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UNIVERSITY OF SAN DIEGO

San Diego

Assessing Ecosystem Health through Contaminants in the
Tijuana River National Estuarine Research Reserve

A thesis submitted in partial satisfaction of the
requirements for the degree of

Master of Science in Environmental and Ocean Sciences

by

Nancy Torres

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2023

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San Diego

2023

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ABSTRACT

Although the Tijuana River Estuary (TRE) remains the largest, most-intact coastal wetland in Southern California, it has a history of major changes, much of this related to its location immediately north of the US / Mexico Border. One of the primary challenges is cross-border flows from the rapidly growing city of Tijuana, Baja California, Mexico, and the delivery of wastewater, debris, and sediment to sensitive coastal wetland ecosystems. There is a need to more fully investigate these environmental changes to assess the ecosystem health of the Tijuana River Estuary over time, especially related to pollution impacts. This can inform an understanding of changes in both species and stressors, and can also help assess the effectiveness of past management strategies. Since 1986, the NOAA Mussel Watch and California Surface Water Ambient Monitoring Programs have periodically collected data on chemical contaminants and biological indicators of water quality in the TRE. This project builds on these past monitoring efforts and established methodologies to assess status and trends of contaminants in sediment and organisms. This work was accomplished by conducting a thorough review of available datasets and literature to document past changes in the estuary and refine sampling approaches. Sampling was conducted at three locations in the Tijuana River Estuary to assess spatial variability. Compared to national thresholds, most contaminant concentrations were at relatively low levels in 2021, with some having decreased from previously higher levels in the 90s. When comparing species groups, fishes had the highest organic concentrations, indicating the ongoing processes of bioaccumulation and biomagnification. Levels of some organics in fish, such as total DDT and PBDEs,

remain at levels which could be of some concern for sensitive piscivorous birds. Also, the highest concentrations were near a local urban outfall point rather than from the Tijuana River itself. Overall, this information improves our ability to document and interpret long-term trajectories of contaminant change in the ambient environment and key taxa. This project's results include communication and management tools depicting the estuary's ecosystem health over time.

Chapter 1 Introduction

1.1 Coastal Wetlands and the Tijuana River Estuary

Embodying the transition zone between fresh and saltwater as well as between terrestrial and aquatic environments, estuarine wetlands play a unique role as dynamic ecosystems that connect watersheds with the ocean. These coastal wetlands provide a wide array of critical ecosystem services, including nursery and breeding habitat for wildlife, natural filtration and detoxification services via suspension feeders, and protection against erosion, flooding, and storm events (Barbier, 2013). Natural wetlands have been often referred to as the “Kidneys of the Earth” due to their capacity to absorb carbon dioxide and reduce pollution (both within the wetland and its surrounding areas) with coastal wetlands, in particular, sequestering and storing carbon up to 55 times faster than tropical rain forests (UN, 2023). Wetlands thus help combat climate change by acting as carbon sinks. Despite their ecological importance on a global scale, wetlands have been disappearing at a concerning rate, with a global loss in habitat of about 25% since the 1940s (Davidson, 2014). Consequently, major ecological and economic losses have ensued, such as an annual global loss of \$7.2 trillion (USD) in ecosystem services from 1997 to 2011 and 0.1 megatons of carbon dioxide released per square kilometer of soil lost (Costanza et al., 2014; Crooks et al., 2011). Coastal wetlands that reside in urbanized watersheds are particularly vulnerable to loss and disturbance, and those that remain in these settings represent opportunities for targeted management and conservation, ideally informed by research and monitoring activities.

The heavily urbanized Tijuana River Watershed spans approximately 1,750 square miles on both sides of the US-Mexico border and is home to a total of ~2,800,000 residents (*Project Clean Water Tijuana Watershed*). The Tijuana River runs through the city of Tijuana before entering the Tijuana River Estuary (TRE) at the mouth of the watershed, which is the location of the Tijuana River National Estuarine Research Reserve (TRNERR), a federal-state partnership aimed at using science to inform decision-making, education, and outreach (Zedler et al. 1992). Contamination from runoff and sewage pollution has long been a concern in this area, both for the health of the ecosystem as well as that of the surrounding communities, such as poor water quality necessitating regular beach closures and leading to public health issues from contamination for both wildlife and people. The Tijuana River Estuary is also critical wildlife habitat as it is along the Pacific Flyway, used as migration and wintering habitat by a variety of waterfowl and shore bird species. The area harbors over 370 species of birds, including threatened and endangered species such as the light-footed Ridgway's rail (*Rallus obsoletus levipes*), California least tern (*Sternula antillarum browni*), western snowy plover (*Charadrius alexandrinus nivosus*), and least Bell's vireo (*Vireo bellii pusillus*) (Safran et al., 2017). As a means of assessing estuarine health, contaminant monitoring has often focused on sediment, bivalves, crustaceans, and fish as sentinels, with implications for the other species, such as birds, as well as ecosystem health more generally (Burger and Gochfeld, 2001; Dodder et al., 2012; Elliott and Elliott, 2013; Zeng et al., 2005). Characterizing contaminants in the Tijuana River Estuary creates knowledge needed for the

preservation of its ecosystem services and best management of the area, improving the surrounding communities' health.

1.2 Project Overview

This project involves sampling for contaminants in the estuary's aquatic biota and sediment. This is coupled with the synthesis and analyses of historical data to provide insight into the current status of the ecosystem's health and how it has changed over time. The outcomes of this project are an improved ability to document and interpret long-term trajectories of change of contaminants in the ambient environment and key taxa within the Tijuana River Estuary. This provides perspective on the effectiveness of past management strategies, such as the initiation of wastewater treatment, and also sets new benchmarks with which to compare to future conditions. This project also aids local resource managers, including those at TRNERR, California State Parks, and the U.S. Fish and Wildlife Service, by assessing contaminants in species important for food chain support of endangered and sensitive species (e.g., marsh birds such as Ridgway's rails). Finally, the results of this project will be used to identify data gaps and research needs, which can then be used to help guide future scientific efforts at TRNERR.

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Chapter 2 Assessing Ecosystem Health Through Contaminants in the Tijuana River Estuary

2.1 Introduction

The Tijuana River Watershed is a binational basin that spans Mexico and the United States, with almost three-quarters of the watershed in Mexico. Virtually all of the flows from the upper watershed pass through the city of Tijuana before entering the United States (Wakida et al., 2008). Due to year-round inputs of untreated wastewaters and discharges of partially treated wastewater from treatment plants in Mexico, the naturally ephemeral streams of this Mediterranean-climate watershed became largely perennial by the 1980's. These continuous flows lead to poor water quality downstream, including the presence of heavy metals (Wakida et al., 2008). In 1999, an international wastewater plant was constructed to capture low flows occurring during dry periods. While this has decreased flows, especially during the summer, transboundary flows that overwhelm this system, typically associated with rainfall or infrastructure failures, are still commonplace (Kargl, 2019). There is currently substantial interest in treating higher volumes of water, to include flows associated with rainfall events (EPA, 2020).

The Tijuana River Estuary (TRE), at the terminus of this binational watershed, sits directly north of the US-Mexico border and has a dynamic history of major ecosystem changes. This includes the cross-border water flows that transport pollution, debris, and sediment (Safran et al., 2017). Despite these stressors, much of the lower river valley is still relatively undeveloped and publicly-owned, and the area continues to be a highly productive ecosystem that

supports a diverse range of wildlife (Safran et al., 2017). The tidal salt marshes are habitats of keen conservation interest, supporting a variety of sensitive and endangered species (e.g., marsh birds such as Light-footed Ridgway's Rail).

Both inorganic and organic contaminants have been found in various places in the watershed, with the latter most likely responsible for in-stream toxicity along the river in Mexico (Gersberg et al., 2004). The flows of contaminated waters into the valley can lead to the accumulation of pollutants in wetland sediments (Becker et al., 2001), and metal concentrations in the top 3 cm were found to have increased approximately four-fold from 1989 to 1997 (Meyer and Gersberg, 1997). A compilation of data on inorganic concentrations in the area's sediment from recent decades was recently developed and offers a solid foundation to build upon (Berry, 2019). However, there has yet to be a comprehensive study examining both inorganics and organics throughout the years to assess how contamination has changed. To inform how to best move forward in managing this ecosystem, we need to understand the Tijuana River Estuary's biotic and abiotic condition through time, particularly related to pollution.

Specific contaminants across both inorganics (heavy metals) and organics have been consistently featured in various monitoring reports and studies. Metals occur naturally in the environment but can be driven to excessive releases due to anthropogenic activity such as fossil fuel and waste burning, mining and ore processing, and chemical production. In the 1990s, the U.S. Fish and Wildlife Service found cadmium, chromium, copper, lead, mercury, and nickel to be

within acceptable threshold levels for inorganics in sediment and fish collected from the Tijuana Slough National Wildlife Refuge (King and Roberts, 2000). Zinc was consistently found over the years to have elevated concentrations, which can impact the growth, reproduction, and survival of fish (Eisler, 1993). Organics, including butyltins, chlordanes, DDTs, dieldrins, PAHs, and PCBs, have been identified by the Mussel Watch Program as contaminants of interest with substantial data (Kimbrough and Lauenstein, 2007). These chemicals are mostly manufactured and can be released in the environment via pesticide use, manufacturing, or disposal processes. PAHs occur both naturally and anthropogenically. DDT is of particular interest due to its known deleterious effect on bird populations from egg shell thinning and possible risk of cancer for humans (Turusov et al., 2002). DDT was found by the U.S. Fish and Wildlife Service to be equal to sediment thresholds which at most would indicate a moderate level of anticipated effects (King and Roberts, 2000). PFCs and PBDEs have been added to the list of Contaminants of Emerging Concern, marking them as chemicals not currently in routine monitoring but may be candidates for future regulation due to potential harmful effects on aquatic life.

One of the primary management concerns related to polluted flows is bioaccumulation in resident organisms. Target species have been previously utilized to assess regional toxicity. Pioneered by Edward D. Goldberg's idea to advance coastal pollution monitoring, bivalve mollusks have proved to serve as effective bioindicators for contaminants because they accumulate metals without suffering excessive mortality and are relatively easy to sample (Boening, 1999;

Farrington et al., 2016). In recent decades, various studies around the world have begun to utilize mussels to monitor organic contaminants as well (Arias et al., 2009; Benali et al., 2017). Since 1986, the NOAA Mussel Watch Program (MWP) has recorded data on the status and trends of chemical contaminants and biological indicators of water quality at the national and regional scale, including some limited sampling in the Tijuana Estuary. Accumulation of contaminants in fish and crabs is also of interest, both because they can serve as bioindicators and because of their role as food chain support for wildlife such as birds (Clatterbuck et al., 2018). In addition to the MWP, there has been periodic sampling of contaminants in fish and shellfish in the Tijuana Estuary as part of the state's Surface Water Ambient Monitoring Program (SWAMP). However, MWP and SWAMP data have not been compiled and analyzed to document long-term trends specific to the Tijuana River Estuary.

Project Goals

For this study, I conducted a suite of sampling for contaminants in the Tijuana River Estuary's sediment and target biota. This was coupled with a synthesis and analyses of historical contaminants data to provide insight into how the system has changed over a time period that has been characterized by various levels of inputs and management interventions, such as treatment plant construction. The current (2021) assessment of condition included consideration of spatial and seasonal contamination in the Tijuana River Estuary (TRE), both to address possible differences associated with locations within the estuary and because large differences in toxicity have been previously observed between the

wet- and dry-seasons elsewhere in the Tijuana River (Riveles and Gersberg, 1999). Contamination levels were also compared across a suite of resident organisms in order to assess possible biomagnification, which has been shown to be evident for major contaminant types throughout the food web in nearby San Diego Bay (Bay et al., 2016). Related to this, various organisms with different life histories (e.g., benthic vs. water column species) and feeding modes (i.e., surface feeders, filter feeders, etc.) provides insight into the level of bioaccumulation that the TRE's wildlife is experiencing.

This study will improve our ability to document and interpret long-term trajectories of change in contaminants in both the ambient environment and in key taxa. It will also establish new baselines against which to compare future changes, and help steer future management action, such as the planned upgrading of sewage treatment infrastructure to treat higher flows (Kargl, 2019). This study also will be used to identify data gaps and research needs, which can then be used to help guide future scientific efforts at the TRNERR.

2.2 Methods

This study consisted of two main components: the synthesis of existing data and the collection of new data on contaminants. Sampling for new data was based on established methodologies and coupled with historical data to result in a comprehensive report of contaminant levels within the Tijuana River Estuary through time.

Synthesis of Existing Data

To document long-term trends in the Tijuana Estuary, historical data on levels of contaminants found in sediment, water, and biota was gathered and compiled into a comprehensive database. This collection focused on the results from past monitoring efforts of the MWP and SWAMP, made accessible through the California Environmental Data Exchange Network (CEDEN). Data synthesis also consisted of a review of published and gray literature on sediment as well as benthic and nektonic fauna to identify key taxa for this project and additional historical data.

Collection of New Sediment and Biotic Data

Three focal areas were chosen for sampling of sediment and resident biota (Figure 2.1). The Oneonta Slough and Boca Rio sites represent the northern arm of the estuary, which has been used by the Mussel Watch Program (MWP) and the state's Surface Water Ambient Monitoring Program (SWAMP) to assess mussels (2007-2009) and forage fish (1985-1992). Grove Avenue is a storm drain outfall immediately adjacent to a residential area in Imperial Beach. Model

Marsh, in the southern arm of the estuary, is a restoration site in a disturbed area (primarily associated with excessive sedimentation from eroding hillsides of Mexico), and is also the site of future marsh restoration. The first sampling round occurred in May 2021, soon after the wet-season and associated rainfall had ended. This round was used to further refine sampling and analysis approaches for the next round that occurred in October 2021, soon after the dry-season (June 1 – September 30) (White and Greer, 2006).

