachemak Bay had the least, 0 NIS (**Table 2**). Although the zeros at the Kachemak Bay sites influenced the regression line, the pattern was still significant when Kachemak's sites were excluded from analysis ( $F = 4.89$ ,  $N = 65$ ,  $P = 0.0306$ ). Similarly, the percent of species that were nonindigenous decreased with increasing latitude (**Fig 4c**,  $Y = 4.24 - 1.03 X$ ,  $r^2 = 0.22$ ;  $F = 19.85$ ,  $N = 73$  sites,  $P < 0.0001$ ; without Kachemak sites,  $F = 7.68$ ,  $P = 0.0073$ ).

Similarly, both the number and percent of nonindigenous Bryozoan species decreased with increasing latitude (**Fig 4d,e** # NIS bryozoa, ln, excluding sites with no bryozoa:  $Y = 8.23 - 2.19 X$ ,  $r^2 = 0.16$ ;  $F = 10.79$ ,  $t = -3.29$ ,  $N = 61$  sites,  $P = 0.0017$ ; proportion NIS bryozoa, ASSR:  $Y = 3.77 - 0.92 X$ ,  $r^2 = 0.15$ ;  $F = 10.59$ ,  $t = -3.25$ ,  $N = 61$  sites,  $P = 0.0019$ ). The number and percent of nonindigenous tunicate species per site also decreased with increasing latitude (**Fig 4d,e**; # NIS tunicates, ln, excluding sites with no tunicates:  $Y = 19.62 - 5.06 X$ ,  $r^2 = 0.65$ ;  $F = 63.24$ ,  $t = -7.95$ ,  $N = 36$  sites,  $P < 0.0001$ ; proportion NIS tunicates, ASSR:  $Y = 8.40 - 2.04 X$ ,  $r^2 = 0.37$ ;  $F = 19.83$ ,  $t = -4.45$ ,  $N = 36$  sites within the 9 protected areas,  $P < 0.0001$ ). The other taxa did not have enough NIS or species across sites to support similar analyses.

The 30 NIS found in or near each protected area by this study plus 24 nonindigenous fouling organisms from other studies are listed in **Appendix D** along with information on the waterways in which they were found, whether they are considered established, and references to relevant literature. Comparison of results from this study with this tabulation of all documented nonindigenous fouling species from the areas in and around these same protected areas shows that this decrease in NIS with increasing latitude is also apparent when a broader range of taxa are included (**Fig. 4a**). **Appendix E** lists 58 NIS from Elkhorn Slough Reserve. It also provides 37 known NIS associated with wetlands from Tijuana River NERR and 87 marine NIS (fouling organisms and also other invertebrates, algae, and plants) known in the broader San Diego area.

