Baseline Characterization of Biodiversity and Target Species in Estuaries along the North Coast of California

Final Report

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Acknowledgments

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Zachariah Badaoui – In Memoriam

During the early summer of 2014 Zach came up from Texas to start his Master's degree with me and join our estuarine MPA team. Sadly, we lost him during January 2015 due to a medical condition. He was so excited to be exploring the estuaries of the north coast. Every alga, invertebrate and fish was new to him and, whether he was kneeling in the mud or back in the lab, he reveled in each discovery. Zach would want me to tell you that it was all neat, but that the algae were the best! Of course. I can still see him with a nose in a quadrat counting infaunal holes; holding a tweezers as he sorted through sieved sediments; pawing through the mud in the seine to find the fish; quietly floating on a kayak as the tide carried him along; helping out in the kitchen to prepare for the field team's evening meal. It's also fortunate that he was so good natured, because there was a lot of bantering and laughing during those field excursions. I can still see him with that whimsical smile as he rolled his eyes in response to some quip. Zach knew he was among friends. The baseline monitoring proceeded along without Zach, but there were many times that I quietly thought – "Look Zach! This is an entire bed of *Gayralia*, not *Ulva*, do you understand what that means about the conditions at this site?!", or I would look up expecting to see him hunched over the sieving table. I am sure that many of the people on our field crew had similar moments. We miss him.

Executive Summary

Despite being one of the most productive ecosystems on the planet, estuaries receive minimal conservation attention relative to terrestrial and ocean spaces even though they are among the most threatened of ecosystems. The extent of this threat is not surprising given the high densities of coastal human populations. While some stressors of estuarine health originate within the estuary, many come from anthropogenic activities around the estuary and in its watersheds. This raises the question of how best to proceed with estuarine conservation. Should a 'no-take' approach be used within the estuary, or should land use practices also be addressed? Perhaps more than for any of the other habitats getting MPA baseline monitoring, answering this question is critical to the ultimate goal of healthy estuaries.

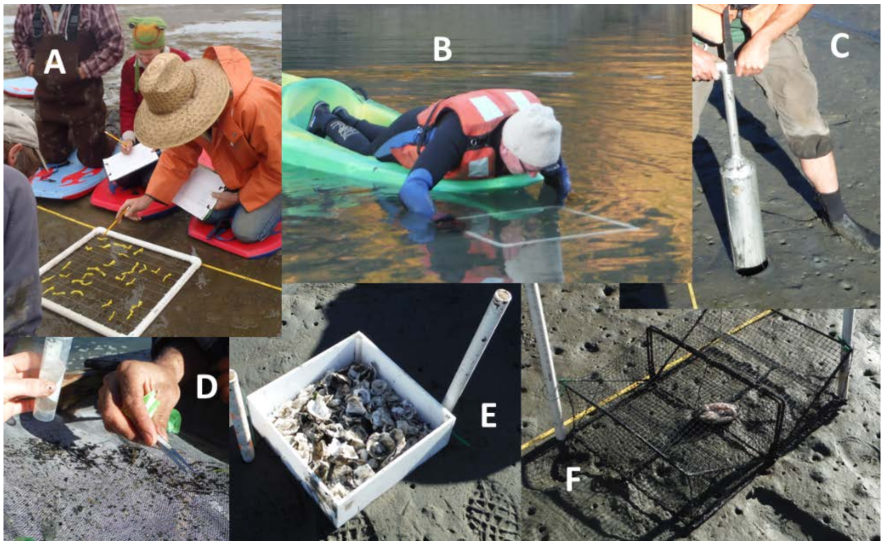
There are 22 estuarine MPAs in California. The present baseline monitoring project for estuaries in the North Coast MPA Region is the first of the MPA projects in the state to study this system. The findings of this study are valuable for providing stakeholders and resource agencies with a picture of biodiversity and target species in estuaries that have received minimal to no study. State and federal agencies need to know if organisms of special interest, like rockfish, salmonids and Dungeness crab, are present in estuaries, as well as knowing the status of critical fish habitats like eelgrass beds. Baseline descriptions are also valuable for understanding how these systems are responding to climate change (MPA Monitoring Enterprise 2012). While this baseline information is valuable to a variety of stakeholders, there is no reason at this time to expect the North Coast Region estuarine MPAs to be immediately effective as a conservation tool. In part, this may be because land use practices should be more highly prioritized for some estuaries, but also because in the North Coast Region most of the 'take' activities are still allowed within estuarine MPA boundaries.

In the interest of providing baseline information to stakeholders, and in enabling future studies that might test for a site-specific event (e.g. change in management, a site level disturbance), the present study had the following goals:

1. To provide contextual descriptions of the physical environment in and around these estuaries if that information already exists,
2. To describe the biodiversity in estuaries from the northern (CA-OR border to Cape Mendocino, CA) and southern (Cape Mendocino to Point Arena, CA) bioregions of the North Coast MPA Region,
3. To provide more detailed information (i.e. abundance, body size, distribution) for target species in these estuaries,
4. To use the data and experiences from the first three goals to make recommendations on how to focus future long-term monitoring efforts.

The final membership of the estuarine project team was a product of a year's worth of traveling up and down the North Coast MPA Region to hear what people wanted to find out about their estuaries, and to find out if and how people wanted to collaborate. Project leader expertise ranged from marine biology faculty from Humboldt State University, environmental resource monitoring from the Wiyot Tribe, fisheries biology from the consulting firm H.T. Harvey & Associates, and physical oceanography from UC Davis. The project leader, Dr. Shaughnessy, also worked with the Intertribal Sinkyone Wilderness Council to recruit a tribal intern, but this was not successful.

The baseline study used four estuaries, two in the northern bioregion (Mad River, Humboldt Bay SMRMA) and two in the southern bioregion (Ten Mile River SMCA, Big River SMCA). Two sites were sampled within each estuary, and for all the organisms except fish, sampling within each site was stratified by mid and low intertidal elevations. Monitoring occurred June 2014, January 2015, June 2015, January 2016 and June 2016. Three trophic levels – macrophytes (i.e. seagrasses, seaweeds), invertebrates, and fish – were described using a variety of sampling methods (ES 1). Descriptions of biodiversity and the abundance and size of target species came from these surveys.

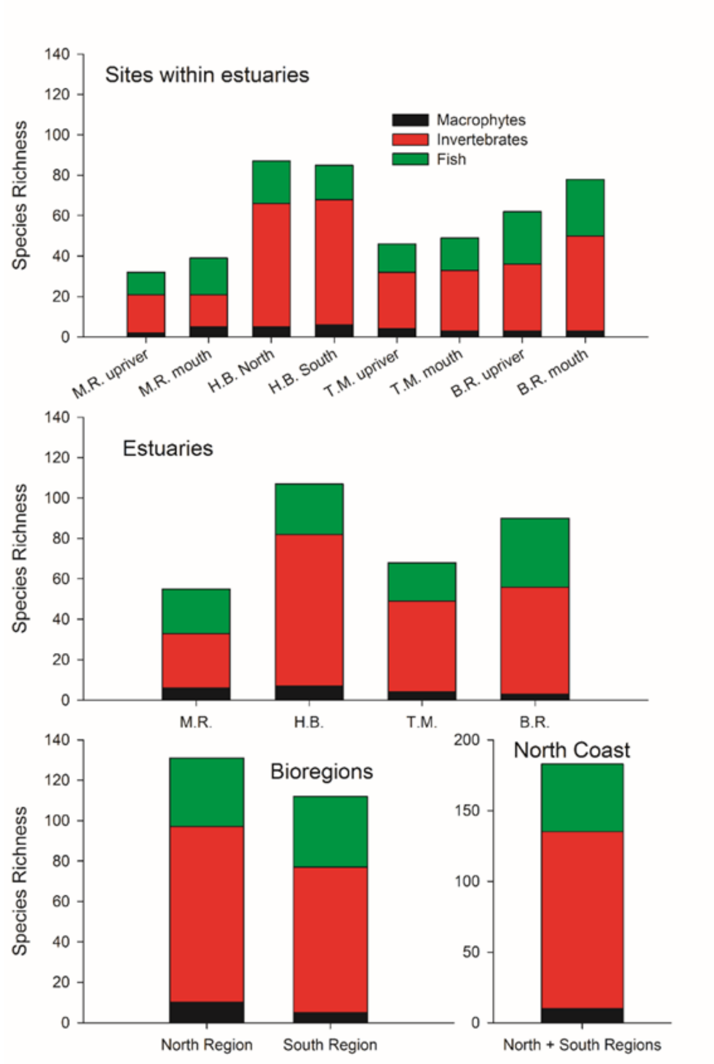


ES 1. Sampling methods for macrophytes and invertebrates, including training (A), using a kayak for quadrat sampling in order to minimize disturbance (B), sampling infauna with the clam gun (C), sorting infauna on the sieving screen (D), box trap with oyster shell (E), and crab trap (F).

Large spatial and temporal scale information about the physical, contextual environment for this study came from the MPA Project: Characterization and Indicators of Oceanographic Conditions (Bjorkstedt, Tissot, Sydeman, Largier, Garcia-Reyes). Of the data products produced by this project, we used sea surface temperature to characterize ocean conditions and river discharges to compare watershed environments. At the finer scale within each estuary, since most estuaries do not have stations for measuring water quality, and the Terms and Conditions of the grant contract for this study prohibited the purchase of instruments for obtaining new data about the physical environment (e.g. water temperature, salinity), there are limitations to comparing the estuarine communities and target species to the physical environment in which they occur. This study did use existing equipment to develop contextual information for the Humboldt Bay SMRMA, the Ten Mile River SMCA, and the Big River SMCA.

The oceanic and watershed conditions during this baseline study were anomalous and of such strength that the typical spatial differences in upwelling and precipitation north and south of Cape Mendocino were partially equalized. There was more variability in sea surface temperature and river discharge among study years. The warm ocean "blob" conditions and the lowest river discharges characterized 2014. With the building of El Niño during 2015, the coastal water became even warmer, strong north to south currents were established, and river discharges increased. These large-scale climate drivers of oceanographic and watershed conditions interacted differently with the geomorphology of each estuary. For example, the Ten Mile River SMCA was converted into a seasonal lagoon as the beach was built up, thereby not always being affected by conditions in the near ocean, whereas the mouth of the Big River SMCA stayed open all summer. The implication of these large-scale changes in oceanographic and watershed climate interacting with the local geomorphology of each estuary is that there is the potential for many types of estuarine habitats to exist, and so foster high estuarine biodiversity within a bioregion (e.g. Pt. Arena to Cape Mendocino).

This is what was found in the biodiversity portion of our study (ES 2). There were important spatial changes in community structure for macrophytes, invertebrates and fish between sites within an estuary, and particularly among estuaries. Each of these three trophic levels appeared to separate on a gradient of salinity. When both sites within an estuary were close to the river mouth their communities were more similar to each other than when two sites, from another estuary, were in very different positions along the salinity gradient. As well, estuaries with strong connections to the ocean contained different communities than those with interrupted connections to the ocean, which is a finding consistent with other studies of estuaries in the state (e.g. Chamberlain 2006). Within each trophic level, ordination



ES 2. The accumulation of estuarine species with spatial scale (M.R. = Mad River estuary, H.B. = Humboldt Bay SMRMA, T.M. = Ten Mile River SMCA, B.R. = Big River SMCA).

comparisons among years did not show community changes although PerMANOVA analyses indicated that some sites did shift more over time than other sites. These sites may have been in estuaries with a stronger watershed influence, like the Mad River and Big River systems.

Using geomorphological diversity as a predictor like other studies have done (e.g. Edgar et al. 2000), the implication of our biodiversity study to the conservation of estuarine marine biodiversity is that the present system of estuarine MPAs in the North Coast Region likely captures a small fraction of the estuarine biota. Our study would likely have described other communities if we had been able to sample sites further upriver within the Ten Mile SMCA. It is also the case that estuarine MPAs encompass small areas of the estuary in which they are located, which is the case for the Humboldt Bay SMRMA and the Big River SMCA. Finally, the major river estuaries and lagoons north of Cape Mendocino contain no MPAs.

The abundance and size of estuarine target species is also presented in this report. These species were divided according to how they use the estuary: **Ocean & Estuary**, **Estuary Residents**, **and Anadromous Fish**. This organization will hopefully facilitate the use of this information for studies on marine habitat connectivity, which would focus on species in the ocean and estuary group, but might also include some estuarine residents like eelgrass, which export detritus to outer coast beaches. Studies of effects of events to specific estuarine sites (e.g. MPA actions, a localized disturbance) should consider the species in the **Estuary Residents** section, and resource managers may want to view all three sections.

Unlike the biodiversity section of this study where the importance of habitat variability is emphasized, some target species demonstrated no abundance or size patterns in space or time, whereas others did, but did so in opposite ways. For example, eelgrass was abundant during 2014 in both the Humboldt Bay SMRMA and the Big River SMCA but it declined in both locations during 2015 and 2016, whereas Staghorn sculpin demonstrated the opposite temporal pattern, being lowest in abundance during 2014 and higher during 2015 and 2016. Interpretation of why the spatial and temporal changes in populations of target species occurred is limited by the lack of pre-MPA data, the short time span of the present study, and the incomplete description of the physical context within each estuary. Our understanding of what is driving the variability of target species would be advanced by extending the time they are monitored and, for some species, coordinating the monitoring among habitats.

Our report contains recommendations to consider for monitoring any of the 22 estuarine MPAs in California, as well as for a revised long-term monitoring program for the estuaries in the North Coast MPA Region. The first is to consider whether or not there is alignment between the threat to an estuary and the nature of the estuarine MPA regulations. Is there a reasonable chance of detecting an MPA effect given this background, or should a design be used that anticipates future site-specific changes? Some of the recommendations for North Coast estuaries, which may also apply to other estuaries in the state, include the purchase of simple instruments for measuring water temperature and salinity; a focus on target species and not biodiversity; choosing target species that have high site fidelity; the use of paired reference sites within the same estuary; strategically expanding baseline monitoring to include a few easily measured salt marsh target species; the use of an unmanned aerial vehicle to remotely sense habitat conditions every three years.

The present study has provided detailed baseline information about the macrophytes, invertebrates and fish for several estuaries in the North Coast Region. Many of these organisms are important to stakeholders and, in some locations like the Mad River estuary and the Ten Mile River SMCA, this is the first time that the biota and physical processes have been comprehensively described. Geomorphological and hydrological differences among estuaries were associated with distinctive plant and animal communities. Species of management interest, such as *Metacarcinus magister*, rockfish, salmonids and seagrasses were found in all of the estuaries, but the differences in abundance and size of each species indicate that some estuaries are more optimal for a particular species than other estuaries. Estuarine MPAs in the North Coast Region may prove to be beneficial by preventing the direct loss of habitats due to future anthropogenic activities, but given the environmental and social context of the North Coast, the more immediate value of the present baseline study may be in how it is used during an outreach process to further inform coastal communities how their backyard activities potentially affect estuarine life.

Introduction

Estuaries are some of the most productive and diverse ecosystems on the planet (Kennish 1990) and they come in many forms. Systems for classifying estuaries use geomorphological features such as how well connected the estuary is to the ocean, and if the estuary is a linear riverine system or if it contains embayments in which lagoon conditions form. Hydrologically, some estuaries receive large rivers whereas others are characterized by seasonal inputs (Hume and Herdendorf 1988, Cooper 2001, Elliot and McLusky 2002, Chuwen et al. 2009, Potter 2010). This physical variability corresponds to distinctive biological communities, and so approaches to estuarine conservation need to address the wide range of habitats that occur within and among estuaries (Edgar et al. 2000). Beta-diversity – the accumulation of new species when additional sites in a region are added (*sensu* Socolar et al. 2015) – is potentially high for estuarine communities because so many types of estuaries can occur within a small region (e.g. Hastie and Smith 2006).

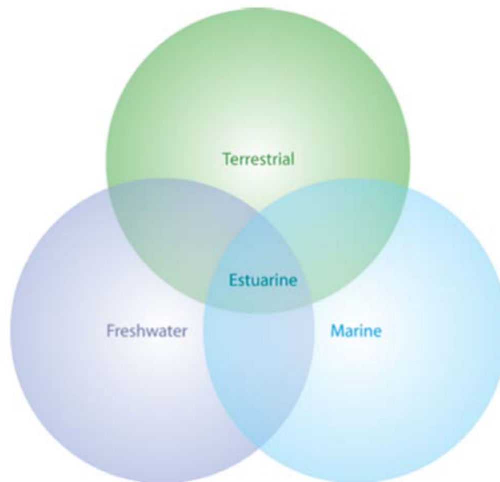


Figure 1. Conceptual model of estuarine ecosystems as influenced by terrestrial, marine and freshwater environments (modified from Gleason et al. 2008).

Estuarine biodiversity and ecosystem functions are affected by processes occurring within estuaries, as well as by terrestrial conditions and activities, freshwater discharges, and nearshore ocean conditions (Figure 1). These coastal ecosystems have always attracted human settlement, and so many have been extensively modified, such as by dredging to allow for navigation, by diking and filling of estuarine habitats to promote other land uses, and by pollutants produced in or carried to estuaries (Gedan et al. 2009). Consequently, estuaries rank among the most threatened of any ecosystem (Kennish 1990).

The types and intensities of threats can be specific to an estuary. In general though, because of the surrounding watershed conditions and activities, conservation of estuarine biodiversity and ecosystem functions often focuses more on the combination of land use practices and within-estuary activities which affect estuarine water quality (e.g. http://www.chesapeakebay.net/), rather than only setting up estuarine no-take areas. Estuarine seagrass beds, for example, would experience less light attenuation if agricultural nutrients and suspended sediments were prevented from reaching estuaries (Ralph et al. 2007). Exceptions to this generalization would include cases where an estuarine MPA could prevent a future activity that might degrade marine habitats, such as dredging or building structures that reduce aquatic light.

Compared to the conservation efforts focused on terrestrial and ocean systems, similar efforts aimed at estuaries have been minimal (Edgar et al. 2000). In California, the Marine Life Protection Act has recognized the importance of estuaries by initiating a public process that resulted in the establishment of 22 estuarine MPAs in the state, four of which are in the North Coast Region. Three of these four are small, linear riverine estuaries, and the Humboldt Bay MPA is an oceanic embayment. Unlike estuaries located on more southern coastlines in the state, these four North Coast estuarine MPAs are not surrounded by dense human populations; e.g. the two largest towns in the Humboldt Bay watershed, Eureka and Arcata, have a combined population of ~ 45,000 people (Draft Transit Dev Plan Humboldt County Systems 2011). But it does not take many people to generate a threat to an estuary. On the North Coast, agricultural water diversions reduce summer freshwater flows into estuaries, and logging/road building practices on steep slopes free up inherently unconsolidated sediments that fill estuaries. The largest threat to estuaries between Cape Mendocino, CA and Cape Blanco, OR is turbidity and sediments, which may contain dioxins (Price-Hall et al. 2015). On the continental shelves, just offshore from rivers between these two capes, the sedimentation rate after 1950 was two to three times greater than from 1000 AD to 1950, which is partly attributable to the logging and road building practices during the second half of the 20th century (Sommerfield et al. 2002, Sommerfield and Wheatcroft 2007). This type of sediment delivery to estuaries and nearshore habitats was more recently demonstrated during the February 2017 floods (Figure 2).

Figure 2. Sediment plumes from rivers along the North Coast Region of California during February 2017.

The present baseline monitoring project for estuaries in the North Coast MPA Region is the first of the MPA projects in the state to study this system. The baseline study undertaken here is valuable for providing stakeholders and resource agencies with a picture of biodiversity and target species in estuaries that have received minimal to no study. State and federal agencies need to know if organisms of special interest, like rockfish, salmonids and Dungeness crab, are present in estuaries, as well as knowing the status of critical fish habitats like eelgrass. Baseline descriptions are also valuable for understanding how these systems are responding to climate change (MPA Monitoring Enterprise 2012).

It is also important to understand what is not being achieved by the estuarine MPAs in the North Coast Region. Most of the 'take' activities that occurred prior to formation of these estuarine MPAs are allowed to continue today (Table 1), even though one of the features of a successful

MPA is considered to be an enforced no-take policy (Edgar et al. 2014). As well, even though there are physical differences among the four estuarine MPAs in the North Coast Region, the large riverine estuaries of the Smith, Klamath and Eel rivers, and lagoon systems that only occasionally breach like Big and Stone lagoons, do not contain MPAs (Figure 3). The Big River SMCA and the Humboldt Bay SMRMA are also small relative to the size of the estuaries in which they occur, and so may not represent more local biodiversity.

Table 1. Permitted/prohibited uses and exemptions in the four estuaries used in the present study. [1] For regulations that apply to all estuaries see http://www.dfg.ca.gov/regulations/. [2] Text from the Guide to the Northern California Marine Protected Areas by the California Department of Fish and Wildlife.

Estuary	MLPA Permitted/Prohibited Uses	MLPA Exemptions
Mad River Estuary	[1]None	[1]None
South Humboldt Bay SMRMA	[2]Take of all living marine resources is prohibited.	[2]Waterfowl, Scientific Collection Permit, Anchoring, Vessel transit, Monitoring, Safety, Tribal take
Ten Mile Estuary SMCA	[2]Take of all living marine resources is prohibited.	[2]Take pursuant to activities authorized in subsection 632(b)(21)(D) is allowed; Waterfowl, Science Collecting Permit, Anchoring, Vessel transit, Monitoring, Safety, Tribal take
Big River Estuary SMCA	[2]Take of all living marine resources is prohibited **except**: -Recreational take of surfperch (family Embiotocidae) by hook and line from shore only. -Recreational take of Dungeness crab by hoop net or hand.	[2]Take pursuant to activities authorized in subsection 632(b)(21)(D) is allowed; Waterfowl, Science Collecting Permit, Anchoring, Vessel transit, Monitoring, Safety, Tribal take

Figure 3. Locations of estuaries, including MPA estuaries (red), along the North Coast Region of California.

Project Goals:

1. To provide contextual descriptions of the physical environment in and around these estuaries if that information already exists,
2. To describe the biodiversity in estuaries from the northern (CA-OR border to Cape Mendocino, CA) and southern (Cape Mendocino to Point Arena, CA) bioregions of the North Coast MPA Region,
3. To provide more detailed information (i.e. abundance, body size, distribution) for target species in these estuaries,
4. To use the data and experiences from the first three goals to make recommendations on how to focus future long-term monitoring efforts.

Methods

Physical Context

Descriptions of broad spatial and temporal scales of oceanic and watershed conditions were provided by the MPA North Coast Project: Characterization and Indicators of Oceanographic Conditions (Bjorkstedt, Tissot, Sydeman, Largier, Garcia-Reyes; 2017). More specifically, discharge data from the Mad and Eel Rivers was used to represent watershed conditions in the northern bioregion. Since Ten Mile River and Big River are not gaged, discharge data from the other rivers in the same bioregion (i.e. Noyo River, Navarro River) were used as proxies. The Sea Surface Temperature (SST) product from this MPA project group was used to understand the spatial changes in oceanic conditions over the 2014 - 2016 span of our study.

For describing the finer scale of physical conditions within each of the four estuaries, no pertinent information is available on watershed biological and chemical loading. For southern Humboldt Bay, high-frequency water property data are available from the CeNCOOS monitoring site, on the eastern shore of this bay. These data include temperature, salinity, pH, chlorophyll fluorescence, turbidity and sub-surface pressure (water level).

The present study deployed instruments to describe the physical environment in the Ten Mile River SMCA and Big River SMCA (hereafter called TM and BR, respectively) estuaries in Mendocino County. For TM, time series data on water level and temperature in the estuary were taken from 27 June 2014 to 3 January 2015 and from 26 June to 30 December 2015. A fixed pressure-temperature recorder (Onsett Water Level Logger) was deployed at 39.54478N and 123.75826W. The water level indexed the degree of tidal exposure, and was used to identify periods of closure (or perched conditions). Spatial surveys were conducted during 27 June 2014 (YSI CastAway for profiles of temperature and salinity; YSI 650 handheld sonde for spot values of dissolved oxygen at depth) and 26 June 2015 (SeaBird 19+ with chlorophyll fluorescence and dissolved oxygen sensors), which was when the estuarine communities were monitored. For BR, Time series data on water level and temperature in the estuary were measured from 28 June 2014 to 4 January 2015 and from 26 June to 30 December 2015. A fixed pressure-temperature recorder (Onsett Water Level Logger) was also deployed at 39.30172N and 123.76871W in BR.

Study Design for Baseline Monitoring

The study design and sampling methods were chosen to meet two of the goals of the present baseline study: 1) a description of estuarine biodiversity, and 2) enumeration of the abundance and size of target species. Estuarine birds and mammals were deliberately omitted from this study because, being so wide ranging, it is difficult to attribute fluctuations in their abundance to the conditions within an MPA. The estuarine MPAs extend up to the Mean High Water (MHW) tidal datum and so include salt marsh habitat. This habitat was not included in the present study due to initial concerns about the time it would take to sample marsh, mudflat and low intertidal habitats.

For the estuarine MPAs in the North Region of California, almost all of the activities that were occurring in each estuary prior to the creation of the MPA are allowed to continue after the estuarine MPA was created (Table 1), so there is no expectation of an MPA effect. This circumstance had a large effect on the design of this baseline study. Rather than creating an MPA versus reference site design, two estuaries from the northern bioregion, Mad River estuary (hereafter MR; Figure 4), which is not an MPA, and the Humboldt Bay SMRMA (hereafter HB, Figure 5), were picked. TM and BR (Figure 6, Figure 7) were monitored in the southern bioregion between Cape Mendocino and Point Arena. The intention with the selection of these four estuaries was to capture some of the estuarine geomorphological variation that exists in these two bioregions and, with sufficient monitoring, provide the foundation for future BACI comparisons (Underwood 1994,

Figure 4. The Mad River estuary showing the locations of the mouth and upriver sites. Saline water has been detected in pools just east of the Hwy 101 bridge.

Murray et al. 2006) that could detect a site, estuary or regional effect on estuarine biodiversity or target species.

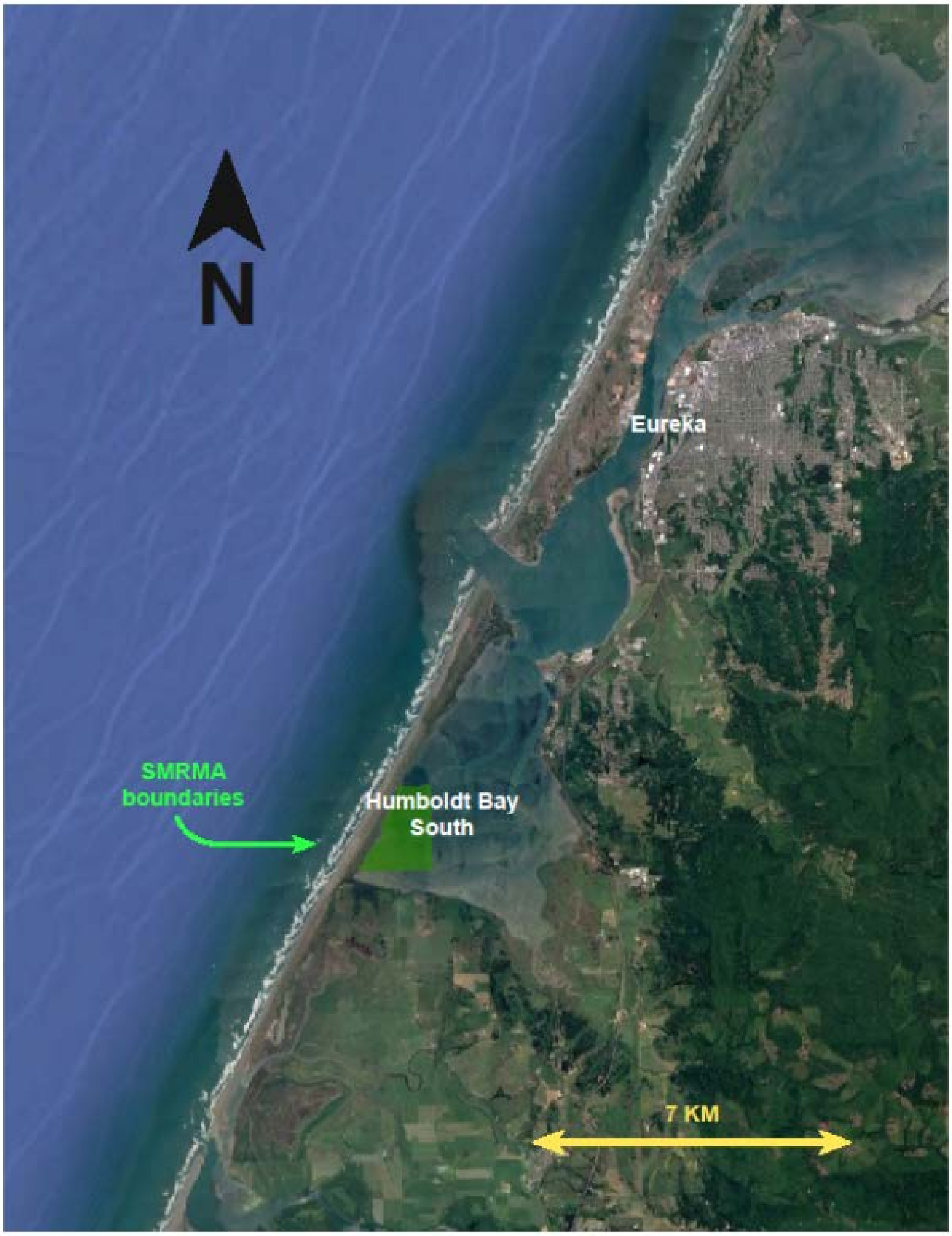


Figure 5. Location of the southern Humboldt Bay SMRMA; the North and South study sites were on the west side of the SMRMA.

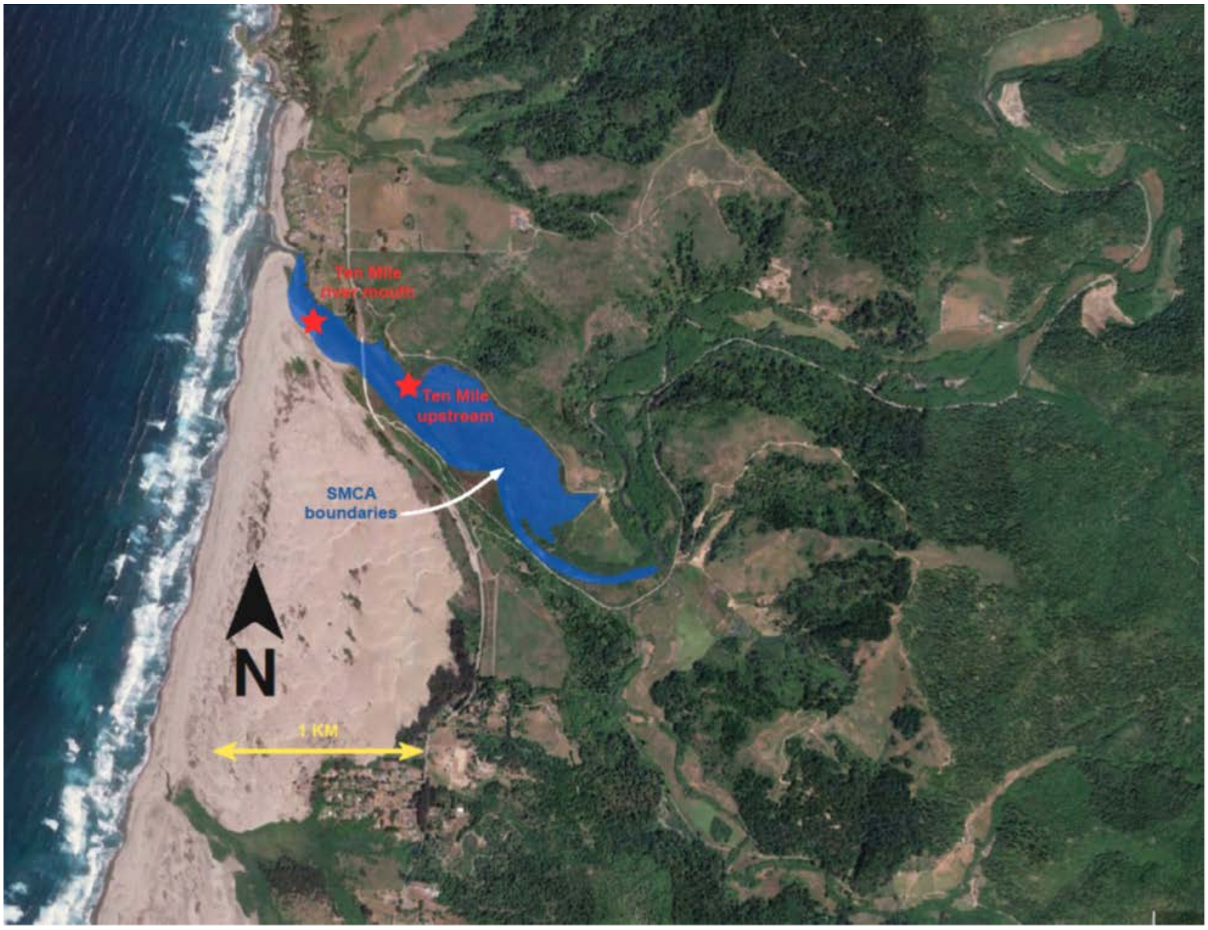


Figure 6. Location of the Ten Mile River estuary SMCA showing the upriver and mouth study sites.