Site Locations

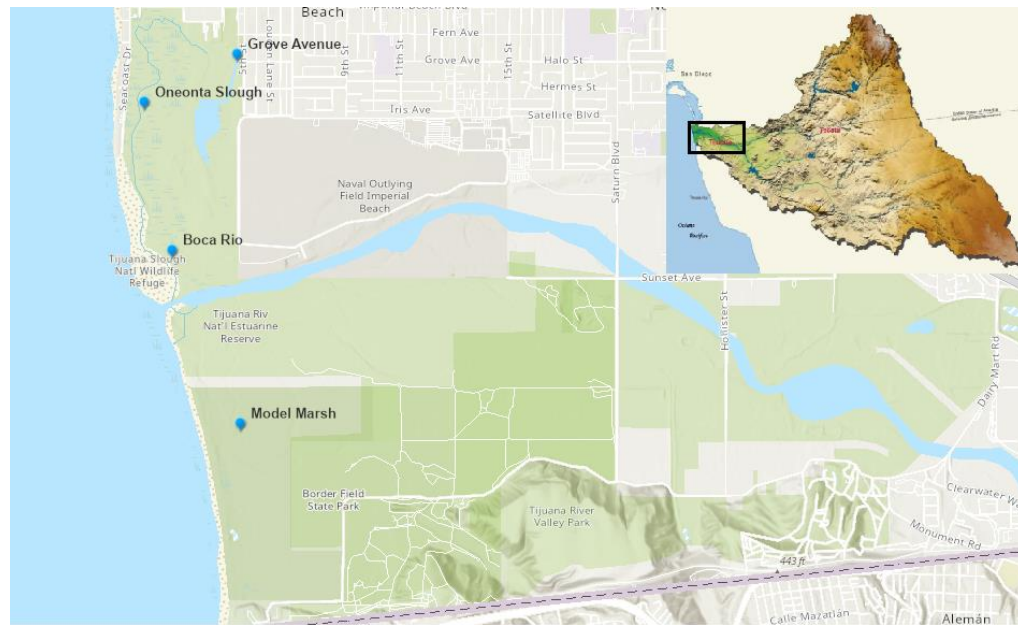


Figure 2.1 Site map of Tijuana River National Estuarine Research Reserve with study sites (blue). The Boca Rio and Oneonta Slough sites are located in the northern arm. Grove Avenue is an urban outfall point by a residential area. Model Marsh is a restoration site in the southern arm. Image of Tijuana River Watershed elevation map as reference adapted from San Diego State University.

Sediment was collected following the MWP's most recently updated standard protocol and U.S. Fish and Wildlife Service's protocol (Apeti et al.,

2012; Zeeman and Roberts, 2014). All samples were processed with stainless steel equipment that was pre-cleaned with Liquinox, followed by rinses with tap water, dilute nitric acid, tap water, acetone, and analyte-free water. Composites of surface sediment from the top 6 inches were collected with ice scoops and homogenized in a mixing bowl before being packaged in Whirl Pak bags.

Sampled biota included the following (Table 2.1): mollusks (mussels and horn snails), fish, and crustaceans (crabs). Two horn snail species were collected to determine whether contamination varies between a native (*Cerithideopsis californica*) and nonnative (*Batillaria attramentaria*) species. Biota collection leveraged ongoing sampling at the Reserve, and sample preparation followed MWP and SWAMP (Stransky et al., 2016) protocols, as appropriate. Upon collection, all biotic samples were immediately placed in labeled Ziploc bags, preserved on ice, and then frozen prior to direct shipping to Physis Environmental Laboratories, Inc. For bivalves, the target species was the bay mussel, *Mytilus* spp., which has been the focus of the MWP. Both bivalves and gastropods were handpicked from intertidal sites. Fish were collected by seine sampling as part of the annual monitoring that occurs at the Reserve, which is conducted both by TRNERR staff and UC Santa Barbara (CCC, 2018). Crabs were either seined with fish or collected via minnow traps. Data from primarily whole-body organisms were included in the data analysis because whole bodies are used to better reflect what is actually ingested by ecological receptors. Using whole bodies rather than individual tissues, has the advantage of obtaining maximum information with limited number of analyses (Bevelhimer et al., 1997; Goldstein and DeWeese,

1999). Horn snail data was processed and analyzed as tissue samples (with shell removed). Horn snail and bivalve samples were of soft tissues only.

Sampling Round	Sample Group	Name	Total Number Sampled	Number of Composites Analyzed	Sites Collected
May 2021 (Post wet-season)	Sediment		N/A	3	Northern Arm - Boca Rio, Grove Avenue, Southern Arm - Model Marsh
	Mollusks	Mytilus spp. - bay mussel	20	1	Northern Arm - Boca Rio
		Batillaria attramentaria - Japanese mud snail	~100s	2	Northern Arm - Boca Rio, Grove Avenue
		Cerithideopsis californica - California horn snail	~100s	2	Northern Arm - Boca Rio, Southern Arm - Model Marsh
	Sediment			2	Northern Arm - Boca Rio, Grove Avenue
October 2021 (Post dry-season)	Forage Fish	Hypsoblennius gentilis - bay blenny	6	1	Northern Arm - Oneonta Slough
		Paralichthys californicus - California halibut	1	1	Northern Arm - Oneonta Slough
		Fundulus parvipinnis - California killifish	6	1	Northern Arm - Oneonta Slough
		Atherinops affinis - Topsmelt	23	3	Northern Arm - Oneonta Slough
	Crustacea	Hemigrapsus oregonensis - yellow shore crab	13	2	Northern Arm - Oneonta Slough, Southern Arm - Model Marsh
		Pachygrapsus crassipes - striped shore crab	4	1	Northern Arm - Oneonta Slough
	Mollusks	Cerithideopsis californica - California horn snail	~100s	1	Northern Arm - Boca Rio
		Mytilus spp. - bay mussel	44	2	Northern Arm - Boca Rio

Table 2.1 Sediment and Biota Sampled in the Tijuana River Estuary in 2021

Target Analytes

Concentrations of inorganic and organic contaminants were determined by Physis Environmental Laboratories, Inc. (Table 2.2; the full list of measured analytes is available in Appendix A). The EPA advises reporting total mercury rather than methylmercury, which is the state that is toxic to humans and wildlife, as most mercury in fish and shellfish tissue is present primarily as methylmercury (U.S.E.P.A, 2000). Following MWP protocol and this EPA advisory, total mercury is reported and analyzed. For biota, there were some cases where tissue weight was insufficient for the full suite of requested chemical contaminants. Thus, not all analytes for organics were measured for certain composites, which were noted. Analytical results are reported as dry weight concentrations unless indicated otherwise.

Inorganics	Organics
Arsenic (As)	Chlordanes
Cadmium (Cd)	Dichlorodiphenyltrichloroethane (DDTs)
Chromium (Cr)	Dieldrin
Copper (Cu)	Polybrominated diphenyl ethers (PBDEs)
Lead (Pb)	Polycyclic aromatic hydrocarbons (PAHs)
Mercury (Hg)	
Nickel (Ni)	
Selenium (Se)	
Tin (Sn)	
Zinc (Zn)	

Table 2.2 Inorganic and Organic Chemical Contaminants Analyzed in 2021

2.3 Results

In the spring and fall of 2021, inorganic (metal) and organic contaminants were sampled in sediment and biota at various locations within the Tijuana Estuary. Assessment of sediment and mussels, viewed in relation to national programs such as NOAA's Screening Quick Reference Tables (SQuiRTs) and the Mussel Watch Program, offer an overall picture of contamination levels in the estuary. One of the most ubiquitous organisms encountered were native and invasive horn snails, which were used to assess spatial and seasonal patterns. Contamination levels in various species of fish and invertebrates (Table 2.1) reveal differences across taxonomic levels and/or life histories, and allow examination of trends over time in the Tijuana Estuary as well as comparisons to nearby San Diego Bay.

Sediment Contaminants in the Tijuana Estuary

In 2021, all inorganic contaminant concentrations were considered low for sediment. When assessing spatial differences in sites across the Tijuana River Estuary (Figure 2.2), all results were well within the Effects Range Low (ERL) or Effects Range Median (ERM) thresholds of NOAA's Screening Quick Reference Table (SQuiRTs) (Buchman, 2008). The most notable difference is that Grove Avenue had the highest levels of copper, lead, and zinc. Model Marsh had the highest arsenic concentrations at $7.98 \mu\text{g (dw)}$, but was still below the Effects Range Low (ERL) value of $8.2 \mu\text{g (dw)}$.

When assessing seasonal differences between the wet- and dry-seasons for sediment sampled in 2021 (Figure 2.2), there was little to no difference across the five contaminants. Contaminants for the wet-season were generally slightly above those of the dry-season but overall, still considered low concentrations when compared to the ERL/ERM thresholds.

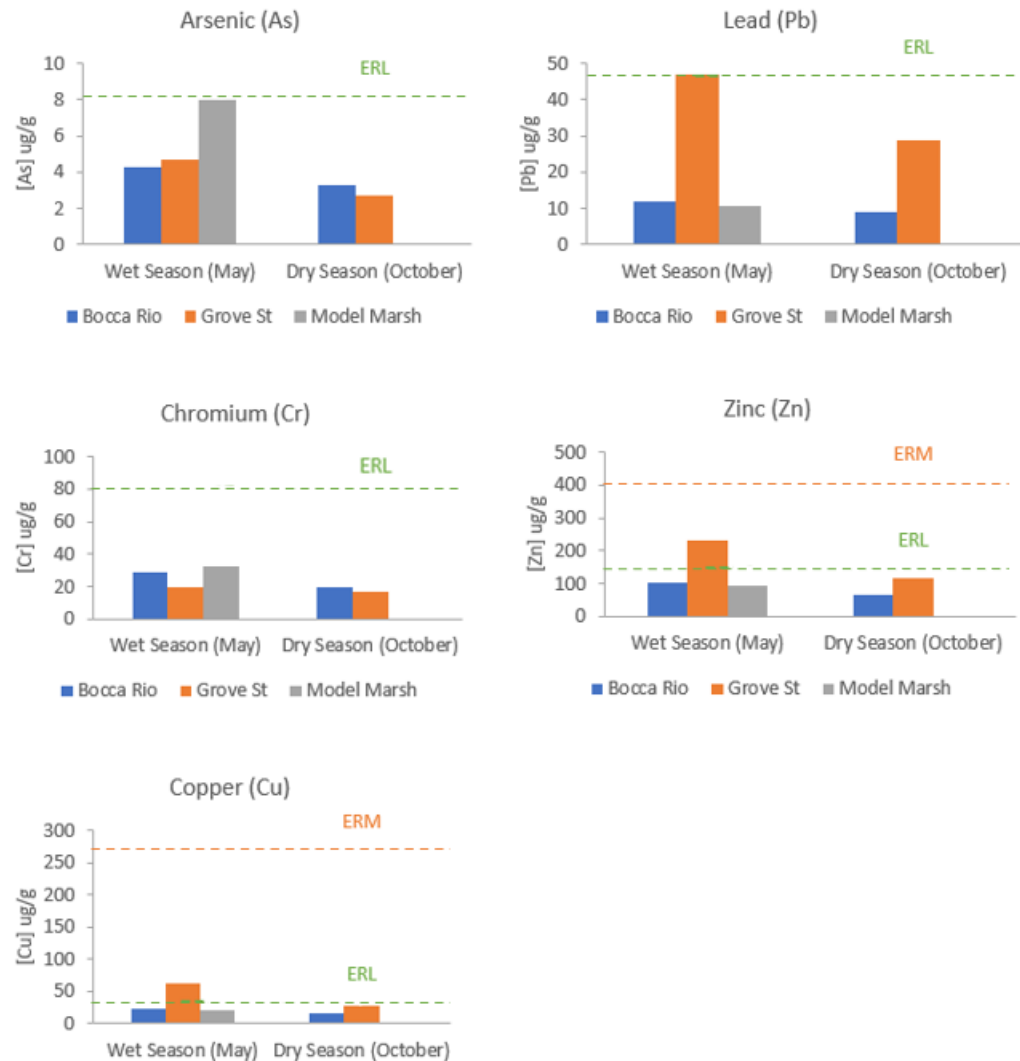


Figure 2.2 Inorganic Contaminant Concentrations ($\mu\text{g dw}$) in Sediment Across Tijuana River Estuary Sites (2021). NOAA SQuiRTs table thresholds shown for ERL (green) and ERM (orange) (Buchman, 2008)

When 2021 data are compared with historical data, as well as the Effects Range Low (ERL) and Effects Range Median (ERM) thresholds from the NOAA SQuiRTs table, inorganics generally were well under the ERM at all points (Figure 2.3). Only zinc had one instance, in 1992, where concentrations indicated potentially harmful effects. Zinc values since then have been closer to ERL, and similar decreases have been seen in lead and copper. Arsenic and chromium were the lowest with all concentrations under the ERL, indicating harmful effects would be rarely observed.