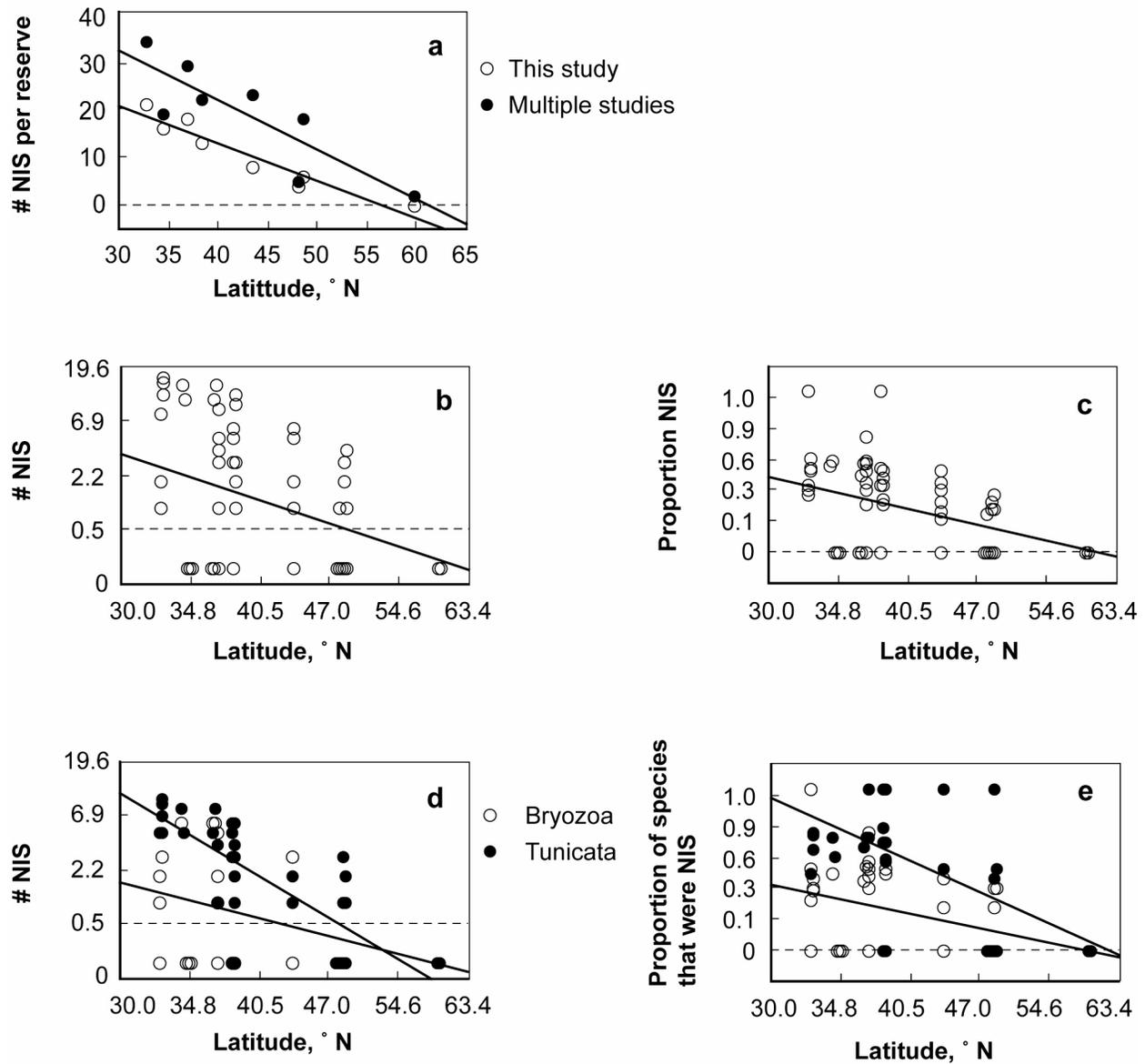


Figure 4. NIS across latitude for a) total number of NIS per protected area, b) number of NIS per site (ln), c) proportion of NIS per site (arcsine square-root transformed), d) number of nonindigenous bryozoan and tunicate species, and e) proportion of bryozoan and tunicate species that were nonindigenous. Equations for lines in text.

The decrease in NIS with increasing latitude is a strong enough pattern and NIS represent such a large proportion of the species at many of the protected areas that NIS are partly driving the pattern of a decrease in species richness from southern to more northern protected areas observed in **Figure 2**. When we removed the NIS from the total number of species and examined the native and cryptogenic species only, we no longer found a decrease in species richness from south to north (**Fig 5 a,b**).