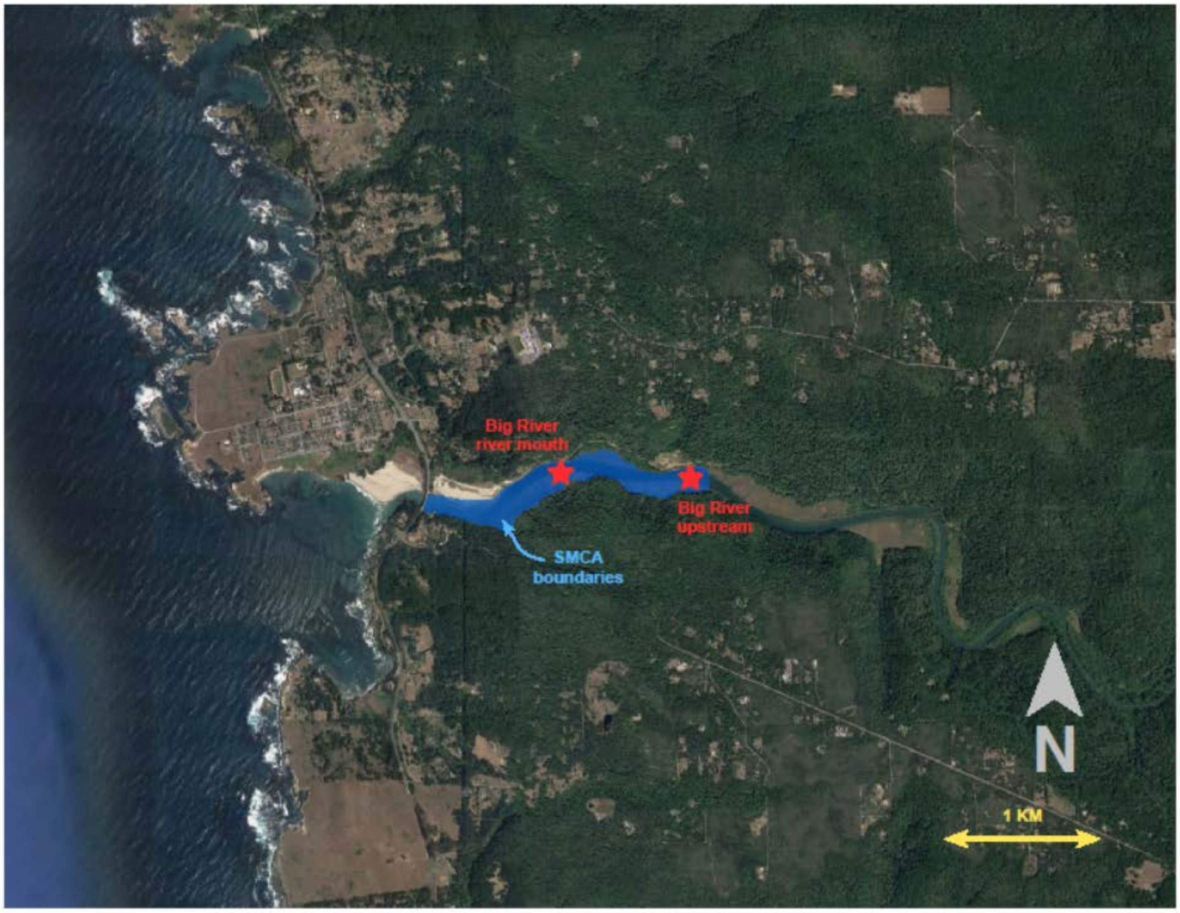


Figure 7. Location of the Big River estuary SMCA showing the upriver and mouth study sites.

Superficially, the four estuaries used in this study appear as three riverine estuaries (MR, TM, BR) and HB, but there are geomorphological and hydrological features that distinguish even the riverine systems (Table 2). The MR estuary flows over a beach that is partially perched during the summer (Figure 4). Of the four estuaries in the present study, MR has the largest and highest watershed (Table 2), therefore receiving both snowmelt and rain. In combination with the Matthews Dam, which forms Ruth Lake, this means that there is water to be released into the MR estuary during the summer which is the time when other estuaries with smaller and lower watersheds (e.g. TM, BR; Table 2), which also lack dams, experience a proportionately greater drop in summer freshwater discharge. Depending upon the slopes of the river valleys, riverine estuaries can experience a summer saltwater wedge that moves far upriver, as in the case of the BR estuary that is 13.3 km long (Warrick and Wilcox 1981). However, a riverine system like TM does not experience an enhanced summer oceanic effect if the beach becomes highly perched, which reduces exchange with the open ocean and results in the formation of lagoon conditions. The TM estuary is similar to the Pescadero Lagoon described by Largier et al. (2015). Relative to the datum NAVD88, the Mean Lower Low Water (MLLW) tidal datum in

Pescadero rises and falls as the barrier beach, respectively, builds during the summer and then potentially gets cut down by peak watershed discharge and wave events during the winter. Systems like Pescadero Lagoon and TM likely have a greater range of interannual physical conditions than estuaries that stay open during the summer like BR (Figure 7).

Table 2. Characteristics of the study estuaries and their associated watersheds. [1] Costa (1982), [2] Barnhart et al. (1992), [3] GMA (2001), [4] Mad River Watershed Assessment (2010), [5] www.wildlife.ca.gov/MPAs, [6] http://www.waterboards.ca.gov/northcoast/water_issues/programs/wpc/10tenmilesec2.pdf, [7] Warrick and Wilcox (1981).

Estuary Name	Estuary Type	Size of Summer Estuary & MPA within the Estuary at MHW (ha)	Relative Study Site Positions on Summer Salinity Gradient from Ocean	Watershed Area (ha)	Highest Watershed Elevation (m)	Average Annual Rainfall at low and high elevations, respectively, in the watershed (m)
Mad River	Riverine, moderately perched beach; water is released during the summer from the Matthews Dam[4]	44,289 (mouth to Route 101 bridge)[4]	Upriver: 70% Mouth: 7%	128,722[4]	[4]1,829	[4]1.02, 2.03
Humboldt Bay	[1]Tide driven coastal lagoon with limited freshwater input	South Bay: 6249. [5]SMRMA: 210	North: 0% South: 0%	[2]Entire Humbolt Bay: 57,757. [2]Salmon Creek: 6087	[2]457	[2]0.91, 1.3 3% of the water entering Humboldt Bay comes into South Bay[2]
Ten Mile River	Riverine, but a tidal lagoon when the beach is perched high	Entire estuary: not known [5]SMCA: 47	Mouth: 18% Upriver: 35%	[6]31,080	[6]977	[6]1.02, 1.8
Big River	[3]Riverine, open year-round	Entire estuary: 77. [5]SMCA: 34	Mouth: 7.5% Upriver: 13.5%	[7]46,879	[7]865	[7]1.02, 2.03

The hydrology of HB differs from the MR, TM, and the BR systems by having a deep-water connection to the open ocean that is maintained by the U.S. Army Corps of Engineers (Figure 5). No large rivers empty directly into HB. Southern HB, where the SMRMA is located (Figure 5), receives 3% of all the freshwater emptying directly into HB (Barnhart et al. 1992). The closest tributary to the HB SMRMA is Salmon Creek located in the SE corner of southern HB; this tributary drains a small watershed (Table 2). The extent to which lagoon conditions form in the HB SMRMA is unknown, but the oceanic effect in southern HB is considered strong; one study estimates that the average tidal prism for southern HB is 60% of the MHW volume (Barnhart et al. 1992). The four systems in the present study therefore have several distinguishing geomorphological and hydrological features that could affect the community structure of macrophytes, invertebrates and fish.

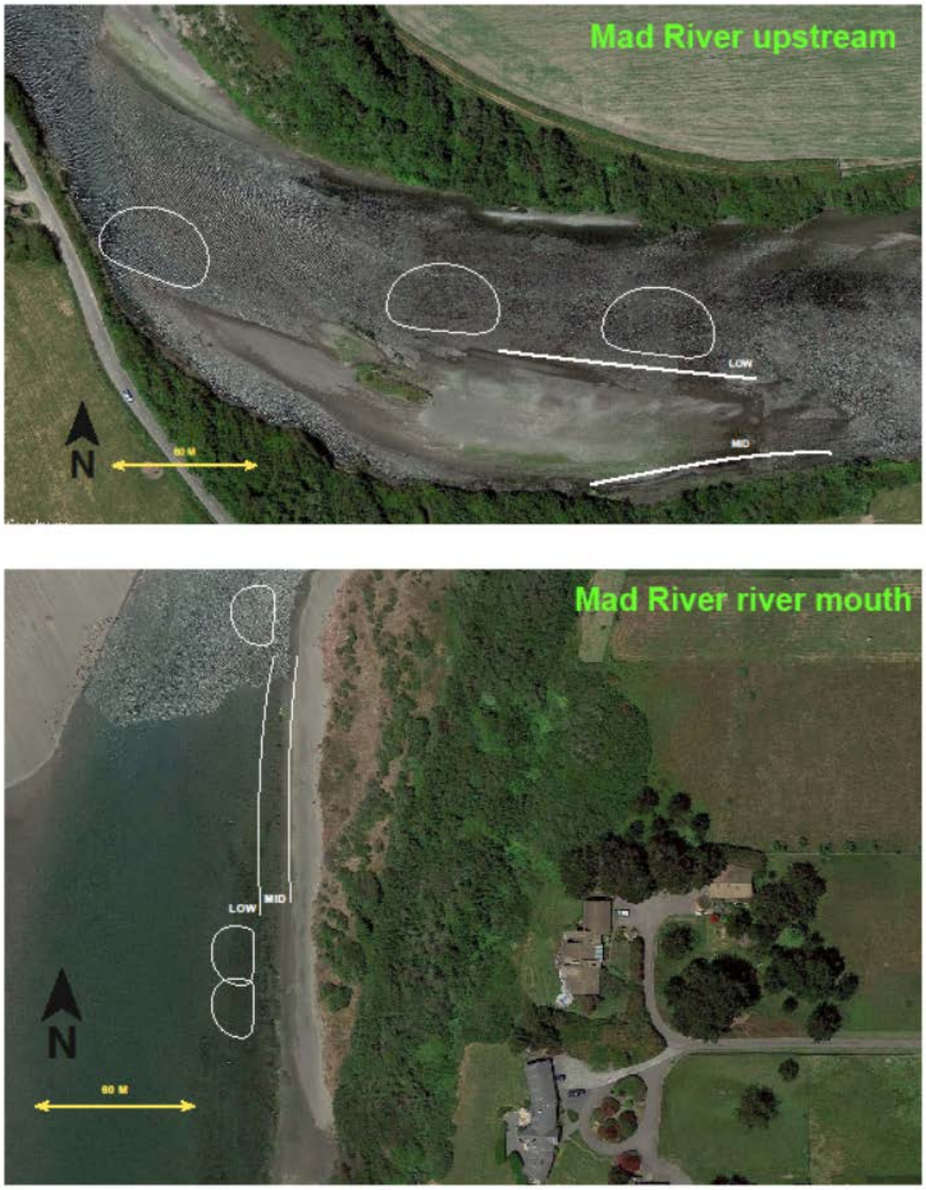


Figure 8. Locations of mid and low transects (white lines) at each Mad River site. The half circles are positions where beach seines were taken.

Figure 9. The Ten Mile River Estuary mouth and upriver study sites with positions of transects (white lines). Seines (white line circles) were used at the mouth site whereas, in the upstream site, a fyke net was placed across a channel coming out of a salt marsh.

Figure 10. The Big River estuary mouth and upriver study sites used; showing transect (white lines) and seining (white line half circles) positions. The orange dots, from left to right, correspond to the “figure point” in Table 16; they are locations where the deep edge of the eelgrass bed was measured.

Figure 11. Locations of mid and low transects (white lines) at each Humboldt Bay SMRMA site. The white line half circles are positions where beach seines were taken. Note the proximity of transects and seines to channels in the North site.

Two sites were sampled within each of the four estuaries. The two sites within the MR (Figure 8), TM (Figure 9) and BR (Figure 10) estuaries were picked to represent a habitat with a strong oceanic influence (i.e mouth site) and a site with more of a watershed effect (i.e. upriver site). The mouth sites used in MR and TM were close to the ocean where they experienced open ocean waves during high tide, and so sediments at these sites were dominated by sand, whereas the mouth site of BR was more protected and so comprised of soft mud. The upriver site in MR was located about 70% of the distance of the summer salt water wedge from the ocean whereas the TM and BR upriver sites were, respectively, 35% and 13.5% up the salt wedge (Table 2). Greater habitat diversity would likely have been captured by placing sites further upriver, but the BR MPA is short relative to the length of the entire estuary (Figure 10, Table 2), and the eastern end of TM (Figure 6) had no place from which to launch vessels or to keep vehicles and people away from the logging truck traffic. However, the upriver site that was chosen for TM was quite different from the mouth site in this estuary since the former site receives less wave activity. The upriver TM site was on a mudflat that fringed a salt marsh and there was a marsh channel emptying onto the northern end of this site (Figure 9). Both sites sampled in HB (North, South; Figure 11) were on the western side of the SMRMA because this location was the most accessible, and fish seines could be pulled on to the shore.

Low and mid intertidal elevations were sampled within each site within each estuary (Figure 8, Figure 9, Figure 10, Figure 11). Permanent 100 m transect lines for each elevation were either marked with pvc pipe or obvious landmarks (Table 3). Low transects were placed at or just below the estimate of 0.0 MLLW (based on the predicted time and height of the tide, and observing when low slack actually occurred). The low transect was placed slightly deeper at BR to describe the fringing eelgrass beds in this estuary that was underwater during most of each tide cycle. The low transects in HB occurred in the upper edge of that eelgrass bed. Mid intertidal transects in HB were set up on the mudflats at a vertical height halfway between the low transect and the edge of the salt marsh.

Table 3. Transect positions for each site and estuary.

Estuary	Site	Elevation	East or North end Transect: Latitude & Longitude	West or South end Transect: Latitude & Longitude
Mad River	Upriver	Mid	40.923042, -124.125091	40.923045, -124.125089
		Low	40.923334, -124.125492	40.923344, -124.125493
	Mouth	Mid	40.966663, -124.120839	40.922962, -124.126279
		Low	40.966681, -124.120975	40.923503, -124.126662
Humboldt Bay	North	Mid	40.716103, -124.257786	40.712209, -124.26057
		Low	40.71596, -124.257486	40.712098, -124.259967

	South	Mid	40.715206, -124.258047	40.711309, -124.260777
		Low	40.715189, -124.257826	40.711205, -124.260194
Ten Mile River	Upriver	Mid	39.547607, -123.760401	39.550772, -123.765759
		Low	39.546906, -123.759667	39.549998, -123.76511
	Mouth	Mid	39.546928, -123.759647	39.550055, -123.765171
		Low	39.547628, -123.760374	39.550731, -123.765907
Big River	Upriver	Mid	39.302986, -123.772091	39.303582, -123.781505
		Low	39.303094, -123.772079	39.303655, -123.781523
	Mouth	Mid	39.303115, -123.773254	39.303452, -123.782578
		Low	39.303246, -123.773252	39.303495, -123.782638

Sampling of Macrophytes, Invertebrates & Fish

Quadrats, which were $0.25m^2$, were subdivided every 0.05m by monofilament line, and 30 intersections were randomly picked so that percent cover could be enumerated used the point intercept technique (ES 1). Fifteen quadrats were placed at randomly selected locations on each transect line for measuring the percent cover of seagrasses, seaweeds and bare space. These general cover categories allow for comparisons to other estuaries and serve as ground truthing information for remote sensing studies. GPS coordinates were not recorded for each quadrat, but the start and end points of each transect are known (Table 3) as well as the position of each quadrat on the transect line. The entire quadrat was used to count the number of mudflat holes of varying diameters (i.e. < 2mm, 2 - 9mm, 10 - 19mm, 20 - 30mm) created by infauna. All of the shoots of *Zostera marina* or *Ruppia maritima* in a quadrat were removed except in the case of the Mad River upriver site where *R. maritima* was subsampled ($0.01m^2$) because shoot densities were high. All *Z. marina* and *R. maritima* shoots were bagged and placed in a cooler for processing in the laboratory. It is important to note that in all calculations of % cover, whether for a general cover category like macroalgae, or for a target species like *Z. marina*, the absence of a cover candidate was entered as a zero data point, not missing data.

Transect lines were also used for sampling infauna and the placement of box, minnow and crab traps (ES 1). A clam gun (12 cm diameter) was pushed 12 cm into the sediment at 5 random locations on a transect line. Trials with the clam gun demonstrated that this relatively shallow coring depth was usually missing the deeper bivalves and ghost shrimp, but these deeper excavations were so destructive and time consuming that it was decided to use the shallower cores along with the diameter classes of mudflat holes as a measure of larger and deeper infaunal presence. Sediment cores from the clam gun were placed in tubs and brought upshore where they were sieved through $1mm^2$ wire mesh (ES 1). Several representatives of each invertebrate encountered from each core were placed in 4% formaldehyde. These species were not counted as the intention of the sampling design was to create a presence-absence invertebrate matrix for the biodiversity analysis. Two minnow, two box (dimensions 30cm * 30cm * 10cm) and two crab traps (mesh size: 1.27cm * 1.27cm) were systematically placed on each transect line. The minnow and box traps were partially filled with old oyster shells to shelter the smaller invertebrates from predation whereas crab traps were baited with squid (ES 1). The minnow traps were dropped after the first year of the study because they were not catching anything that was not also being caught by the box and crab traps. All traps were deployed for 24 hrs. Small fish and crabs caught by these traps were counted, measured for size and sex, and released. Representative examples of smaller invertebrates, mostly isopods, amphipods and some shrimp, were removed from the traps and placed in 5% formaldehyde and then transferred a week later to 40% isopropyl alcohol.

Figure 12. Beach seining an eelgrass bed at the mouth site of Big River (A), at the upriver site in Mad River (B), and measuring fish caught in a seine (C).

Fish were sampled by doing two to three beach seines at each site within each estuary using a 45.7m (150') by 1.8m (6') seine with 6.4mm (1/4") mesh (Figure 12). Deep mud prevented the use of a seine at the upriver site in TM and so a fyke net (0.7 *0.7m wings and lead; two 0.7 * 1.0m frames with internal fykes; 6.4mm ($^1/_4$") mesh) was placed across a marsh channel at one end of this site (Figure 9). All fish were identified to species in the field (Figure 12); up to 30 individuals of each species were measured for length before releasing all of the fish.

Seasonal and interannual variation of macrophytes, invertebrates and fish was described by doing most of the fieldwork during June 2014, January 2015, June 2015, January 2016 and June 2016 (Table 4).

Table 4. Sampling dates for each estuary.

Year & Season	Estuary	Dates
2014 Summer	Mad River	6/15/2014 - 6/17/2014
	Humboldt Bay	6/13/2014 - 6/14/2014
	Ten Mile River	6/27/2014 - 6/28/2014
	Big River	6/29/2014 - 6/30/2014
2015 Winter	Mad River	1/14/2015 - 1/15/2015
	Humboldt Bay	1/16/2015 - 1/17/2015
	Ten Mile River	1/3/2015 - 1/4/2015
	Big River	1/5/2015 - 1/6/2015
2015 Summer	Mad River	6/9/2015 - 6/10/2015
	Humboldt Bay	6/4/2015 - 6/5/2015
	Ten Mile River	6/17/2015 - 6/18/2015
	Big River	6/15/2015 - 6/16/2015
2016 Winter	Mad River	2/14/2016 - 2/15/2016
	Humboldt Bay	1/20/2016 - 1/21/2016
	Ten Mile River	1/5/2016 - 1/6/2016
	Big River	1/7/2016 - 1/8/2016
2016 Summer	Mad River	6/24/2016 - 6/25/2016
	Humboldt Bay	6/22/2016 - 6/23/2016
	Ten Mile River	6/5/2016 - 6/6/2016
	Big River	6/7/2016 - 6/8/2016

Laboratory Processing

For each quadrat collection of the seagrasses *Z. marina* and *R. maritima*, the length of each shoot (i.e. turion) was measured, thus also providing shoot density for each quadrat. The dry weight of all the seagrass in a quadrat was measured after oven drying for 72 hours at 70 ^{o}C.

Invertebrates from all sampling sources were identified to species using Light (1954). Kozlov (1987) and the Oregon Institute of Marine Biology (https://library.uoregon.edu/scilib/oimb/OEI) were also used. WoRMS (http://www.marinespecies.org/) was used to find the current name for each species.

Data Analyses

Biodiversity

Lists of macrophyte, invertebrate and fish species were developed and include taxa from all the quadrat, seagrass epifauna, infaunal cores, traps, seine, and fyke net sampling techniques used over the course of the study. Sampling modes for determining the species richness of each trophic level were as follows: quadrat surveys for macrophytes; cores, box and crab traps for invertebrates; seines and fyke net for fish. All five sampling times were included in the counts of species richness.

In order to visually portray the variation in species identities among sites and times (i.e. beta-diversity; *sensu* Anderson et al. 2011), ordinations using Nonmetric Multidimensional Scaling (NMDS) were applied to each trophic level. Only the summer data for each trophic level were used in the ordinations since many rows in the ordination matrix (as quadrats, cores, traps, seines, fyke) contained no species during the two winters. PC-ORD (v. 5.1; McCune and Medford 2011) was used to further trim each matrix to remove the effects of rare species and sample units on the ordination. This step was followed by applying an arcsine squareroot transformation to the macrophyte % cover data. For the invertebrate presence-absence data and the fish count data, the trimming step was followed by applying Beal's smoothing using PC-ORD. This transformation can generate pattern when none exists if the number of species and sampling units in the matrix is too small (i.e. 10 species by 40 sample units; Caceres and Legendre 2008). All of the invertebrate and fish matrices used by the NMDS procedure surpassed the minimum sample sizes for species and units described by Cáceres and Legendre (2008). Specifics on the size of each matrix, and the types of transformation used, are reported in the figure caption of each ordination. Matrices ready for analysis were imported into *R* where the vegan Community Ecology Package (v. 2.4-2; Oksanen et al. 2017) was used to perform the NMDS ordinations, all of which used the Bray-Curtis distance measure. All of the ordinations settled on a 2-dimensional solution and stress levels were always less than 0.2; the latter are reported within each ordination figure.

There were two sets of ordinations performed for each trophic level. The intent of the first set of ordinations was to understand how much estuarine biodiversity is being captured by the estuarine MPAs. These ordinations therefore compared community structure among estuaries, and sites within estuaries, for a total of 8 groups (i.e. 4 estuaries * 2 sites / estuary). In this ordination, each group is represented by the summer data from 2014, 2015 and 2016. The purpose of the second set of ordinations was to see if the communities could be responding to the switch from drought to El Niño conditions, or the shift in ocean temperatures, that occurred during the study.

This second set of ordinations therefore compared the three summers where each summer was represented by each estuary for a total of 12 groups (i.e. 3 summers * 4 estuaries / summer). Each site within an estuary was included in this second set but was not pulled out as a separate group.

Permutation Multivariate Analysis of Variance (PerMANOVA) in *R* was used to test for an effect of Year and Site, the latter nested within Estuary, on the multivariate community structure of each trophic level. There were eight groups from each trophic level, as previously described. The exception was TM where, for the invertebrate analysis, the mouth site at TM was dropped because it contained no animals. The general formula used in *R* was: taxonomic group ~ Year * Estuary / Site. Because PERMANOVA may be sensitive to the heterogeneity of multivariate dispersion (Anderson 2006; Anderson and Walsh 2013), we also tested for the degree of dispersion among sites and years for all three trophic levles using PERMDISP2 (betadisper() from the R package vegan). Where there was evidence of dispersion, we used pairwise comparisons (permutest.betadisper() from the R package vegan) to identify the pairs of sites or years between which dispersion differed. These results affect the interpretation of the PERMANOVAs in that, where there is evidence for dispersion (significant results from PERMDISP2), then species diversity differences among sites or years, as indicated by significant results from the PERMANOVA, may or may not be valid. We also completed pairwise comparisons from the PERMANOVA (adonis ()); the validity of these are also affected by dispersion. Since a statistically significant PerMANOVA result has implications for how well MPAs are representing estuarine biodiversity, it is important to be aware of the limitations of this method.

Indicator Species Analyses (ISA) were performed by PC-ORD in order to determine if the species themselves suggest that environmental conditions among sites and estuaries differ (McCune and Grace 2002). A high indicator value (i.e. 0 = no indication, 100 = perfect indication) is interpreted as a taxon or functional group having high fidelity to a particular site. Computationally, ISA considers the frequency that a taxon occurs among samples within a site, as well as the abundance value of a sample. For example, if *Z. marina* occurred in all 15 quadrats within a site, and had high percent cover values in each quadrat, and only occurred at one site, then it would receive an IV score close to 100. The ISA tests compared the same groups used in the NMDS and PerMANOVA analyses. The ISA tests were also run on the same matrices as the ordinations for comparability, even though ISA is computationally independent of NMDS. The transformations, or lack of them, used for each ISA test are described in the caption for each ISA table.

Target Species

The set of macrophyte, invertebrate and fish species that were described more completely (i.e. abundance, size) by this study (Table 5) were chosen for several reasons; their known keystone and bioindicator value (*sensu* Bortone 2005); if they were identified by stakeholders during the outreach portion of MPA planning; sampling practicality. The intent of the present study with respect to target species was twofold: 1) to describe these species in sufficient detail in order to allow future sampling to detect a management or climate event, and 2) to collect enough species level information to recommend which species should be dropped, which should be retained, and which sampling methods should be modified for future estuarine monitoring.

Table 5. Target species proposed for study compared to what was able to target variables that the study used.

Proposed	Actual
Bivalves: density & size by species Green algae: biomass for functional groups - all ulvoids, all green algal filaments *Z. marina*: Shoot density, Leaf Area Index, Inflorescence density, actual and estimated above ground biomass, depths relative to MLLW for the deep and shallow edges of the eelgrass bed, GPS positions for bed edges *Phyllaplysia taylori*: # / leaf area, length then size class Crabs: Abundance by species (# / trapping effort), size and life history stage by species Bivalves: Abundance by species (density / volume), size by species Fish: Abundance and size of all species caught, with particular attention to salmonids and rockfish.	Bivalves: Could not excavate, but densities of holes of varying diameters were counted. The larger holes were produced by species of bivalves and ghost shrimp. Green algae: Total algal cover, which was dominated by green algae, was calculated instead. Green algal cover could be split out from total algal cover. *Z. marina*: Shoot Density, Shoot Length, Above ground biomass, maximum depths and GPS positions taken for the eelgrass beds in Big River SMCA. *R. maritima*: not anticipated, but found in the Mad River and Ten Mile estuaries; shoot density, shoot lengths, above ground biomass. *P. taylori*: None were found. Crabs: Abundance by species and sex, carapace width sizes. Traps were subject to predation and people. Fish: Abundance and size of all species caught, with particular attention to salmonids and rockfish.

Some of the variables used for describing each target species (Table 5) warrant further explanation. Eelgrass maximum depths, which are a strong indicator of water quality (Biber et al. 2005), were only measured at the two BR sites. Latitude, longitude, date, time and water depth were recorded at multiple locations on the deep edge of the two beds but eelgrass depths could only be expressed relative to the MLLW tidal datum at Arena Cove, CA because, while an adjustment for low tide time and height is estimated for the town of Mendocino just outside of the mouth of BR, there are no time and water level adjustments for within the BR estuary.

For presenting the relative abundance of a fish species over time at a particular estuary, the number of individuals for a particular species counted at one site at one time was divided by the total number of individuals of that species counted at both sites over all times in a particular estuary. Relative abundance values are therefore directly comparable within an estuary, but only patterns of abundance can be compared among estuaries.

For crab species, catch numbers from box and fish traps from low and mid elevations were combined due to the motility of crabs and, in most cases, the low and mid transect lines were horizontally close to each other. Seagrass shoot lengths are displayed as box plots as calculated by SigmaPlot (v. 11); the line in the box is the median; the low and high value ends of the box are 25th and 75th percentiles; the low and high whiskers are the 10th and 90th percentiles, which require at least nine samples to be calculated.

Results & Discussion

Physical Context

Over the broad scales of space and time in the North Coast MPA region, data products from the MPA project Characterization and Indicators of Oceanographic Conditions (Bjorkstedt, Tissot, Sydeman, Largier, Garcia-Reyes; 2017) make it clear that the physical conditions in the ocean and watersheds were unusual over the course of this estuarine baseline study from June 2014 to June 2016. Extreme drought and the warm water "blob" conditions prevailed during 2014, and were gradually modified by the El Niño conditions initiated during 2015. The El Niño conditions kept coastal waters in the North Coast Region warm – relative to La Niña periods – but this particular El Niño was also characterized by strong south to north currents. Based on studies of the coastal ocean when the "blob" had not yet formed, the expectation during the baseline study was that the strong upwelling center on the south side of Cape Mendocino would have produced cooler, more productive conditions than on the north side of the cape (Magnell et al. 1990, Largier et al. 1993). However, both the "blob" and the following El Niño events were so strong that, during a particular year, the nearshore oceanic conditions in the MPA bioregions on each side of the cape were more similar than usual, as indicated by mean monthly sea surface temperature (Figure 13). As well, when river discharges were low north of Cape Mendocino (Figure 14, Figure 15) they were also low south of the cape (Figure 16, Figure 17).

The strength of the "blob" and El Niño events means that any ocean effects on the estuarine biota should have been greater across years rather than between bioregions. In Humboldt Bay, with its deep connection to the open ocean (Table 2), this oceanic SST signal was evident during the late summer of 2015 when coastal waters were also the warmest (Figure 18). The particular temporal changes in ocean conditions documented by the MPA project Characterization and Indicators of Oceanographic Conditions (Bjorkstedt, Tissot, Sydeman, Largier, Garcia-Reyes; 2017) have the potential to thermally stress estuarine organisms, and the changes to nearshore circulation due to the El Niño event could alter patterns of invertebrate and fish recruitment to estuaries.

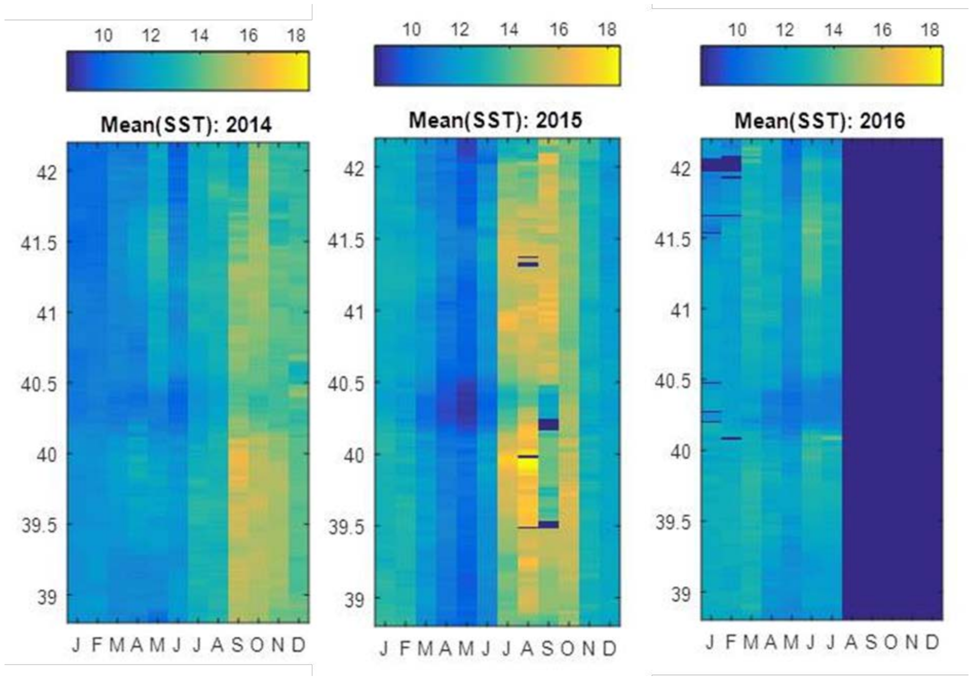


Figure 13. Monthly sea surface temperatures (SST) for the North Coast MPA region. Cape Mendocino is at 40.4 latitude (produced by the Ocean Characterization MPA project).

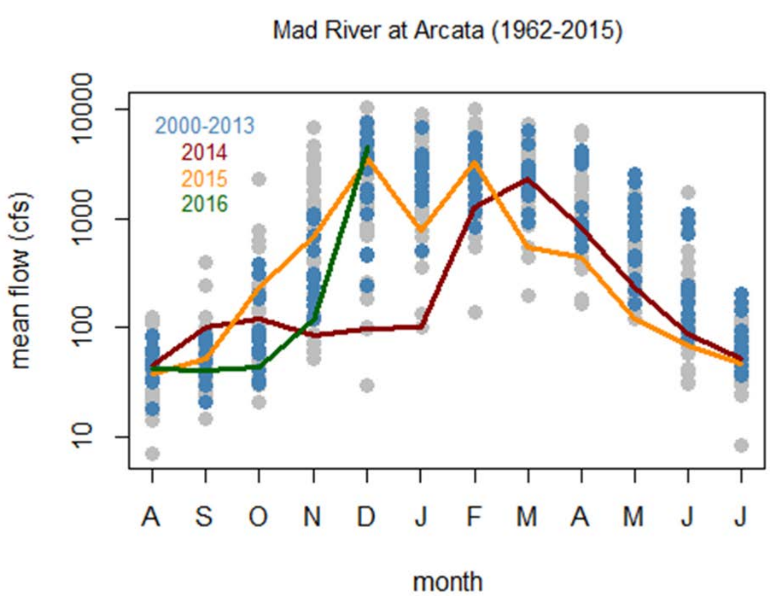


Figure 14. Mean monthly flow from the Mad River USGS gaging station 1148100 (produced by the Ocean Characterization MPA project) representing the northern bioregion.

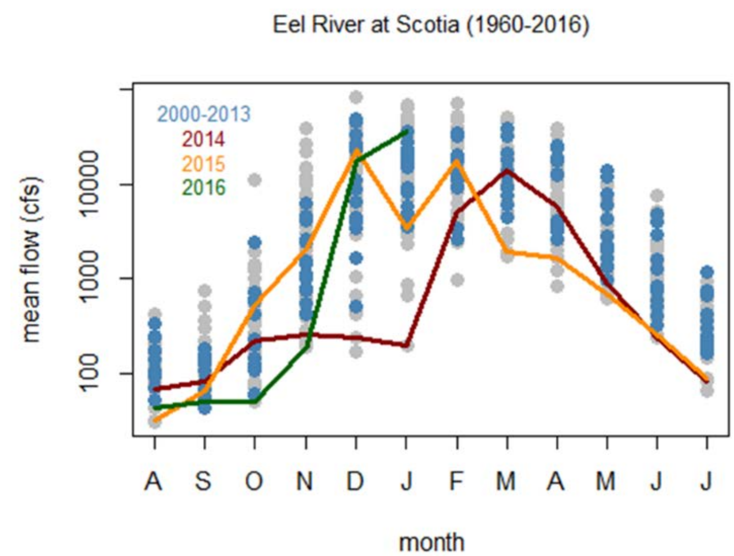


Figure 15. Mean monthly flow for the Eel River USGS gaging station 11477000 (produced by the Ocean Environment MPA project) representing the northern bioregion.