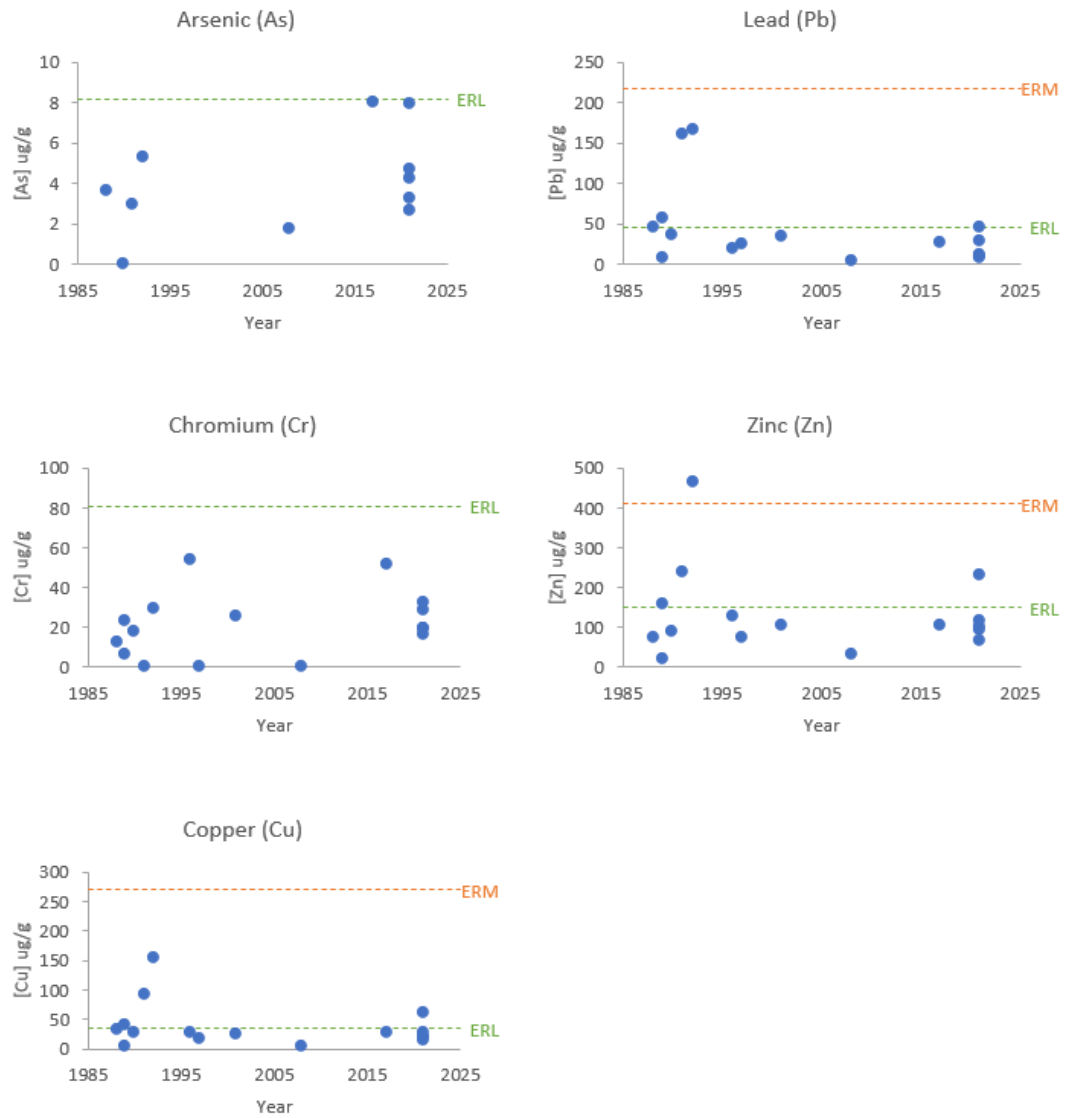


Figure 2.3 Status of Sediment Inorganic Contaminant Concentrations ($\mu\text{g/g dw}$) within Tijuana River Estuary (1985 – 2021). ERL (green) and ERM (orange) (Buchman, 2008). Historical data gathered from (Berry, 2019).

In 2021, some organic contaminant concentrations in sediment were at elevated levels for total chlordanes and DDTs while LPAHs, and HPAHs were low. When assessing spatial differences in sites across the Tijuana River Estuary (Figure 2.4), Grove Avenue consistently had the highest contaminant concentration relative to other sites. Total chlordanes were especially high, largely surpassing the Effects Range Median (ERM) threshold by 14.1 ng/g (dw). Total DDTs at Grove Avenue were 13.9 ng/g dw short of reaching the ERM of 46.1 ng/g dw.

When assessing seasonal differences between the wet- and dry-seasons for sediment sampled in 2021 (Figure 2.4), substantial differences were observed for total chlordanes and DDTs. Grove Avenue's DDT concentrations went from largely surpassing the ERM in the spring to not being detected in the fall. Total DDT concentrations across all sites went from above the ERL in the spring to not detected in the fall. Although total PBDEs do not have a standard threshold to contextualize the difference, Boca Rio and Grove Avenue concentrations decreased by at least half from spring to fall. Site differences were minimal for PAHs as concentrations were close to or far below the ERL threshold.

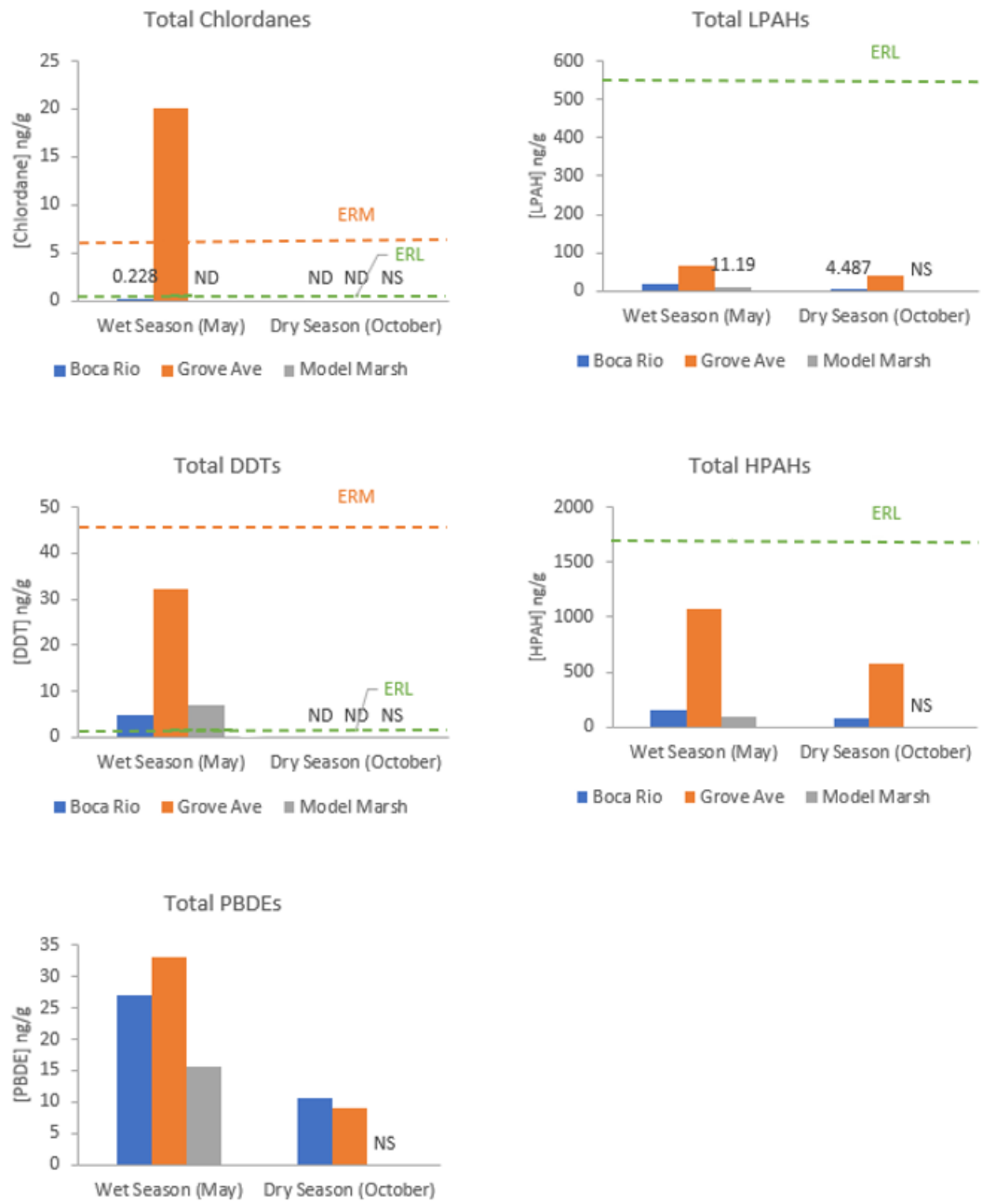


Figure 2.4 Organic Contaminant Concentrations (ng/g dw) in Sediment Across Tijuana River Estuary Sites (2021). NOAA SQUIRTs table thresholds shown for ERL (green) and ERM (orange) (Buchman, 2008). ND = No Detect. NS = Not Sampled.

Historical organic contaminant concentrations in sediment were only available from NOAA’s National Status and Trends data in 1992, which was insufficient data to compile a long-term timeline such as was done for inorganics (Figure 2.3). The closest temporal comparison that could be performed was comparing the ranges between two periods, one from the 90s and the most current 2021 dataset. A decrease was observed across total Chlordanes, DDTs, and dieldrin (Table 2.3). Values that were either above the ERL or ERM in 1992 had decreased to either below the ERM or ERL in 2021, now mostly under the ERL threshold at which toxic effects are infrequently observed. Total DDTs in 1992 had an especially high maximum of 287 ng/g dw, which was above the ERM, and in 2021 it was still found to be above the ERL.

Analyte	1992 Range (ng/g)	2021 Range (ng/g)	ERL (ng/g)	ERM (ng/g)
Chlordanes	ND - 2.21	ND - 0.228	0.50	6.00
DDTs	ND - 287	ND - 32.3	1.58	46.1
Dieldrin	ND - 4.26	ND	0.02	8.00
PCBs	ND - 20.9	0.98 - 39.2	22.7	180

Table 2.3 Status of Sediment Organic Contaminant Concentrations (ng/g dw) within Tijuana River Estuary (1980 – 2021). ERL (green / italicized) and ERM (orange / bolded) (Buchman, 2008).

Mussel Bioindicator Assessment

The national Mussel Watch Program (MWP) provides a good basis for comparison due to its delineation of contaminant categories (low, medium, and high) based on national patterns (Kimbrough et al., 2008), although local data for

the blue mussel *M. galloprovincialis* from the estuary only dated back to 2007. According to MWP categorizations for regional species across the West Coast and Northeast United States, current (2021) mussel contaminant concentrations were all categorized as “low” (Tables 2.4 and 2.5). In 2007-2009 samples, most values were also in the “low” range, although some “medium” ranges were found. The only metal above “low” was zinc, with a value of 145 $\mu\text{g dw}$. Total DDTs in 2007-2009 indicated consistently higher “medium” values, while some samples of total chlordanes and PAHs exceeded “low” ranges.

Analyte	2007 - 2009	2021
Arsenic (As)	9.46 - 10.6	8.76 - 9.5
Cadmium (Cd)	2.05 - 2.12	1.5 - 1.99
Chromium (Cr)	0.88 - 1.05	0.9 - 1.09
Copper (Cu)	7.11 - 8.85	4.57 - 5.95
Lead (Pb)	1.45 - 2.27	0.89 - 1.48
Mercury (Hg)	0.09 - 0.1	0.09 - 0.1
Nickel (Ni)	2.04 - 2.33	0.89 - 2.1
Selenium (Se)	2.28 - 2.79	2.54 - 2.84
Tin (Sn)	ND - 0.118	0.13 - 0.16
Zinc (Zn)	129 - 145	92 - 118

Table 2.4 Inorganic Contaminant Concentrations Found in Mussels ($\mu\text{g dw}$). Results are compared to “Mussel Watch Program: An Assessment of Two Decades” (Kimbrough et al., 2008). Numbers in black are categorized as low while those orange / bolded are categorized as medium.

Analyte	2007 - 2009	2021
Total Chlordanes	5.95 - 9.85	ND - 2.38
Total DDTs	128 - 147	10 - 48
Total Dieldrin	0.91 - 3.32	ND
Total PAHs	939 - 2774	654

Table 2.5 Organic Contaminant Concentrations Found in Mussels (ng/g dw). Results are compared to “Mussel Watch Program: An Assessment of Two Decades” (Kimbrough et al., 2008). Numbers in black are categorized as low while those orange / bolded are categorized as medium.

Spatial Relationship Between Sediment and Horn Snails

Spatial patterns were assessed in the Tijuana River Estuary for sediment and horn snails as samples were available for Boca Rio, Grove Avenue, and Model Marsh. For both sediment and horn snails (Figure 2.5A), Grove Avenue had the highest concentrations by far for copper, lead, and zinc as compared with the remaining sites. For organics (Figure 2.5B), total chlordanes and PAHs at Grove Avenue again had the highest concentrations for both sediment and snails when compared to Boca Rio and Model Marsh.

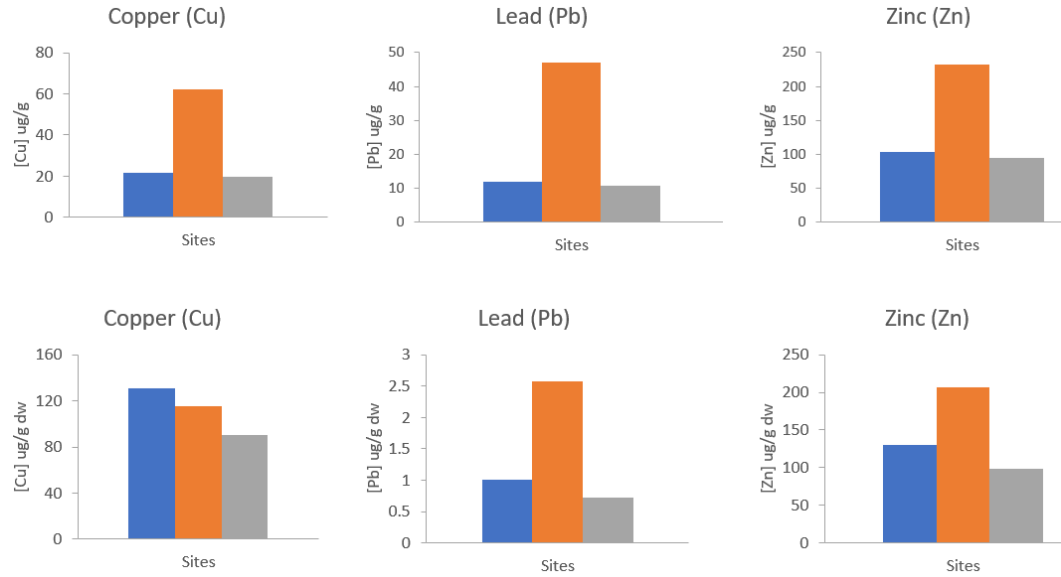


Figure 2.5A Inorganic Contaminant Concentrations ($\mu\text{g/g dw}$) Found in Sediment (top row) and Horn Snails (bottom row). Sites displayed are Boca Rio (blue), Grove Avenue (orange), and Model Marsh (grey).