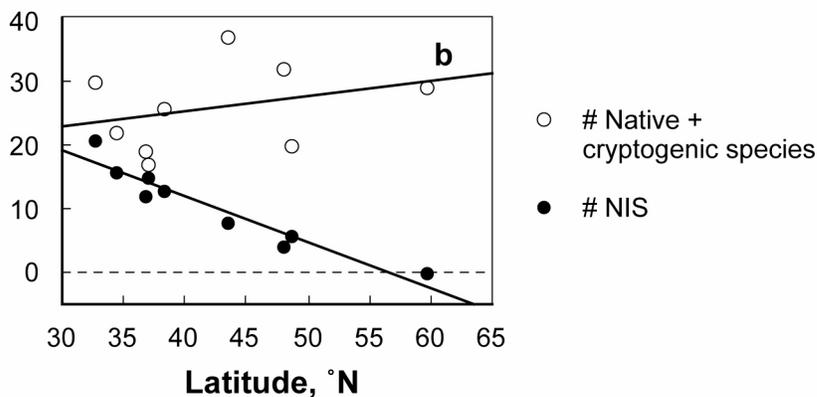
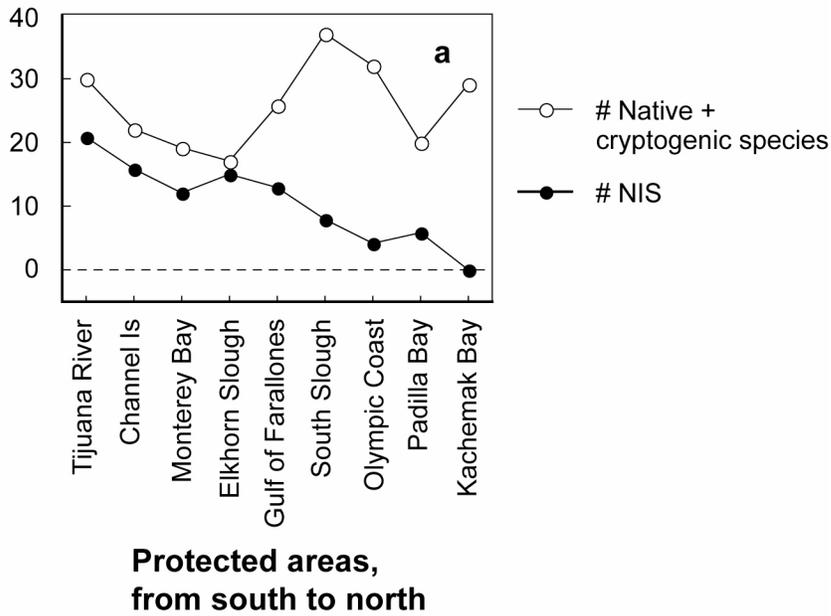


Figure 5. Number of NIS and native plus cryptogenic species across protected areas and latitudes.

### *Diversity between habitats*

Only a fraction of the species overlapped between marinas and natural estuarine and offshore habitats (**Table 4**). For example, in Padilla Bay, where the number of species found in marinas was similar to the number found in the Reserve on plates suspended from frames and buoys, less than a third (29.6%) of the species were found in both types of habitats (marina versus in the Reserve). Plates in Olympic Coast marinas and offshore buoys also had similar numbers of species (22 versus 20) but only one sixth (16.7%) of these species were found in both habitats. In Elkhorn Slough and the Gulf of Farallones area, NIS represented 60% of the overlapping species (**Table 4**). However, NIS did not substantially increase the homogeneity between habitats in the other protected areas. Nonindigenous Bryozoa, Cirripedia, and Tunicata species were found in marinas as well as other sites.

More species were found at marina sites than at the more natural sites, those requiring buoys or frames for plate deployment. Species richness was three times higher in marinas ( $18.9 \pm 1.6$  species per site,  $N = 20$  sites) than in natural sites ( $6.3 \pm 0.5$ ,  $N = 51$  sites) overall. Across protected areas, this pattern was found everywhere except the Olympic Coast (**Fig 6**). Similarly, marinas had higher species richness ( $21.2 \pm 1.9$  species per site,  $N = 20$  sites) on 5 plates than natural sites ( $9.7 \pm 1.1$  species per site,  $N = 19$ ). The highest species richness per 5 plates in marinas was found in Tijuana River area marinas, with  $28.5 \pm 2.8$  species per site ( $N = 4$  sites), whereas Tijuana River's in-estuary plates had just  $7 \pm 2$  species on 5 plates per site ( $N = 2$ ). The lowest marina diversity was found in the Padilla Bay marinas ( $12.0 \pm 4.0$  species per site,  $N = 2$ ) and Padilla's non-marina sites were slightly less diverse ( $9.5 \pm 1.5$ ,  $N = 2$ ; **Fig 6**). In contrast, the Olympic Coast had higher species richness on 5 plates deployed from offshore buoys with 22 species than on its marina plates ( $14 \pm 6$ ,  $N = 2$ ). Monterey Bay buoys had the lowest number, 6 species on 5 plates. Of the non-marina sites, the in-estuary frame-deployed plates had the lowest number of species on 5 plates ( $8.2 \pm 1.0$  species per site,  $N = 13$  sites). The offshore buoy sites averaged  $12.5 \pm 3.4$  species on 5 plates ( $N = 4$ ), and the one in-bay buoy-deployed set (Tomales Bay) had 18 species.