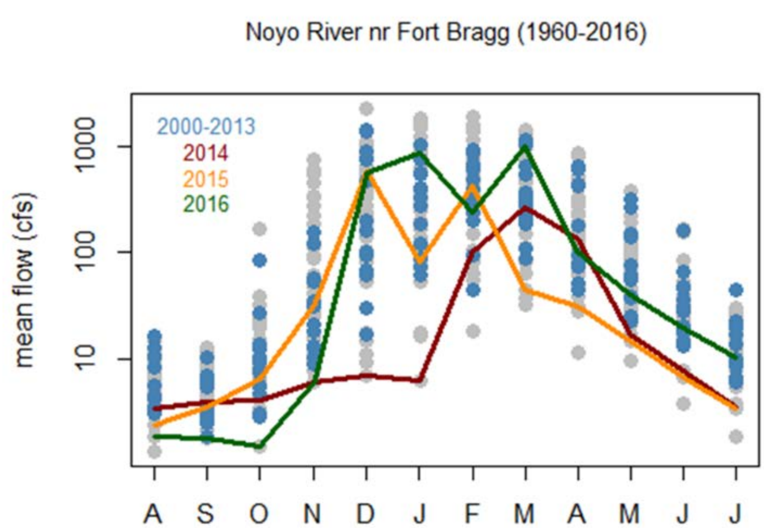


Figure 16. Mean Monthly flow for Noyo River from USGS gaging station 11468500 (produced by the Ocean Environment MPA project) representing the southern bioregion.

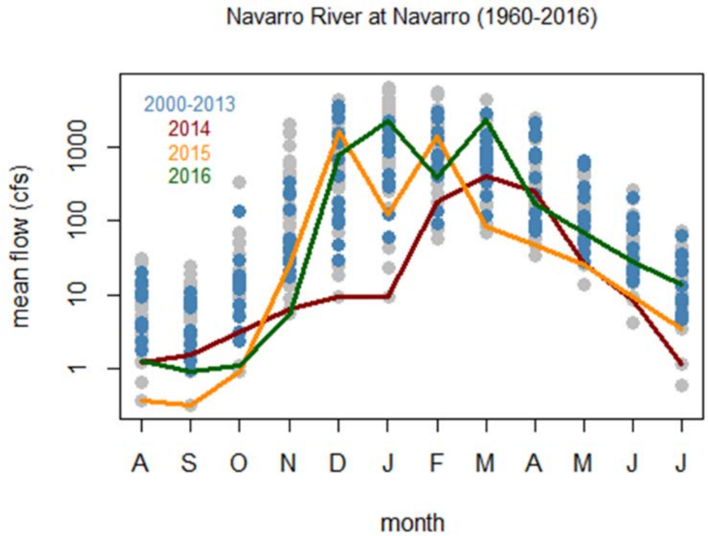


Figure 17. Mean monthly flow by the Navarro River from USGS gaging station 11468000 (produced by the Ocean Environment MPA project) representing the southern bioregion.

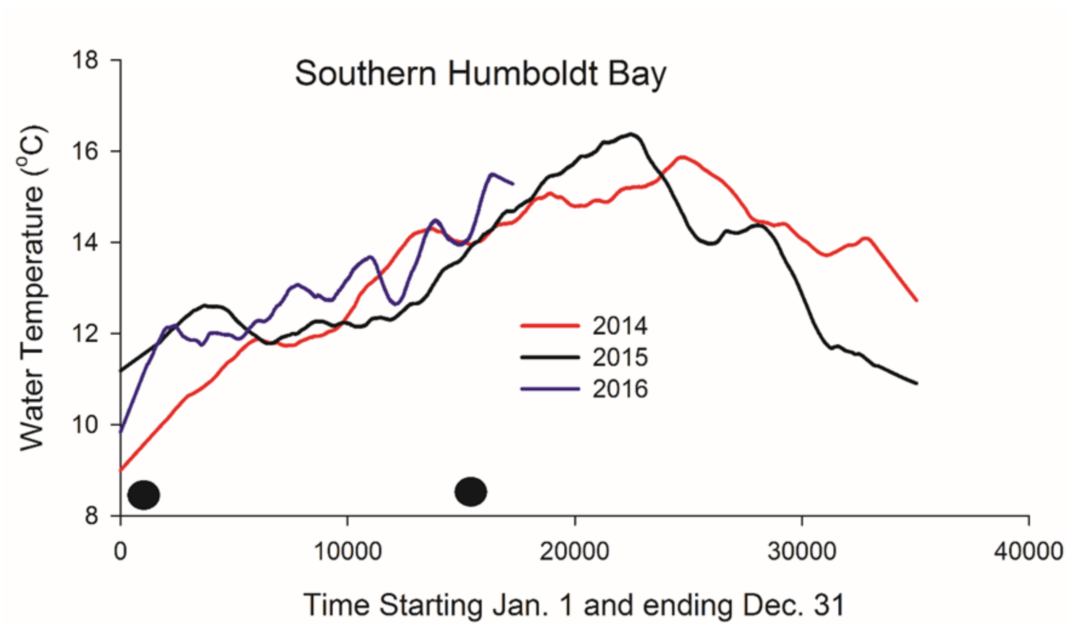


Figure 18. Sea Surface Temperatures recorded by CeNCOOS from the NE corner of southern Humboldt Bay. The black circles are the months when the baseline monitoring occurred.

Despite the relatively unusual oceanic environment, it is not necessarily the case that the estuarine biota will only be driven by oceanic forcing variables. The geomorphology and hydrology of each estuary affects its connectivity to the open ocean. The contrast made in the present study of the physical conditions in the TM and BR estuaries demonstrates how local conditions modify the oceanic connection.

The TM estuary is more typical of smaller estuaries that may close during summer. The mouth is surrounded by beaches and is not sheltered from wave action. The tidal area is also much smaller, so that tidal action alone is not strong enough to keep the mouth open. Tidal fluctuations in water level were strong in the summer (July-August 2014), although the low tide was truncated as the bar-built sill of sand across the mouth became higher during the second half of 2014 (Figure 19). The mouth channel shoaled during an early fall wave event on 25th September, and the estuary transitioned to a perched state by the end of the month (i.e. outflow only) – this is corroborated by photographs of the mouth (Figure 20), and also the absence of tidal fluctuations in water temperature (only a day-night cycle is evident – Figure 19). A sequence of wave events in October further closed the mouth and built the berm so that the mouth closed completely, and water level rose more than 1m above high tide levels in late October following a major wave event on 26th October (Figure 19, Figure 20). Evidently freshwater discharge into the small lagoon was large enough to overfill the basin, perhaps also due to wave overwash on 26th October, 2014. The mouth breached at the end of October, returning to tidal conditions in early November (Figure 19). The mouth shoaled again in mid-November, but did not close completely. The mouth was scoured more deeply, and lower low-tide levels were observed following the strong rains and river flows in mid-December 2014 (Figure 20). The estuary became warmer when the mouth closed during October, but it appears that wave overwash on 12 October cooled the estuary and presumably increased salinity. This occurred again after it breached at the end of October 2014. A similar seasonal pattern occurred during 2015 (Figure 21), with tidal conditions being muted as the mouth shoaled in September and eventually closed on 10 October. With less river flow, the mouth remained closed until December 2015.

TM depth profile data in June 2014 and 2015 (Figure 22, Figure 23) showed a marked 2-layer structure with high-salinity water trapped in deeper sections and an over-flowing low-salinity layer. Top-to-bottom salinity differences were as big as 30ppt in less than 2m, with a tendency for low oxygen levels at depth. This deep water was also warmer at some stations, likely due to the penetration of solar radiation to depth. The surface layer was about ½ m deep.

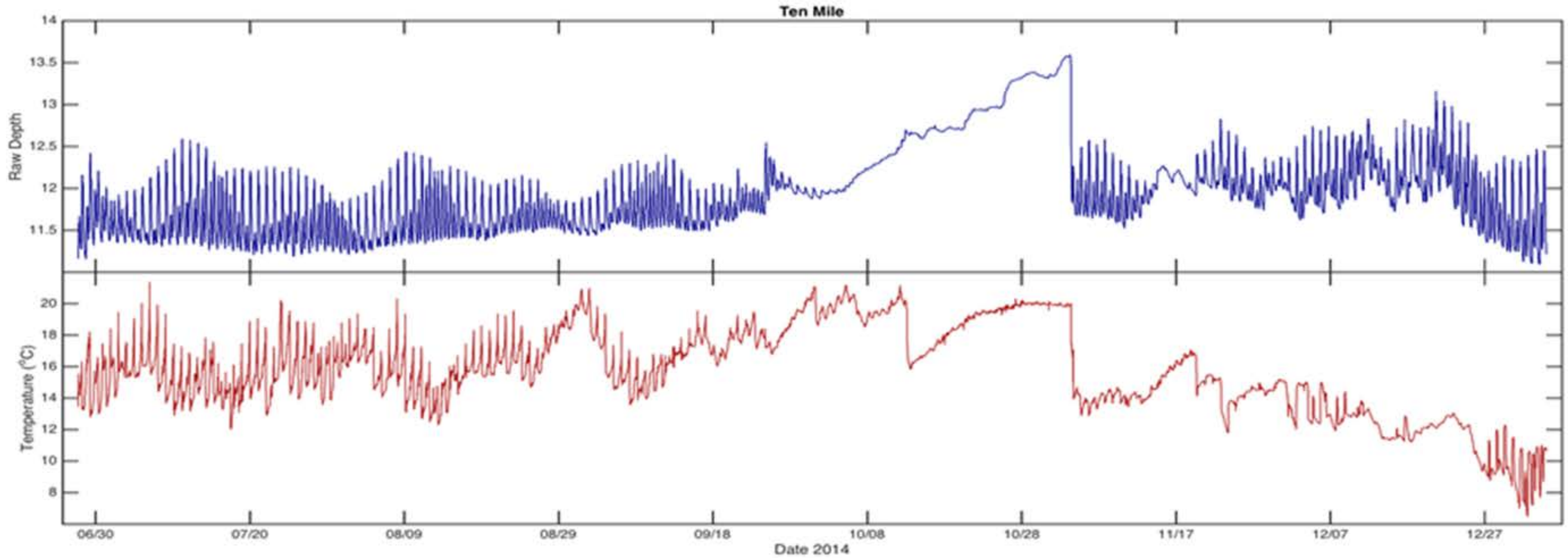


Figure 19. Water level (top panel; arbitrary datum) and water temperature (bottom panel) in the Ten Mile River SMCA during the second half of 2014.

Figure 20. Photographic progression of beach building and erosion at the mouth of the Ten Mile River SMCA during 2014.

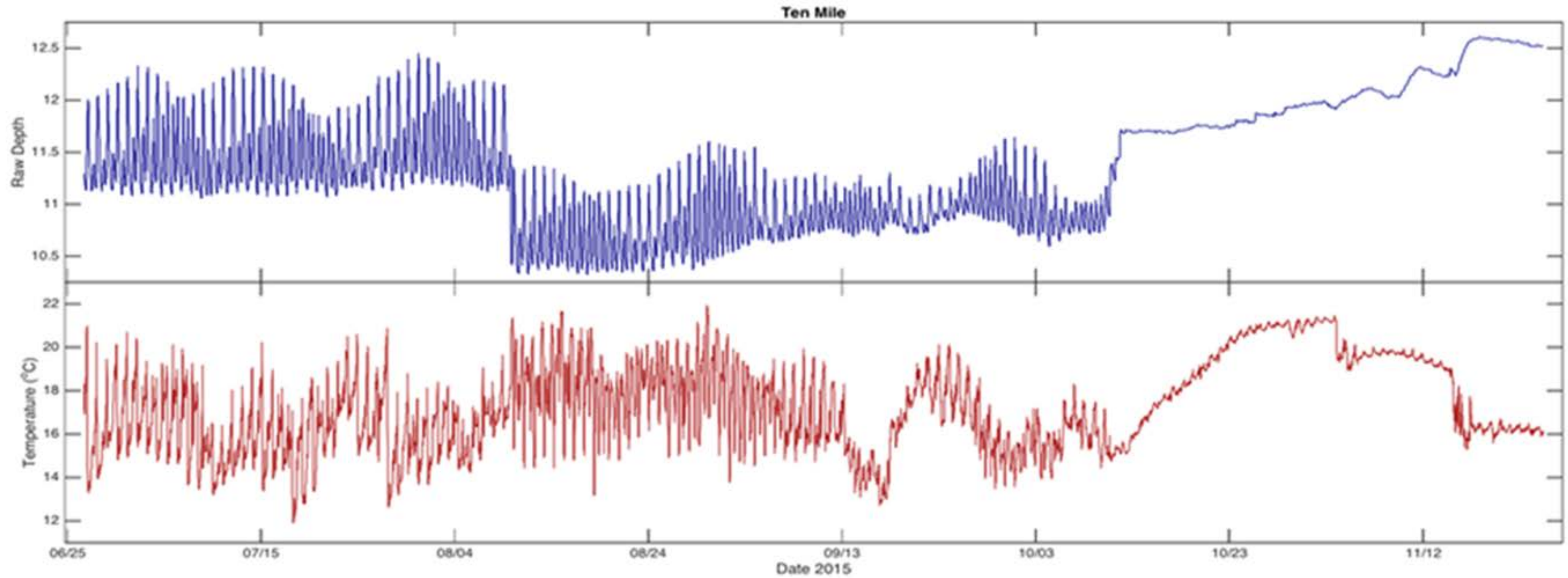


Figure 21. Water level (top panel; arbitrary datum) and water temperature (bottom panel) in the Ten Mile River SMCA during the second half of 2015.

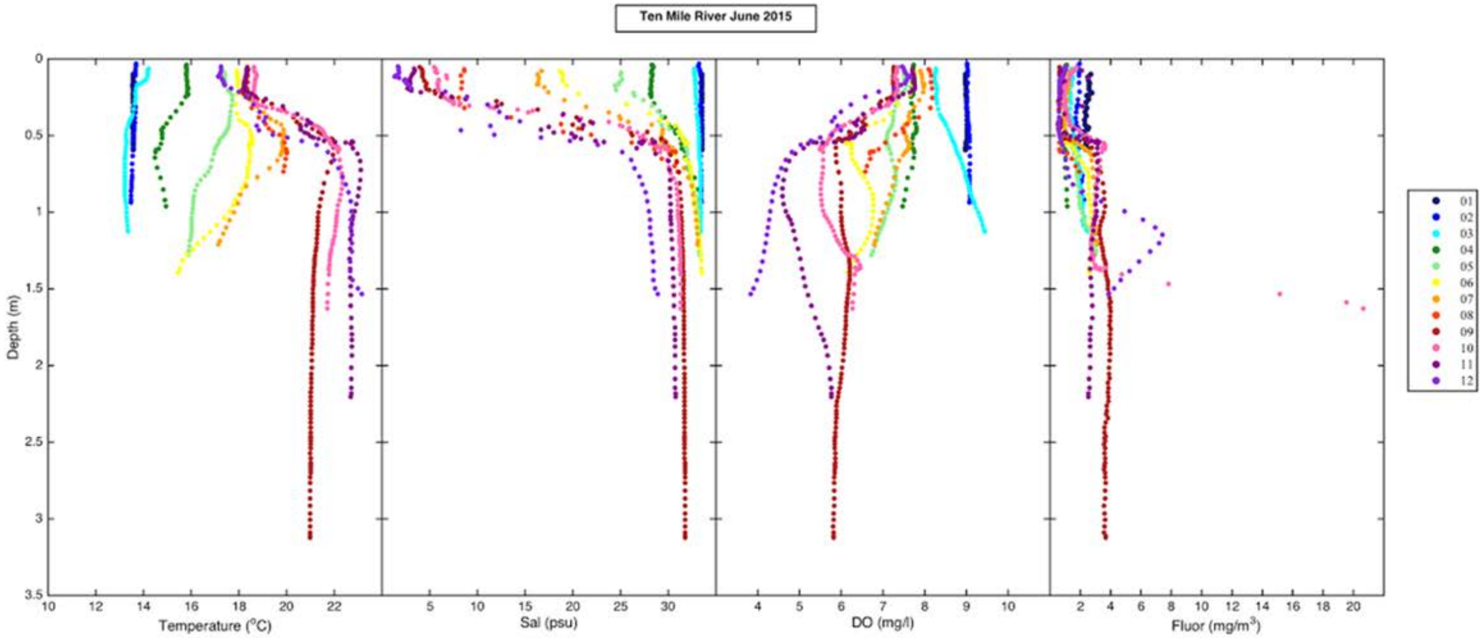


Figure 22. Vertical profiles of water quality conditions (temperature, salinity, dissolved oxygen, chlorophyll fluorescence) taken along a transect of 12 stations in Ten Mile River estuary during June 2015. See Fig. 23 for a map of station positions. Stations 3 and 4 are close to the "Mouth" site whereas station 6 is just downstream from the "Up" site.

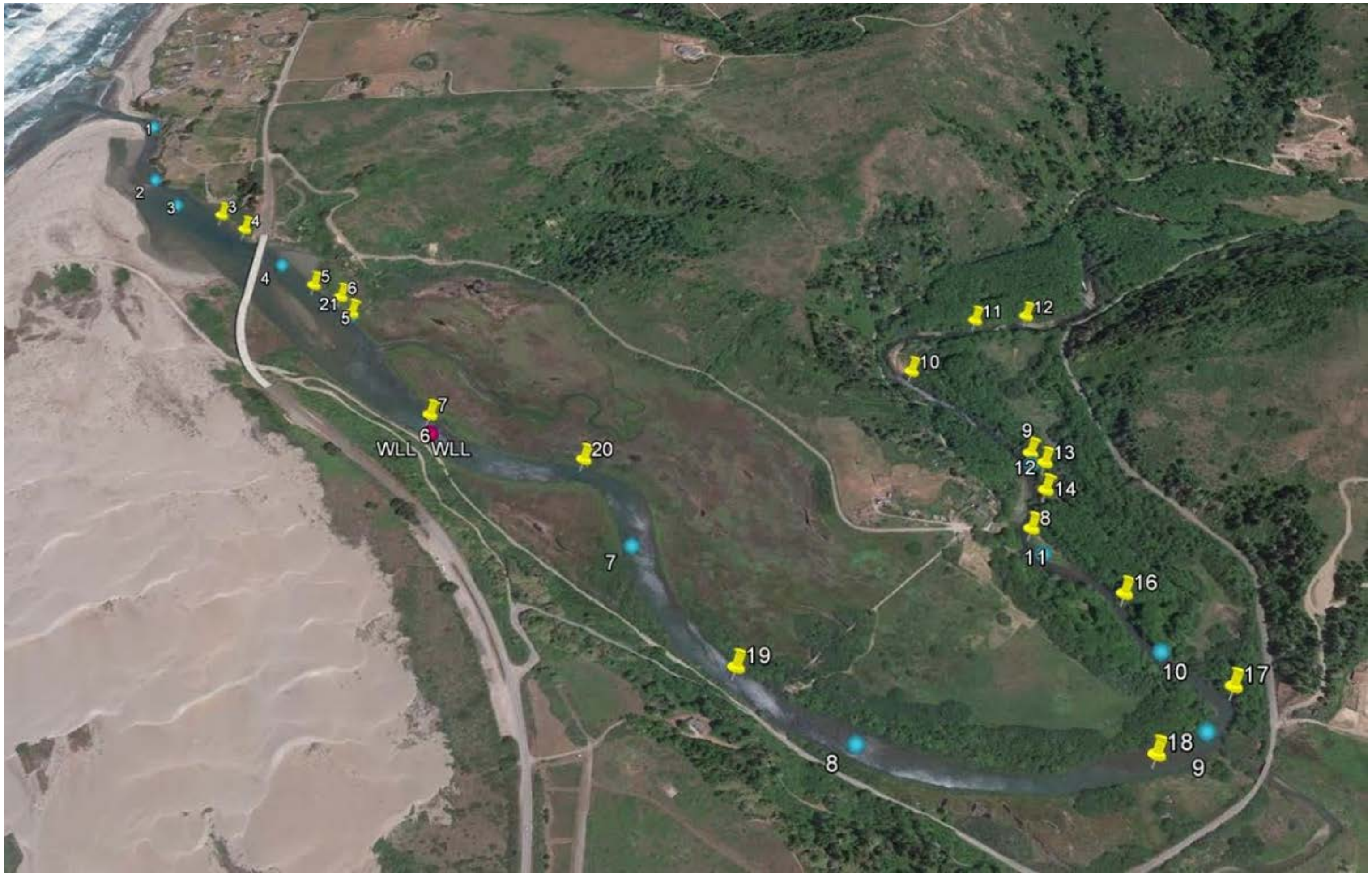


Figure 23. Vertical profiling stations in Ten Mile River estuary during June 2015. The 12 blue dots are where measures of water quality were taken; stations 5 and 6 are under the yellow pins.

In contrast to TM, the mouth of BR is bracketed by rocky headlands, and so it remained open and tidal from June to December during 2014 and 2015, as it usually does (Warrick & Wilcox 1981). The mouth is sheltered from the direct action of waves, and the long tidal reach ensures a large tidal prism that continuously scours the mouth channel even during times of minimal freshwater discharge. The most evident signal in water level was tidal, including a well-pronounced spring-neap cycle during 2014 and 2015 (Figure 24, Figure 25). The other noticeable feature was elevated water levels during strong river flow in mid-December 2014 (i.e. low tide water levels up to 1m above normal). Estuary water temperatures varied tidally, with a range of about 6^{o}C in summer, when the ocean is both cold (i.e. typically below 12^{o}C) and replete in upwelled nitrate and plankton, while the river and back-estuary water were warm (i.e. above 20^{o}C; Figure 24, Figure 25). After September, the estuary and river cooled down and tidal variations in temperature were weak, but tidal variations in salinity may become more important as river flow increases in winter.

BR profile data collected in June 2014 and 2015 showed weak stratification (i.e. salinity differences of 5ppt or less) and well-oxygenated conditions with moderate levels of water column chlorophyll (Figure 26, Figure 27). Given the strong tidal action and weak freshwater inflow, these conditions are expected to be typical and persistent through summer. The warmer and lower salinity surface layer was about 1m thick, and it appeared that light could penetrate below that depth, where a chlorophyll maximum was observed between 2m and 3m in 2014, concurrent with a sub-surface oxygen maximum (i.e. super-saturated concentrations).

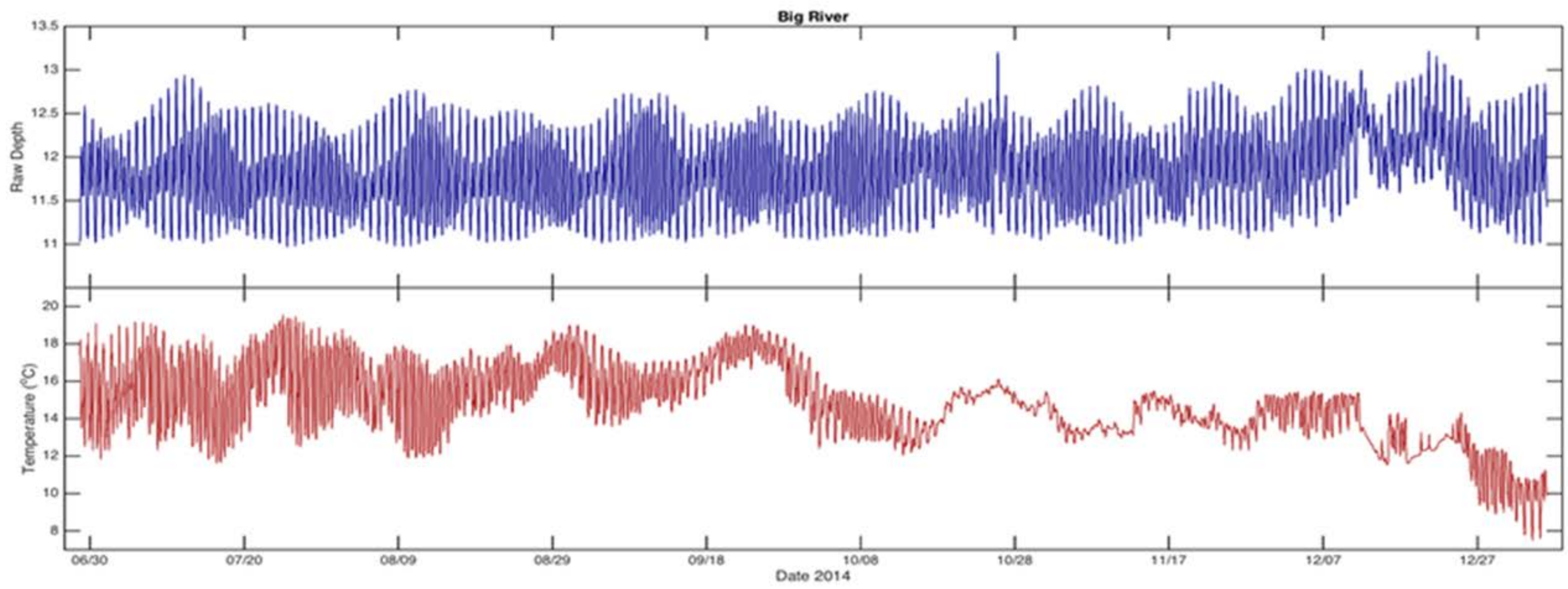


Figure 24. Water level (top panel; arbitrary datum) and water temperature (bottom panel) in the Big River SMCA during the second half of 2014.

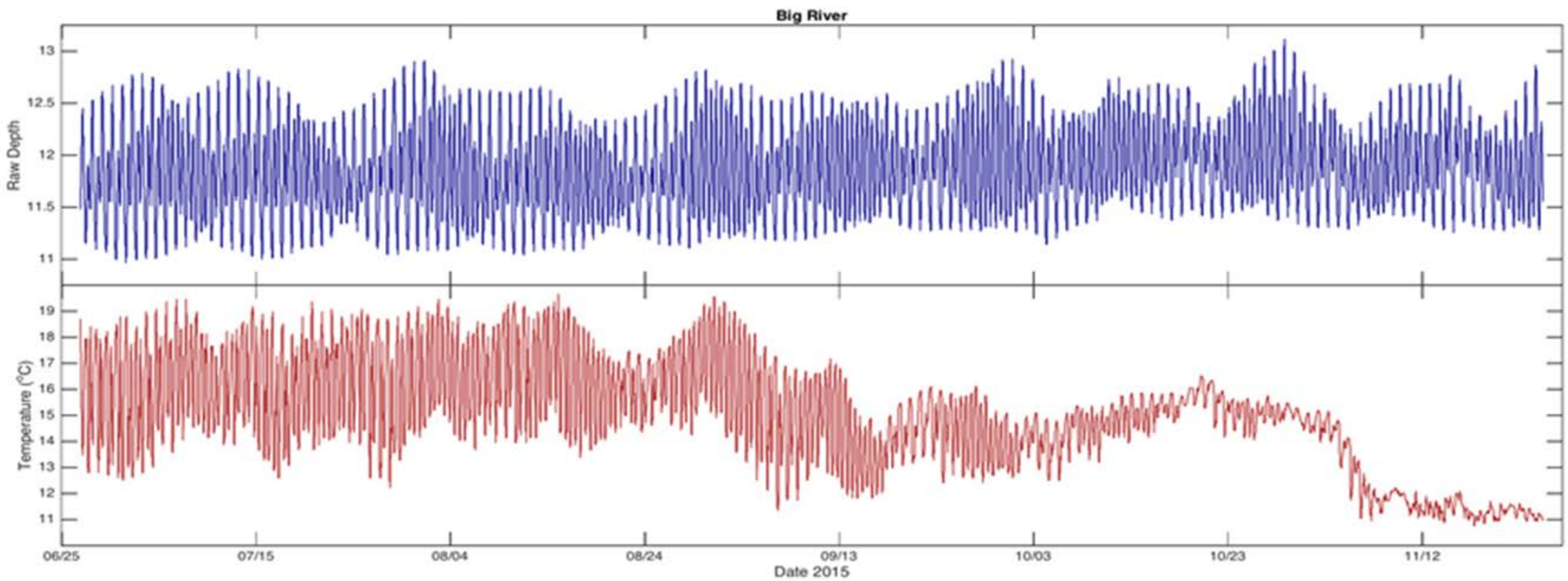


Figure 25. Water level (top panel; arbitrary datum) and water temperature (bottom panel) in the Big River SMCA during the second half of 2015.

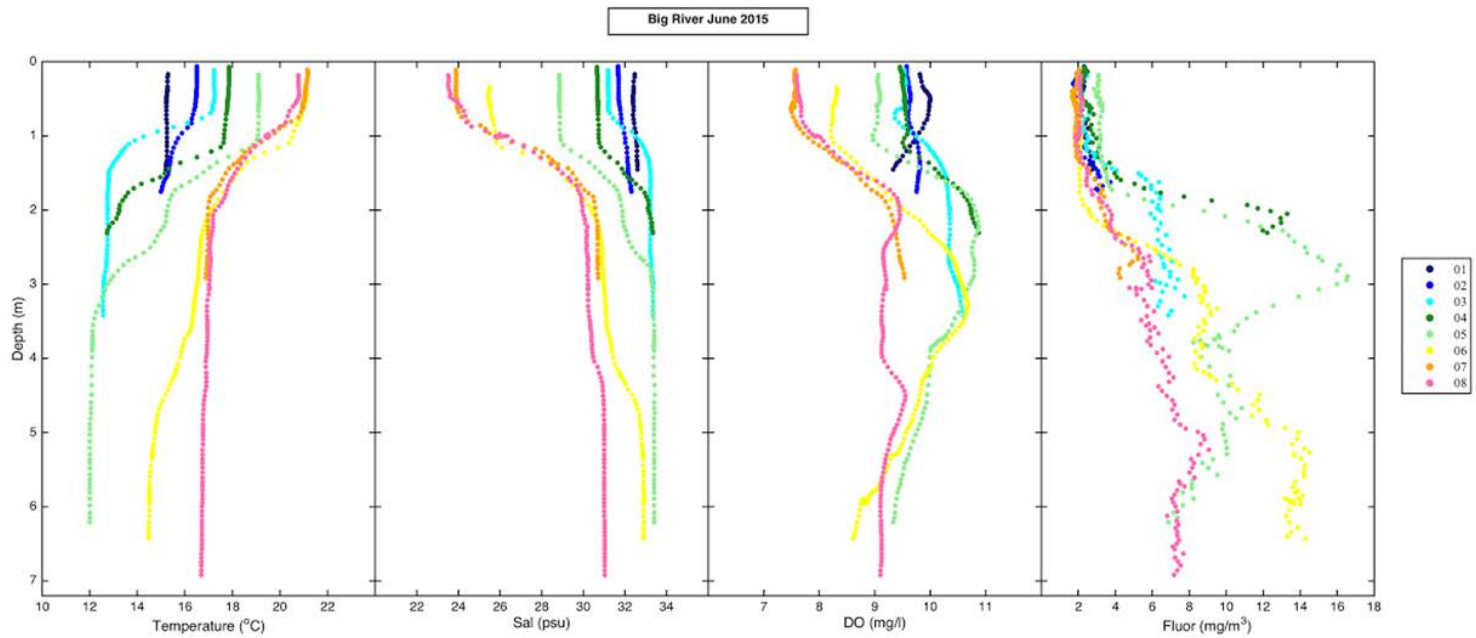


Figure 26. Vertical profiles of water quality conditions taken along a transect of 8 stations in Big River estuary during June 2015. See Fig. 27 for a map of station positions. Stations 4 and 6 are close to the "Mouth" and "Up" sites, respectively.

Figure 27. Vertical profiling stations in Big Mile River estuary during June 2015. The 8 blue dots are where measures of water quality were taken.

Biodiversity

Species Richness

Most of the species encountered during the study had either marine affinities, since they occur in the marine end of estuaries and the nearshore ocean, or they were species that spend the majority if not all their life in estuaries. Examples of species found in nearshore habitats and the lower, marine end of estuaries were the green alga *Ulva californica*, the brown alga *Fucus distichus*, the crab *Metacarcinus magister*, as well as juvenile flatfish, shiner surfperch, rockfish and cabezon (ES 2, Table 6, Table 7, Table 8). The seagrass *Z. marina*, the heterokont alga *Vaucheria littorea*, several of the annelids, isopods and amphipods, as well as fish like prickly sculpin and three-spined stickleback occur only in estuaries. Other taxa occurred that are known to require brackish conditions, or are able to withstand short periods of higher salinities, such as the seagrass *Ruppia maritima*, the green algae *Gayralia oxysperma* and *Cladophora glomerata*, chironimid and dipteran larvae, and tidewater goby. Coho and chinook salmon as well as steelhead trout also occurred and span freshwater, estuarine and oceanic habitats.

Table 6. Algal and seagrass species found in each estuary. *Gracilaria vermiculophylla* was identified using sequencing techniques carried out by S.A. Krueger-Hadfield (U. Alabama, Birmingham).

Phylum	Class	Order	Family	Species	Common Name	Mad River	Humboldt Bay	Ten Mile River	Big River	Notes
Charophyta	Conjugatophyceae	Zygnematales	Zygnematophyceae	*Spirogyra wrightiana*		x				Epiphytic on *Ruppia maritima*
Chlorophyta	Chlorophyceae	Chaetophorales	Chaetophoraceae	*Stigeoclonium lubricum*		x				Epiphytic on *Ulva intestinalis*
	Ulvophyceae	Cladophorales	Cladophoraceae	*Cladophora glomerata*	river weed	x				Epilithic & entangled at upriver site, a freshwater species
				Rhizoclonium tortuosum			x			Skeins on mid intertidal mudflats
		Ulotrichales	Gayraliaceae	*Gayralia oxysperma*		x		x		Epilithic & entangled, mostly at upriver sites in each estuary, low intertidal to shallow subtidal
		Ulvales	Ulvaceae	*Ulva californica*	sea lettuce			x	x	Epilithic & epiphytic, for Ten Mile River only drift at mouth site
				Ulva compressa		x				Attached to rocks and wood, upriver site
				Ulva intestinalis	gut weed	x	x	x	x	Epilithic & epiphytic

Phylum	Class	Order	Family	Species	Common Name	Mad River	Humboldt Bay	Ten Mile River	Big River	Notes
				Ulva linza		x		x	x	Abundant mouth site of Mad River, on rocks and wood
				Ulva torta		x	x	x	x	Skeins on mid intertidal mudflats
Rhodophyta	Bangiophyceae	Bangiales	Bangiaceae	*Pyropia nereocystis*	nori			x		Drift
	Florideophyceae	Ceramiales	Ceramiaceae	*Ceramium pacificum*			x			Mid intertidal mudflats, unattached
			Rhodomelaceae	*Polysiphonia hendryi*				x		Epilithic & epiphytic, shallow subtidal
				Polysiphonia paniculata				x		Epilithic
				Pterochondria woodii				x		Drift
		Gracilariales	Gracilariaceae	*Gracilaria vermiculophylla*			x			Mid intertidal mudflats, unattached
		Halymeniales	Halymeniaceae	*Grateloupia doryphora*				x		Drift
Ochrophyta	Phaeophyceae	Desmarestiales	Desmarestiaceae	*Desmarestia ligulata*	acid weed		x	x	x	Drift
				Desmarestia latissima	acid weed			x	x	Drift
		Fucales	Fucaceae	*Fucus distichus*	rockweed			x	x	Drift & attached to rocks & wood
		Laminariales	Lessoniaceae	*Egregia menziesii*	feather boa kelp			x	x	Drift
	Xanthophyceae	Vaucheriales	Vaucheriaceae	*Vaucheria littorea*			x	x	x	Forms mats on mudflats just

Phylum	**Class**	**Order**	**Family**	**Species**	**Common Name**	**Mad River**	**Humboldt Bay**	**Ten Mile River**	**Big River**	**Notes**
										below marsh plants
Tracheophyta	Monocots	Alismatales	Potamogetonaceae	*Ruppia maritima*	beaked tasselweed , widgeon grass	x		x		Mid intertidal mud - sand beds, sparse in Humboldt Bay
			Zosteraceae	*Zostera marina*	eelgrass		x	x	x	Low intertidal to shallow subtidal, very sporadic at upriver site in 10 Mile River

Table 7. Invertebrate species found in each estuary.