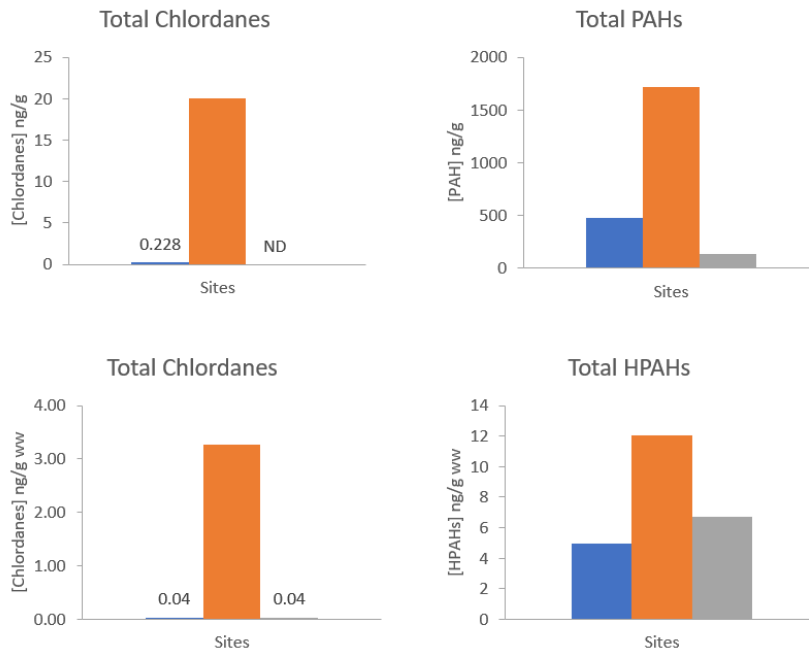


Figure 2.5B Organic Contaminant Concentrations Found in Sediment (top row; ng/g dw) and Horn Snails (bottom row; ng/g ww). Sites displayed are Boca Rio (blue), Grove Avenue (orange), and Model Marsh (grey).

Contamination Levels Across Nine Species of Fish and Invertebrates

For consistency, the same five inorganics from the sediment analysis were selected for species comparison. When comparing species groups within the northern arm of the Tijuana River Estuary (Figure 2.6), species within groups tended to generally stay in similar ranges, except for one anomaly in chromium for halibut. Arsenic, copper, and zinc were the most consistent in ranges within species groups. Across groups, snails were among the highest for metals with the composites having much higher concentrations for copper, lead, and zinc. Crabs also tended to be higher for copper and arsenic. Halibut is the only fish sample that had a sharp contrast to the other organisms with its chromium concentration having about a ten-fold difference. Topsmelt had higher lead concentrations compared to the other fish species. For organic contaminants, differences across species groups were greater than what was seen for inorganics. Fishes consistently had the highest concentrations (Figure 2.7) for total DDTs and chlordane.

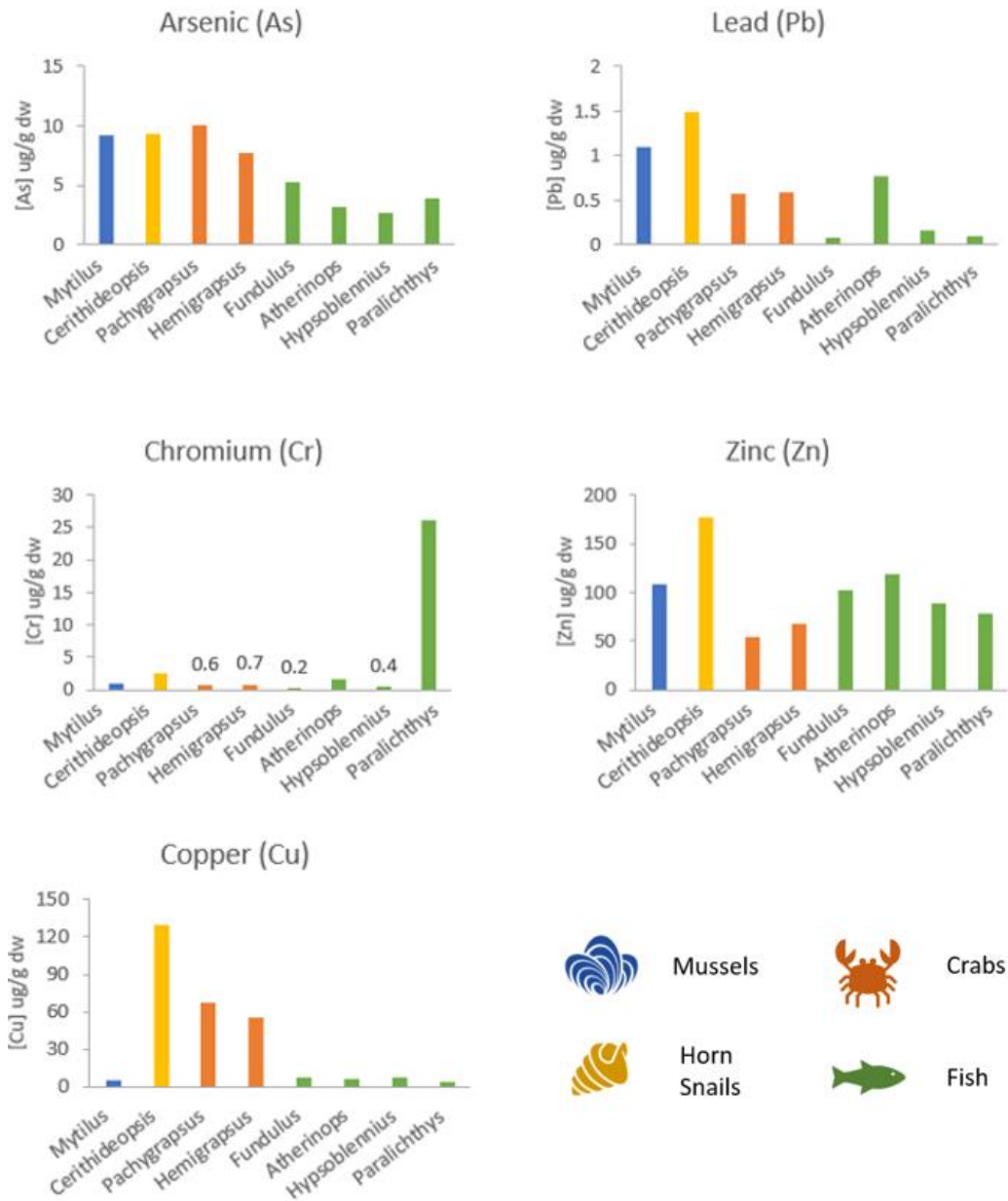


Figure 2.6 Inorganic Contaminant Concentrations Across Species Groups in the Northern Arm of the Tijuana River Estuary.

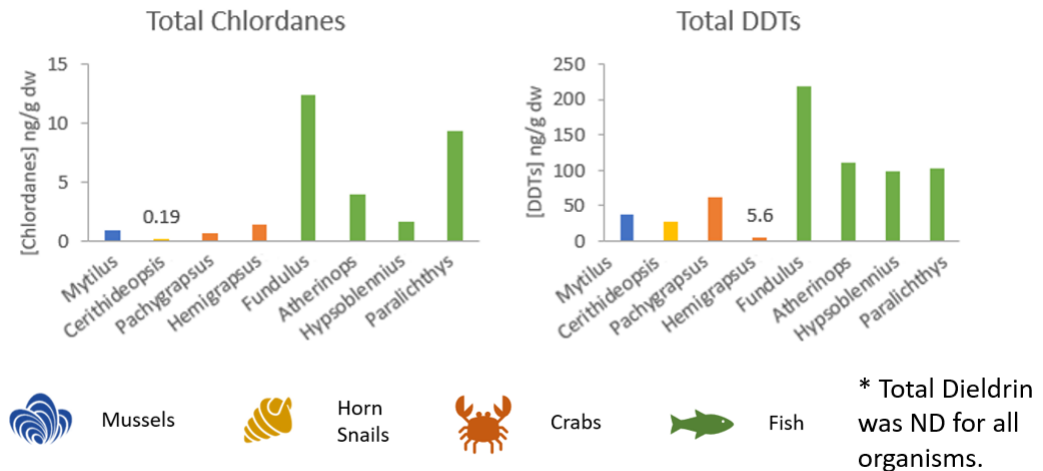


Figure 2.7 Organic Contaminant Concentrations Across Species Groups in the Tijuana River Estuary.

Contaminants in fish have been sampled several times over the last 35 years, and it is possible to look at trends over time in these taxa. For most contaminants, patterns indicated little change, or perhaps a slight decrease, over time (Figures 2.8A and 2.8B). There were only thresholds for four inorganics in fish species. For inorganic contaminants that had established thresholds from U.S.D.O.I. Guidelines, all 2021 samples were at low levels (Figures 2.8A and 2.8B). Arsenic concentrations in fish were above the threshold of which there would be no effect, but still well below the toxicity threshold. Copper had some historical samples that hovered just above and below the toxicity threshold, but all 2021 samples were under the “No Effect” threshold. Mercury and selenium concentrations in fish have consistently stayed below the “No Effect” threshold since 1985. Zinc has previously been up to almost double “Normal Background” levels, but had lowered in 2021. For metals without a threshold, chromium and

nickel each had an outlier for the 2021 samples while the rest clustered closer together.

For organics, all 2021 results were at low levels, and some decreases from previously higher concentrations were seen (Figure 2.9). Organic concentrations were all found to be under the most sensitive dietary screening values for a small avian piscivores (Zeeman, 2016) except for DDTs. DDT concentrations were above the less sensitive dietary screening level for avian species (which are relatively sensitive to toxic effects of DDT), but still just below the FDA action level for human health. No graph is shown for dieldrin as there has been none detected in fish throughout records since 1985.

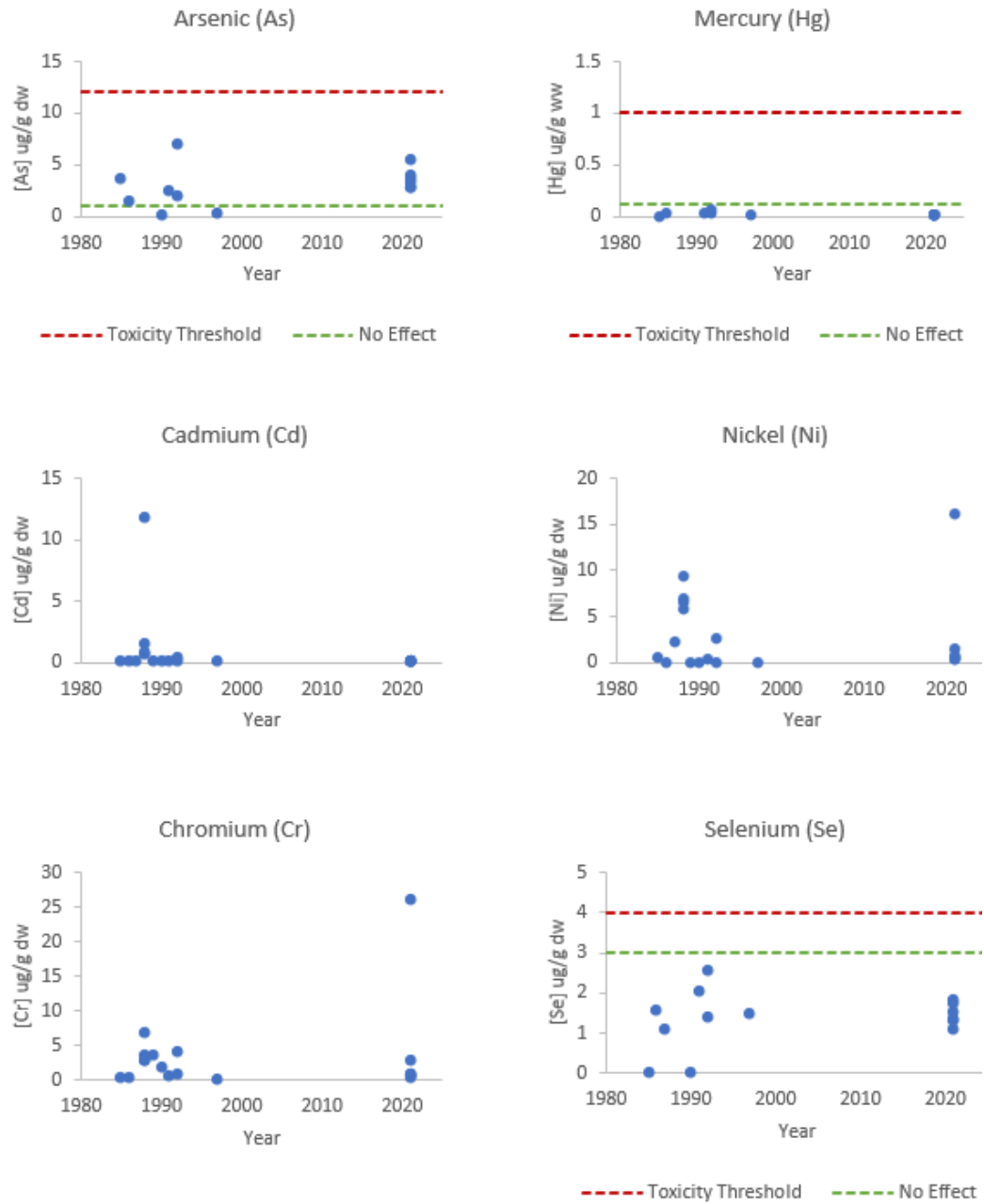


Figure 2.8A Inorganic Contaminant Concentrations from Whole-Body Fish in the Tijuana River Estuary with Thresholds (1985-2021). Where thresholds were available in USDOJ Guidelines (U.S.D.O.I., 1998) for fish (As, Cu, Se, and Zn) and FDA Action Level for human health (Hg), above Toxicity Threshold (red) is where harmful effects appear to likely occur and below No Effect threshold (green) is where no harmful effects appear to occur. Mercury is in wet weight.