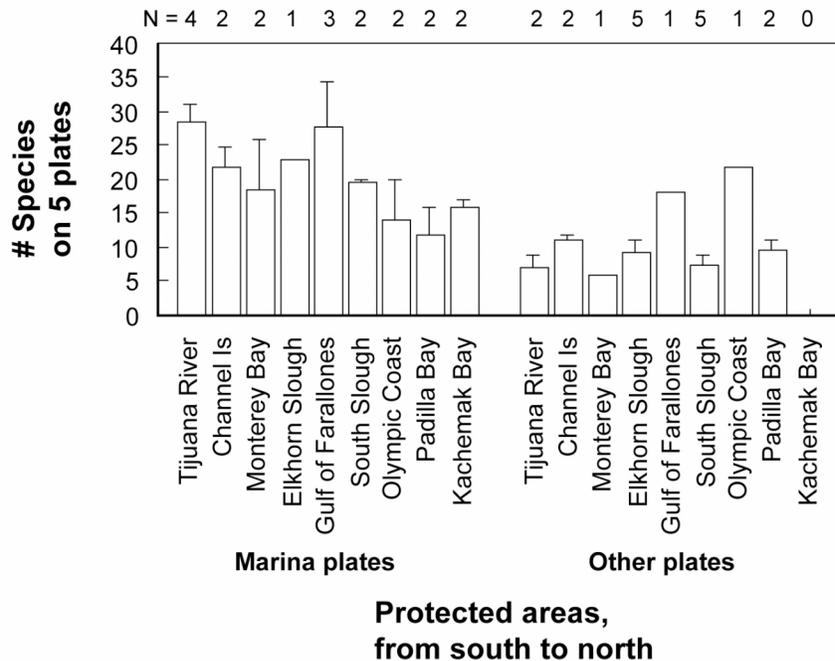


Figure 6. The number of species on 5 plates in marina sites versus frame or buoy sites across protected areas. Bars show mean  $\pm$  1 SE; N, number of sites in given along the top.

Species diversity was almost twice as high on the marina plates ( $H = 1.07 \pm 0.04$ ,  $N = 158$ ) than other plates ( $0.59 \pm 0.04$ ,  $N = 151$ ) at all the protected areas. The marina at the mouth of Elkhorn Slough had the highest diversity ( $1.57 \pm 0.06$ ,  $N = 12$  plates), as measured by the Shannon Index, despite moderate species richness, while the Olympic Coast marinas had the lowest diversity ( $0.67 \pm 0.14$ ,  $N = 12$ ). South Slough marina plates had high species richness but relatively low diversity once evenness was considered ( $0.87 \pm 0.09$ ,  $N = 30$ ). South Slough's plates deployed from frames had much higher evenness than plates in the nearby marinas. Tomales Bay buoys

(Gulf of Farallones) had the highest diversity of non-marina plates ( $0.78 \pm 0.23$ ,  $N = 7$ ), but this was still low diversity compared to marina plates. Kachemak Bay had the lowest diversity for non-marina plates ( $0.37 \pm 0.11$ ,  $N = 14$ ). Biovolume was also higher for marina plates ( $188.0 \pm 6.6$  ml,  $N = 204$ ) than non-marina ones ( $146.3 \pm 4.4$  ml,  $N = 200$ ), but not as different as diversity or richness were between the types of sites. The biovolume was high in Elkhorn Slough marina given the relatively low species richness, indicating large colonies or individuals.