Phylum	Class	Order	Family	*Species*	Big River	Humboldt Bay	Mad River	Ten Mile River
Annelida	Polychaeta	Echiuroidea	Urechidae	*Urechis caupo*	X			
		Eunicida	Dorvilleidae	*Schistomeringos longicornis*		X		
			Lumbrineridae	*Lumbrineris zonata*		X		
		Phyllodocida	Nephtyidae	*Nephtys caecoides*		X		
				Alitta brandti	X			
				Alitta succinea		X		
				Neanthes lighti	X	X	X	X
				Nereis latescens		X	X	
				Nereis procera	X			X
				Platynereis bicanaliculata	X	X		
			Phyllodocidae	*Eteone californica*	X		X	X
				Eulalia quadrioculata	X	X		
			Polynoidae	*Harmothoe imbricata*		X		

Phylum	Class	Order	Family	*Species*	Big River	Humboldt Bay	Mad River	Ten Mile River
				Hesperonoe complanata	X			
			Syllidae	*Exogone lourei*		X		
				Exogone molesta	X			
		Spionida	Spionidae	*Boccardia proboscidea*		X		
				Boccardiella hamata		X		
				Boccardiella ligerica		X		X
				Dipolydora socialis		X		
				Polydora nuchalis	X	X		
				Pseudopolydora kempi	X	X		
				Pseudopolydora paucibranchiata		X		
				Pygospio elegans				X
				Scolelepis tridentata		X		
			Arenicolidae	*Arenicola cristata*	X			
			Capitellidae	*Mediomastus ambiseta*		X	X	X
				Mediomastus californiensis	X	X	X	
				Notomastus magnus		X		
			Maldanidae	*Axiothella rubrocincta*		X	X	
			Orbiniidae	*Leitoscoloplos pugettensis*		X		
Arthropoda	Arachnida	Araneae	Linyphiidae	*Spirembolus mundus*			X	
				Arachnid - Terrestrial				X
	Hexanauplia	Sessilia	Balanidae	*Balanus glandula*			X	
	Insecta	Coleoptera		*Coleoptera adult*		X	X	
				Coleoptera larvae				X
		Diptera	Chironomidae	*Chironomidae Larvae*	X		X	
				Diptera Larvae	X	X	X	X
				Diptera pupae			X	
		Hemiptera	Corixidae	*Trichocorixa reticulata*			X	
			Naucoridae	*Ambrysus sp.*			X	
		Odonata	Gomphidae	*Ophiogomphus bison*			X	
				Dragonfly Larvae			X	X
	Malacostraca	Amphipoda	Ampithoidae	*Ampithoe lacertosa*		X		
				Ampithoe valida		X		
				Peramphithoe mea		X		
			Anisogammaridae	*Anisogammarus pugettensis*		X		
				Eogammarus confervicolus	X	X	X	X
				Ramellogammarus ramellus	X			X

Phylum	Class	Order	Family	*Species*	Big River	Humboldt Bay	Mad River	Ten Mile River
			Aoridae	*Grandidierella japonica*		X		
				Microdeutopus gryllotalpa		X		
				Paramicrodeutopus schmitti		X		
			Caprellidae	*Caprella californica*		X		
				Caprella drepanochir		X		
				Caprella natalensis		X		
			Corophiidae	*Americorophium salmonis*	X	X	X	
				Americorophium spinicorne	X	X	X	X
				Paracorophium sp.		X		
			Dogielinotidae	*Allorchestes angusta*		X		
			Gammaridae	*Gammarus daiberi*	X	X		
			Hyalellidae	*Hyalella azteca*			X	
			Photidae	*Photis brevipes*		X		
			Talitridae	*Megalorchestia californiana*				X
		Cumacea	Leuconidae	*Nippoleucon hinumensis*	X			
		Decapoda	Callianassidae	*Neotrypaea californiensis*	X			X
			Cancridae	*Cancer productus*	X	X		X
				Metacarcinus magister	X	X	X	X
				Carcinus maenas		X		
			Crangonidae	*Crangon franciscorum*	X	X	X	
				Crangon nigricauda		X		X
				Crangon nigromaculata	X	X		
			Epialtidae	*Pugettia producta*		X		X
			Grapsidae	*Pachygrapsus crassipes*		X		
			Hippidae	*Emerita analoga*				X
			Paguridae	*Pagurus hirsutiusculus*		X		
			Palaemon	*Palaemon macrodactylus*	X			
			Pandalidae	*Pandalus danae*	X			
			Thoridae	*Heptacarpus paludicola*		X		
				Heptacarpus pugettensis			X	
				Heptacarpus sitchensis		X		
			Varunidae	*Hemigrapsus oregonensis*	X	X	X	X
				Crab Megalopa			X	
		Isopoda	Aegidae	*Rocinela signata*		X		
			Cirolanidae	*Excirolana chiltoni*			X	X
			Cymothoidae	*Elthusa californica*		X		

Phylum	Class	Order	Family	*Species*	Big River	Humboldt Bay	Mad River	Ten Mile River
			Cymothoidae	*Elthusa vulgaris*		X		
			Halophilosciidae	*Littorophiloscia richardsonae*		X		
			Idoteidae	*Idotea fewkesi*	X			
				Idotea ochotensis	X			
				Idotea rufescens		X		
				Pentidotea resecata	X	X		
			Limnoriidae	*Limnoria tripunctata*		X		
			Porcellionidae	*Porcellio laevis*				X
				Porcellio scaber				X
			Sphaeromatidae	*Gnorimosphaeroma noblei*	X	X	X	X
				Gnorimosphaeroma oregonensis	X	X	X	X
		Leptostraca	Nebaliidae	*Nebalia kensleyi*		X	X	
		Mysida	Mysidae	*Neomysis mercedis*	X			X
		Tanaidacea	Leptocheliidae	*Leptochelia sp.*		X		
			Tanaididae	*Zeuxo normani*		X		
	Pycnogonida	Pantopoda	Ammotheidae	*Achelia chelata*		X		
Chordata	Thaliacea	Pyrosomida	Pyrosomatidae	*Pyrosome*				X
		Salpida	Salpidae	*Salp*				X
Cnidaria	Scyphozoa	Semaeostomeae	Ulmaridae	*Aurelia*			X	
Ctenophora				*Ctenophora*				X
Echinodermata	Echinoidea	Camarodonta	Strongylocentrotidae	*Mesocentrotus franciscanus*	X			
Mollusca	Bivalvia	Cardiida	Cardiidae	*Clinocardium nuttallii*	X			X
			Tellinidae	*Limecola balthica*	X			X
				Macoma inquinata	X			
				Macoma nasuta	X	X		X
		Myida	Myidae	*Cryptomya californica*	X			X
				Mya arenaria	X			X
		Mytilida	Mytilidae	*Mytilus edulis*		X		X
		Pectinida	Pectinidae	*Chlamys rubida*			X	
		Venerida	Veneridae	*Leukoma staminea*	X			X
				Nutricola tantilla	X	X		
	Gastropoda	Cephalaspidea	Haminoeidae	*Haminoea vesicula*				X
				Haminoea vesicula eggs				X
			Philinidae	*Philine auriformis*				X
		Littorinimorpha	Littorinidae	*Lacuna marmorata*	X	X		X
				Lacuna porrecta	X			X

Phylum	Class	Order	Family	*Species*	Big River	Humboldt Bay	Mad River	Ten Mile River
				Lacuna unifasciata	X		X	X
				Lacuna variegata				X
				Littorina keenae	X			
				Littorina littorea	X			X
		Neogastropoda	Columbellidae	*Mitrella tuberosa*	X			
		Nudibranchia	Facelinidae	*Hermissenda crassicornis*	X			X
Nemertea	Enopla	Monostilifera	Emplectonematidae	*Paranemertes peregrina*	X	X	X	
Phoronida			Phoronidae	*Phoronis pallida*		X		X
				Phoronopsis harmeri	X	X	X	X
Platyhelminthes	Rhabditophora	Polycladida	Stylochidae	*Imogine exiguus*		X		

Table 8. Fish species found in each estuary.

Family	Species Name	Common Name	Big River Estuary	Mad River Estuary	South Humboldt Bay	Ten Mile River Estuary
Atherinopsidae	*Atherinops affinis*	Topsmelt	x	x	x	x
Atherinopsidae	*Atherinopsis californiensis*	Jacksmelt		x		
Catostomidae	*Catostomus occidentalis*	Sacramento Sucker		x		
Gasterosteidae	*Gasterosteus aculeatus*	Three-Spined Stickleback	x	x	x	x
Aulorhynchidae	*Aulorhynchus flavidus*	Tubesnout			x	
Syngnathidae	*Syngnathus leptorhynchus*	Bay Pipefish	x	x	x	x
Clupeidae	*Clupea pallasi*	Pacific Herring	x	x	x	x
Engraulidae	*Engraulis mordax*	Northern Anchovy		x	x	
Cottidae	*Artedius fenestralis*	Padded Sculpin	x			
Cottidae	*Artedius notospilotus*	Bonyhead Sculpin	x			
Cottidae	*Clinocottus acuticeps*	Sharpnose Sculpin	x	x		
Cottidae	*Cottus aleuticus*	Coastrange Sculpin	x	x		
Cottidae	*Cottus asper*	Prickly Sculpin	x	x	x	x
Cottidae	*Cottus asperrimus*	Rough sculpin		x		
Cottidae	*Enophrys bison*	Buffalo Sculpin	x	x	x	
Cottidae	*Hemilepidotus hemilepidotus*	Red Irish Lord	x			
Cottidae	*Leptocottus armatus*	Pacific Staghorn Sculpin	x	x	x	x

Cottidae	*Scorpaenichthys marmoratus*	Cabezon	x	x	x	
Cottidae	Oligocottus maculosus	Tidepool sculpin	x	x		x
Hemitripteridae	*Blepsias cirrhosus*	Silverspot Sculpin	x			
Embiotocidae	*Cymatogaster aggregata*	Shiner Surfperch	x	x	x	x
Embiotocidae	*Embiotica lateralis*	Striped Surfperch	x			
Embiotocidae	*Phanerodon furcatus*	White Surfperch	x			
Gobiidae	*Clevelandia ios*	Arrow Goby	x		x	x
Gobiidae	*Eucyclogobius newberryi*	Tidewater Goby			x	
Gobiidae	*Lepidogobius lepidus*	Bay Goby		x	x	
Gobiesocidae	*Gobiesox naeabdricus*	Northern Clingfish		x		
Pholidae	*Apodichthys flavidus*	Penpoint Gunnel	x	x	x	x
Pholidae	*Pholis ornata*	Saddleback Gunnel		x	x	
Osmeridae	*Hypomesus pretiosis*	Surf Smelt	x	x	x	
Osmeridae	*Spirinchus starski*	Night Smelt	x	x	x	x
Salmonidae	*Oncorhynchus kisutch*	Coho Salmon	x	x		x
Salmonidae	*Oncorhynchus mykiss*	Steelhead		x		x
Salmonidae	*Oncorhynchus tshawytscha*	Chinook Salmon	x	x		x
Paralichthyidae	*Citharichthys sordidus*	Pacific Sanddab			x	
Paralichthyidae	*Citharichthys stigmaeus*	Speckled Sanddab	x	x	x	x
Pleuronectidae	*Platichthys stellatus*	Starry Flounder	x	x	x	x
Pleuronectidae	*Pleuronectes vetulus*	English Sole	x	x	x	x
Sebastes	*Sebastes caurinus*	Copper Rockfish	x	x		
Sebastes	*Sebastes melanops*	Black Rockfish	x	x		
Sebastes	*Sebastes rastrelliger*	Grass Rockfish		x		
Hexagrammidae	*Hexogrammos decagrammus*	Kelp Greenling	x	x		
Hexagrammidae	*Ophiodon elongatus*	Lingcod	x			
Clinidae	*Heterostichus rostratus*	Giant Kelpfish	x	x		
Batrachoididae	*Porichthys notatus*	Plainfin Midshipman			x	

Macrophyte (i.e. seagrasses & seaweeds) species richness was low and similar across the spatial scales from site within estuary to the entire North Coast MPA Region (ES 2). Relative to temperate outer coast rocky habitats, macrophyte diversity in estuaries is much lower. Infaunal, epifaunal and more mobile invertebrates were the most numerous species surveyed, and their richness was highest at the most marine estuaries of HB and BR. Invertebrate richness increased with spatial scale as did fish richness. The more marine estuaries of HB and BR had the highest overall richness mostly due to the number of invertebrate species (ES 2).

Community Structure among Sites

Macrophyte community structure as visualized by a NMDS ordination separates estuaries from the least to most (respectively, left and right side; Figure 28) marine influenced. The upriver site in MR is about 70% of the way up the summer saltwater wedge (Table 2) followed by the next most freshwater influenced site – the upriver site at TM. The mouth sites at MR and TM are close to each other followed to the right by the two BR sites, which are both less than 15% up the wedge (Table 2). The HB macrophytes, with effectively no freshwater influence and a dredged Entrance Channel, are furthest to the right in the ordination. Of the two HB sites, HB North is next to a channel, and in the ordination, was pulled furthest to the right by the red algae *Ceramium pacificum* and *Gracilaria vermiculophylla*, which only occurred on the mudflats at this site. The Indicator Species Analysis (ISA) found that tubes (i.e. *Ulva linza* at this site) typified the environmental conditions at the MR mouth site, suggesting high disturbance, whereas *R. maritima*, known for requiring brackish waters, scored the highest at the MR upriver site (Table 9). *G. vermiculophylla* was a strong indicator of HB North whereas *C. pacificum* was not because, while it only occurred at HB North, it was not abundant in the few quadrats in which it did occur. *Gayralia oxysperma*, a green alga that requires brackish water, was a significant indicator for TM upriver where it occurred consistently and in abundance

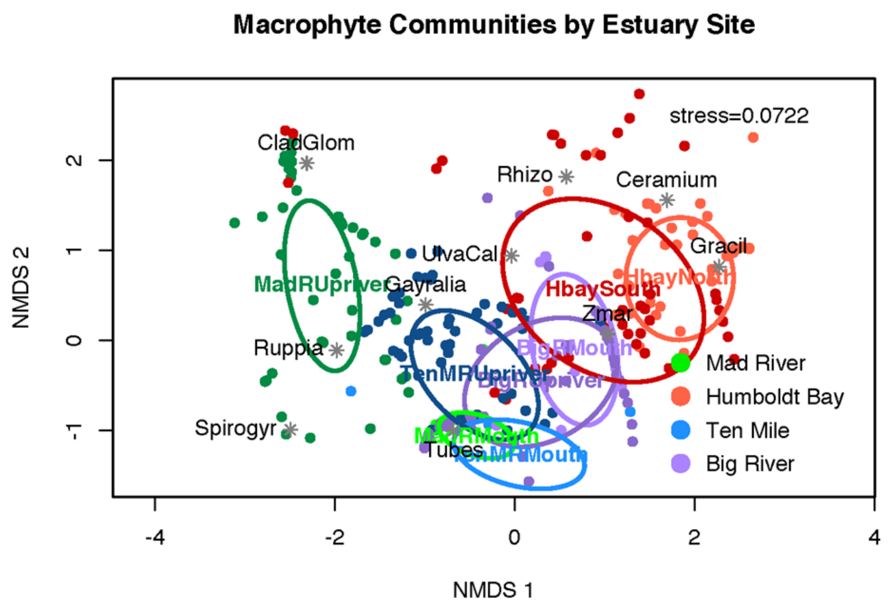


Figure 28. NMDS ordination of summer macrophytes for the 8 estuarine sites. Sites from the same estuary are shades of the same color. The final matrix was 10 species by 425 quadrats; the % cover data were arcsine square root transformed. Ellipses are standard deviations for each group centroid.

versus the MR upriver site where it occurred sporadically. The BR mouth site did not have taxa with high Indicator Value (IV) scores because *Ulva californica* was not abundant and *Z. marina* occurred in three of the estuaries. Although IV values were not always high, the taxa the ISA identified as significant indicate a range of different salinity preferences, which supports the interpretation that salinity is the primary gradient producing the macrophyte community differences visualized in the ordination.

Table 9. Indicator species analysis of summer macrophytes for the 8 estuarine sites (IV: indicator value). The % cover data were not transformed.

			IV from Randomized Groups		
Site	Species	Observed Indicator Value (IV)	Mean	St. Dev.	P
MR, Mouth	*Ulva* tubes	42.4	9.2	2.5	0.0002
MR, Upriver	*Cladophora glomerata*	34.5	4.2	2.28	0.0002
MR, Upriver	*Ruppia maritima*	47.2	5.2	2.34	0.0002
MR, Upriver	*Spirogyra wrightinana*	8.1	2.3	1.8	0.0096
HB, North	*Gracilaria vermiculophylla*	67.5	6.5	2.68	0.0002
HB, North	*Ceramium pacificum*	18.7	3	2.22	0.0012
HB, South	*Rhizoclonium riparium*	18.5	3.8	2.13	0.0014
TM, Upriver	*Gayralia oxysperma*	41.6	5.1	2.46	0.0002
BR, Mouth	*Ulva californica*	10	2.9	2.01	0.0164
BR, Mouth	*Zostera marina*	22.4	7.4	2.4	0.001

Invertebrate community structure was the same for the two HB sites, and both were separate from all three of the riverine estuaries (Figure 29). Similar to the macrophyte ordination, the mouth site for TM and MR are closer to each other than they are to their respective upriver sites. BR invertebrate communities were, like their macrophyte communities, very similar. Despite being so close to the ocean, the BR invertebrate communities were distinct from those in HB. The isopods (Sphaeromatidae) *Gnorimosphaeroma noblei* and *Gnorimosphaeroma oregonensis* had the highest IV scores for MR upriver whereas the polychaete *Lumbrineris zonata* was the one and only indicator for HB south (Table 10).

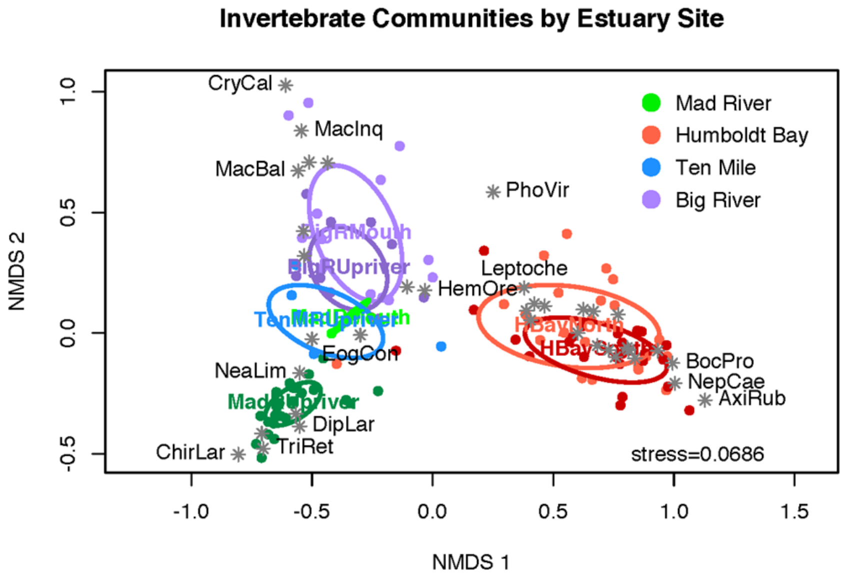


Figure 29. NMDS ordination of summer invertebrates at the 8 estuarine sites. Only invertebrates from the box and infaunal cores were included. The final matrix was 41 invertebrate species by 118 traps. The presence-absence data were transformed using the Beals smoothing function in PC-ORD. Ellipses are standard deviations for the centroid of each group.

Table 10. Indicator species analysis of summer invertebrates in the 8 estuarine sites (IV: indicator value). The data were not smoothed by the Beal's function.

			IV from Randomized Groups		
Site	Species	Observed Indicator Value (IV)	Mean	St. Dev.	P
Mad River, Mouth	Diptera Larvae	37.3	12.5	8.06	0.025
Mad River, Mouth	*Gnorimosphaeroma noblei*	56.7	12.3	7.86	0.0034
Mad River, Mouth	*Gnorimosphaeroma oregonensis*	56.3	13	7.68	0.0002
Mad River, Upriver	*Americorophium salmonis*	32.3	14	7.02	0.0206
Humboldt Bay, South	*Lumbrineris zonata*	38.3	13.5	7.34	0.016
Big River, Mouth	*Neotrypaea californiensis*	26.2	11.4	8.87	0.0464

The fish communities were ordinated in almost the exact same gradient pattern as for macrophytes (Figure 30). The upriver sites of MR and TM were on one side of the ordination whereas the two HB sites were on the other. Since both of the BR sites are so oceanic, the two BR fish communities were similar to each other and close to HB. Two fish, the coastrange sculpin and prickly sculpin, which are known to prefer estuarine reaches with a strong freshwater component, had high IV scores in the upriver site of MR. Fish with high IV scores from the channel draining the salt marsh at TM upriver (Figure 9) included shiner surfperch and three-spined stickleback (Table 11). IV scores for these fish are partially inflated because, while they were abundant in the fyke net used to capture them (i.e. catch numbers were relativized before doing ordination and ISA analyses), only one fyke net was deployed per survey time and so, from the perspective of an ISA, 'frequency' would have been high. English sole and arrow goby had high scores at, respectively, the BR mouth and upriver sites (Table 11).

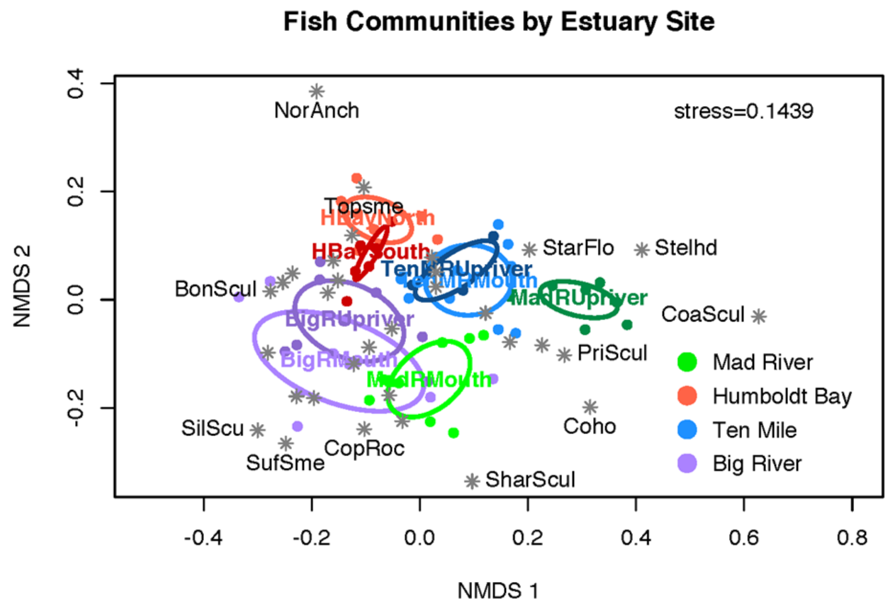


Figure 30. NMDS ordination of summer fish at the 8 estuarine sites. Species occurring in 2 or fewer seine/fyke nets were removed leaving a matrix of 32 species by 61 netting events. Catch data underwent General Relativization (*sensu* McCune and Grace 2002) by species before undergoing Beal's smoothing. Ellipses are standard deviations for each group centroid.

Table 11. Indicator species analysis of summer fish from the 8 estuarine sites (IV: indicator value). The catch data were General Relativized (McCune and Grace 1992) by species.

			IV from Randomized Groups		
Site	Species	Observed Indicator Value (IV)	Mean	St. Dev.	P
Mad River Mouth	Saddleback Gunnel (*Pholis ornata*)	58.5	12.9	7.62	0.001
Mad River Upriver	Coastrange Sculpin (*Cottus aleuticus*)	62.5	13.9	8.33	0.0018
Mad River Upriver	Prickly Sculpin (*Cottus asper*)	88.1	20	8.91	0.0002
Mad River Upriver	Starry Flounder (*Platichthys stellatus*)	38.1	19.5	8.4	0.0402

Mad River Upriver	Steelhead Trout (*Oncorhynchus mykiss*)	27	11.8	6.54	0.0426
Ten Mile River, Upriver	Pacific Staghorn Sculpin (*Leptocottus armatus*)	71.1	36.7	7.18	0.001
Ten Mile River, Upriver	Shiner Surfperch (*Cymatogaster aggregata*)	92.2	26.3	10.67	0.0002
Ten Mile River, Upriver	Three-spined stickleback (*Gasterosteus aculeatus*)	88.3	42.3	13.07	0.002
Big River, Mouth	Buffalo Sculpin (*Enophrys bison*)	32.1	11.7	7.06	0.0254
Big River, Mouth	Cabezon (*Scorpaenichthys marmoratus*)	44.4	16.1	7.97	0.0074
Big River, Mouth	English Sole (*Pleuronectes vetulus*)	47.2	21.4	8.11	0.014
Big River, Upriver	Arrow Goby (*Clevelandia ios*)	70.1	20.5	8.71	0.001

For all three trophic levels, the degree of ocean connectivity (i.e. the extent to which the mouth of the estuary remains open to the ocean), and so presumably the extent of the salt water wedge, appears to have a stronger effect on community structure than MPA bioregion. The similar SST and watershed discharge patterns between the two MPA bioregions (Figure 14, Figure 15, Figure 16, Figure 17, Figure 18) did not make estuarine community structures similar, particularly in the case of macrophytes and fish.

Community Structure among Years

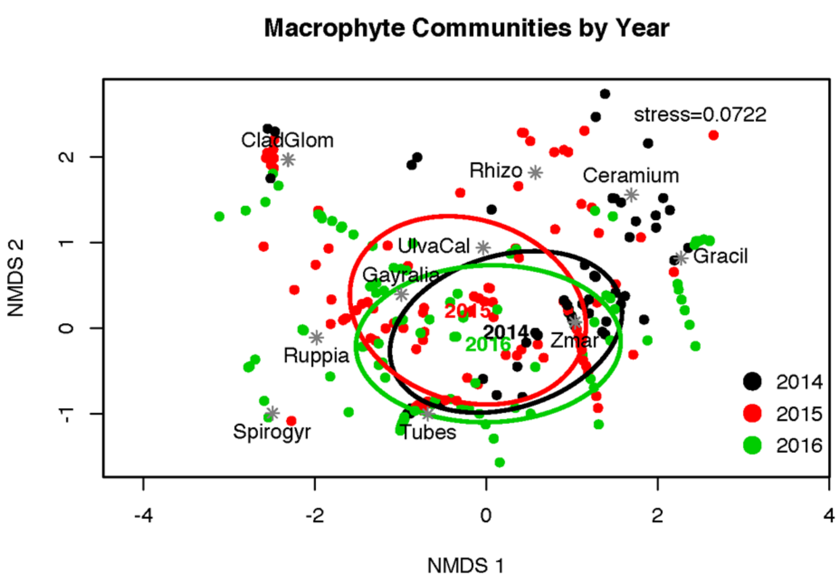


Figure 31. NMDS ordination of the three summer macrophyte communities. The final matrix was 10 species by 425 quadrats. The percent cover data were then arcsine square root transformed. Ellipses are standard deviations for group centroids.

Macrophyte community structure among the three summers was not different despite the oceanic SST and watershed discharge environments that did change from Su14 to Su15 (Figure 31). However, ISA suggests that particular species may have been responding to the interannual climate changes. *G. oxysperma* in TM, which needs freshwater to create a brackish habitat, and *G. vermiculophylla* in HB, which occurs on mid intertidal mudflats, may have positively responded to, respectively, more summer freshwater and less desiccation (Table 12). Overall invertebrate communities also did not change over the three years (Figure 32) and, while many invertebrates had significant IV scores, the scores themselves are generally low with the exception of the clam *Macoma inquinata* during 2015 at BR (Table 13). Fish communities also did not vary among summers (Figure 33). Many fish had high IV scores for particular years (Table 14). Because of the way IV scores are calculated, just because

Table 12. Indicator species analysis of summer macrophytes occurring in 12 groups (3 summers * 4 estuaries / summer; IV: indicator value). The % cover data were not transformed.

			IV from Randomized Groups		
Site	Species	Observed Indicator Value (IV)	Mean	St. Dev.	P
2014, Mad River	*Ruppia maritima*	15.8	4	1.23	0.0002
2016, Mad River	*Spirogyra wrightiana*	15	2.2	1.37	0.0002
2014, Humboldt Bay	*Ceramium pacificum*	21.6	3	1.68	0.0002
2014, Big River	*Zostera marina*	33.5	5.5	1.32	0.0002
2015, Mad River	*Cladophora glomerata*	21.9	3.3	1.28	0.0002
2015, Humboldt Bay	*Rhizoclonium riparium*	32	3.1	1.37	0.0002
2015, Ten Mile River	*Gayralia oxysperma*	37.2	3.9	1.38	0.0002
2016, Humboldt Bay	*Gracilaria vermiculophylla*	53.3	5	1.55	0.0002
2016, Big River	*Ulva* tubes	19	6.8	1.45	0.0002
2016, Big River	*Ulva californica*	5.7	2.6	1.37	0.0354

three-spined stickleback had a high IV during summer 2016 in TM does not mean it was absent in another estuary like MR, it only indicates that this fish was present in all the seines at TM and its abundance in those seines was relatively high. The significant IV fish species for BR were, across the three summers, all outer coast rocky reef fish (e.g. cabezon, juvenile rockfish) or outer coast beach-sandy bottom fish like striped surfperch and English sole. In contrast, the more freshwater influenced MR and TM had high IV scores for prickly sculpin and three-spined stickleback, which are fish with a preference for lower salinities.

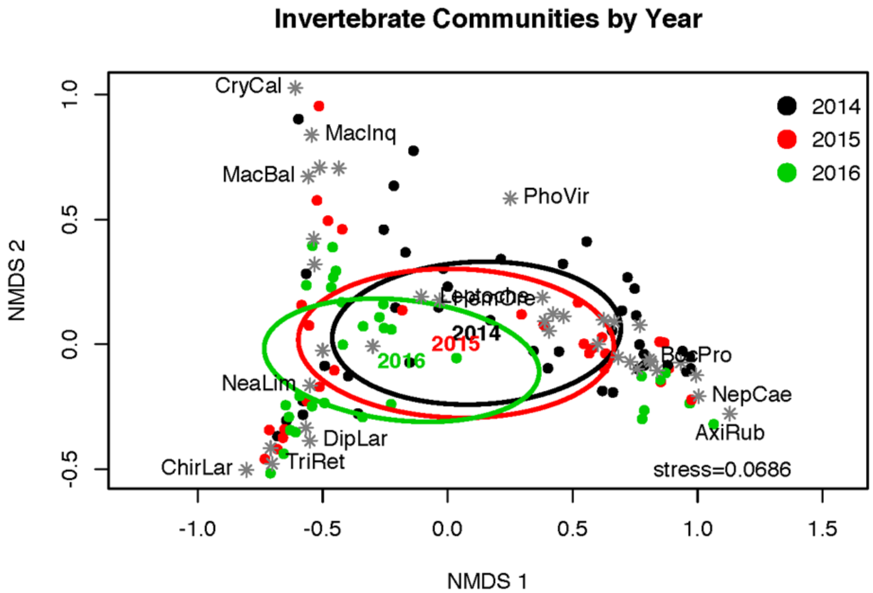


Figure 32. NMDS ordination of invertebrates during the 3 summers. Species occurring in 3 or less traps were removed leaving a matrix of 41 species by 246 traps. These presence/absence data were then transformed using the Beal's smoothing function. Ellipses are standard deviations for group centroids.

Table 13. Indicator species analysis of summer invertebrates in 12 groups (3 summers * 4 estuaries / summer; IV: indicator value). The presence-absence data were not transformed.