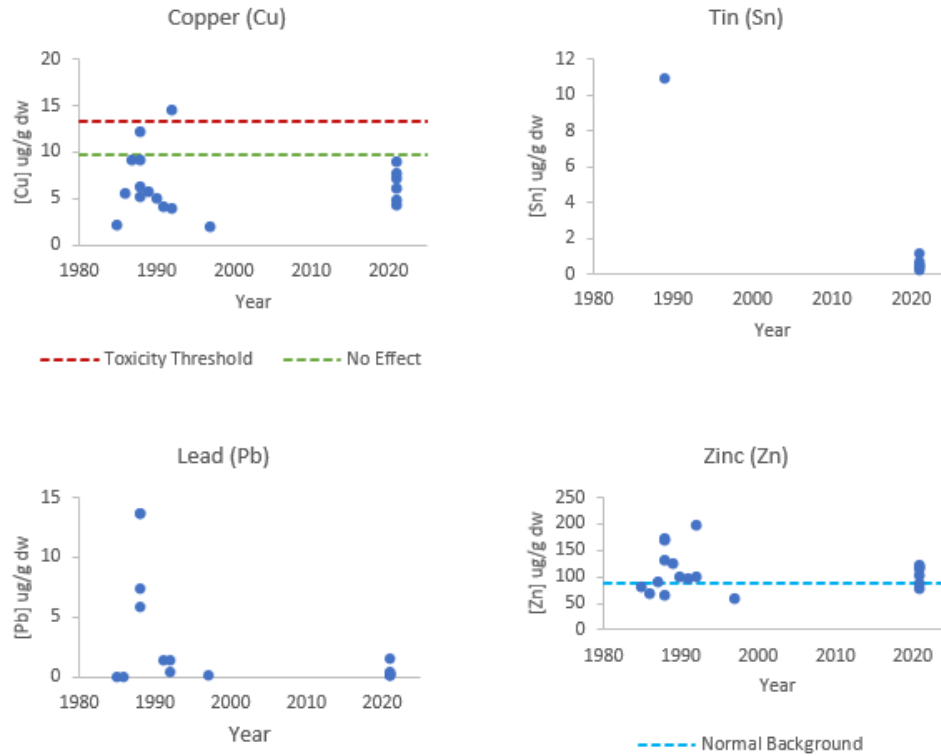


Figure 2.8B Inorganic Contaminant Concentrations from Whole-Body Fish in the Tijuana River Estuary with Thresholds (1985-2021). Where thresholds were available in USDOJ Guidelines (U.S.D.O.I., 1998), above Toxicity Threshold (red) is where harmful effects appear to likely occur and below No Effect threshold (green) is where no harmful effects appear to occur. Zinc has normal background concentrations for fish displayed (blue). Thresholds are for impacts on fish.

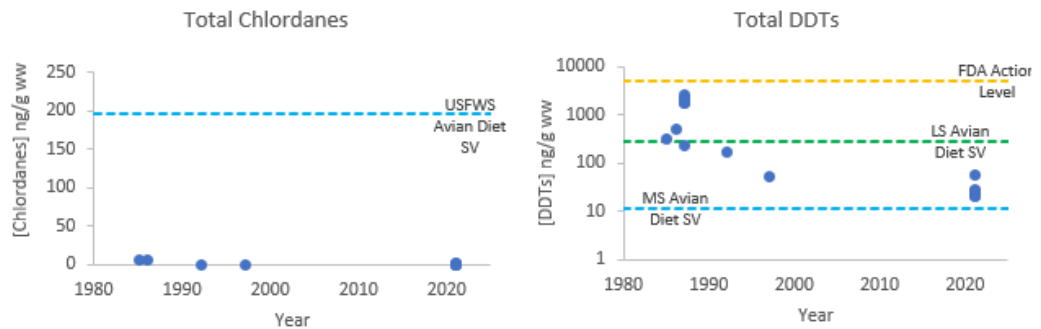


Figure 2.9 Organic Concentrations Found in Whole-Body Fish in the Tijuana River Estuary (1985-2021). Thresholds of MS (Most Sensitive) or LS (Least Sensitive) Observed effect level (EL) for most sensitive species) Avian Receptor dietary screening values (Zeeman, 2016). FDA = Food & Drug Administration.

Comparison to San Diego Bay

When comparing to recent data from the San Diego Bay Bioaccumulation Study (Stransky et al., 2016) to that in 2021 from the Tijuana Estuary, some differences in contaminant concentrations were noted. Inorganic contaminant concentrations in the Tijuana River Estuary were generally lower than those found in San Diego Bay (Figures 2.10 and 2.11). For fish species (Figure 2.10), mercury concentrations in San Diego Bay were higher for halibut, killifish, and topsmelt, but were all still well under the threshold at which harmful effects in fish are likely to occur. Concentrations in some fish samples at least are greater than dietary screening levels, including observed effect levels for sensitive avian piscivores (Zeeman, 2016). When comparing invertebrates, mollusks, fish, and crustacea from San Diego Bay (Figure 2.11), mercury and selenium were slightly higher than Tijuana River Estuary.

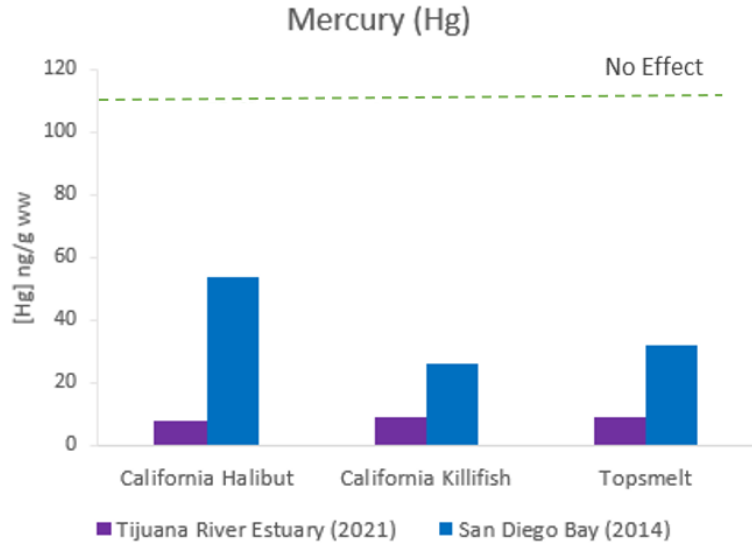


Figure 2.10 Mercury Concentrations in Fish From the Tijuana River Estuary (2021) and San Diego Bay (2014). Compared to the “No Effect” Threshold from the Department of the Interior for fish (U.S.D.O.I., 1998)

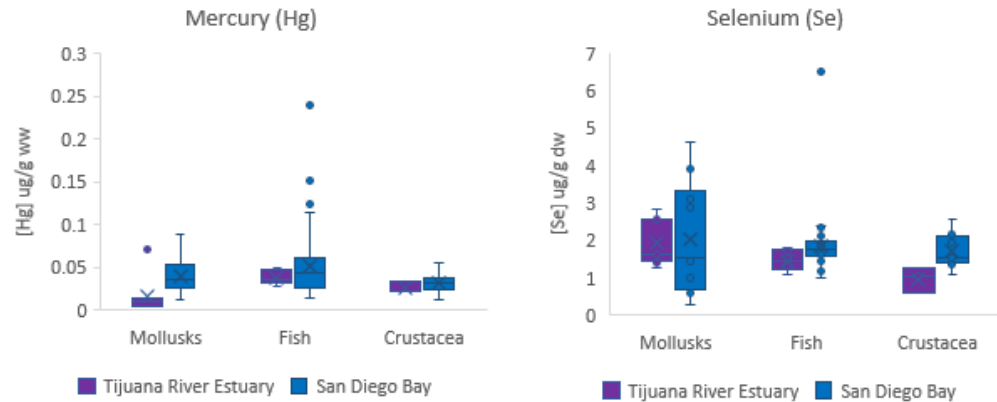


Figure 2.11 Inorganic Concentrations Across Organisms in Tijuana River Estuary (2021) and San Diego Bay (Stransky et al., 2016)

For organics (Figure 2.12), concentrations of total DDTs and PBDEs in fish species from the Tijuana River Estuary are greater than concentrations of OCs in fish from San Diego Bay, and these may exceed levels of concern for sensitive avian species eating these fish. Total chlordanes had minimal differences between the two sites, and both sites were far below potentially harmful dietary levels for avian receptors for total chlordanes. Across different species groups (Figure 2.13), DDT concentrations were much higher in the Tijuana River Estuary when compared to San Diego Bay. Both sites were more aligned for total chlordanes and PBDEs.



Figure 2.12 Organic Contaminant Concentrations Found in Fish Species from the Tijuana River Estuary (2021) and San Diego Bay (2014). Dietary screening values for avian receptors for small piscivores (Zeeman, 2016). MS = Most Sensitive.

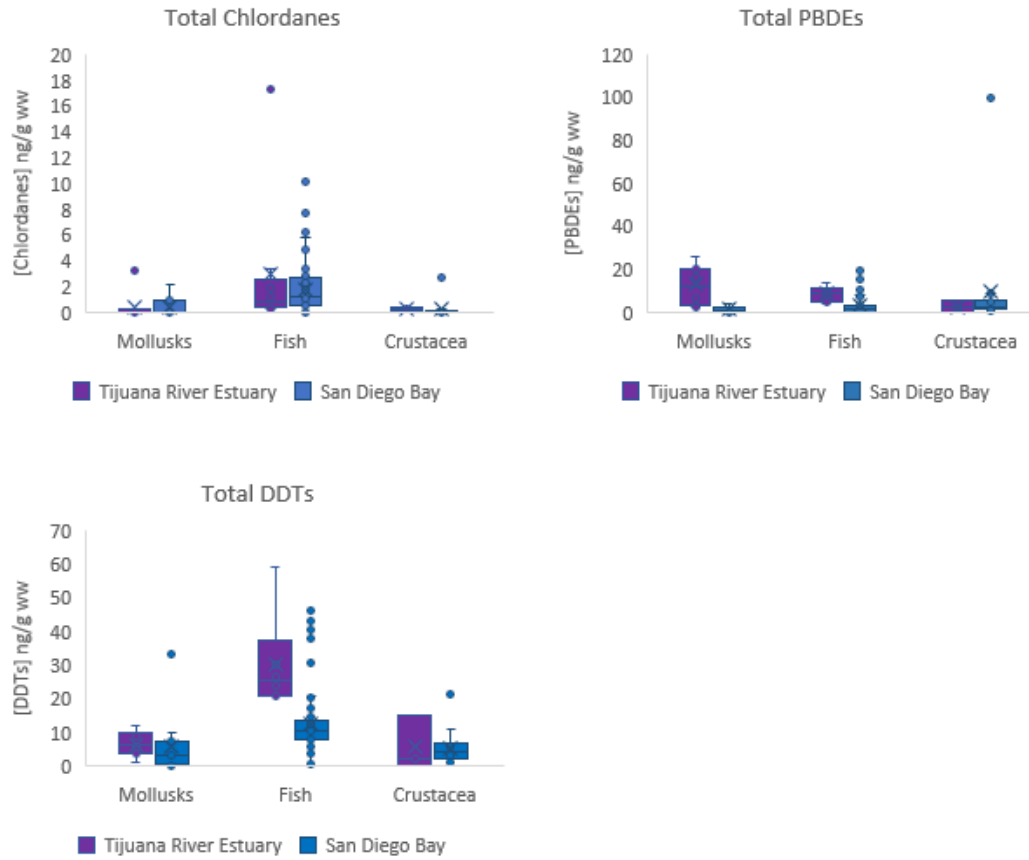


Figure 2.13 Organic Contaminant Concentrations from Species Groups in Tijuana River Estuary and San Diego Bay (Stransky et al., 2016)

2.4 Discussion

A suite of sediment and biotic indicators were used to assess the conditions of the Tijuana Estuary, using established thresholds where available (e.g., NOAA's sediment Screening Quick Reference Tables and National Status and Trends Mussel Watch Program), comparisons to other systems, and trends over time. Some differences across sites and seasons were noted, but, in general, the Tijuana River Estuary's ecosystem health in 2021 has improved over time with respect to the contaminants addressed. For most of these contaminants, levels appear relatively low, which perhaps might be unexpected for a heavily urbanized area such as the Tijuana Estuary. However, while risks to wildlife have lessened (perhaps below levels detectable by conspicuous die-offs or other failures), there is evidence of bioaccumulation and biomagnification, leading to potential concern for some contaminants (e.g., total DDT and PBDEs). For other contaminants (e.g. PAHs), the relative lack of thresholds makes it difficult to assess current risk to wildlife.

Contamination Levels in the Tijuana Estuary

Zinc

Zinc is an essential nutrient, although toxic effects are associated with excessively high doses. Toxic levels are uncommon, but can cause anemia or damage to the pancreas and kidneys. Previous assessments of the Southern California Bight have identified zinc as a contaminant of concern (Kimbrough et

al., 2008). In both past studies and this work, zinc stood out as one contaminant with somewhat elevated values in the Tijuana Estuary. In sediment, one site (Grove Avenue, Figures 2.2 and 2.3) had concentrations above the ERL. For fish, zinc also had values above background levels (Figure 2.8B). Mussels had previously (2007-2009) exhibited somewhat elevated values, but 2021 levels had decreased to low levels (Tables 2.4 and 2.5). From the Southern California Bight's most common sources of pollution (Maruya and Schiff, 2009), the Tijuana River Estuary's was likely derived from stormwater, outfall pipes, and potentially erosion of metal-rich soil. Interestingly, the MWP's 2007-2009 assessment analyzed the entire West Coast and found only seven sites to have zinc concentrations in the medium range, six of which were in San Diego County. This indicates that this is a unique, regional issue worthy of continued consideration.

Copper

Copper is a naturally occurring element that is ubiquitous in the environment, especially with trace amounts being essential nutrients for plants and animals. At high enough levels, copper can be toxic, altering freshwater benthic community abundance and species richness (Gardham et al., 2014) as well as leading to the mortality of diatoms, copepods, and fish larvae in marine waters (Bao et al., 2018). In water, copper is mostly found bound with organic matter and transported to coastal and estuarine waters via runoff and river transport. Anthropogenic sources include mining, manufacturing, agriculture, and sewage sludge (Kimbrough et al., 2008; van der Gon et al., 2007). Like zinc,

copper in Tijuana Estuary sediments has shown some elevated values, most recently associated with the Grove site (Figures 2.2 and 2.3). Copper in mussels has remained at low levels (Table 2.4), however, and concentrations in fish have decreased over the last 30 years (Figure 2.8B).