### *NIS differences between habitats*

The difference in species richness between marina and natural sites was even greater for NIS. The occurrence of NIS in marinas ( $7.8 \pm 1.2$  NIS per site,  $N = 20$  sites) was 7 times higher than the occurrence of NIS in other sites ( $1.1 \pm 0.2$  NIS per site,  $N = 51$  sites). **Figure 7** shows that, averaged across protected areas, almost 80% of NIS per site were found at marinas only, and most of the remaining 20% of NIS were found at both marinas and other sites. There was only one occurrence of an NIS found only at a non-marina site, the sites with buoys or frames. The native plus cryptogenic species also had higher species richness per site at marinas only, but had a much larger proportion of species per site at non-marina areas than did the NIS.

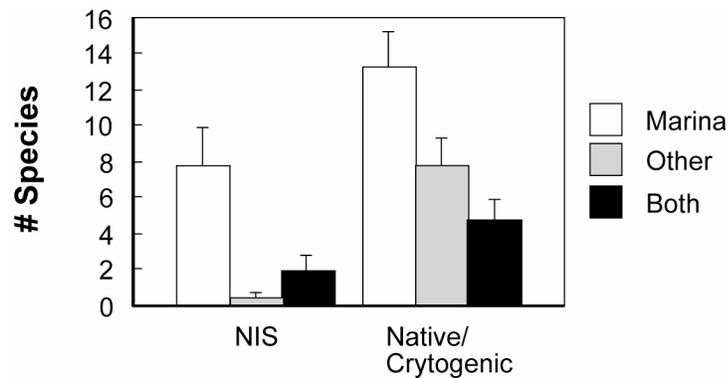


Figure 7. Number of NIS versus native plus cryptogenic species at marinas only, at frames and buoys only, or at all types of sites. Bars show mean  $\pm$  1 SE; N, N = 9 protected areas for all bars.

This ratio of NIS at marinas compared to other sites was less skewed towards marinas only when we examine number of species per habitat instead of per site. We found 14 NIS in marinas only (plus 2 NIS Hydrozoa, shown in Table 2, at a port from South Slough's site-specific study that is not yet completely analyzed and so was not included in these analyses). 14 NIS colonized plates both at marinas and other sites. Just 1 NIS was only in other sites.

We found more nonindigenous Tunicata species in marinas only than other NIS taxa, whereas more nonindigenous Bryozoa species than other taxa overlapped between marinas and the frames, buoys, and other in-estuary monitoring sites (the SWMP YSI sites of the NERRS). One nonindigenous Nudibranchia was found at non-marina sites only. The NIS at marinas only included 9 nonindigenous Tunicata species, 4 NIS Bryozoa, and 1 NIS Cirripedia. In contrast, the NIS at both marinas and other sites included 5 Tunicates, 7 Bryozoa, and 2 Cirripedia. Therefore, around 1/3 of the Tunicate and 2/3 of the Bryozoa and Cirripedia NIS dispersed to non-marina sites.

Further subdividing the non-marina plates into in-estuary frames (the lower four NERRS; Kachemak was excluded from the analysis as it had no NIS at any type of site), in-bay buoys (Gulf of Farallones area buoy sites), and offshore buoys (other three Sanctuaries) shows the non-marina plates differ in NIS by habitat or deployment method. No NIS were found on offshore buoys (N = 15 sites),  $1.5 \pm 0.28$  NIS per site were found on in-estuary frame-deployed plates (N = 24 sites), and  $3.0 \pm 0.9$  NIS per site were found on the in-estuary buoy-deployed plates (N = 6 sites).

## Trapping

We trapped the invasive green crab, *Carcinus maenas* (henceforth *Carcinus*) in Elkhorn Slough. *Carcinus* were only found in Elkhorn Slough although they were previously found near our South Slough trapping sites and they have been found as far north as British Columbia. These crabs were common in Elkhorn Slough. They represented from 10 to 99% of the crab catch at 6 of the 7 sites we trapped, at all sites but the one at the mouth of the slough (**Fig 8**). They were the numerically dominant crab in the Hummingbird Is, North Marsh, and Azevedo Pond sites. The carapace width of these *Carcinus* averaged  $51.4 \pm 8.4$  mm (mean  $\pm$  SD, N = 904), and ranged from 28 to 86 mm wide. *Carcinus* was the only verified nonindigenous species in our traps. We trapped 3 fish in Tijuana River that may have been *Tridentiger trigonocephalus*, but we did not verify the field identification; this non-native goby is already documented in this area.