			IV from Randomized Groups		
Site	Species	Observed Indicator Value (IV)	Mean	St. Dev.	P
2014, Mad River	*Gnorimosphaeroma noblei*	16.6	4.3	2.43	0.007
2014, Mad River	*Gnorimosphaeroma oregonensis*	31.8	5.1	2.33	0.0002
2014, Humboldt Bay	*Ampithoe lacertosa*	11.1	3.8	2.87	0.0132
2014, Humboldt Bay	*Boccardiella ligerica*	22.3	4	2.41	0.0008
2014, Humboldt Bay	*Boccardia proboscidea*	14.8	3.9	2.74	0.0118
2014, Humboldt Bay	*Leptochelia* sp.	11.6	3.9	2.67	0.0196

2014, Humboldt Bay	*Lumbrineris zonata*	19.8	5.5	2.37	0.0016
2014, Humboldt Bay	*Macoma nasuta*	25.9	4	2.55	0.0008
2014, Humboldt Bay	*Mediomastus ambiseta*	25	5.3	2.34	0.0004
2014, Humboldt Bay	*Mediomastus californiensis*	32.3	5.3	2.29	0.0002
2014, Humboldt Bay	*Nebalia kensleyi*	23.4	4.1	2.57	0.0002
2014, Humboldt Bay	*Notomastus magnus*	14.8	3.9	2.68	0.0108
2014, Humboldt Bay	*Nutricola tantilla*	20.7	4.8	2.43	0.0026
2014, Humboldt Bay	*Paracorophium* sp.	25.9	4	2.5	0.0008
2014, Humboldt Bay	*Peramphithoe mea*	11.6	3.9	2.56	0.0188
2014, Humboldt Bay	*Platynereis bicanaliculata*	15	4	2.44	0.0094
2014, Humboldt Bay	*Schistomeringos longicornis*	14.2	4	2.47	0.0062
2014, Ten Mile River	*Americorophium spinicorne*	17.3	8	1.82	0.002
2014, Big River	*Cryptomya californica*	9.6	3.9	2.39	0.028
2015, Mad River	*Chironomidae Larvae*	13.3	4	2.55	0.0152
2015, Humboldt Bay	*Caprella californica*	9.6	3.9	2.76	0.0216
2015, Ten Mile River	*Eogammarus confervicolus*	15	6.3	2.21	0.0102
2015, Ten Mile River	*Limecola balthica*	10	4.1	2.64	0.0438
2015, Ten Mile River	*Mya arenaria*	14.3	4	2.44	0.0116
2015, Big River	*Hemigrapsus oregonensis*	21.5	5.5	2.31	0.0004
2015, Big River	*Macoma inquinata*	44.4	3.9	2.77	0.0002
2015, Big River	*Alitta brandti*	19.9	4	2.49	0.002
2015, Big River	*Neotrypaea californiensis*	19.3	4.3	2.47	0.0008
2016, Mad River	*Diptera Larvae*	14.7	4.6	2.54	0.0144
2016, Mad River	*Excirolana chiltoni*	14.3	4	2.49	0.0142
2016, Mad River	*Trichocorixa reticulata*	19.2	3.9	2.54	0.0062
2016, Humboldt Bay	*Axiothella rubrocincta*	23	4	2.37	0.0004

2016, Big River	*Americorophium salmonis*	14.3	6.2	2.19	0.014
2016, Big River	*Neomysis mercedis*	22	4	2.42	0.0018

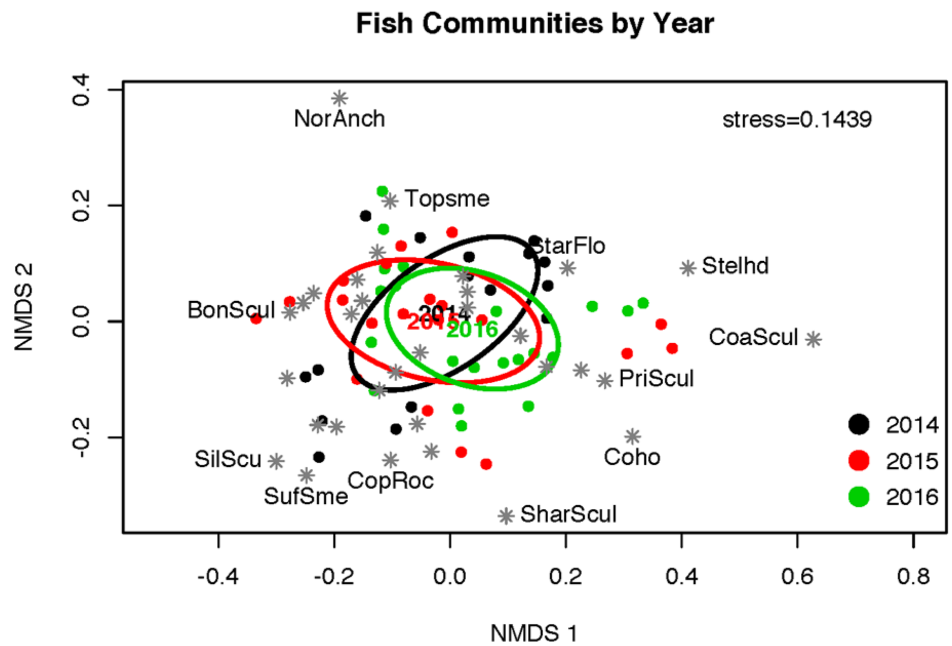


Figure 33. NMDS ordination of the three summer fish communities. Removal of rare species resulted in a matrix of 26 fish species in 60 netting events. The data were transformed using the Beals smoothing function. Ellipses are standard deviations for group centroids.

Table 14. Indicator species analysis of summer fish communities in 12 groups (3 summers * 4 estuaries / summer; IV: indicator value). The data underwent General Relativizing (McCune and Grace 1992).

			IV from Randomized Groups		
Site	Species	Observed Indicator Value (IV)	Mean	St. Dev.	P
2014, Big River	Buffalo Sculpin (*Enophrys bison*)	37.5	13.9	8.12	0.0272
2014, Big River	Copper Rockfish *(Sebastes caurinus*)	41.7	14.6	7.13	0.0062
2014, Big River	Striped Surfperch (*Embiotica Lateralis*)	66.2	18	9.06	0.0016
2015, Mad River	Coastrange Sculpin (*Cottus aleuticus*)	47.9	17.1	8.86	0.0088
2015, Mad River	Prickly Sculpin (*Cottus asper*)	52.9	20.8	9.37	0.0092
2015, Mad River	Saddleback Gunnel (*Pholis ornata*)	37.9	14.9	7.81	0.014
2015, Big River	Arrow Goby (*Clevelandia ios*)	60.8	20.7	9.14	0.0016
2015, Big River	Cabezon (*Scorpaenichthys marmoratus*)	83.5	17.3	8.48	0.0002
2015, Big River	English Sole (*Pleuronectes vetulus*)	67.7	20.8	7.72	0.0002
2015, Big River	Juvenile Rockfish (*Sebastes sp.*)	79.5	22	10.98	0.0004
2015, Big River	Kelp Greenling (*Hexogrammos decagrammus*)	50	14.5	7.68	0.005
2015, Big River	White Surfperch (*Phanerodon furcatus*)	50	13.1	7.38	0.0052
2016, Ten Mile River	Pacific Staghorn Sculpin (*Leptocottus armatus*)	45.4	33.6	6.14	0.0492
2016, Ten Mile River	Speckled Sanddab (*Citharichthys stigmaeus*)	48.5	14.1	8.17	0.0102
2016, Ten Mile River	Three-spined stickleback (*Gasterosteus aculeatus*)	74.9	44.6	15.15	0.045
2016, Big River	Pacific Herring (*Clupea pallasii*)	42.8	19	9.66	0.0274

PerMANOVA Analyses

The PerMANOVA models for each trophic level (Table 15) generally supported the visual representation of community structure in the ordinations. Given that there was often statistically significant multivariate dispersion among groups, we placed more emphasis in these analyses on R^2 values than p values. Variation in macrophyte community structure was most affected by Estuary, as shown in Figure 28, but also by the interaction between Year and Site nested within Estuary, which is not evident in Figure 31because sites within each year are not labelled. Some species that would have high ordination weight at particular sites, and varied in abundance across years, could have resulted in this interaction. One example of this kind of species is the large changes in *Z. marina* abundance across years at four of the eight sites (see

Target Species below). In contrast, Estuary had the largest effect on invertebrate community structure, which is also the case for the fish communities (Table 15). However, variation in the latter community also depended upon the site and year that the community was sampled.

Table 15. PerMANOVA analyses of the effects of estuary, site (nested within estuary) and year on the macrophyte, invertebrate and fish distances measures used in the NMDS ordination of each of these trophic levels. The p values for each PerMANOVA should be used with caution as each group showed evidence of heterogeneity of multivariate dispersion among sites.

Macrophytes						
	DF	SS	MS	F model	R^2	p
Year	2	4.005	2.0025	7.833	0.0224	0.001
Estuary	3	30.594	10.1979	39.890	0.1710	0.001
Site(Estuary)	6	15.043	2.5072	9.807	0.0841	0.001
Year * Site(Estuary)	12	26.763	2.2303	8.724	0.1496	0.001
Residuals	401	102.516	0.2557		0.5730	
Total	424	178.921			1.0000	

Invertebrates						
	DF	SS	MS	F model	R^2	p
Year	2	0.6595	0.3297	9.104	0.0439	0.001
Estuary	3	9.5858	3.1953	88.217	0.6388	0.001
Site(Estuary)	6	0.5675	0.0946	2.611	0.0378	0.001
Year * Site(Estuary)	9	0.7518	0.0835	2.306	0.0501	0.003
Residuals	95	3.4410	0.0362		0.2293	
Total	115	15.0056			1.0000	

Fishes						
	DF	SS	MS	F model	R^2	p
Year	2	0.0971	0.0485	11.080	0.0878	0.001
Estuary	3	0.4835	0.1612	36.762	0.4368	0.001
Site(Estuary)	6	0.1466	0.0244	5.573	0.1324	0.001
Year * Site(Estuary)	12	0.2175	0.0181	4.135	0.1965	0.001
Residuals	37	0.1622	0.0044		0.1465	
Total	60	1.10698			1.00000	

Target Species – a Preface

Most of the species and metrics described in the project proposal (Table 5) were able to be measured in this baseline study. There were some exceptions, like enumerating the mesograzer

Phyllaplysia taylori, and measuring the depth of the deep edge of *Z. marina* at all the sites in which it occurred. More explanation of all of these target variables is given below.

Presentation of the target species information in this study is organized according to how organisms use the estuary. There are **Ocean & Estuary** species that spend a part of their lives in each system. This includes many invertebrate and fish species; e.g. rockfish and certain crabs. There are also **Estuary Residents** that include macrophyte communities, but also some infaunal species and fish like the Three-spine Stickleback. Finally, there are the **Anadromous Fish** that use the freshwater, estuarine and ocean ecosystems.

Organizing the target species information into these three categories of estuarine use will hopefully assist future studies about ecosystem connections among marine habitats, or which organisms to focus on for measuring site specific events. **Ocean & Estuary** target species are the obvious list to start with for those interested in ecosystem connections, but an **Estuary Resident** like *Z. marina* is also connected to other systems because the plant is exported as detritus. In order to attribute variation of a species at a particular estuarine site to an event at the same site, the species considered should spend the majority of its life history at that site, and so those are the kinds of species included in the section on **Estuary Residents**. Finally, management priorities often focus on species of commercial importance, and so will consider all of the **Anadromous Fish** in this study, along with macrophytes like *Z. marina* that form critical fish, crab, clam and waterfowl habitat, as well as **Ocean & Estuary** species like rockfish.

Ocean & Estuary Species

Except for HB, the relative abundance of *M. magister* was higher during Su14 and Wi15 than the following three sample times, and body sizes tended to be 100 mm or less north of Cape Mendocino and 100 to 150 mm in the estuaries south of the cape (Figure 34). However, this may not be an accurate sample of this crab because traps were often pulled up by people even though the traps were identified as being part of a monitoring study. Crab traps also showed evidence of being attacked by seals, raccoons and gulls.

Juvenile rockfish were more abundant at the mouth sites of the MR and BR estuaries (Figure 35). Almost no rockfish were found in HB, which is contrast to previous studies that have documented the presence of black rockfish juveniles in HB eelgrass beds (Frimodig 2007). Most of the rockfish identified in the present study were copper rockfish, or they were specimens that were too small to be identified to species.

Night smelt was one of the few species that had its highest relative abundance during Su14 in all four estuaries (Figure 36). In contrast, English sole, surf smelt, top smelt and bay pipefish all peaked in abundance Su15 or later (Figure 37, Figure 38, Figure 39, Figure 40). English sole and bay pipefish were more common at mouth sites (the North site in HB was adjacent to a channel and the entire site was closer to Entrance Channel) whereas surf and top smelt were more abundant at up-river sites or, in the case of HB, the south site. Body lengths of these species among estuaries were similar except for top smelt, which were smaller in HB than the two estuaries south of Cape Mendocino.

Pacific herring, shiner surfperch, and starry flounder did not show parallel patterns of temporal abundance in the estuaries in which they occurred (Figure 41, Figure 42, Figure 43). Some of these species used the sites within the estuary differently. Shiner surfperch in TM only occurred at the up-river site. Even though this location was warm, saline, and with a low DO at depth, the prey coming out of the salt marsh channel may have attracted these fish. In contrast, the benthic feeding starry flounder juveniles avoided the anoxic up-river site in TM, and were most abundant at the mouth site where the water was less stratified, and DO was higher (Figure 22). The shiner surfperch and starry flounder north of Cape Mendocino were usually smaller than their counterparts south of the cape (Figure 42, Figure 43).

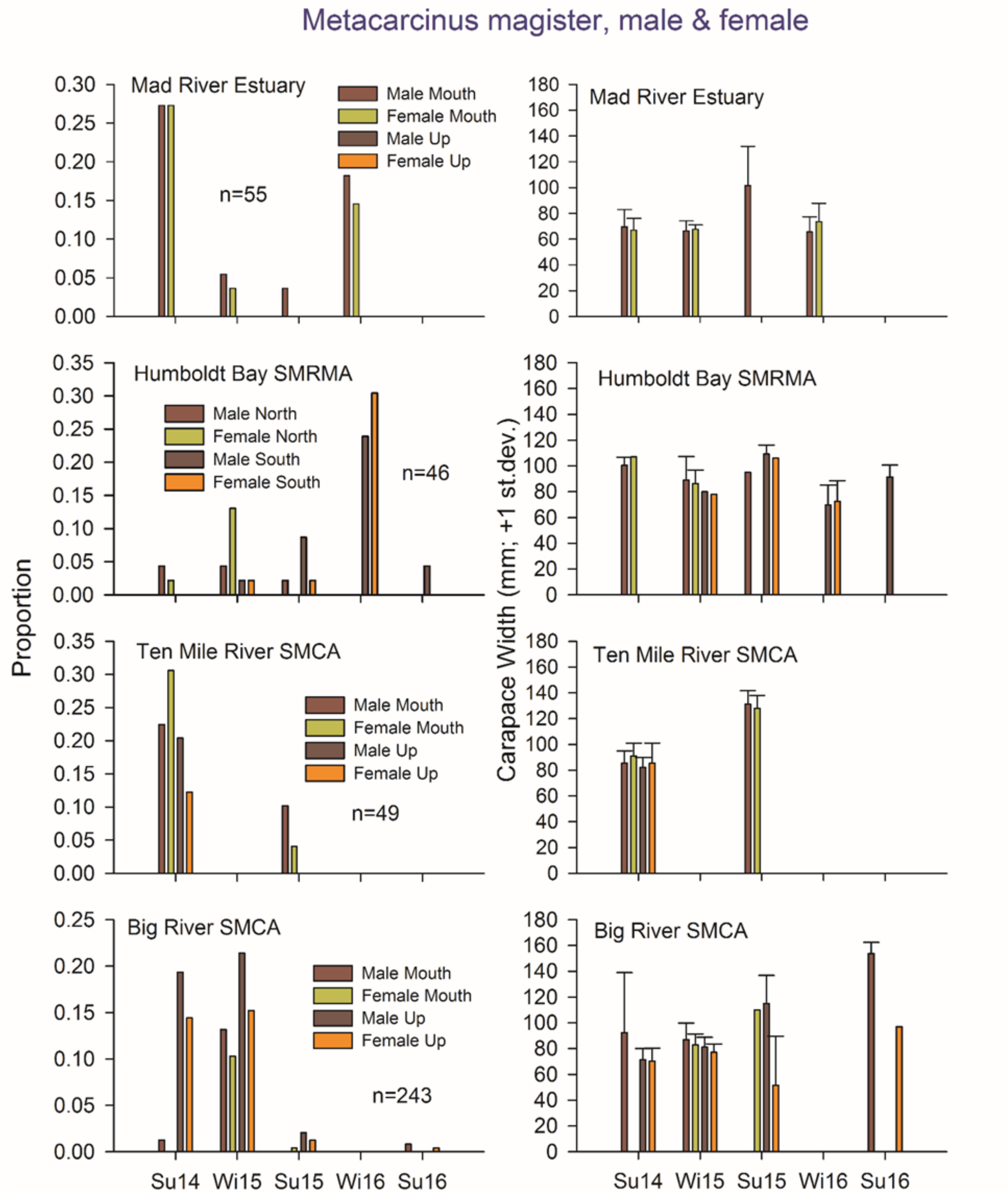


Figure 34. The relative abundance (proportion: # individuals at a site / total # individuals from all sites and times in one estuary; the total # is indicated in the left side panels as "n=") and body size (i.e. carapace width) of *Metacarcinus magister* in each estuary.

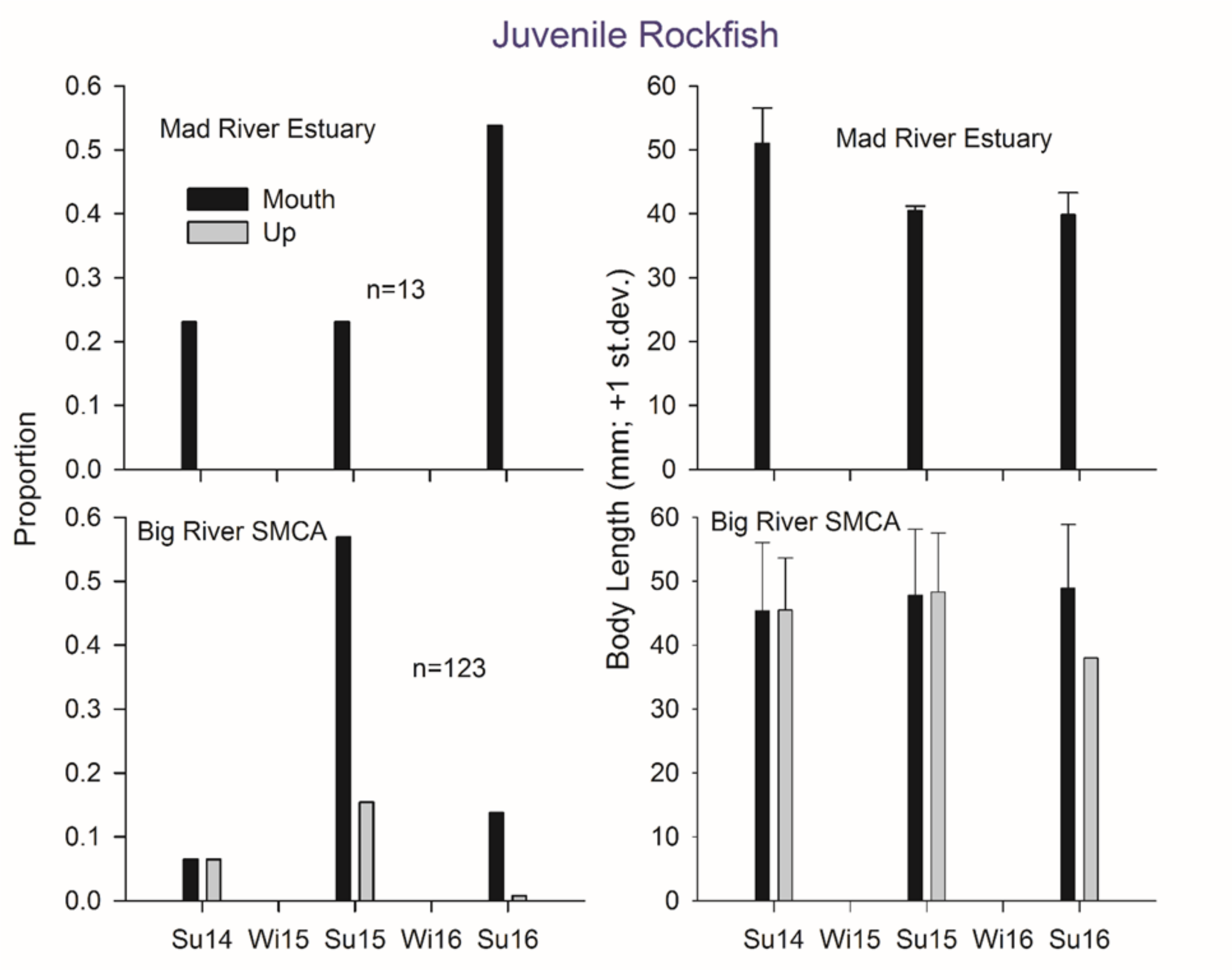


Figure 35. The relative abundance (proportion: # individuals at a site / total # individuals from all sites and times in one estuary; the total # is indicated in the left side panels as "n=") and body length of juvenile rockfish in each estuary. Over 90% of the rockfish were either copper rockfish or juveniles that could not be identified to species.

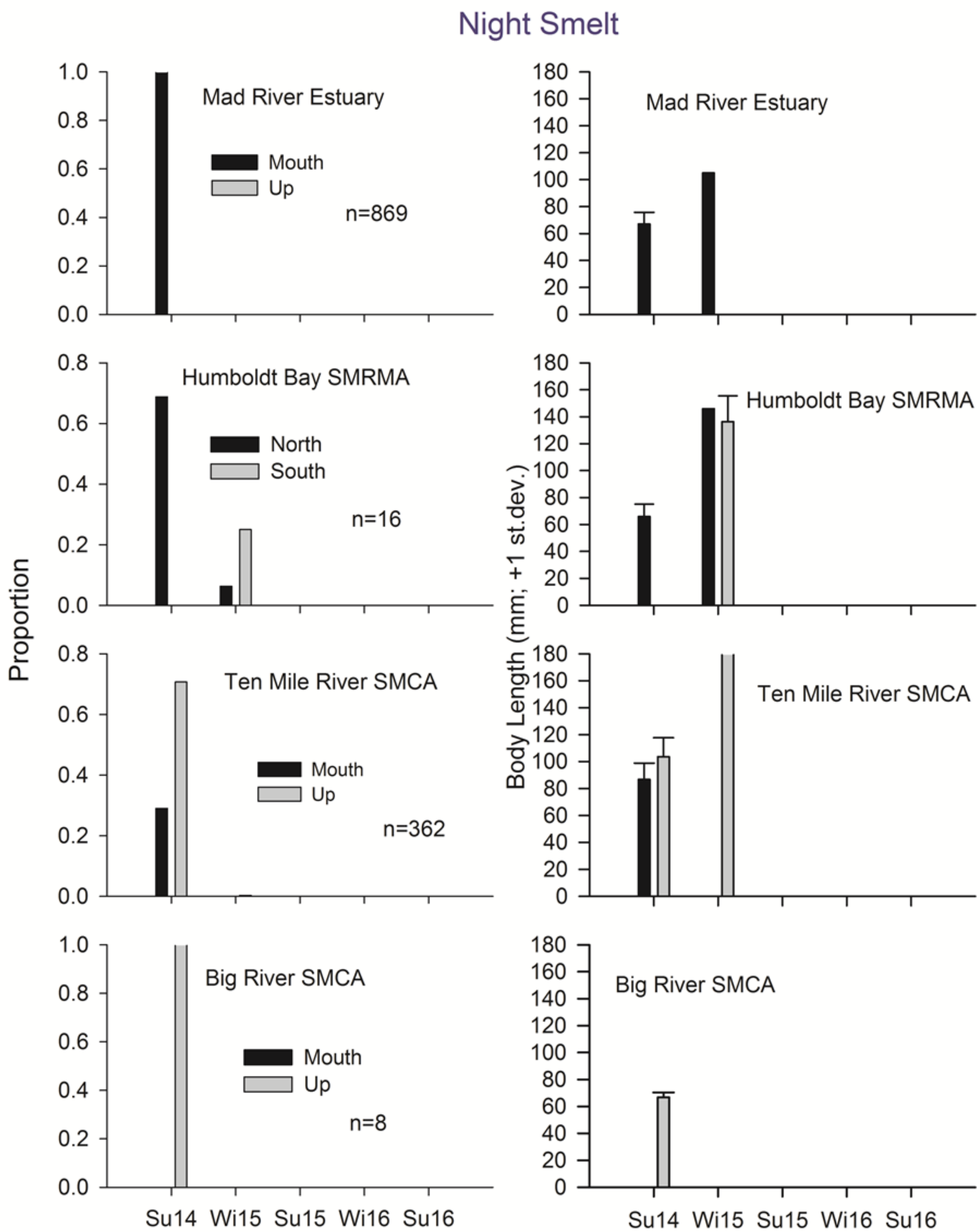


Figure 36. The relative abundance (proportion: # individuals at a site / total # individuals from all sites and times in one estuary; the total # is indicated in the left side panels as "n=") and body length of night smelt in each estuary.

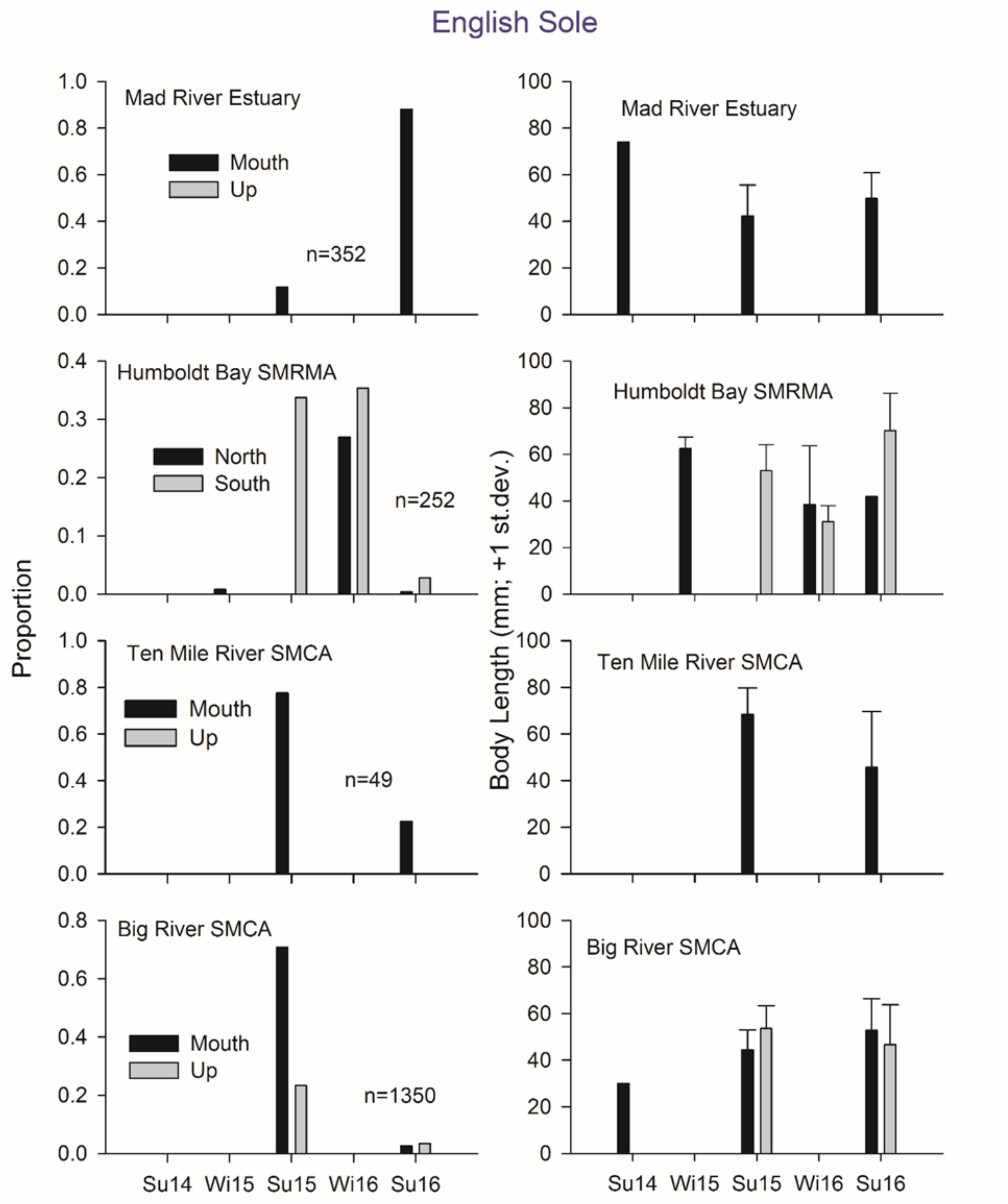


Figure 37. The relative abundance (proportion: # individuals at a site / total # individuals from all sites and times in one estuary; the total # is indicated in the left side panels as "n=") and body length of English sole in each estuary.

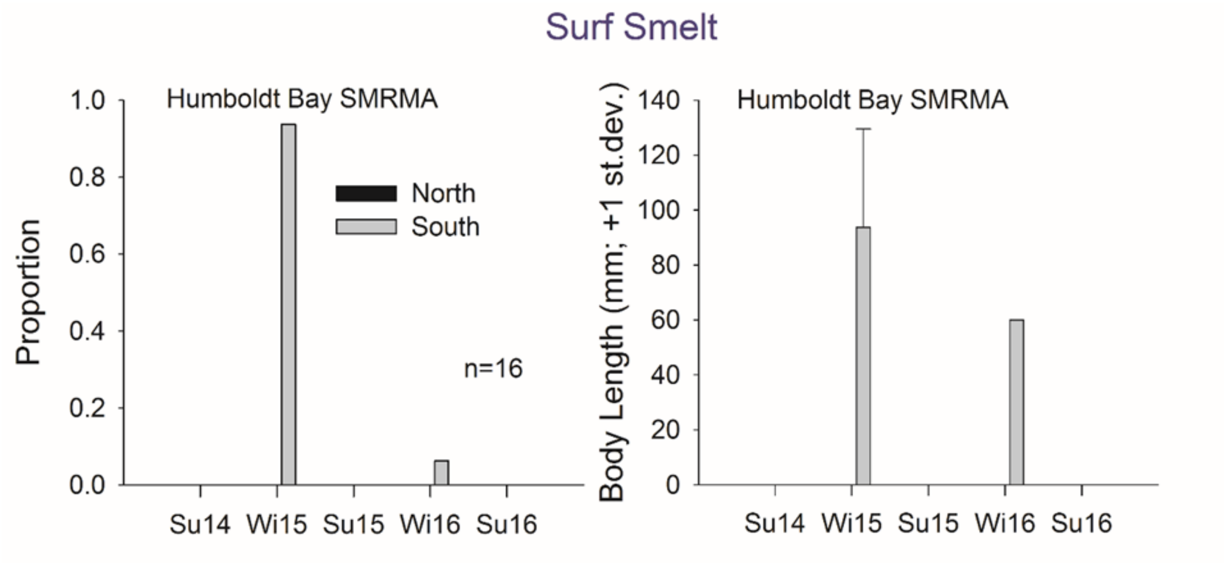


Figure 38. The relative abundance (proportion: # individuals at a site / total # individuals from all sites and times in one estuary; the total # is indicated in the left side panels as "n=") and body length of surf smelt in the Humboldt Bay SMRMA.

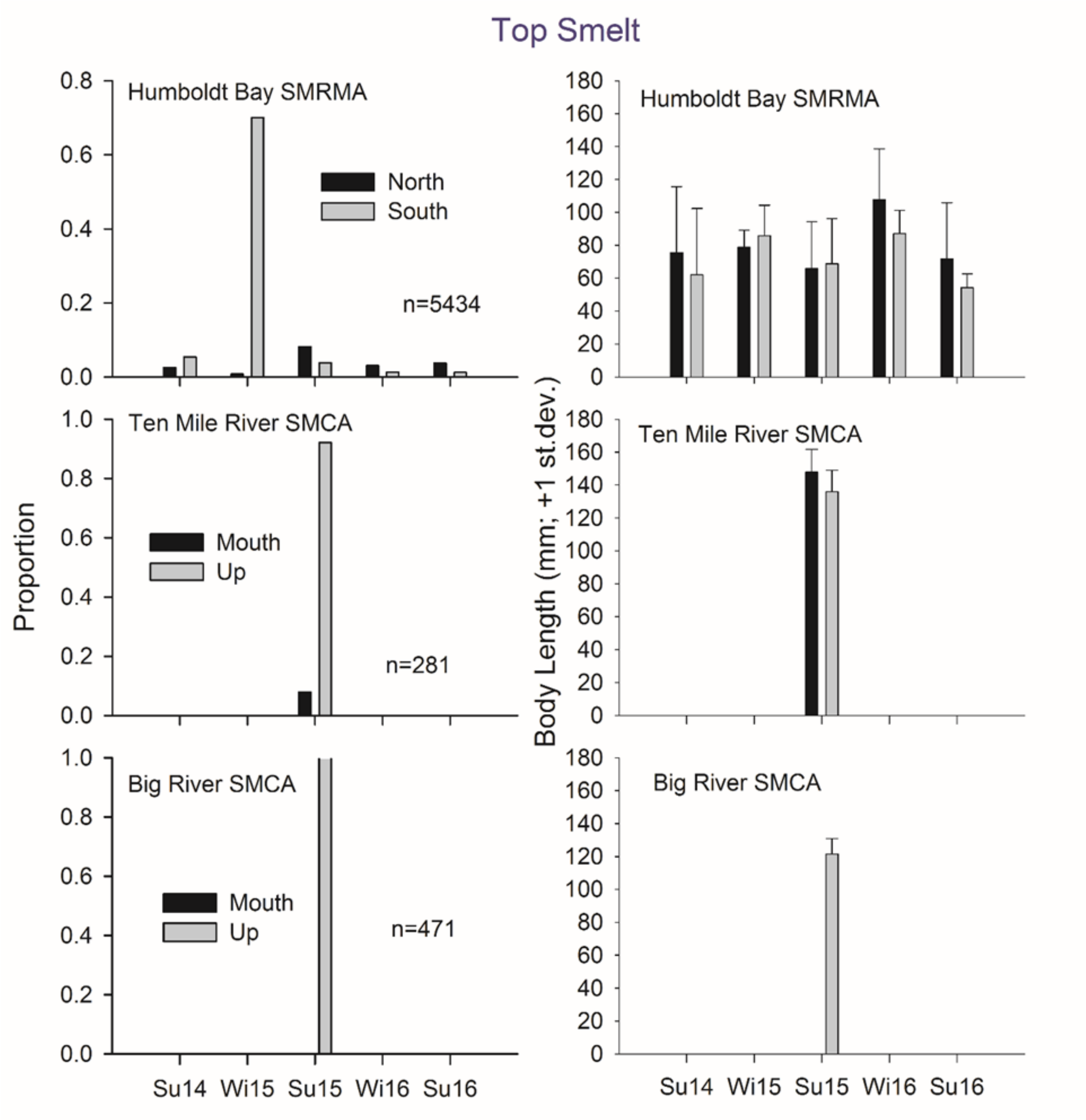


Figure 39. The relative abundance (proportion: # individuals at a site / total # individuals from all sites and times in one estuary; the total # is indicated in the left side panels as "n=") and body length of top smelt in each estuary.