Chromium

Chromium(III) is a naturally-occurring essential nutrient for normal energy metabolism, and the human population is exposed by ambient air, food, drinking water, soil, and consumer products. There are standards for drinking water, soil, and food, but none for aquatic life. Chromium(VI) compounds are generally more toxic than chromium(III) due to the generation of free radicals formed during reduction to chromium(III) in biological systems and are almost always related to anthropogenic activity. Consistent exposure to high levels of chromium or its compounds, primarily chromium(VI), can result in respiratory, cardiovascular, gastrointestinal and hematological, and liver and kidney effects, and increased risks of death from lung cancer (Wilbur, 2000). Although chromium contamination in the Tijuana River Estuary is unlikely to result in human ingestion leading to these effects, high doses have resulted in similar detrimental effects in animals. Chromium levels in both sediment and mussels appeared relatively low (Figures 2.2 and 2.3, Table 2.4). Notably, however, one fish sample, for halibut, was found with very high levels (Figure 2.6). Although this is abnormally high compared to average levels of $< 1 \mu\text{g ww}$ in fish of other regions (Aslam and Yousafzai, 2017; Davis et al., 2004), levels of chromium concentrations exceeding $26.1 \mu\text{g dw}$ (or $5.33 \mu\text{g ww}$) have been previously

observed in whole-body fish in other areas (Holdway, 1988). High chromium concentrations in fish have even led to fish scales to be assessed as a bioadsorbent (Teshale et al., 2020). It is important to note that the halibut sample, consisting of a single fish, is the only one of four fish composites that experienced a high level of chromium, meaning that this outlier could be the result of some error. No sample collected from the Tijuana River from 2018-2019 by the U.S. Boundary and International Water Commission detected toxic parameters of concern for Hexavalent Chromium (IBWC, 2020). Nonetheless, it would be beneficial to continue to analyze for total chromium concentrations in future biomonitoring in the case that it does need to be a contaminant of concern for the Tijuana River Estuary.

Arsenic

Arsenic comes from both natural sources and industrial production, generally resulting in high levels found in the environment. Sources can come from products such as preserved wood, semiconductors, pesticides, and pigments as well as from the atmosphere via smelting, fossil fuel combustion, and power generation. Human activities have altered the natural biogeochemical cycle of arsenic, leading to contamination of land, air, and water. Movement of arsenic to coastal and estuarine waters is primarily from river runoff and atmospheric deposition, dependent on the geochemical breakdown of arsenic host minerals and mechanisms. For example, a local historic artisanal mine site in the Rancho Peñasquitos area is strongly enriched in arsenic (Wright et al., 2022), which has the potential to be highly toxic with linkages to hyperkeratosis, pulmonary

disease, and various cancers (WHO, 2018). Arsenic concentrations in sediments and mussels were all relatively low, but those in fishes tended to be slightly above the “No Effect” threshold for fishes (Figure 2.8A). Although this could be the result of the local geological environment (Wright et al., 2022), arsenic could be a contaminant of interest.

Mercury and Selenium

Mercury occurs naturally in the environment but is driven to excessive levels from human activities such as coal fired-electric turbines, mining, and sewage sludge in the United States (Kimbrough et al., 2008). It can change forms between elemental, inorganic and organic though its organic state (in this case, methylmercury) is what is of greatest concern for humans and wildlife. As recommended by the EPA, total mercury is reported here as methylmercury is what is most present in fish and shellfish tissue (U.S.E.P.A, 2000). Selenium also occurs naturally in the environment but can enter the air from burning coal or oil (ATSDR). Mercury and selenium concentrations tended to be low in the Tijuana Estuary, both relative to U.S.D.O.I. guidelines and in comparison to nearby San Diego Bay, where values were slightly higher (Figures 2.10 and 2.11). A few fish composites in San Diego Bay exceeded the “No Effect” threshold of 0.11 $\mu\text{g ww}$ for mercury while fish composites neared the “Level of Concern” range of 3-4 $\mu\text{g dw}$ for selenium at which concentrations rarely produce discernible adverse effects in fish but are elevated above typical background. One outlier for mercury in San Diego Bay far exceeded the “Toxicity” threshold at which concentrations

above this value appear to produce adverse effects on some fish and wildlife species.

Chlordanes

Total chlordanes are typically the sum of a few prominent compounds, but technically chlordane consists of at least fifty compounds. The insecticide was mainly used for agricultural and urban settings before being completely banned by 1988. Dietary exposure to high levels of chlordane in fish and shellfish from contaminated waters can be carcinogenic for humans and the pesticide is highly toxic to invertebrates and fish. Chlordane levels in Tijuana Estuary sediments tended to be relatively low in 2021, with the notable exception of the Grove site during the wet season (Figure 2.4). This was also reflected in high concentrations in horn snails during that time period (Figure 2.5B). In mussels, some medium values were found in 2007-2009, but these have since decreased to low levels. Chlordane concentrations in fish were well below the FDA action level for humans of 0.3 ppm wet weight. They were also below dietary screening levels for avian consumers of fish.

DDTs

Total DDTs are widely known to be extremely toxic, particularly to birds, having been banned in the United States in 1972 and halted in Mexico in 2000 (CEC, 2003). Despite this, DDT and its transformation products are very persistent and are still detected at elevated levels today. MWP findings previously indicated much higher proportions of medium and high DDT measurements in the

Southern California Coast relative to the rest of the nation (with most sites categorized as high). Southern California's high concentrations can be attributed at least in part to previous DDT manufacturing plants such as the former Montrose Chemical Corporation in Torrance, California. The Tijuana River Estuary was medium in comparison to the rest of southern California, but a cluster of results once neared the FDA Action Level, which would have necessitated regulatory action from the Food & Drug Administration if these levels were found in a fish or fishery product.

Total DDT concentrations in Tijuana Estuary sediments in 2021 ranged from below detection limits in the dry season to above ERL during the wet season, particularly at the Grove Site. For mussels, 2007-2009 data were consistently categorized as "medium" relative to Mussel Watch Program data from other regions, but DDT levels have now decreased to low in 2021, indicating that the phasing out of DDT manufacturing is having some effect in reducing the harmful contaminant concentrations bioaccumulating in resident organisms. However, the full range of 2021 DDT concentrations for mussels remained above dietary screening levels for sensitive avian receptors screening value (but below the Observed Adverse Effect Level screening value for sensitive avian species). Similarly, for fish, although DDT concentrations across species in the Tijuana River Estuary were above the "Most Sensitive" Avian Receptor Diet screening value, all concentrations are still well below the "Observed Adverse Effect Level dietary screening value for sensitive avian species" Avian Receptor Diet screening value (Zeeman, 2016). Contaminants that exceed these initial screening

levels are considered to be contaminants of potential concern (Bay et al., 2016). If DDT concentrations remain at these levels, this contaminant may be of potential concern for the more sensitive bird species in the area.

PAHs

Total PAHs are made up of a suite of hundreds of compounds and are the only organic contaminants the MWP analyzes that can come from both anthropogenic and natural sources. Human exposure mainly is due to smoke from forest fires, automobile exhaust, grilling, and cigarettes while aquatic organism exposure is from oil spills, boat exhaust and urban runoff. Despite the wide range of toxicities and potential to cause reproduction inhibition, mutations, liver abnormalities and mortality in aquatic organisms, there is no FDA recommended safety level for PAHs in fish and fish products. Sediment standards showed PAH concentrations to be below the ERL, indicating that levels appear to be low in the Tijuana River Estuary compared with other Mussel Watch stations. However, absent information on levels of concern (e.g., screening values) for humans, bottom fish, and wildlife that rely on benthic biota for food, it is difficult to determine whether these levels are of concern for the ecosystem.

PDBEs

PBDEs have not been historically documented throughout reports and studies but are now looked at more consistently since being listed as a contaminant of emerging concern. These chemicals are flame retardants that can be found in products such as furniture foam padding, wire insulation, and small

appliances. Total PBDEs were higher in the Tijuana River Estuary than those in San Diego Bay with one species (*Fundulus*) exceeding the avian diet receptor for small piscivores. Higher PBDEs in the Tijuana River Estuary aligns with the previously found geographic pattern of contamination in piscivorous seabird eggs with DDTs increasing northwards and PBDEs southwards along the California coast (Clatterbuck et al., 2018).

Temporal Trends Across Sediment, Mussels, and Fish

Overall, contaminant concentrations for sediment, mussels, and fish had generally decreased over time, and closer to safer levels when compared to national thresholds. When comparing inorganics across arsenic, chromium, copper, lead, and zinc (Figure 2.2), most concentrations in sediment were under the ERL threshold at which toxic effects are scarcely observed or predicted among most species of benthic invertebrates. Any that were previously near or above the ERM in the 90s decreased closer to the ERL in 2021. When comparing organics in sediment (Figure 2.3) across total chlordanes, DDTs, and dieldrin, organics generally decreased from the 90s to the present and are now mostly under the ERL threshold. When comparing 2007-2009 mussels, any inorganic or organic contaminant previously categorized as “medium” decreased to “low” in 2021. Fish concentrations had similar decreases in arsenic, copper, mercury, selenium, zinc, and total DDTs. The Tijuana River Estuary’s ecosystem health in 2021 was thus not in poor condition in terms of these inorganics and organics. This demonstrates the resiliency of the estuary despite the ongoing overall pollution issue of untreated wastewater reaching the wetland throughout the years,

although more work is needed on loading of these contaminants into the Tijuana River Valley and the degree to which these might be attenuated by wetland processes (Allsing et al., 2022). Some contaminants (most notably DDTs and PAHs) are locally at potential levels of concern for fish and wildlife, and there are ongoing sources associated with wastewater releases and land use practices (both historic and ongoing) in the watershed.

Spatial Relationship Between Sediment and Horn Snails

Despite the need for a broader investigation of the critical ecosystem service of natural filtration helping to preserve the area's ecosystem health, this study does provide evidence of decreasing contamination away from a point source. The Grove site, which was away from the main stem of the river but adjacent to an urban outfall in the City of Imperial Beach (Figure 2.1), was relatively distinct and tended to have higher concentrations in both sediments and snails (the indicators assessed for spatial differences) relative to other locations. For sediment, it had the highest copper, lead, zinc, total chlordanes, and HPAH concentrations (Figures 2.5A and 2.5B), with concentrations in some cases greater than the ERL, and in the case of chlordanes, substantially exceeding the ERM (Figure 2.4). Higher levels of these five contaminants were also reflected in the horn snails that live and feed at the sediment surface, although dietary screening values for PAHs and chlordanes from (Zeeman, 2016) indicated that the horn snail organics were below levels of concern for avian species that forage in the marsh habitat. This suggests that the contamination in the estuary is not only coming from the Tijuana River, but from local land use and discharge at an urban

outfall point. A clear relationship has been previously found between proximity to trans-border sites (versus estuarine) and pollution in the form of fecal contamination for genomes of pathogenic species that have persisted out to the mouth of the river (Allsing et al., 2022), but this study's analysis of contaminants show there might be attention needed at local urban outfall points such as Grove Avenue.

Contamination Across Taxonomic Levels

Contamination across different species groups can give an indication of major exposure pathways, such as the bioaccumulation and biomagnification from birds feeding on sediment-dwelling organisms (benthic pathway) or water column-dwelling organisms (pelagic) (Bay et al., 2016). For the inorganics evaluated in this study, crabs and snails tended to have higher levels of copper concentrations compared to other species groups. Diagnostic levels are not available for copper in invertebrates partly because Cu is generally homeostatically regulated. Some species groups (e.g., crustaceans and mollusks) require Cu for hemocyanin and may normally have higher levels than other species (Furness and Rainbow 1990). Overall, however, there little evidence of increased concentrations for organisms further up the food chain (i.e., fish) observed for inorganic contaminants. This is likely due to the fact that certain heavy metals are essential for survival and metabolic processes, while others may be non-essential but are readily eliminated (Khayat-zadeh and Abbasi, 2010). Some inorganics may be non-essential, but are more readily eliminated than other metals.

While little evidence for biomagnification was seen for inorganics, organic contaminants are synthetic, man-made chemicals that living organisms are not naturally equipped to metabolize efficiently (Khayatzadeh and Abbasi, 2010). Across taxonomic groups in the Tijuana Estuary, fishes tended to have the highest organic contaminant concentrations compared to other species groups. Over the life span of these fish, which are higher up in the food chain and regularly feeding upon species such as the invertebrates sampled here, contaminant concentrations biomagnify. Although birds were not sampled as part of this study, they are of particular conservation and management interest. As they are higher in the food chain, birds are likely experiencing even higher contamination levels in their diet. In a region-wide study of bird eggs, which included TRE, Clatterbuck et al. (2018) found that piscivorous species that consume larger, upper trophic level fish species had higher contaminant concentrations than species that consumer smaller, lower trophic level fish. Also, PBDE levels in bird eggs at TRE were the highest across all sites, a pattern consistent with concentrations seen in invertebrates and fish in TRE compared to San Diego Bay (Figure 2.13).

DDT is a contaminant of particular interest given its role in eggshell thinning and reduced nest productivity (Bay et al., 2016; Carson, 2002). In TRE, although DDT concentrations appear to be decreasing over time, some levels still exceeded the screening levels for most sensitive birds (Figure 2.9). In the past, DDT has been a factor in the proposed management of ospreys at the TRE. During the 1980s, unpublished records indicate that there were no osprey populations in the TRE, likely due to the levels of DDT concentrations in the area.

During that time, the U.S.F.W.S. collected DDT data for a local prey species for osprey (mullet - *Mugil cephalus*) to determine the feasibility of reintroducing osprey into the area again, but DDT concentrations were considered too high for active re-introduction. As DDT concentrations naturally lowered over time due to their ban in the U.S. and Mexico in the years 1972 and 2000, ospreys came back into the estuary on their own and are now a conspicuous part of the bird fauna.

Comparison To Other Systems

As indicated with comparisons to San Diego Bay, differences in contaminant levels are possible in distinct systems even over short geographic distances. Even within systems, differences can be apparent and depend on local inputs and processes (Bay et al., 2016; Talley et al., 2022). The Tijuana River Estuary had lower concentrations in mercury and selenium when compared to San Diego Bay, which has a long history of industrial and military uses, but had substantially higher DDT concentrations than San Diego Bay. This likely reflects the historical context of the Tijuana River Valley, with extensive agricultural land in the mid-20th century (Safran et al., 2017) and likely use of DDT as a pesticide.

On a regional and national scale, the Tijuana River Estuary was found to have relatively low or moderate concentrations of measured contaminants. When assessing contaminant trends across estuarine systems in the United States, the National Estuarine Research Reserves represent some of the least contaminated estuaries (Lauenstein and Cantillo, 2002). The Mussel Watch Program analyzed 2007-2009 concentrations in mussels along the California coast at Areas of Special Biological Significance (ASBS) and non-ASBS sites for several

contaminants including copper, silver, arsenic, lead, cadmium, chromium, nickel, zinc, mercury, total DDTs, total PAHs, total Chlordanes and dieldrin (SWRCB, 2010). When compared to the California coast, this study's results were found to be either around or under the median (Appendix D).