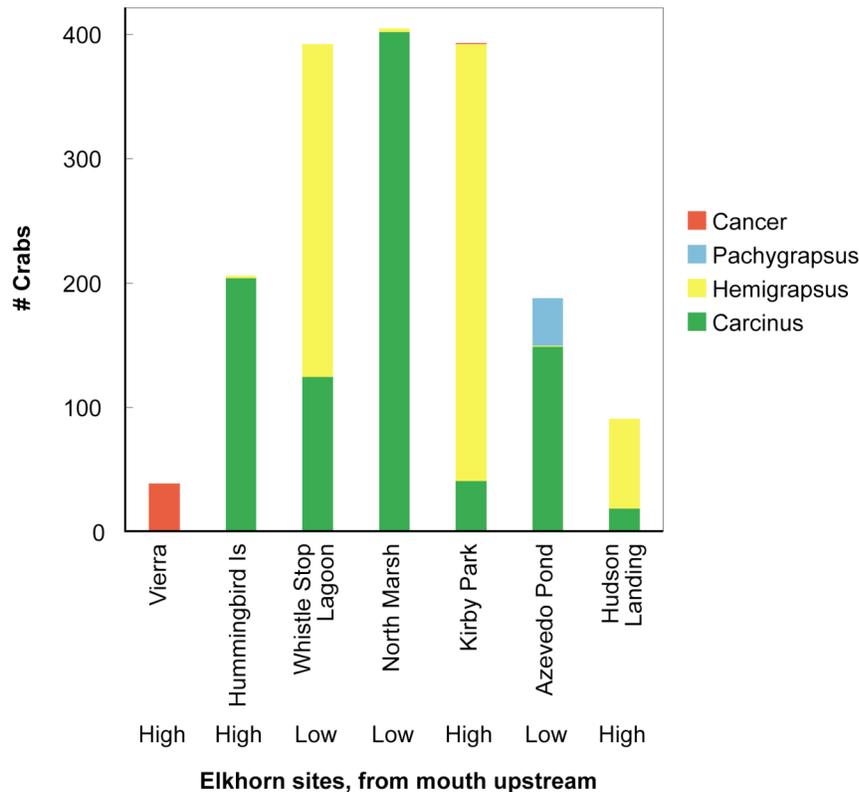


Figure 8. The number of trapped nonindigenous *Carcinus maenas* and native crabs across 7 sites in Elkhorn Slough NERR.

Across the protected areas, we caught 7 crab and 10 fish species in our traps (**Table 5, Appendix B**). The native shore crab *Hemigrapsus oregonensis* was most abundant in the Tijuana River traps. Both *H. oregonensis* and *Carcinus* were abundant in the Elkhorn Slough traps. These crabs were absent and *Cancer magister* was abundant in the South Slough traps. Fewer crabs were caught at the trapping sites in the more northern protected areas. The most common fish we caught were *Gillichthys mirabilis* and *Leptocottus armatus*. We caught more crabs and fish per trap in Elkhorn Slough than in the other protected areas.

## **Discussion, Broad-Scale Project**

To determine the impact and spatial patterns of nonindigenous species (NIS), we identified to species the four most common fouling community taxa, Bryozoa, Cirripedia, Hydrozoa, and Tunicata, plus Nudibranchia, from settling plates in and around nine US west coast protected areas. NIS accounted for more than one quarter of species richness in the 5 taxa under study at these protected areas. Tunicate species richness was especially influenced by NIS: 53.8% of the tunicate species found on our settling plates were introduced. Bryozoa, which represented the highest number of NIS in a taxa, and Cirripedia also had high proportions of NIS. Hydrozoa and Nudibranchia had just one NIS each. The most common NIS were Tunicate and Bryozoa species. These findings of many NIS Tunicates and Bryozoa deviates from the literature review finding (Ruiz et al. 2000) that most NIS identified in previous studies were crustaceans or mollusks. In fact, the mollusc order we identified, Nudibranchia, had the lowest percent NIS (5.9%) out of the five taxa under study. This difference illustrates why it is imperative to conduct broad geographic surveys using uniform protocols rather than relying on literature reviews. As more of the taxa on our fouling plates are identified, we will be able to evaluate the relative risk of invasions for a number of taxonomic groups.