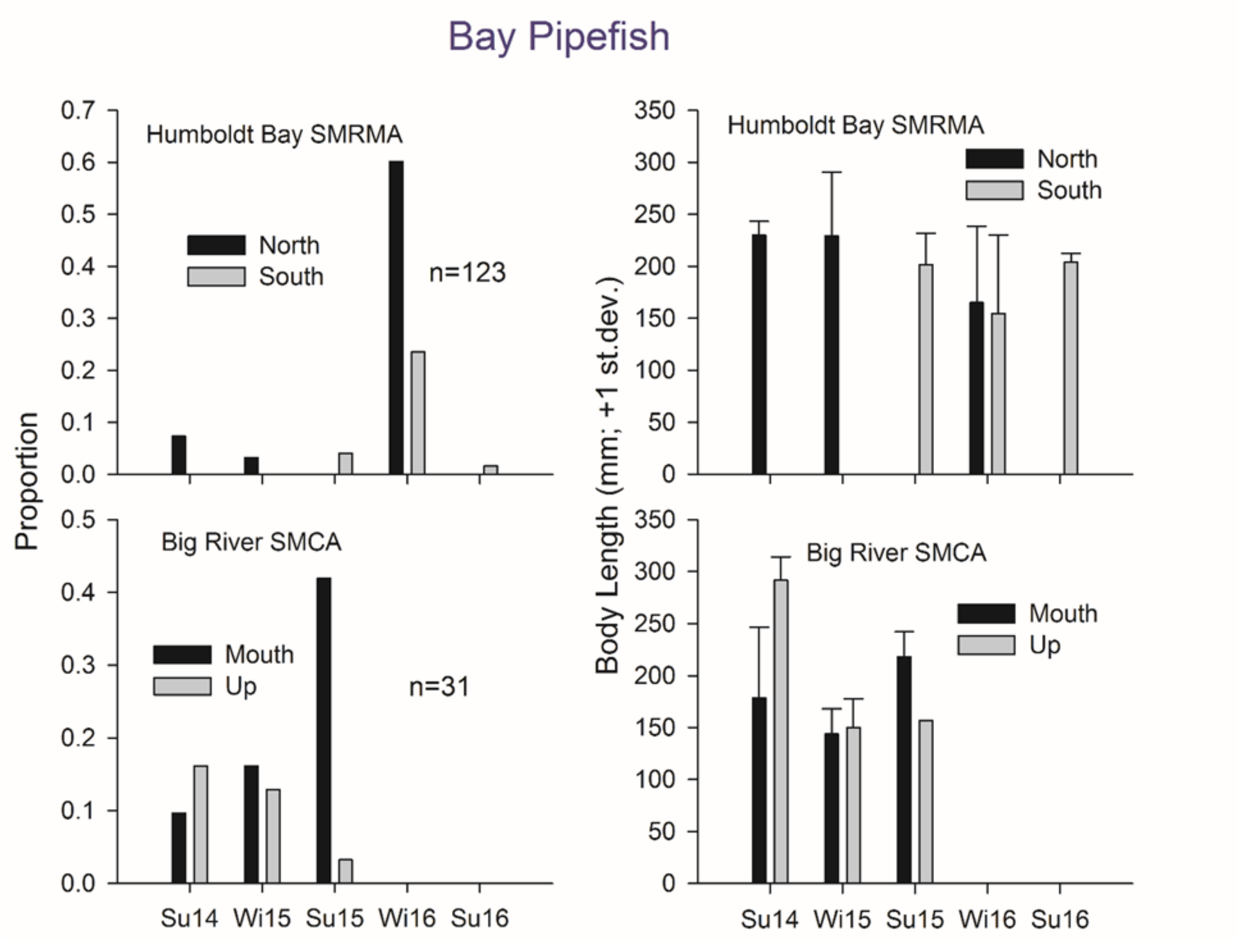


Figure 40. The relative abundance (proportion: # individuals at a site / total # individuals from all sites and times in one estuary; the total # is indicated in the left side panels as “n=”) and body length of bay pipefish in each estuary.

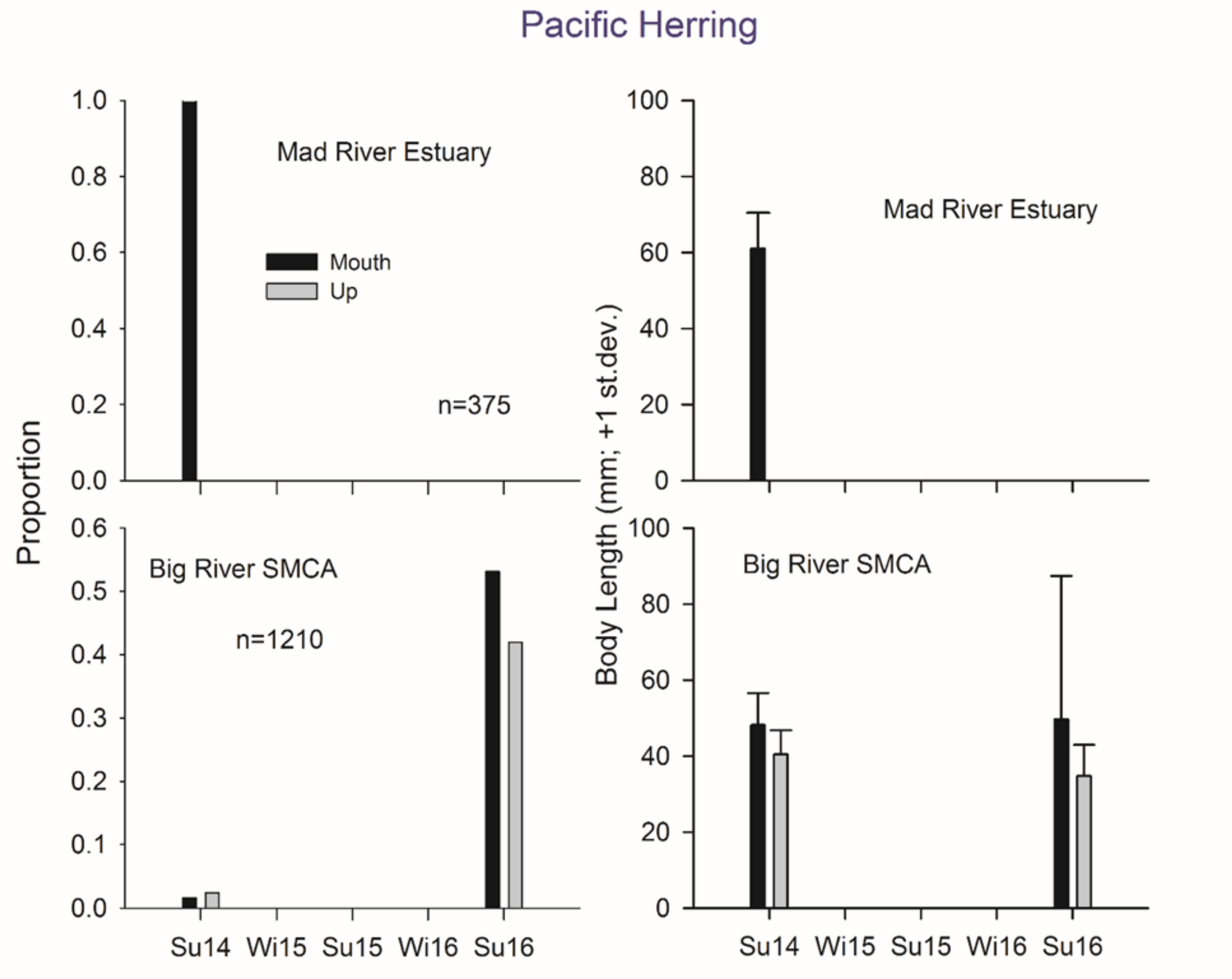


Figure 41. The relative abundance (proportion: # individuals at a site / total # individuals from all sites and times in one estuary; the total # is indicated in the left side panels as “n=”) and body length of pacific herring in each estuary.

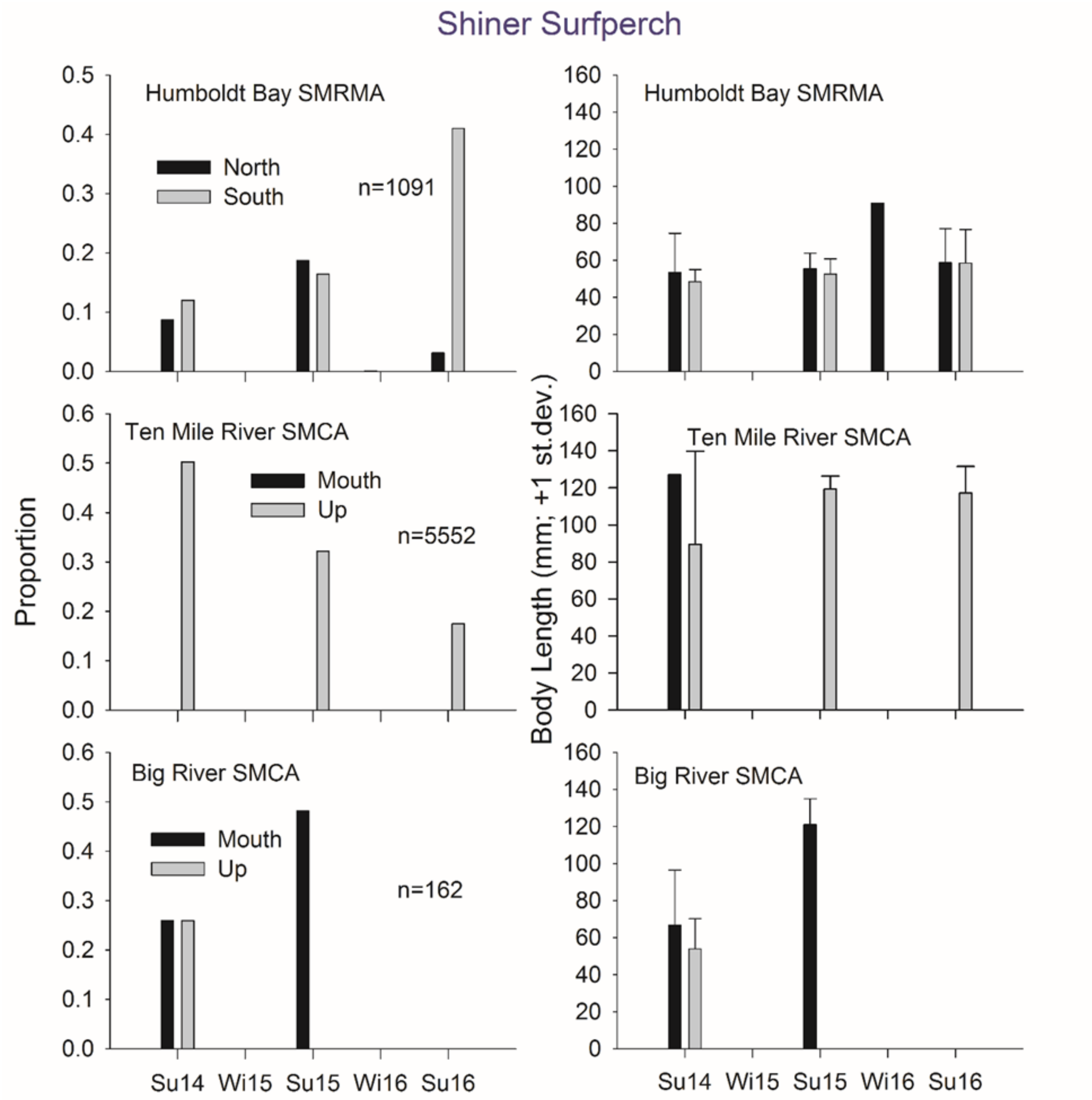


Figure 42. The relative abundance (proportion: # individuals at a site / total # individuals from all sites and times in one estuary; the total # is indicated in the left side panels as "n=") and body length of shiner surfperch in each estuary.

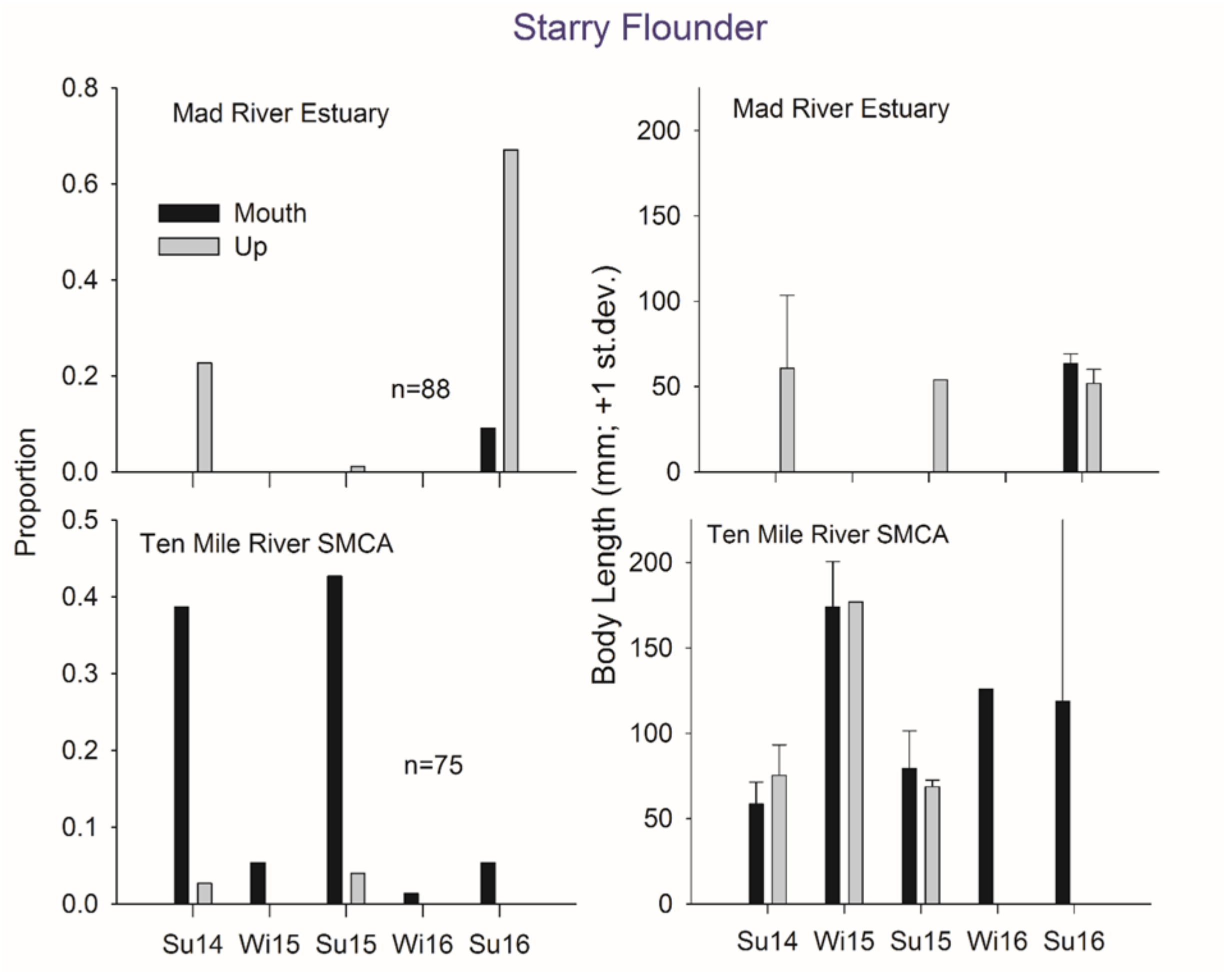


Figure 43. The relative abundance (proportion: # individuals at a site / total # individuals from all sites and times in one estuary; the total # is indicated in the left side panels as “n=”) and body length of starry flounder in each estuary.

Estuary Residents

Of the three general cover classes – macroalgae, seagrasses, bare substratum – it was bare substratum that dominated during most winter sampling periods, and in most mid intertidal transects (Figure 45, Figure 44, Figure 47, Figure 46). Perennial seagrasses are less abundant during the winter and most estuarine macroalgae are not firmly attached to a substratum, and so get swept out of the system by higher winter flows. Mouth sites at MR and TM are best described as estuarine beaches because of the high energy of ocean waves reaching these sites. When seagrass cover was higher, it was during the summer. In the low intertidal of HB and BR this cover was *Z. marina*, but in TM it was a mix of *Z. marina* and *R. maritima*. In the mid intertidal of MR and TM this was *R. maritima*. Summer increases in macroalgal cover, which could occur in the low or mid intertidal, were primarily *Ulva linza* at the MR mouth site, *C. glomerata* at the MR upriver site, *G. vermiculophylla* at the HB North site, *Rhizoclonium riparium* at the HB South site, *G. oxxysperma* and *Ulva intestinalis* at the TM upriver site, and *Ulva torta* at the BR upriver site. The summer surveys were done in June and so the tubular ulvoids (*U. linza, U. intestinalis, U. torta*) would continue to grow during July and August, but their generally low June cover suggests a lack of eutrophication in these estuaries (Valiela et al. 1997).

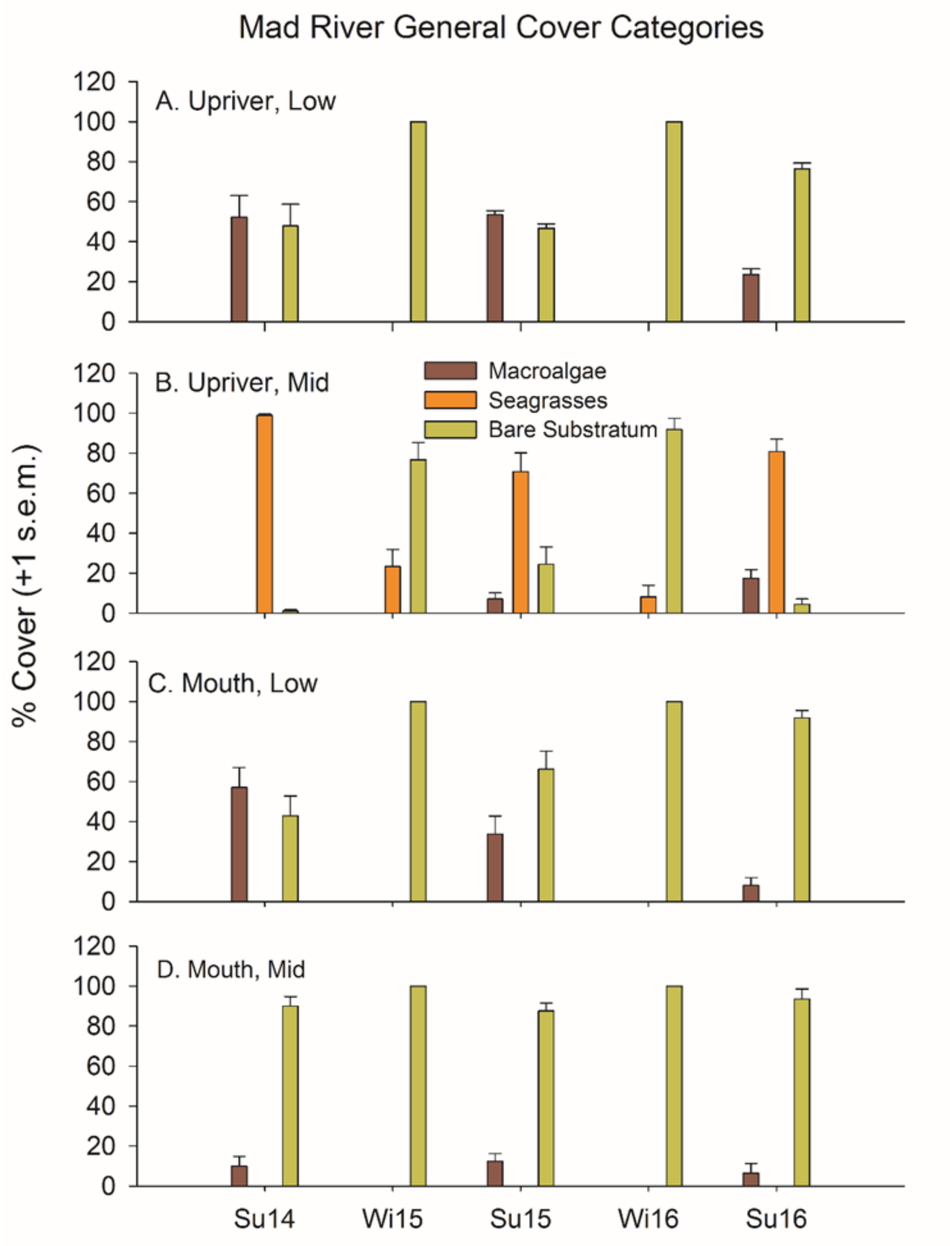


Figure 44. The general cover classes of macroalgae, seagrasses and bare substratum for the mid and low transects within the Mad River estuary mouth and upriver sites.

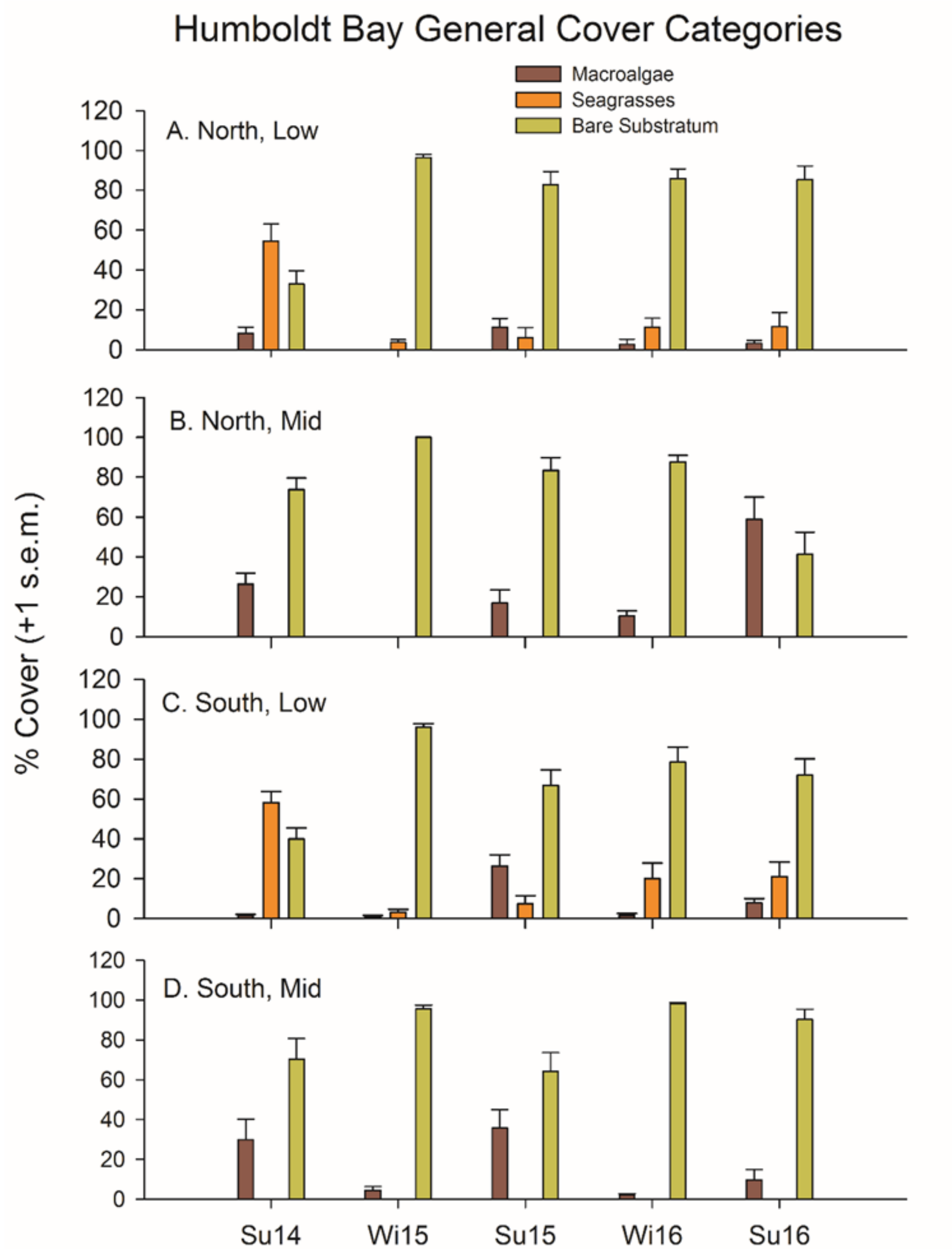


Figure 45. The general cover classes of macroalgae, seagrasses and bare substratum for the mid and low transects within the Humboldt Bay SMRMA North and South sites.

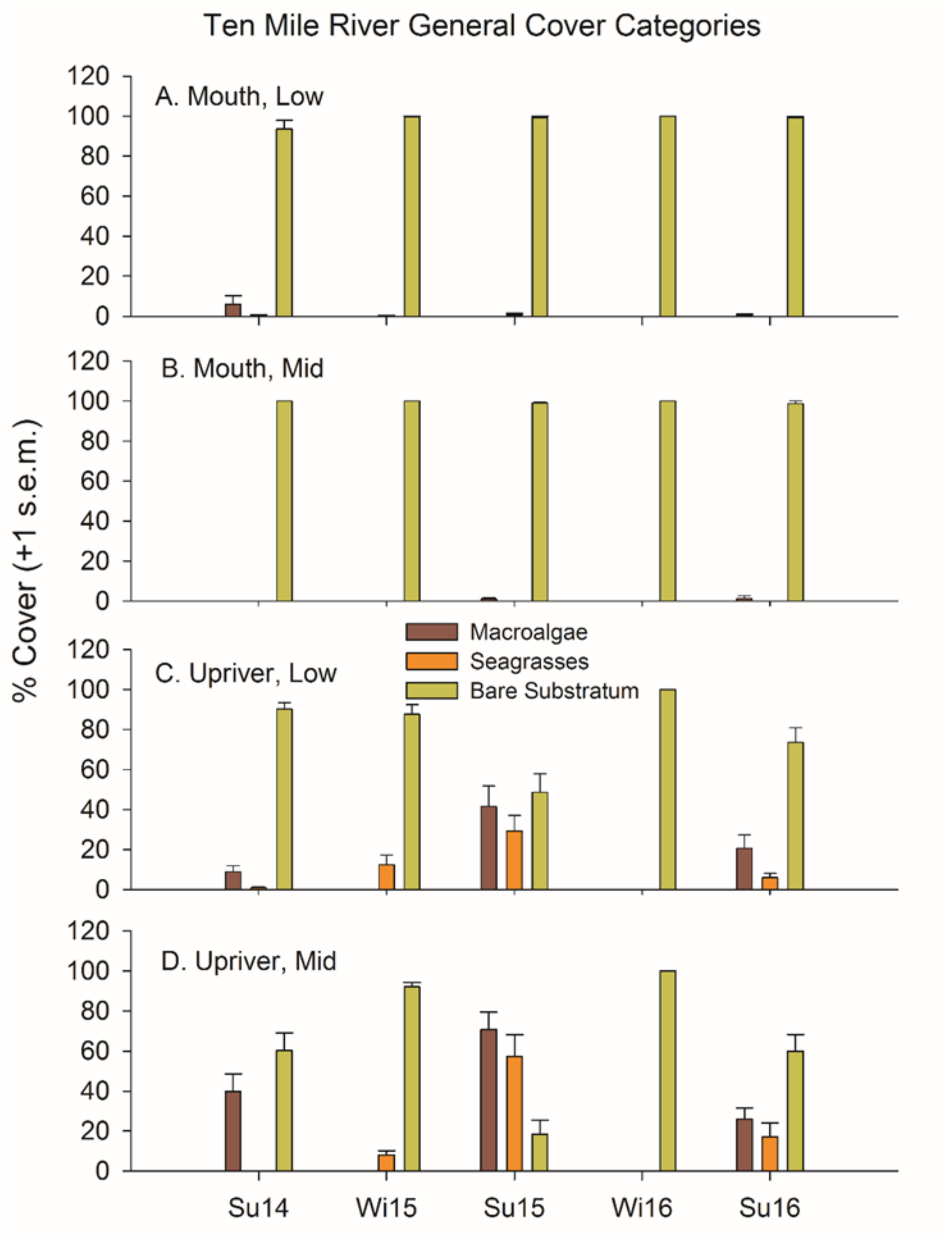


Figure 46. The general cover classes of macroalgae, seagrasses and bare substratum for the mid and low transects within the Ten Mile River SMCA mouth and upriver sites.

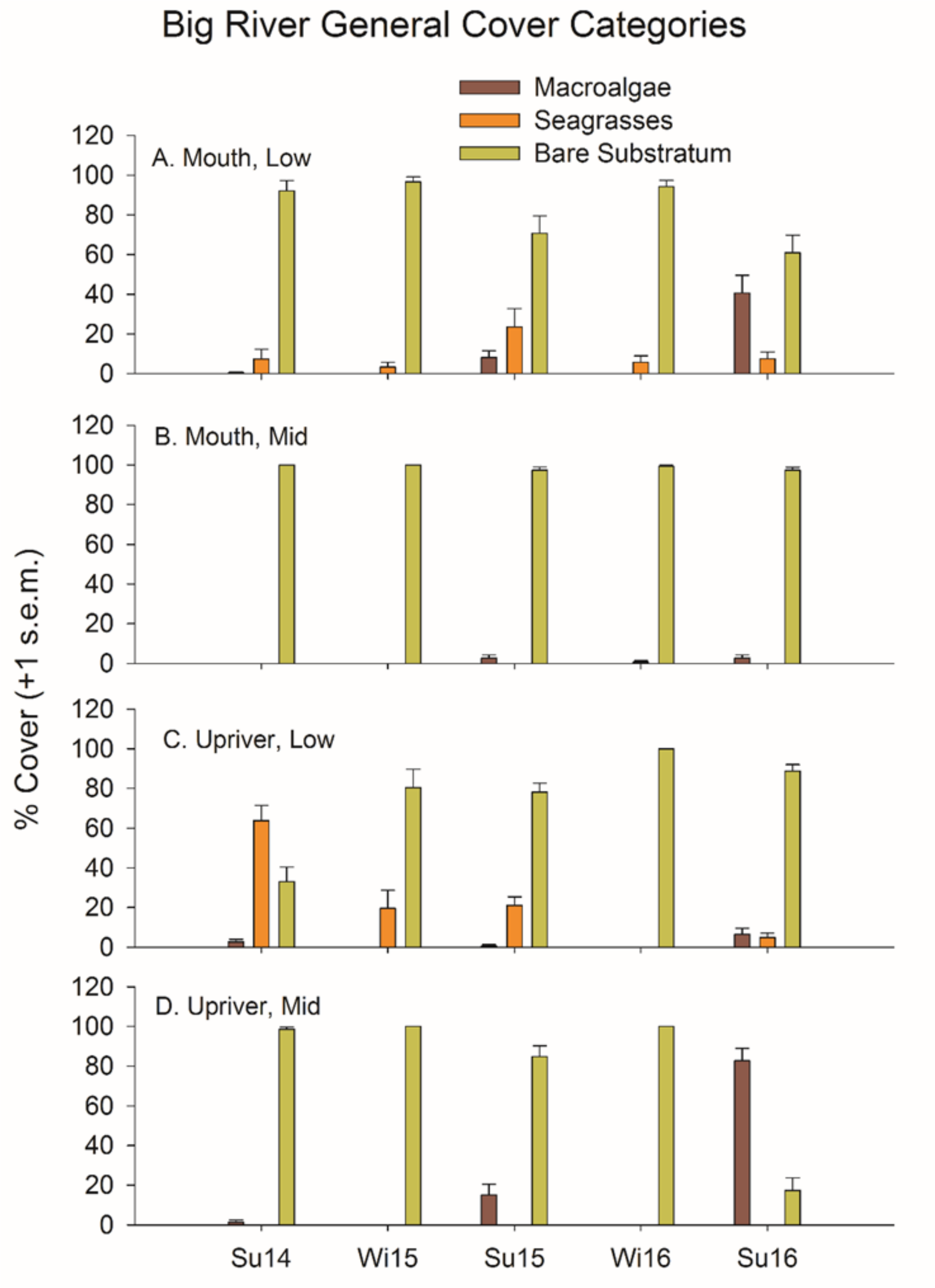


Figure 47. The general cover classes of macroalgae, seagrasses and bare substratum for the mid and low transects within the Big River estuary mouth and upriver sites.

The North and South sites in HB demonstrated the same pattern of *Z. marina* abundance through time (Figure 48). Cover and shoot densities were highest during Su14, and then dropped and stayed low for the next two summers. Since these *Z. marina* permanent transects were at the top edge of the continuous eelgrass when the sites were established in Su14, they may have experienced more heating and desiccation during the later summer and fall of 2014 when the drought conditions occurred. *Z. marina* shoot lengths distributions did not vary as much over time in HB as the % cover and density of the plant (Figure 48). At the upriver site in TM, *Z. marina* did not parallel the HB *Z. marina* (Figure 49). The plant was either absent at TM or sparsely present. The unstable MLLW tidal datum in TM in combination with potentially switching back and forth between lagoon and oceanic conditions may be why *Z. marina* abundance was so low in this estuary. The *Z. marina* at the BR sites demonstrated the temporal pattern seen in HB (Figure 50). The sites surveyed in HB and BR are either oceanic or close to the estuary mouth, and both should have a more stable MLLW tidal datum than TM. The reason for the HB and BR decline in eelgrass abundance during Su15 and Su16 is unclear. The BR eelgrass beds were almost completely subtidal – barely out of the water even when there was a prediction of -1' MLLW on the outer coast. Either winter discharge events, or increasing ocean temperatures, could have stressed the *Z. marina* plants in BR. In contrast, the eelgrass transects monitored in HB (i.e. upper edge of eelgrass bed) were out of the water for most tide cycles. These plants would also have experienced the high oceanic SST values during Su15 (Figure 13), and that signal is evident for the late Su15 SST curve in HB (Figure 18), but it also possible that they were stressed by more desiccation and heating while emerged during Su14 and Su15.