Conclusions

In general, the Tijuana River Estuary's state is contrary to what might be hypothesized from the sewage flows that reach the estuary associated with rainfall. Although more information is needed on levels of pollutants entering the estuary from the watershed, the relatively low levels of most contaminants aligns with what would be expected from wetland processes (e.g., biofiltration and changing flow characteristics) that are known to reduce pollution reaching coastal waters (Allsing et al., 2022; Barbier et al., 2011; Gudimov, 2021). This is a crucial ecosystem service to continue protecting with entities such as the Tijuana River National Estuarine Research Reserve, CA State Parks, the U.S.F.W.S. and their partners. This research has also set a new benchmark with which to assess future changes to the system, such as the planned expansion of wastewater treatment associated with the U.S. Mexico Canada Agreement (USMCA). Continued monitoring of both abiotic and biotic indicators will be critical for informing responses to management actions such as these and continuing to track ecosystem health at the US / Mexico border.

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Chapter 3 Conclusions and Future Work

3.1 Contaminants in the Tijuana River Estuary

Assessment of heavy metal and organic contaminants in the sediment and biota of the Tijuana Estuary suggest that levels in 2021 are of less concern than anticipated, and that there is a general trend towards decreases from previously higher concentrations (Chapter 2). This study also supported the accepted reliability of using organisms such as mussels, snails, and fish as bioindicators for coastal wetlands and marine waters, given their relationships to ambient conditions (Fig. 2.5) and availability of comparative data (Benali et al., 2017; Boening, 1999; Kimbrough et al. 2008, Stransky et al. 2016). This data has set a new baseline and benchmark to evaluate future changes.

Some patterns were observed in the study, which are of both scientific and management interest. There was typically a good correlation between sediment and horn snails, indicating that one can be a proxy for the other for these specific contaminants. More generally, the higher levels seen in sediment and snail contaminants at the urban outfall point into the estuary rather than closer to the main stem of the river suggest that preservation of natural areas is critical as they can naturally mitigate inputs of contaminants. Biomagnification, although not at high levels for forage species, was also observed for organic contaminants within the Tijuana River Estuary. In particular, the historical context of the Tijuana River Estuary as agricultural lands have led to DDT concentrations remaining in the area to this day, despite a decades-long official ban of the product. This speaks to the long-term lasting anthropogenic effects on natural lands and emphasizes that

continued monitoring every decade or so, including assessments of birds where possible (Clatterbuck et al. 2018), would help measure our progress with future management strategies in limiting and mitigating contaminants.

3.2 Broader Socio-ecological Setting of the Tijuana River Watershed

Chapter 2 helped characterize an ecosystem service provided by the Tijuana River Estuary, namely water quality improvement associated with processes operating in wetlands (Zedler and Kercher 2002). Assessing ecosystem health also informs public health, as people are inextricably linked to the environment, especially for the illnesses that can arise from high levels of pollution. In Mexico, there are communities that are living and working near the sewage-contaminated flows that don't have the benefit of water quality improvements afforded by healthy natural systems. In the United States, there are also equity barriers for who is able to benefit from the coast, such as cost and lack of transportation. This study has demonstrated the ecosystem services a wetland provides, but there remains a disconnect for who can access and benefit from them. The lessons learned from this study would be beneficial to apply across the border region, assessing water quality in the urban creeks leading to the estuary and the need to enhance ecosystem health with nature-based solutions. A holistic approach is necessary that integrates the perspectives and knowledge of all affected communities.

Uniquely, the Tijuana River Watershed encompasses one border and three nations, Mexico, the United States, and the Kumeyaay nation, requiring

collaboration for the most effective, equitable environmental management to build resilience for natural areas and the people that live there. The border cities of San Diego and Tijuana have naturally experienced increasing economic, political, and social interdependencies, leading to cross-border public, private, and nonprofit organizations emerging (Mendoza & Dupeyron, 2020). The “CaliBaja” region, stretching 150 miles from California to Baja California, has a gross domestic product of between \$250 and \$300 billion, and more than 90 million people annually crossing the border for commuting / tourism (SDWP, 2022). This dynamic region has developed innovative approaches to best navigate complex issues associated with both natural and community resilience, including philanthropic efforts filling in crucial capacity gaps. This includes the San Diego Foundation’s Binational Resilience Initiative, which supports binational, community-driven projects and programs that work towards regional resilience to climate and other stressors. Multi-sector collaboration has also been built between academia and Tijuana community partners such as the University of San Diego establishing multiple satellite offices to support their wide range of efforts in cross-border collaboration that focus on education, arts, entrepreneurship, social innovation, migration, and human rights. In order to best address the social impacts of missing ecosystem services, data-backed solutions should be co-developed with the affected communities. Long-term impact would result from bridging the gap between science and policy in the form of boundary spanning, creating actionable knowledge that best informs and advances equitable public policy and management (Goodrich et al., 2020).

3.3 Future Work

This study has been able to document trends in select contaminants by assessing current conditions and comparing them to results from different points in time, and it will be critical to track changes in the system with continued sampling. Beyond this study's focus on heavy metals and select legacy organics, (some of which, while at lower concentrations than before, are still present at levels of concern), microbial contaminants (e.g. protozoa, bacteria and viruses) and other contaminants of emerging concern (e.g. PCBs, PFAS) should continue to receive attention both in the estuary and more broadly (e.g., Allsing et al. 2022). Further investigating the relationship between sediment characteristics and contamination, including grain size and organic matter (e.g., Berry 2019), would also offer insight into the patterns and variability observed, as well as help shape approaches dealing with sediment management in the Tijuana River Valley (Dudek 2023). As highlighted above, analyzing species across taxonomic levels, life histories, and life stages could improve our understanding of bioaccumulation and biomagnification in the food web. Ultimately, coastal wetlands continue to prove to be an invaluable ecosystem best preserved through data-driven resource management.

With increasing information on the Tijuana Estuary, as well as along the California coast in general, a vital next step is to expand the focus on the estuary to the rest of the shared watershed and coastline. Data and monitoring upstream in the Tijuana River, or on the shorelines of Mexico, could highlight the need for preservation and restoration of natural lands that can mitigate contamination and

inform future management efforts, such as the upcoming \$300 million provided by the U.S. Environmental Protection Agency for upgrading the international wastewater treatment system. To shape research such as this to be more community-centered, it would be beneficial to work directly with the communities throughout the development and implementation of projects and programs. An example of future efforts that could result in community benefits are looking into the specific sources of contaminants to mitigate and better manage the area's surrounding environmental and public health. Such multi-benefit solutions would then help to simultaneously resolve both socioeconomic and environmental issues.

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Appendix A

Full List of Analytes Determined by Physis Laboratories

Metals	PBDEs	Pesticides
Al (Aluminum)	Mono-	4,4'-DDT
Sb (Antimony)	Di-	2,4'-DDT
As (Arsenic)	Tri-	4,4'-DDD
B (Boron)	Tetra-	2,4'-DDD
Ba (Barium)	Penta-	4,4'-DDE
Be (Beryllium)	Hexa-	2,4'-DDE
Cd (Cadmium)	Hepta-	4,4'-DDMU
Cr (Chromium)		alpha-Chlordane
Cu (Copper)		gamma-Chlordane
Fe (Iron)		cis-nonachlor
Pb (Lead)		trans-nonachlor
Hg (Mercury)		oxychlordane
Mg (Magnesium)		Aldrin
Mo (Molybdenum)		Dieldrin
Ni (Nickel)		Endrin
Se (Selenium)		
Ag (Silver)		
Sr (Strontium)		
V (Vanadium)		
Zn (Zinc)		
	PFCs	
	PFOS	
	PFAS	
		Phenols
		Alkyphenols
		Alkylphenol ethoxylates
		Nonylphenols

PCBs
Mono-Cl
Di-Cl
Tri-Cl
Tetra-Cl
Penta-Cl
Hexa-Cl
Hepta-Cl
Octa-Cl
Nona-Cl
Deca-Cl
Cl1 Homologs
Cl2 Homologs
Cl3 Homologs
Cl4 Homologs
Cl5 Homologs
Cl6 Homologs
Cl7 Homologs
Cl8 Homologs
Cl9 Homologs
Cl10 Homologs

Pyrethroids
bifenthrin
cyfluthrin
cypermethrin
permethrin
lambda-cyhalothrin
esfenvalerate (also fenvalerate)
Di(2-ethylhexyl)phthalate (DEHP)
Di-n-butyl phthalate (DBP)
Butyl benzyl phthalate (BBP)
Di-n-octyl phthalate (DOP)
Dimethyl phthalate (DMP)
Diethyl phthalate (DEP)

PAHs		
Two-ring	Napthalene	1,6,7-Trimethylnaphthalene 1-Methylnaphthalene 1-Methylphenanthrene 2,6-Dimethylnaphthalene 2-Methylnaphthalene C1-napthalenes C2-napthalenes C3-napthalenes C4-napthalenes
	Biphenyl	
Three-ring	Acenaphthalene Acenaphthene Fluorene	C1-fluorenes C2-fluorenes C3-fluorenes
	Phenanthrene	1-methylphenanthrene
	Anthracene	C1-Phenanthrenes & Anthracenes C2-Phenanthrenes & Anthracenes C3-Phenanthrenes & Anthracenes C4-Phenanthrenes & Anthracenes
Four-ring	Fluoranthene Pyrene Chrysene	C1-Fluoranthenes & Pyrenes C1-chrysenes C2-chrysenes C3-chrysenes C4-chrysenes
Five-ring		Benzo[a]anthracene Benzo[a]pyrene Benzo[b]fluoranthene Benzo[e]pyrene Benzo[g,h,i]perylene Benzo[k]fluoranthene Perylene
Six-ring		Dibenz[a,h]anthracene Indeno[1,2,3-c,d]pyrene Benzo[g,h,i]perylene

Appendix B

Comparable Thresholds

Resources:

- Mussel Watch Program: An Assessment of Two Decades (Kimbrough et al., 2008)
- NOAA's Screening Quick Reference Table (SQuiRTs) table (Buchman, 2008)
- Guidelines for Interpretation of the Biological Effects of Selected Constituents in Biota, Water, and Sediment (U.S.D.O.I., 1998): inorganic contaminants in horn snails
 - Selenium "Toxicity Threshold" for diet (red): > 3 mg/kg dw
 - Selenium "Background" for terrestrial invertebrates (blue): 0.1 to 2.5 mg/kg dw
 - Selenium "No Effect" for diet (green): < 2 mg/kg dw
 - Arsenic "No Effect" for invertebrates (green): 30 mg/kg dw
- Inorganics for arsenic, copper, mercury, selenium, zinc (toxicity, normal background, no effect) in fish
- Zeeman 2016, Draft dietary screening levels, San Diego Bay Bioaccumulation Study (Stransky et al., 2016)
 - Chlordanes "Avian Diet Screening Value" for Generalist (small piscivores): 513 ng/g ww

- DDTs “Most Sensitive Avian Diet Screening Value” for small piscivores: 11 ng/g ww
- PBDEs “Avian Diet Screening Value” for small piscivores: 12 ng/g ww
- LPAHs “Avian Diet Screening Value” for consumer of benthic invertebrates: 946 ng/g ww
- HPAHs “Avian Diet Screening Value” for consumer of benthic invertebrates: 46 ng/g ww
- Chlordanes Avian Diet SV for small piscivores: 196 ng/g ww
- Food & Drug Administration (FDA) Action Level (Kimbrough et al., 2008)

Threshold Resource References

- Buchman, M.F., 2008. Screening Quick Reference Tables (SQuiRTs), NOAA OR & R Report. United States, National Ocean Service., Office of Response and Restoration;United States, National Ocean Service,;United States, National Oceanic and Atmospheric Administration,;
- Kimbrough, K.L., Lauenstein, G., Christensen, J., Apeti, D., 2008. An assessment of two decades of contaminant monitoring in the Nation’s Coastal Zone.
- Stransky, C., Tait, K., Sheredy, C., Schottle, R., Kolb, R., Bernstein, B., 2016. Aquatic Food Web Bioaccumulation Study of San Diego Bay, Final Report, pp. 20-23.
- U.S.D.O.I., 1998. Guidelines for Interpretation of the Biological Effects of Selected Constituents in Biota, Water, and Sediment, in: Babbitt, B. (Ed.), National Irrigation Water Quality Program Information Report No. 3. U.S. Department of the Interior, Bureau of Reclamation, U.S. Fish and Wildlife Service, U.S. Geological Survey, Bureau of Indian Affairs, Denver, CO.