The species richness at many of the protected areas was highly influenced by NIS. For example, Tijuana River Reserve had 21 NIS (51.2% of species). Even larger proportions of species were introduced in Monterey Bay Sanctuary (57.1%, from 12 NIS) and adjacent Elkhorn Slough Reserve (53.6%, from 15 NIS). Due to facilitation of new NIS by already established NIS, these protected areas have an especially high risk of new invasions as well as ecological or economic impacts from the NIS already established. Kachemak Bay Reserve is at the other extreme, with no NIS found in this study, yet is not free of NIS and is also vulnerable to new introductions. While no NIS in the five taxa under study were found in Kachemak Bay, Alaska, our previous studies identified two NIS in Kachemak, and two additional NIS in nearby Prince William Sound.

Our results supported the hypothesis that NIS decrease with increasing latitude and that they serve to drive a similar pattern apparent in overall species richness across latitudes. We found a strong pattern of decreasing species diversity and abundance along the US west coast from Tijuana River in the south to Kachemak Bay in the north. Gulf of Farallones and Olympic Coast sites had high diversity and abundance given their relative latitudes. The pattern of decreasing species richness with increasing latitude was largely driven by nonindigenous species. NIS showed this same pattern of decreasing species richness across increasing latitudes. However,

when NIS were subtracted from the total species count, the latitudinal pattern was no longer evident for the native plus cryptogenic species.

These results combined with data from SERC's additional studies of NIS in the fouling community of ports and marinas in 24 US bays will be used to identify whether the strong latitudinal pattern identified here exists across coasts. We have already compared NIS across latitudes from this study with NIS from this plus other studies (Appendix D). This comparison showed the points were less clustered about the regression line for the combination tally, perhaps because methods varied across the studies used in this compilation and taxonomic group may have been studied unequally. Moreover, the latitudinal pattern of all studies combined would have been less apparent without the datapoints from this study.

Ships serve as transport vectors for many NIS, especially ones in the fouling community (Carlton 1986, Ruiz et al. 2000). Therefore, commercial ports and also marinas likely serve as a point of entry for many NIS, including several tunicates and the kelp *Undaria* (Lambert & Lambert 1998, Thornber et al. 2004). We found the marinas in and near the west coast protected areas were highly impacted by NIS. While half the NIS found in this study lived only in marinas, all the NIS in non-marina areas were also found in marinas except one Nudibranch species. This further supports the idea that marinas and ports are the entry point for NIS into more natural habitats in estuaries, bays, and offshore. However, this study found little overlap between the occurrence of NIS in marinas and their occurrence further in the estuaries or, especially offshore, where no NIS were found. This lack of overlap could be due to the fact that marinas provided hard substrate near the plates whereas very few of the plates in more natural areas were set near hard substrate. Areas with natural hard substrate may have a much higher overlap in community structure with marinas. A large proportion of the NIS in marinas were tunicates (9/14 NIS species), which have brief, local larval dispersal. Clearly, the question of dispersal of tunicate and other NIS from marinas, and more importantly from commercial ports, to nearby natural areas needs to be studied further.

This study already identified five new range expansions, including two new US West Coast introductions. Four of these species were found at marinas only and the fifth was found at both marina and non-marina sites.

The *Carcinus maenas* data from our trapping illustrate the impact and high abundance NIS can reach after a short timespan. This species is dominant in the crab community in Elkhorn Slough. Similarly, *Carcinus* have impacted native crabs at other west coast sites. Other studies have shown reciprocal abundance from native crabs due to *Carcinus*'s impacts on small native crabs and also to large native *Cancer* crabs impacting *Carcinus* (Grosholz et al. 2000, Hunt & Behrens Yamada 2003). The absence of *Carcinus* in South Slough during this study may be due to the large number of *Cancer* crabs caught there.