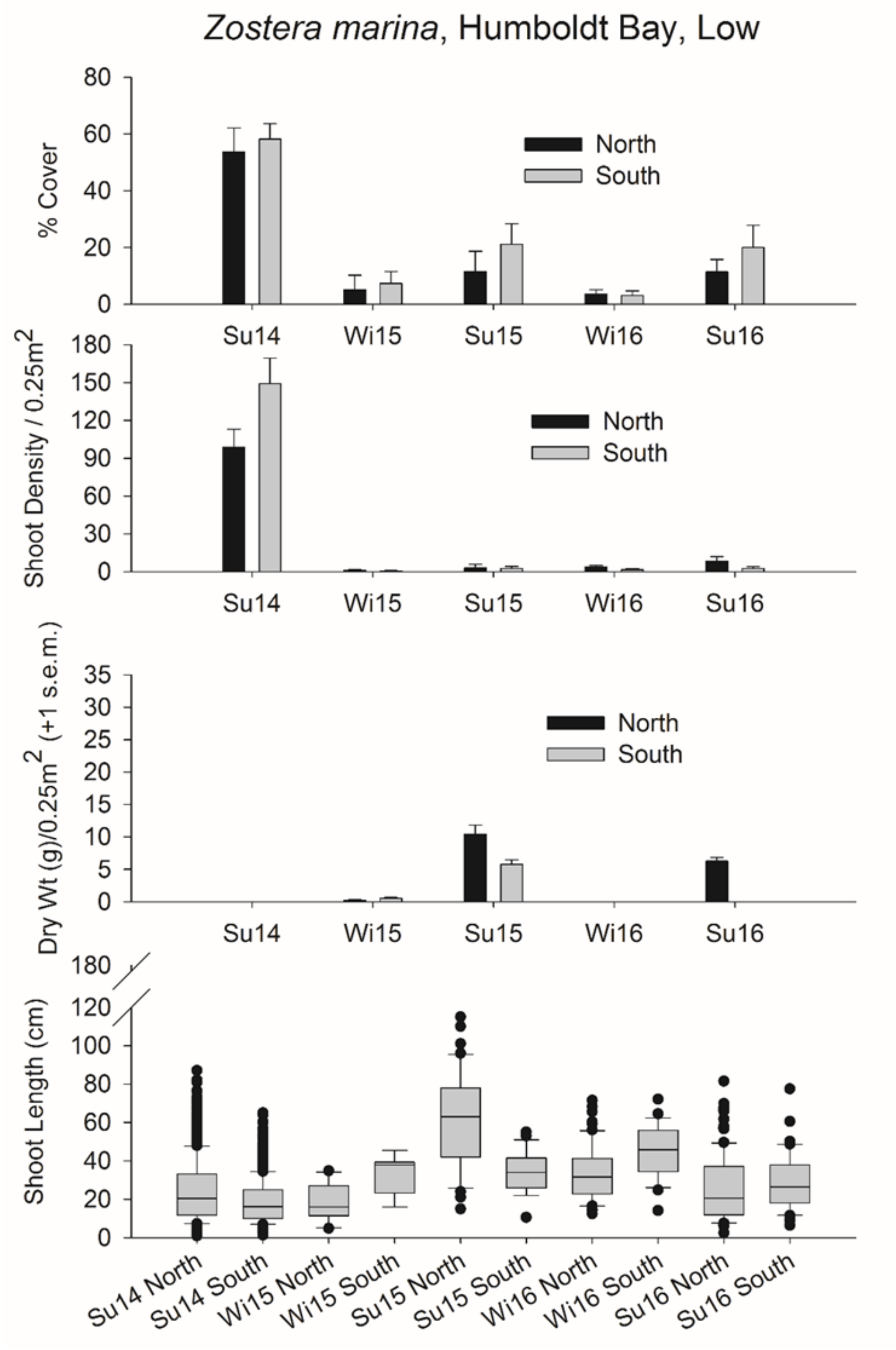


Figure 48. The mean (error bars: + 1 s.e.m.) percent cover, shoot density, above ground dry weight and median shoot lengths of *Zostera marina* (eelgrass) in the Humboldt Bay SMRMA. Dry weights during Su14 were lost.

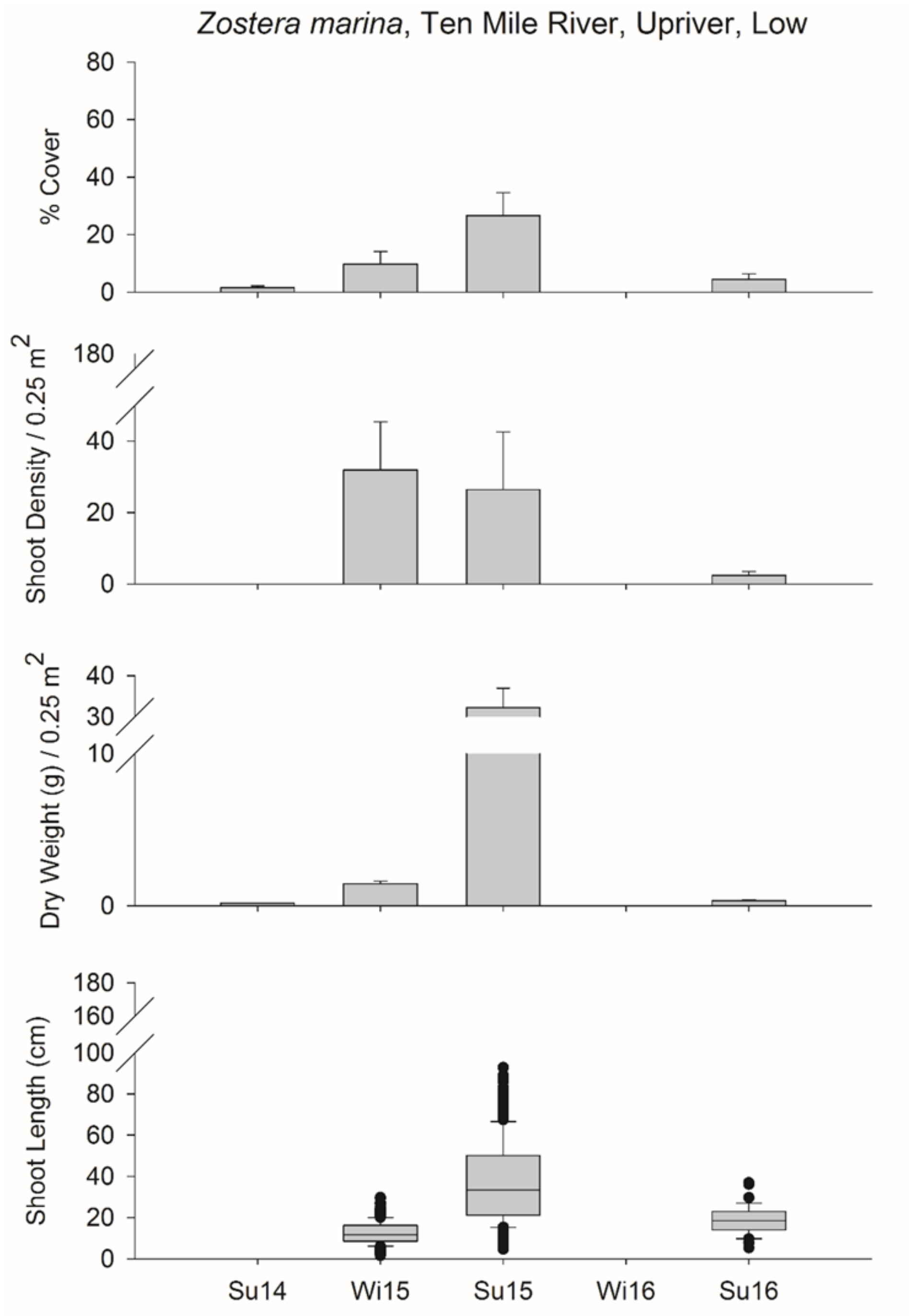


Figure 49. The mean (error bars: +1 s.e.m.) percent cover, shoot density, above ground dry weight and median shoot lengths of *Zostera marina* (eelgrass) in the Ten Mile River SMCA.

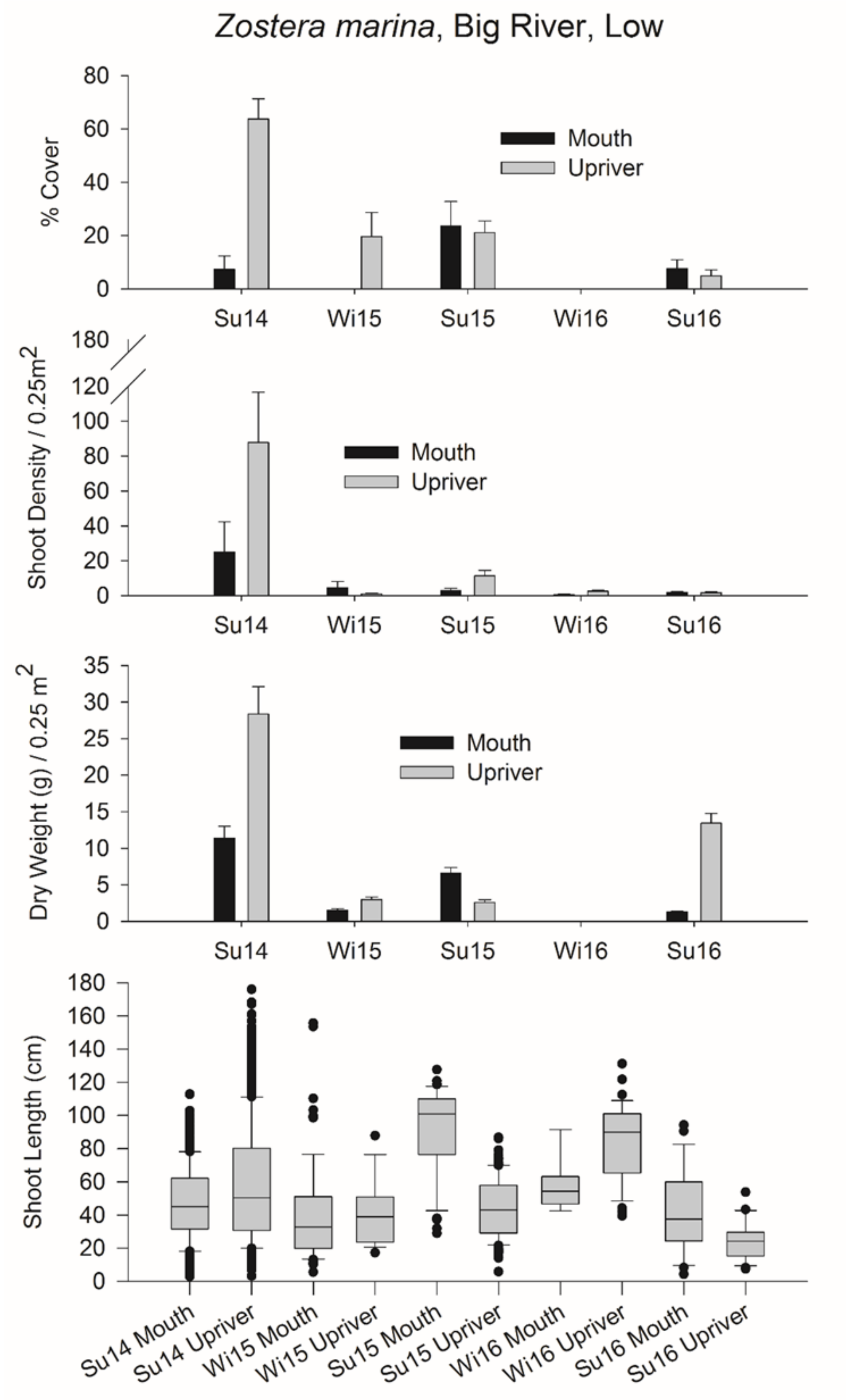


Figure 50. The mean (error bars: +1 s.e.m.) percent cover, shoot density, above ground dry weight and median shoot lengths of *Zostera marina* (eelgrass) in the Big River SMCA.

Z. marina maximum depths, which are strong indicator of estuarine water quality conditions, were sampled in BR but not expressed relative to a BR tidal datum. The water depth at the deep edge of *Z. marina* at both BR sites was recorded during summer 2016; see "figure points" in Table 16. Eelgrass maximum depths taken on 6/9/2016 at both BR sites correspond to the orange circles in Figure 10. The maximum depth of *Z. marina* in BR could only be expressed relative to Arena Cove MLLW. Since the absolute elevation of BR MLLW is likely to be higher than for Arena Cove, the Elevations in Table 16 need to be shifted down. It would be possible to use a depth gage to derive a time offset for the observed tides in Arena Cove, but without a terrestrial datum benchmark from which to reference BR water levels, it is not possible to estimate the vertical adjustment needed to estimate the BR MLLW tidal datum.

Maximum depths of *Z. marin*a did not get sampled in HB due to the lack of an underwater camera for locating the deep edge of the bed. This would still be a good metric of ecosystem health in HB because it is possible to express those depths relative to the MLLW tidal datum. However, this tidal datum in TM, where there is patchy *Z. marina*, turned out to be transitory because of the building and eroding of the barrier beach (Figure 20).

Table 16. Eelgrass maximum depths taken 6/9/2016 at both of the Big River SMCA sites.

Site	Figure point	Latitude	Longitude	Time (Local Time)	Water Depth (m)	Elevation (m Arena Cove MLLW)
Upriver	1	39.303234	-123.773242	9:44	0.14	0.804
	2	39.303267	-123.773164	9:46	0.73	0.214
	3	39.30329	-123.77308	9:47	0.595	0.349
	4	39.303307	-123.77301	9:48	0.85	0.125
	5	39.30332	-123.772922	9:50	0.88	0.126
	6	39.303338	-123.772804	9:51	0.57	0.436
	7	39.303366	-123.772757	9:52	0.87	0.136
	8	39.303363	-123.772658	9:53	0.425	0.581
	9	39.303403	-123.772533	9:54	0.43	0.606
	10	39.303401	-123.772399	9:55	0.45	0.616
	11	39.303434	-123.772311	9:57	0.74	0.326
	12	39.303416	-123.77221	9:58	0.79	0.276
	13	39.30341	-123.772117	9:59	0.81	0.256
	14	39.30336	-123.772022	10:00	0.765	0.313
	15	39.303225	-123.771897	10:02	0.62	0.47
	16	39.303177	-123.771805	10:04	0.6	0.49
	17	39.303228	-123.771671	10:05	0.81	0.28
	18	39.303168	-123.771613	10:07	0.815	0.318
	19	39.303106	-123.771554	10:08	0.82	0.313
	20	39.303065	-123.771462	10:09	0.82	0.313
	21	39.30305	-123.771383	10:11	0.77	0.363
	22	39.303054	-123.771309	10:12	0.935	0.211
	23	39.303019	-123.771228	10:13	0.9	0.259

	24	39.302974	-123.771174	10:14	0.9	0.259
	25	39.302983	-123.771061	10:16	1.12	0.039
	26	39.302937	-123.771012	10:17	1.06	0.099
	27	39.302902	-123.770984	10:18	1	0.1815
Mouth	1	39.303658	-123.781837	8:38	1.1	-0.628
	2	39.303658	-123.781773	8:44	1.21	-0.712
	3	39.303679	-123.7817	8:45	1.15	-0.652
	4	39.303694	-123.781647	8:47	1.035	-0.537
	5	39.303689	-123.78156	8:49	0.82	-0.291
	6	39.303725	-123.7815	8:51	0.89	-0.33
	7	39.303721	-123.781448	8:52	0.83	-0.27
	8	39.303715	-123.781365	8:54	0.45	0.11
	9	39.303689	-123.781294	8:55	0.53	0.052
	10	39.303698	-123.781233	8:56	0.585	0.019
	11	39.303694	-123.78116	8:58	0.585	0.019
	12	39.303718	-123.781107	8:59	0.85	-0.246
	13	39.303705	-123.781043	9:01	0.72	-0.0935
	14	39.303705	-123.780995	9:02	0.805	-0.156
	15	39.303645	-123.780889	9:04	0.48	0.169
	16	39.303633	-123.780789	9:05	0.74	-0.091

The seagrass *R. maritima* had not previously been documented in MR or TM. A known indicator of brackish water (Levings et al. 2002, Mathieson et al. 2009), it was more prevalent on the mid intertidal transects at the upriver sites at TM and MR (Figure 51, Figure 52). In MR, with its thin shoot (i.e. 1 – 2 mm), it also demonstrated extremely high shoot densities. The occurrence of *R. maritima* in both zones at TM may again reflect the unstable MLLW tidal datum, and the metrics showing low abundance of *R. maritima* suggest that it is more physiologically stressed at TM.

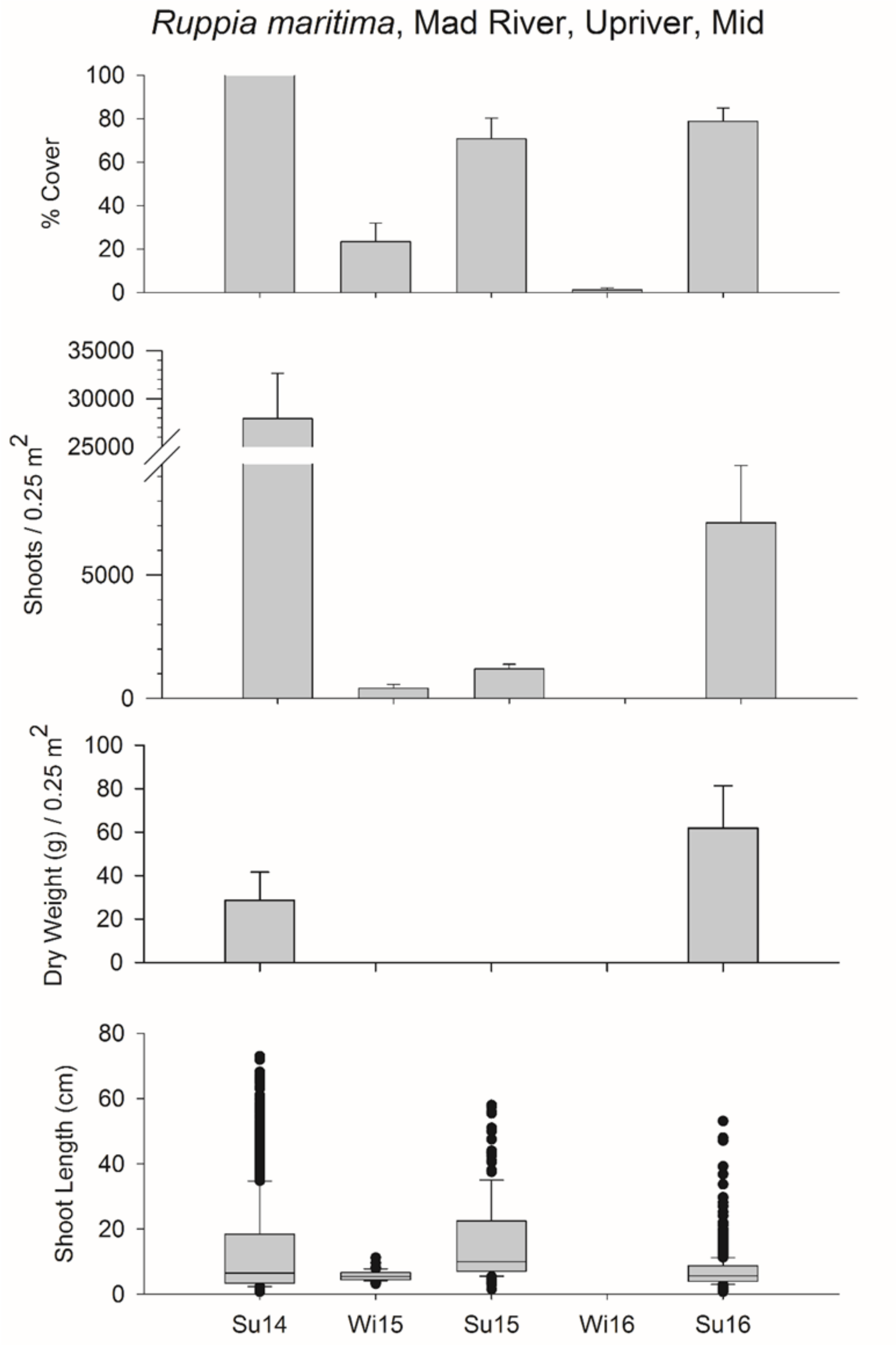


Figure 51. The mean (error bars: +1 s.e.m.) percent cover, shoot density, above ground dry weight and median shoot lengths of *Ruppia maritima* (widgeon grass) in the Mad River Estuary.

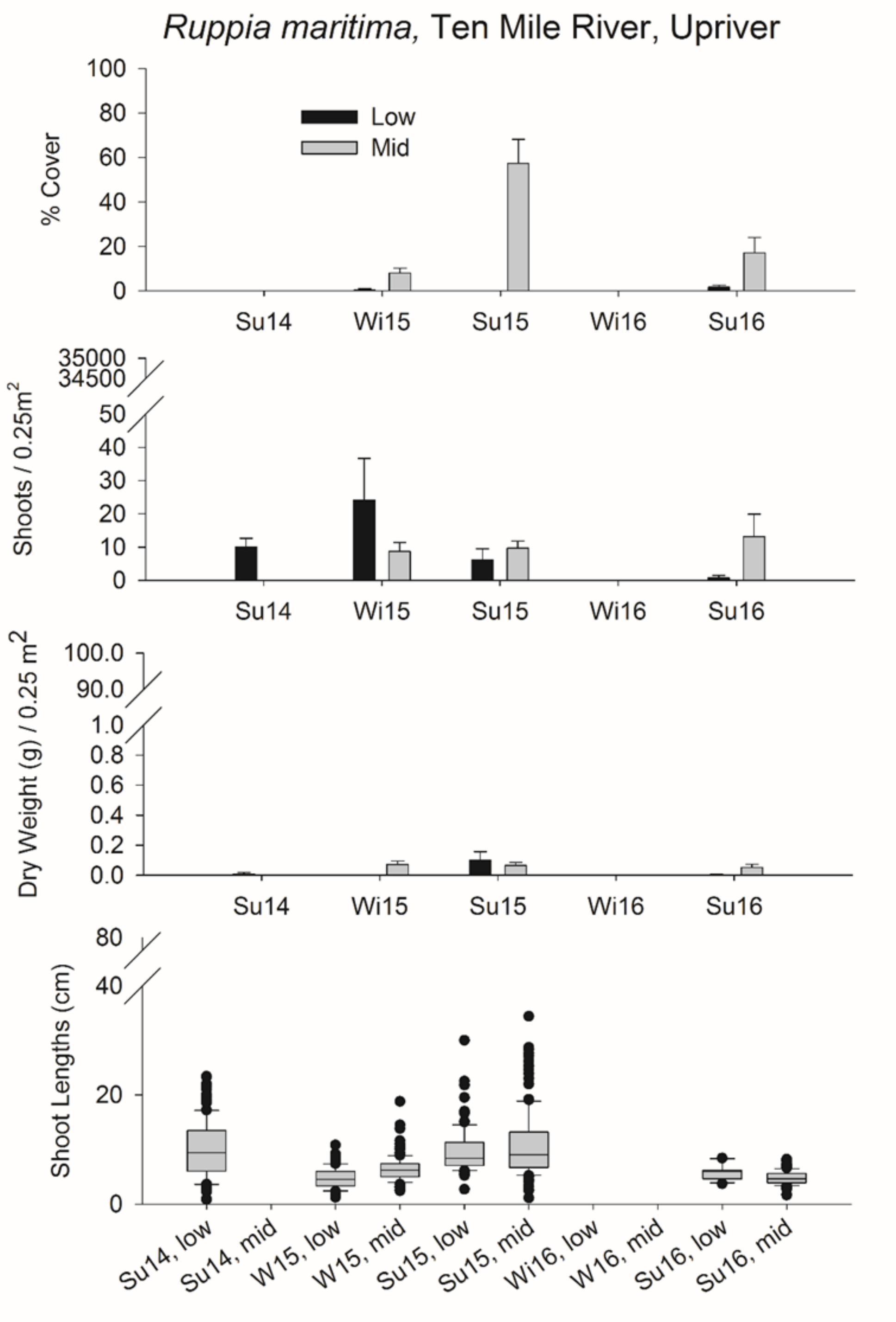


Figure 52. The mean (error bars: +1 s.e.m.) percent cover, shoot density, above ground dry weight and median shoot lengths of *Ruppia maritima* (widgeon grass) in the Ten Mile River SMCA.

Holes of different diameter classes were used to indicate the degree of infaunal presence beyond what could be described using sediment cores. With the exception of the upriver site in MR, which was cobble, gravel and course sand, all of the other sites in the study were either soft mud or some combination of mud and sand. All of these sites with soft bottoms had infaunal communities whose species made holes that were apparent on the surface. Most of these sites had many holes less than 2 mm across, which were possibly made by phoronid worms and a range of corophid and gammarid amphipods. We attempted to quantify these 2 mm holes, but they could only be reliably counted when floating over the quadrat while there was little turbidity. These counts are therefore not presented, but under ideal viewing conditions numbers ranged from 100 – 500 0.25 m^2. In order to identify the occupants of the larger holes, separate excavations were undertaken. These yielded the clam *Macoma nasuta* in HB, the clam *Mya arenaria* at TM and BR, and the larger holes in BR were associated with two other bivalves (i.e. *Cryptomya californica, Limecola balthica*) and the ghost shrimp *Neotrypaea californiensis*. The two estuarine beach sites where the sand was reworked so often – the mouth sites of MR and TM – had the least number of holes, and those site had the smallest diameter class reported (2 – 9 mm; Figure 53, Figure 54). HB, and especially BR, had the greatest number and size of holes indicating a relatively abundant infaunal community of larger bivalves and ghost shrimp (Figure 55, Figure 56).

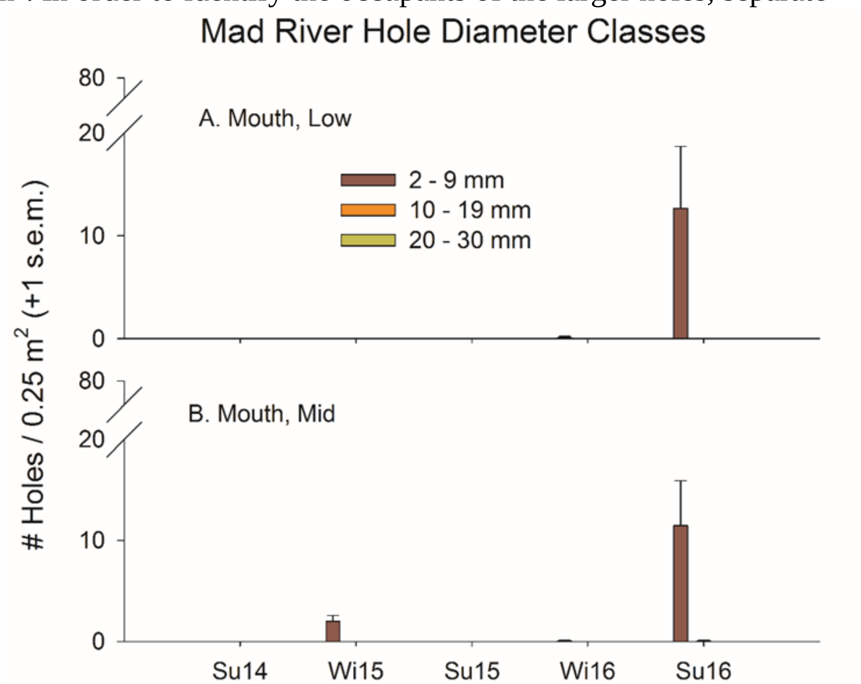


Figure 53. The mean number of holes in the mud substratum for each diameter class in the Mad River Estuary.

Phyllaplysia taylorii, an opisthobranch and mesograzer of *Z. marina* epiphytes, did not occur in HB over the three years of monitoring even though it is found in multiple other *Z. marina* beds in HB (Frimodig 2007). Sampling of *Z. marina* in HB occurred at the upper edge of its distribution which may not be suitable habitat for this animal. However, *P. taylorii* was also not found on the permanently submerged plants of *Z. marina* in TM or BR.

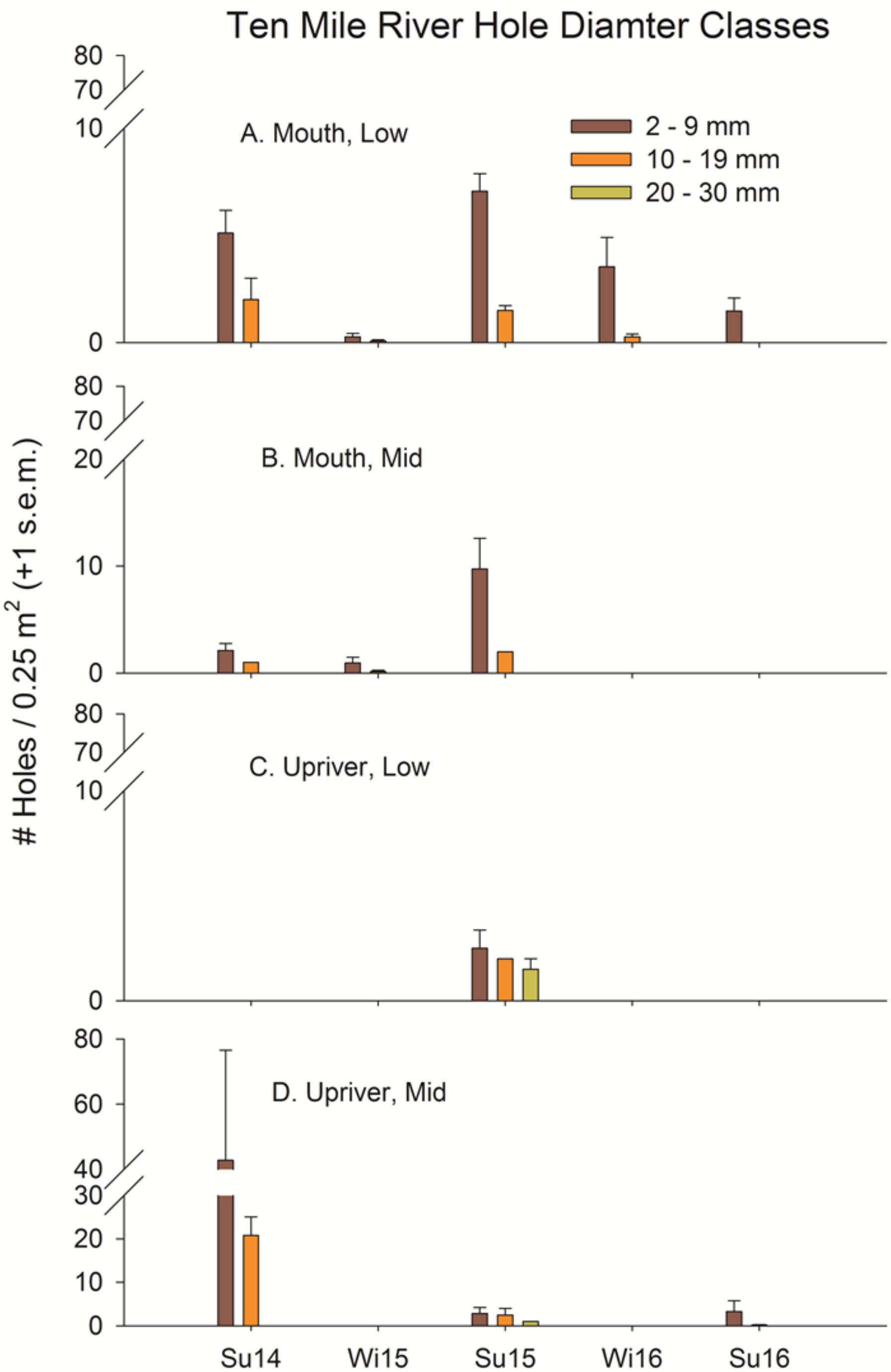


Figure 54. The mean number of holes in the mud substratum for each diameter class in the Ten Mile SMCA.

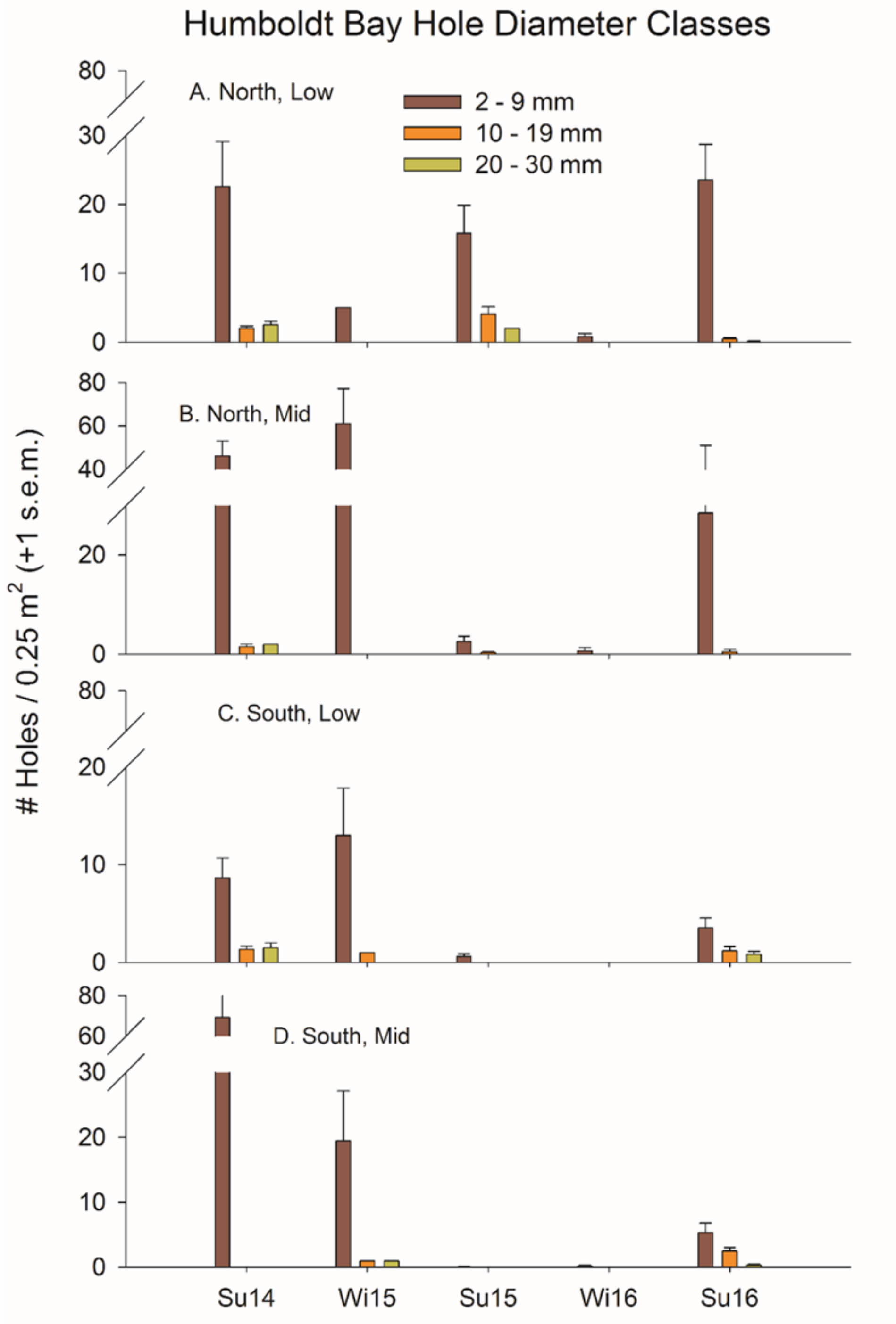


Figure 55. The mean number of holes in the mud substratum for each diameter class in the Humboldt Bay SMRMA.

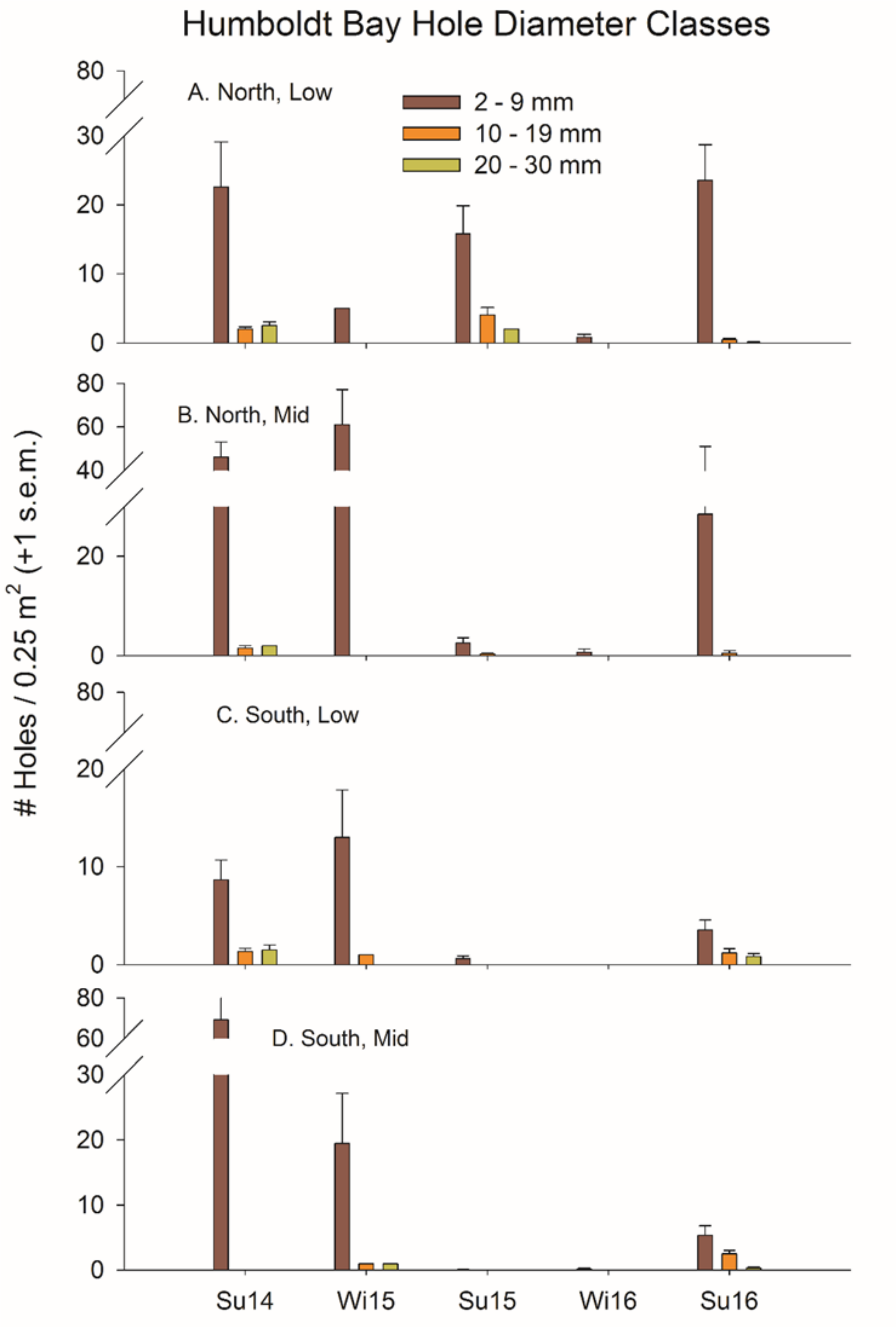


Figure 56. The mean number of holes in the mud substratum for each diameter class in the Big River SMCA.

The distribution through time of the crab *Hemigrapsus oregonensis* was sporadic. It occurred most consistently at sites in HB (Figure 57). A pit trap, as opposed to the box traps with shells used in the present study (ES 1), may have been a more effective way of assessing the abundance of this crab that likely links the seagrass and algal mesograzer community to higher trophic levels (Moksnes et al. 2008, Hughes et al. 2013).

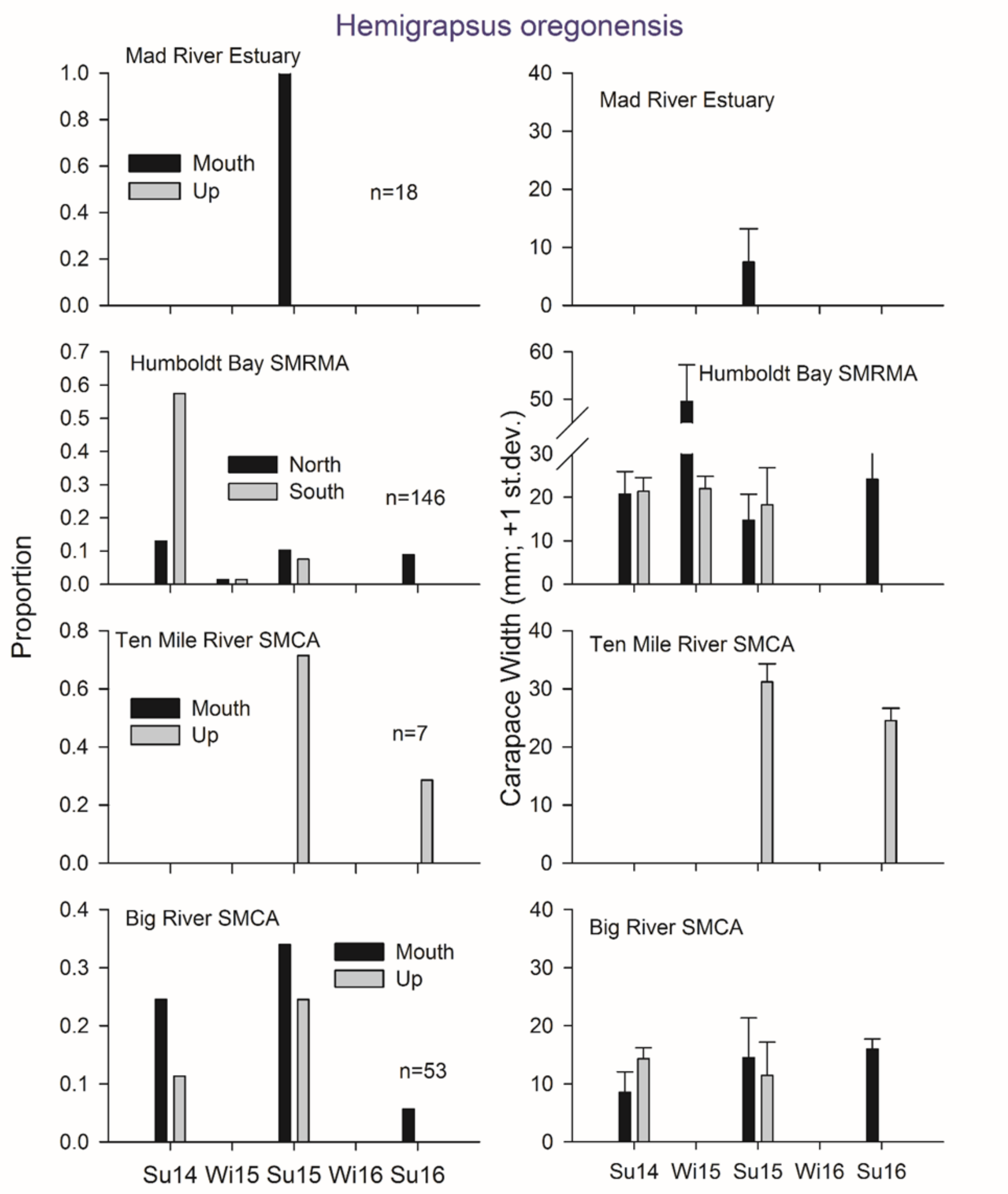


Figure 57. The relative abundance (proportion: # individuals at a site / total # individuals from all sites and times in one estuary; the total # is indicated in the left side panels as "n=") and body length of *Hemigrapsus oregonensis* in each estuary.

Prickly sculpin were relatively more abundant at the upriver sites in MR and TM; body sizes of the TM population were larger than those of MR (Figure 58). Staghorn sculpin spend about the first two years of their life in an estuary before moving to outer coast habitats (Moyle 2002), and so are treated as Estuary Residents in this report. Very few of these fish were present in Su14 relative to Su15 and Su16 (Figure 59). This pattern was evident in all four estuaries, possibly indicating increased recruitment with the setting up of the strong south to north El Niño current during 2015. TM, the estuary with the most spatially and temporally variable physical conditions, also contained the highest number and largest size of staghorn sculpin (Figure 59).

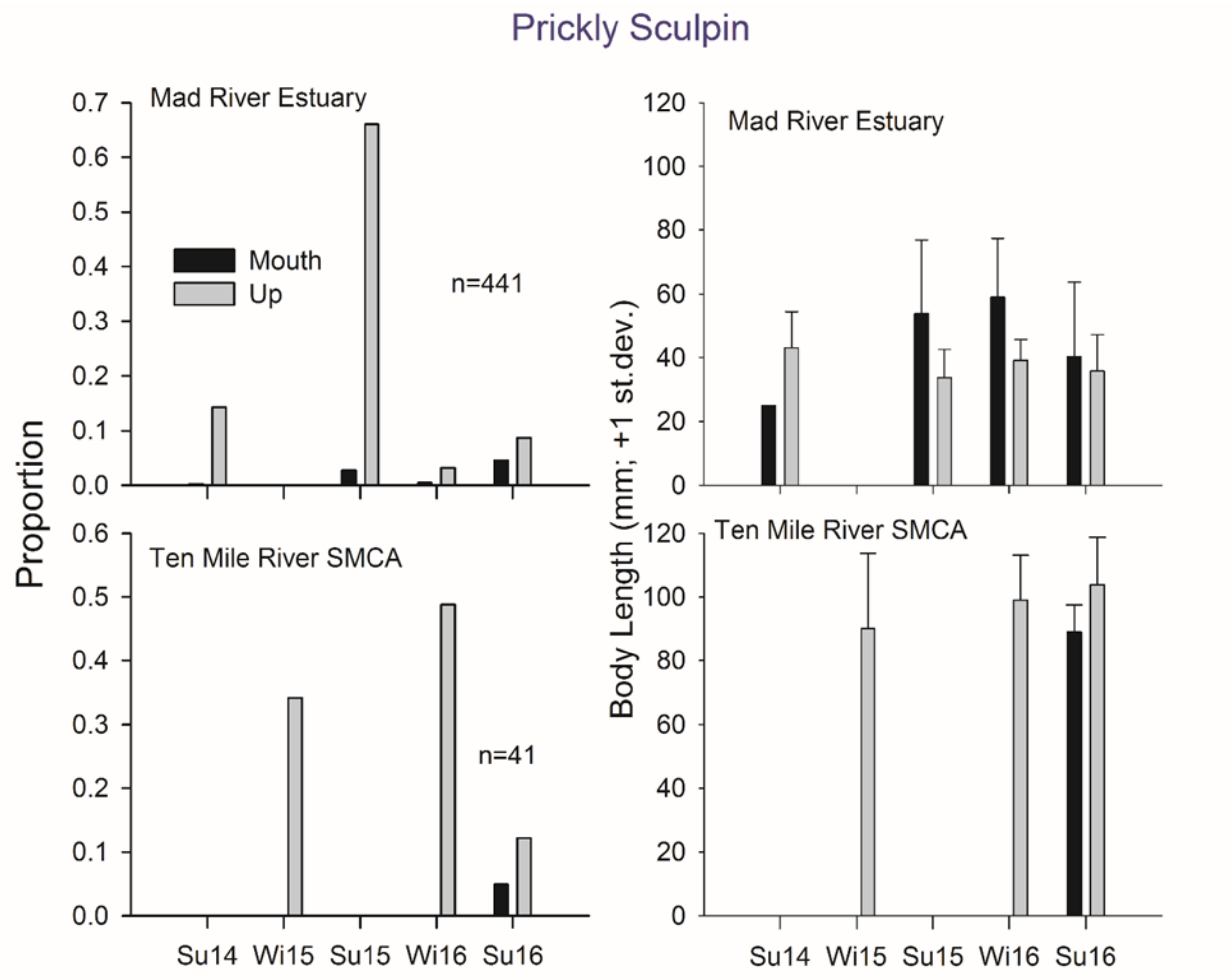


Figure 58. The relative abundance (proportion: # individuals at a site / total # individuals from all sites and times in one estuary; the total # is indicated in the left side panels as “n=”) and body length of prickly sculpin in each estuary.

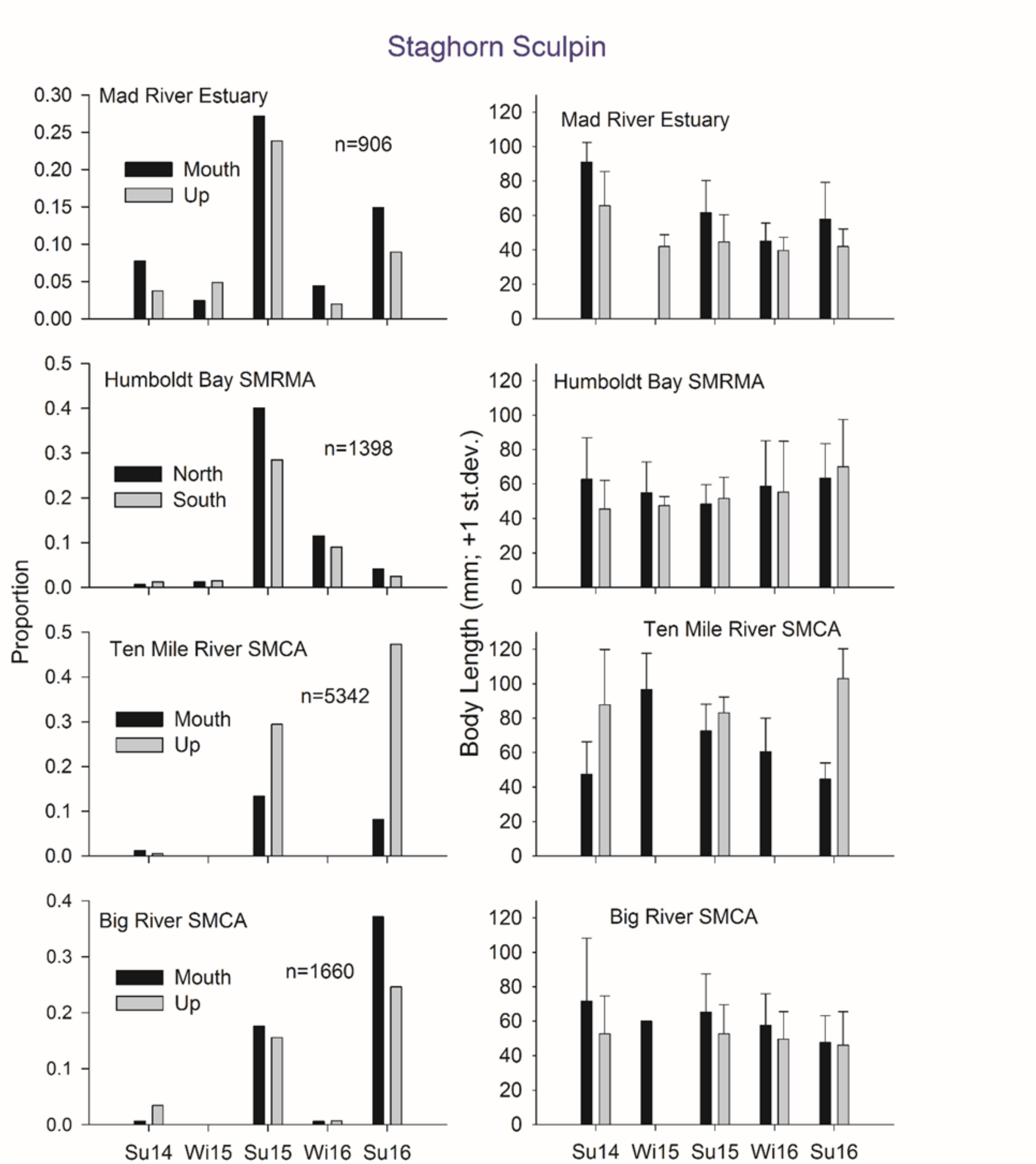


Figure 59. The relative abundance (proportion: # individuals at a site / total # individuals from all sites and times in one estuary; the total # is indicated in the left side panels as "n=") and body length of staghorn sculpin in each estuary.

Two tidewater goby were found in HB North during June 2016 during a flooding tide (Table 8). Although this fish is considered to prefer lower salinity ranges (i.e. brackish), it has been found across a wide range of salinities (Chamberlain 2006). The only tidewater goby previously reported in HB occurred in the Arcata Marsh and on the lee side of HB levees (Chamberlain 2006). This species was also found by Chamberlain (2006) in the Ten Mile River estuary, but our study did not find this species in TM.

In the estuaries north of Cape Mendocino, three-spined stickleback peaked in relative abundance during Su14, whereas below the cape they peaked during Wi16 or Su16 (Figure 60). In the three riverine estuaries, they were more abundant in the up-river sites whereas, within HB, their relative abundances were similar at the North and South sites that should not differ with respect to salinity. Body sizes of the three-spined sticklebacks were similar across all estuaries except for BR where they were consistently shorter (Figure 60).

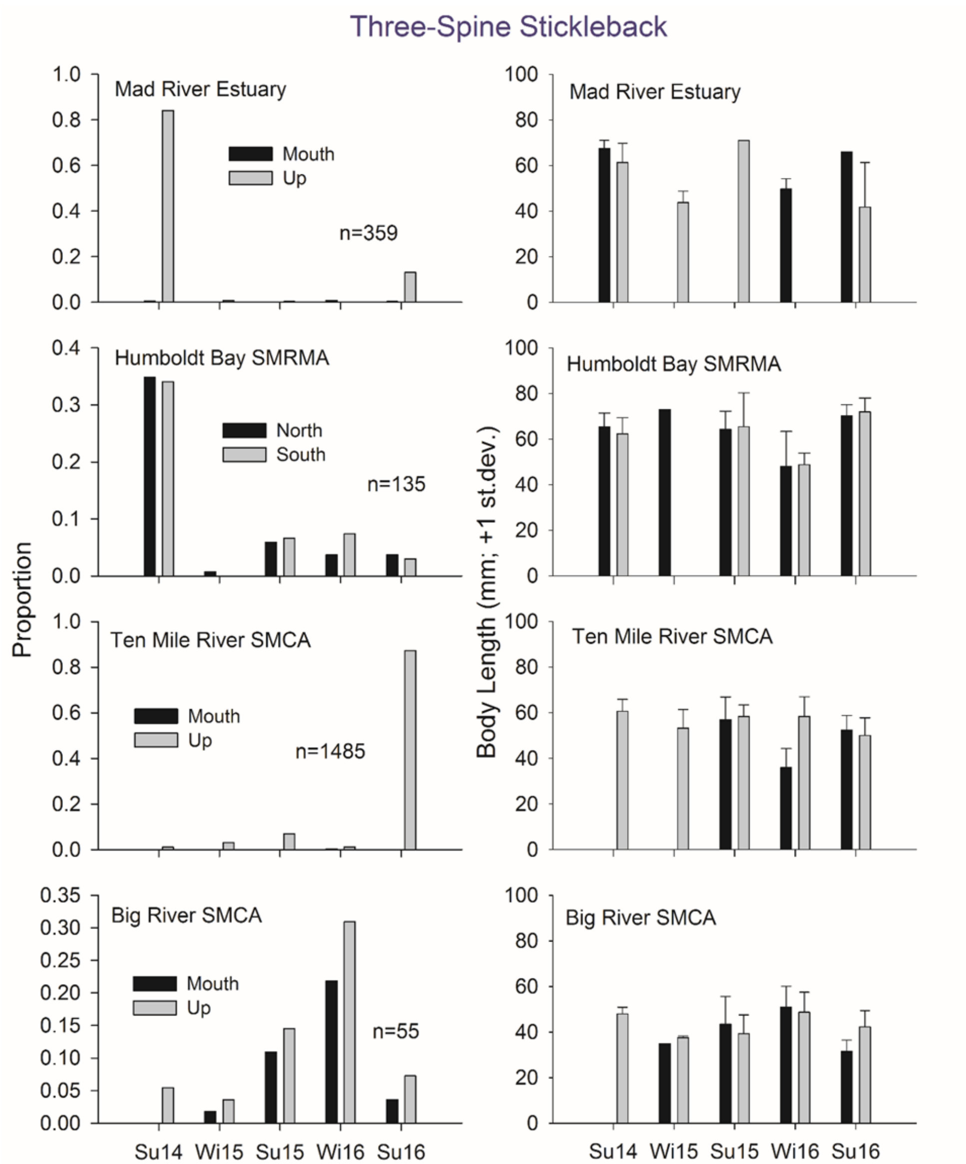


Figure 60. The relative abundance (proportion: # individuals at a site / total # individuals from all sites and times in one estuary; the total # is indicated in the left side panels as "n=") and body length of three-spined stickleback in each estuary.

Anadromous Fish

The presence of coho salmon, chinook salmon and steelhead trout in each estuary is indicated in Table 8. Only the abundance and body size data for coho and chinook are presented here because the sample sizes for steelhead trout were so low.

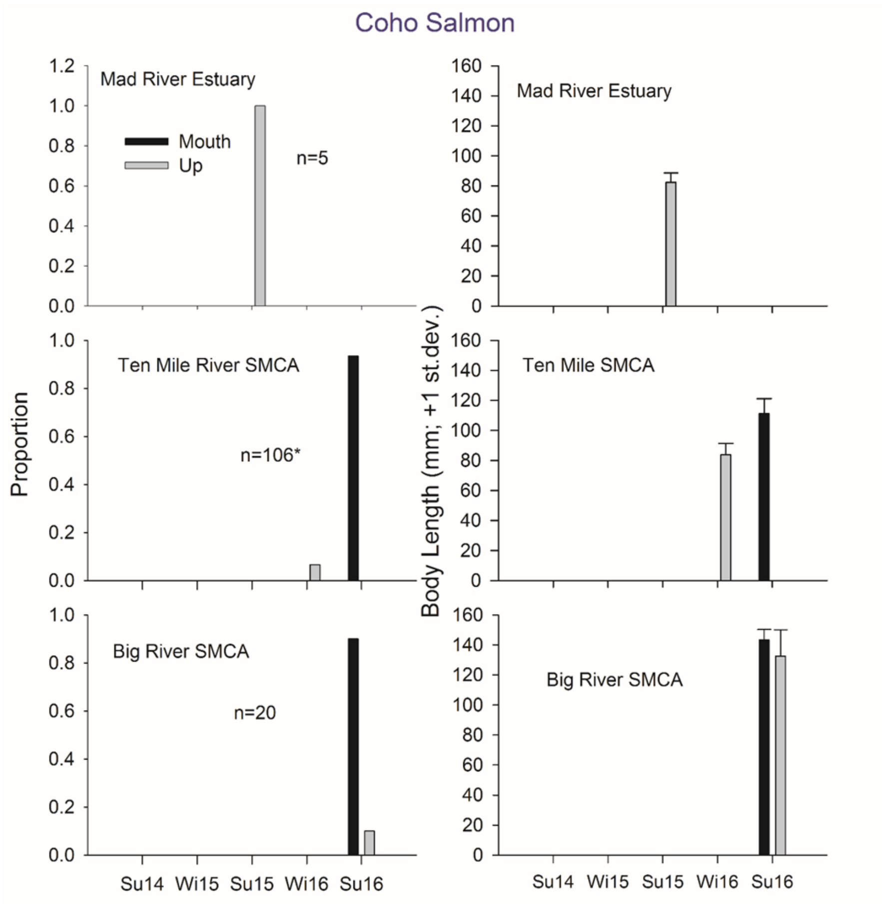


Figure 61. The relative abundance (proportion: # individuals at a site / total # individuals from all sites and times in one estuary; the total # is indicated in the left side panels as "n=") and body length of coho salmon in each estuary.

Coho salmon peaked in relative abundance in MR, TM and BR during either Su15 or Su16, again coincident with the onset of El Niño oceanic and watershed conditions (Figure 61). Very few coho were caught in the up-river site in MR (n=5). TM, the estuary with likely the most variable and extreme physical conditions, had the most coho caught across the entire study (n=106), and this number is lower than it could have been because the federal permit limit was reached during the Su16 sampling event. Coho in TM and BR were almost all located in the mouth site of each estuary.

Chinook salmon were only present during summer (i.e. June) sampling events but, among estuaries, did not have a parallel pattern of year to year relative abundance (Figure 62). They only showed a preference for the mouth site when in TM, possibly due to the lower DO at the upriver site in this estuary (Figure 22).

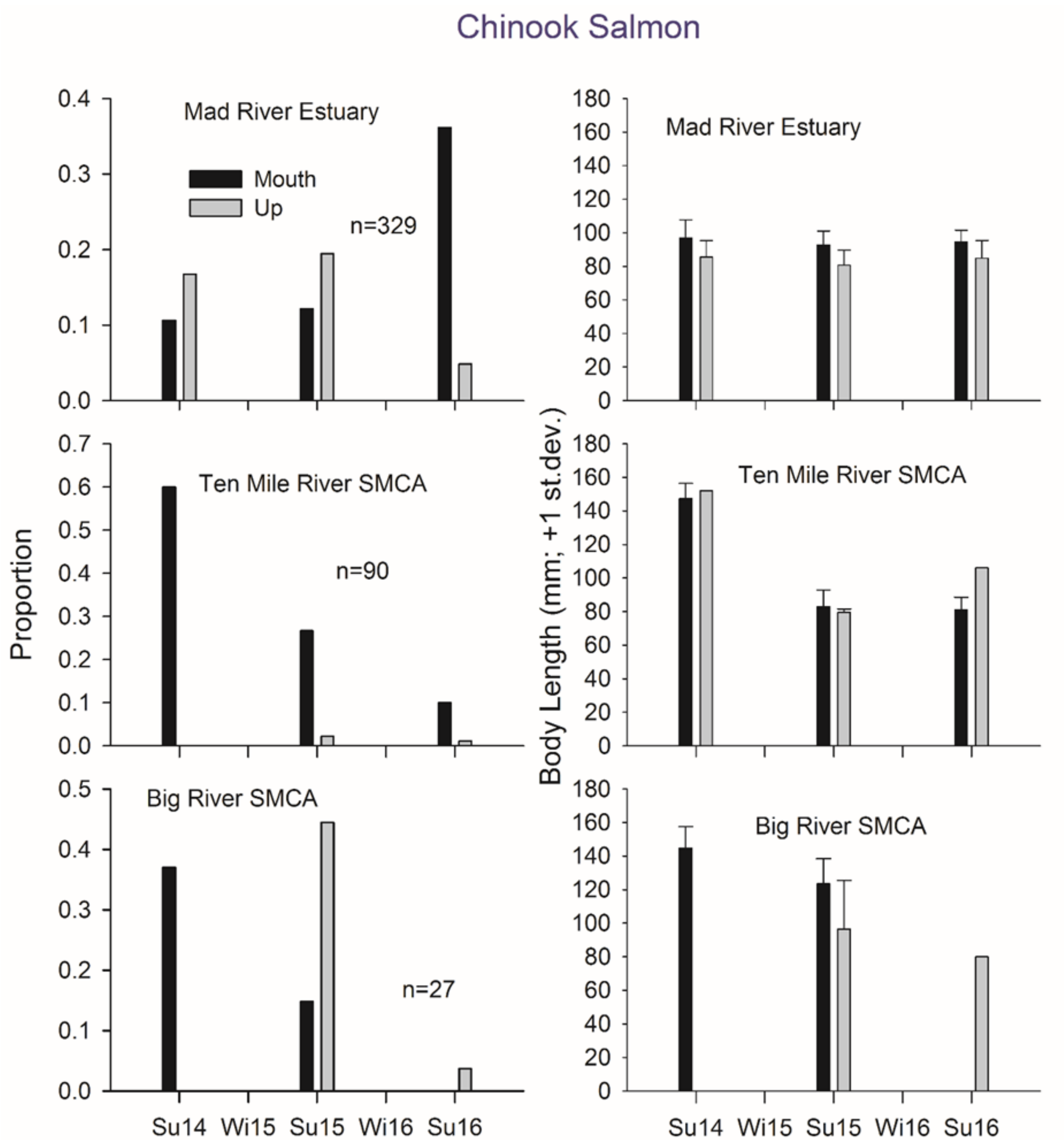


Figure 62. The relative abundance (proportion: # individuals at a site / total # individuals from all sites and times in one estuary; the total # is indicated in the left side panels as "n=") and body length of chinook salmon in each estuary.

Recommendations for the Long-Term Monitoring of Estuaries

1. Decide on the larger goal of the monitoring program. Is it to track community and population bioindicators of estuarine ecosystem health? Should the design also be able to detect the effect of an event happening at a particular site in the future (e.g. MPA regulations, oil spill, fishing/hunting, eutrophication)?

2. If the intention is to test for an estuarine MPA effect, then monitoring should only proceed if several conditions are met: 1) there is a realistic match between the nature of the threat to estuarine health versus the activities that are being regulated in the MPA – i.e. the right tool; 2) there should not be a list of exemptions that make the MPA regulations meaningless – i.e. a broken tool; 3) there is likely to be enforcement of disallowed MPA activities.

3. For detecting an MPA effect, the organisms monitored should have a high life history affinity to the MPA site. This means that marine mammals as well as most birds and fish like salmonids and rockfish juveniles are poor target species because fluctuations in their abundance and size are due to environmental conditions spanning much broader spatial scales than a small estuarine MPA. MPAs on the outer coast have affected the number and size of particular outer coast fish species (Hilborn et al. 2004), but these affects more often apply to species that, upon becoming larger adults, also switch to having high site fidelity – e.g. adult rockfish and lingcod. This recommendation for detecting an MPA effect is potentially at odds with the fact that estuaries are critical habitat for these organisms, and that state and federal agencies as well as the public have policy, economic and recreational reasons for being interested in them. There are separate justifications for monitoring these higher trophic levels in estuaries, but testing highly mobile species for MPA effects will be more challenging than focusing on more permanent residents of the estuary.

4. If an MPA and reference site design is justified, then the experience from the present baseline study is that reference sites should occur within the same estuary as a paired design rather than using separate reference estuaries. The list and extent of covariate differences among estuaries due to geomorphology, hydrology and anthropogenic uses is so extensive that statistically controlling for them is not feasible, even if those covariates could be quantified. Similar challenges will exist for a paired design within the same estuary, but the degree of environmental differences between paired sites should be less than for among estuaries. A paired design will be more possible in larger than smaller estuaries. For example, an almost unchanging horizontal gradient of salinity in HB allows for southern HB to also hold a reference site, as does the long, linear estuary of BR. In a small estuary where most of the estuary is within the MPA, such as TM, paired sites are more likely to occur at different points along steeper estuarine gradients.

5. It is critical that long-term monitoring of estuaries funds the purchase of simple data recorders for describing the physical context *within an estuary*. Continuous measurements of temperature and salinity in each of the estuaries used in the present study would have accomplished the following:
   a. The physical environments of each estuary could have been more easily compared.
   b. The strength of the connection between the estuary and the ocean, and the estuary and the watershed, could have been assessed more rigorously.
   c. The relationship between the physical context and biota in each estuary could have been more rigorously described and the biological responses more clearly interpreted.
   d. If the present study had included a reference site to test for an MPA effect, or if a before-after comparison of the same estuaries was undertaken, then physical measurements of temperature and salinity would need to be among the covariates used to either statistically control for those differences among sites or times, and/or those covariates would identify drivers of biological variation that could be more important than management actions.

6. Given limited funding and the monitoring goal to detect a site-specific effect in a North Coast estuary, and based on the data and experiences from the present study, the following features of a long-term estuarine study are recommended:
   a. Focus on target species and not biodiversity descriptions. For the estuaries in the present study, a subset of **Estuary Residents** should be selected. From low to higher trophic levels, the following variables can be rapidly and accurately enumerated: algal cover, seagrass abundance (as cover, shoot density), cover of seagrass leaf lesions, densities of infaunal hole diameter classes, number and size of *H. oregonensis* crabs assessed with unbaited pit traps (i.e. the cover provided by these traps attracts this crab and protects it from intense predation) rather than box traps, number and size of prickly and staghorn sculpins, and three-spined stickleback; all fish assessed with beach seines. *M. magiste*r and *C. productus* crabs are not recommended for monitoring because their life histories span multiple habitats and, even when present, abundance data are unreliable due to trap vandalism and predation from a variety of animals.

   b. Include target species from salt marsh, mudflat and seagrass habitats. Salt marshes are included in the estuarine MPAs. Most of the salt marsh area in the world, and California, has been lost to development (Gedan et al. 2009). This is also the habitat that is most immediately being affected by sea level rise since anthropogenic barriers are partly responsible for preventing the upslope migration of estuarine habitats.

c. Shift the sampling times from once in January and once in June (present study), to June and late summer. These sample times match when the suggested target species are peaking in abundance and growth.

d. In each site, continuously measure salinity and water temperature, and place temperature sensors in the mid intertidal mudflat and high salt marsh to describe the air-substratum temperature environment when the habitat is emerged. The exceptional oceanic and drought conditions that occurred during this study suggest that not all of the stresses experienced by the estuarine biota occur when the organisms are in the water (e.g. the yearly decrease in *Z. marina* in HB).

e. Remote sensing with unmanned aerial vehicles (UAVs) should be used every three years to map the migration of habitat boundaries, which are changing in response to climate drivers (Shaughnessy et al. 2012), and could change in response to management actions. Data on the abundance of target species (see *a*) should be used to ground truth the UAV imagery.

   Flying at 65m with cm^2 resolution, at far below the flying and post-processing costs of traditional remote sensing, the UAV technology can distinguish between plants (e.g. eelgrass versus green algae) and map habitat boundaries. Many of the estuarine MPAs are small. Even so, a decision could be made to only fly a portion of an MPA in order to track a particular eelgrass bed or salt marsh. Other types of remote sensing fail on the North Coast because 1) the imagery is not gathered during a low enough tide, which is necessary because the high turbidity of the water prevents sensors from ‘seeing’ into the water, 2) the resolution is too course for delineating plant types and habitat boundaries, 3) coastal fog and clouds degrade the imagery of high flying platforms – there are very few days on the North Coast when both the sky is clear and the tide is very low. UAVs provide more flexibility in the timing of the flight along with imagery that is useful for monitoring habitats and individual species.

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