Appendix C

Definitions for Totals of Organic Compounds

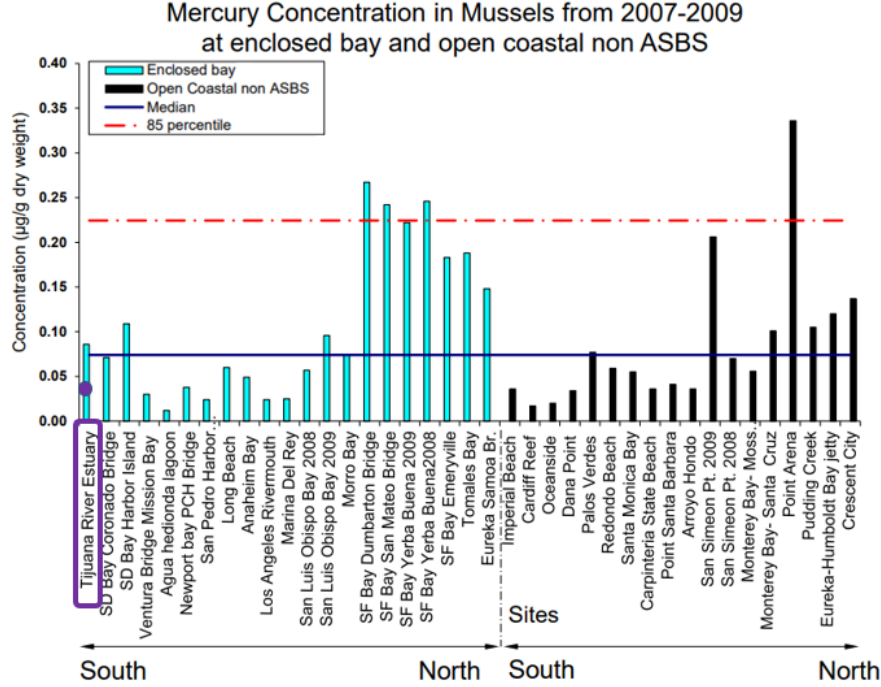
This study's definitions followed as close as possible to the NOAA National Status and Trends Mussel Watch Report and the San Diego Bay Bioaccumulation Study:

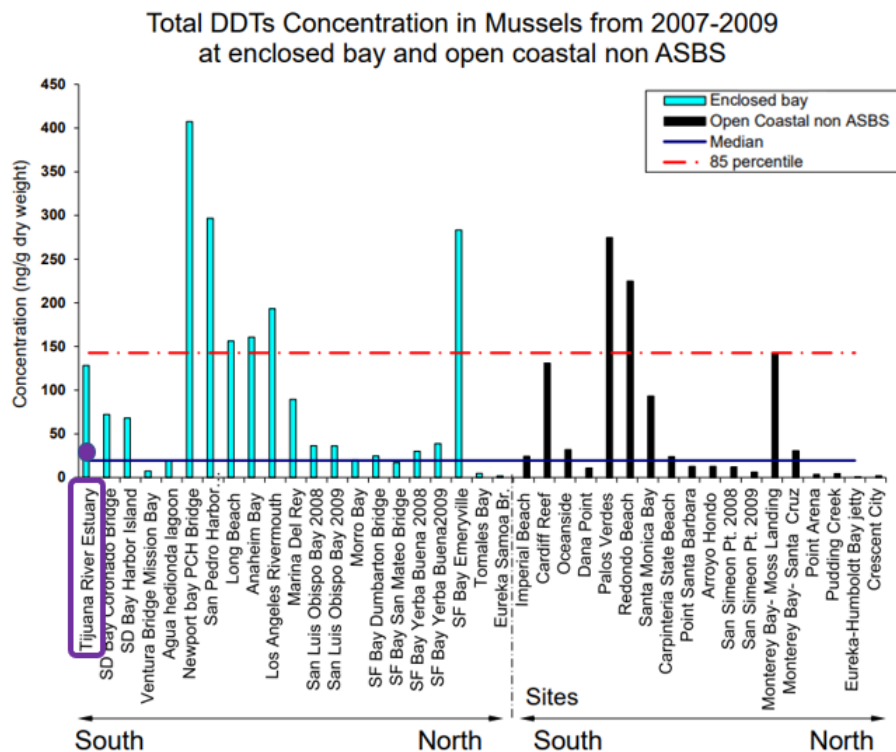
- NOAA Total Chlordanes (sum of 4 compounds): Alpha-Chlordane, Heptachlor, Heptachlor-Epoxyde, Trans-Nonachlor
- NOAA Total DDTs (sum of 6 compounds): 2,4'-DDD; 2,4'-DDE; 2,4'-DDT; 4,4'-DDD; 4,4'-DDE; 4,4'-DDT
- NOAA Total Dieldrin (Sum of 2 compounds): Aldrin, Dieldrin
- NOAA Total PAHs (sum of 19 parent PAH compounds plus 19 groups of alkylated PAHs):
 - Sum of 7 parent low molecular weight PAHs (with 2 or 3 rings): naphthalene, biphenyl, acenaphthene, acenaphthylene, fluorene, phenanthrene, anthracene
 - plus the sum of 12 parent high molecular weight PAHs (4 or more rings): fluoranthene, pyrene, benz[a]anthracene, chrysene, benzo[b]fluoranthene, benz[k]fluoranthene, benzo[e]pyrene, benzo[a]pyrene, perylene, dibenz[a,h]anthracene, indeno[1,2,3-cd]pyrene, benzo[ghi]perylene
 - plus the sum of 19 groups of alkylated PAHs: C1-Chrysenes, C1-Dibenzothiophenes, C1-Fluoranthenes/Pyrenes, C1-Fluorenes, C1-Naphthalenes, C1-Phenanthrenes/Anthracenes, C2-Chrysenes, C2-Dibenzothiophenes, C2-Fluorenes, C2-Naphthalenes, C2-Phenanthrenes/Anthracenes, C3-Chrysenes, C3-Dibenzothiophenes, C3-Fluorenes, C3-Naphthalenes, C3-Phenanthrenes/Anthracenes, C4-Chrysenes, C4-Naphthalenes, C4-Phenanthrenes/Anthracenes
- San Diego Bay Low Molecular Weight PAHs (LPAHs):
 - Two Ring: Naphthalene; 1-methylnaphthalene; 2-methylnaphthalene; 2,6-dimethylnaphthalene; 1,6,7-Trimethylnaphthalene; Biphenyl
 - Three Ring: Acenaphthylene; Acenaphthene; Fluorene; Phenanthrene; 1-methylphenanthrene; Anthracene
- San Diego Bay High Molecular Weight PAHs (HPAHs):
 - Four Ring: Fluoranthene; Pyrene; Chrysene; Benzo(a)anthracene
 - Five Ring: Benzo(b)fluoranthene; Benzo(k)fluoranthene; Benzo(a)pyrene; Benzo(e)pyrene; Perylene; Dibenz(a,h)anthracene
 - Six Ring: Indeno(1,2,3-cd)pyrene; Benzo(g,h,i)perylene
- Physis Total PBDEs: PBDE017; PBDE028; PBDE047; PBDE066; PBDE071; PBDE085; PBDE099; PBDE100; PBDE138; PBDE153; PBDE190; PBDE183; PBDE154

Note: Actual specific analytes utilized in sum varied by contaminant and organic dependent upon availability; analysis available upon request

Appendix D

Example of Mussel Watch California Coast Comparison with Mercury and DDTs





Figures adapted from (SWRCB, 2010; full citation on page 58); 2021 average data point for the Tijuana River Estuary overlaid onto graph

Appendix E

Seasonal and Spatial Comparisons in Horn Snails

When assessing spatial and seasonal differences within the Tijuana River Estuary’s biota, horn snails were the organism available at each of the sites sampled during the spring and fall. The horn snail species collected from each site varied depending on which was available, but both native (*Cerithideopsis californica*) and invasive (*Batillaria attramentaria*) species were determined to be comparable as they resulted in similar concentrations when both were collected from Boca Rio. Standards for inorganic contaminants in invertebrates were scarce with only the metalloids (arsenic and selenium) having thresholds from federal guidelines assessing biota contamination (U.S.D.O.I., 1998), which limited the ability to assess whether results could be considered high or low in horn snails.

Out of the two available thresholds for horn snails, arsenic concentrations were low while selenium had one toxic result. All arsenic concentrations were well below the level at which “No Effect” takes place for invertebrates (30 μg dw), ranging from 6.4 to 9.9 μg dry weight (Figure below). Selenium concentrations in snails were slightly below the “No Effect” threshold value for fish and wildlife diets (2 μg dw) for all May 2021 samples, but differed in

October 2021 as the concentration for snails from Boca Rio was 0.29 $\mu\text{g dw}$ above the “Toxicity Threshold” of 3 $\mu\text{g dw}$ (U.S.D.O.I., 1998). Selenium is also notable as the only inorganic contaminant with a substantial difference between the dry- and wet-seasons for Boca Rio. The post-wet-season has all-around low levels of selenium while toxic levels are observed for the post-dry-season. Although there is no threshold to give context for chromium, a similar larger increase of 1.9 $\mu\text{g dw}$ is seen after the dry-season. Nickel was the only inorganic with a substantial decrease of 9.9 $\mu\text{g dw}$ after the dry-season. Organic contaminant concentrations in snails were all far below the screening values for which avian receptors could potentially experience harmful effects. Total DDTs, PBDEs, and HPAHs had higher concentrations in the wet-season but are not substantially different as the values are all far below the avian diet screening values (Figure below).

Discussion On Seasonal Differences

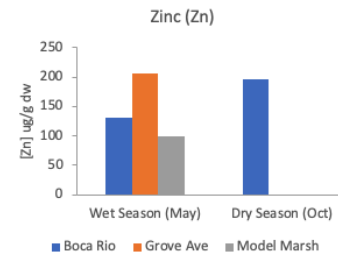
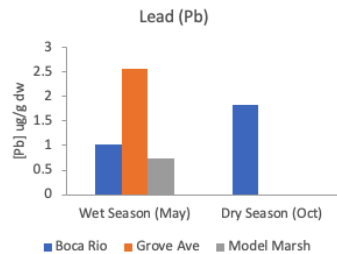
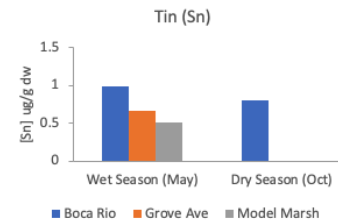
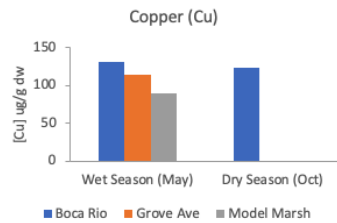
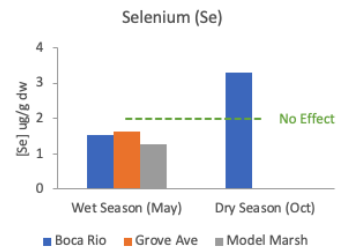
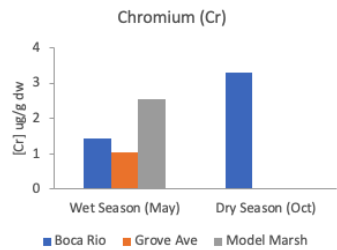
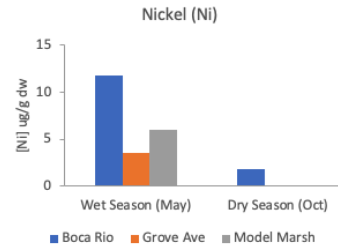
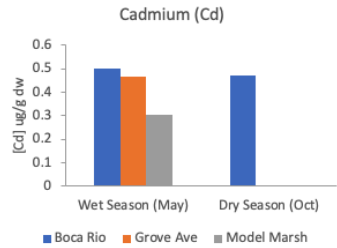
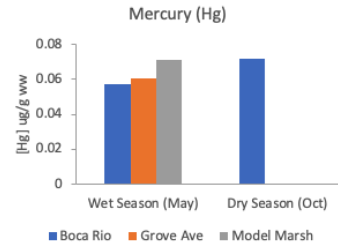
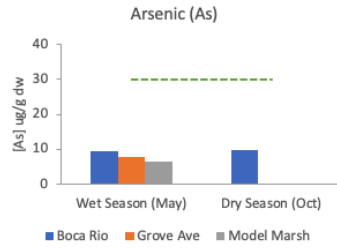
Like spatial differences, seasonal differences within an estuary are possible (Riveles & Gersberg, 1999; Talley et al., 2022). For sediment, two sites were sampled from to assess seasonal differences: Boca Rio and Grove Avenue. Both sites consistently had decreases in inorganics from spring to fall, with more substantial decreases for copper, lead, and zinc, going from above the ERL to below. Organics experienced substantial decreases at both sites as well, particularly for chlordanes, DDTs, PBDEs, and PCBs. The only contaminant in sediment that experienced an increase from spring to fall was total PCBs at Boca Rio, going from below the ERL to above. As was observed in a European estuary, the seasonal distribution of metals in surface sediment might be attributed as a flushing and retention mechanism from seasonally fluctuating environmental conditions (Buggy & Tobin, 2008).

Differences in biota can also be observed within an estuary, such as oysters in San Diego Bay that varied in contamination levels between the summer and winter, specifically for copper, zinc, chlordane, PCB and PAH thresholds (Talley et al., 2022). In the Tijuana Estuary, horn snails were collected from Boca Rio to assess seasonal differences. Patterns were not as distinct, but this could be due to only one site being assessed (Grove, which tended to have the highest concentrations, was not sampled in the fall). However, a marked increase in selenium occurred during the dry season, and the horn snail composite rose above the toxicity threshold at which there could be adverse effects on some fish and wildlife species. Though unlikely, this suggests that there could still be contaminated flows into the Tijuana River Estuary occurring during the summer that accumulate higher concentrations of selenium, but more consistent data would be needed to determine whether this pattern occurs every season. Overall, stronger patterns in the Tijuana River Estuary were observed across sites though seasonal differences were also present. Dominance of spatial over seasonal

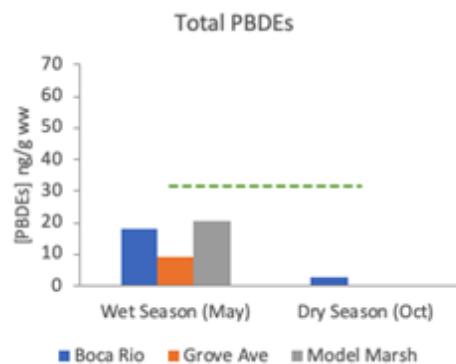
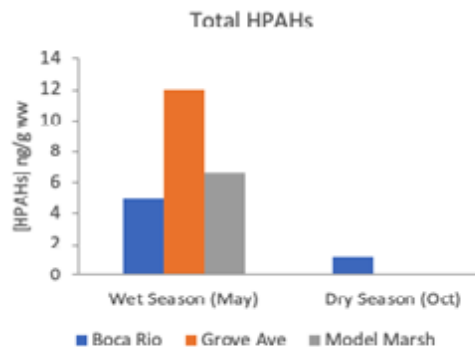
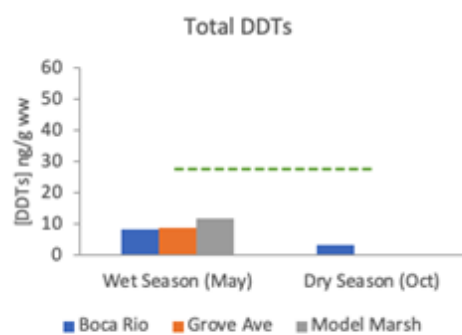
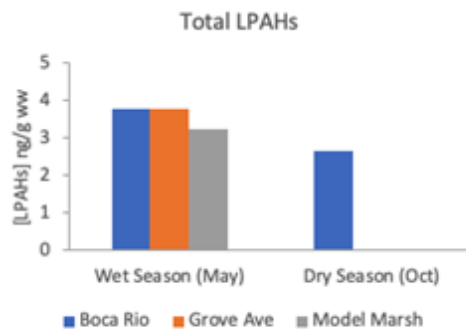
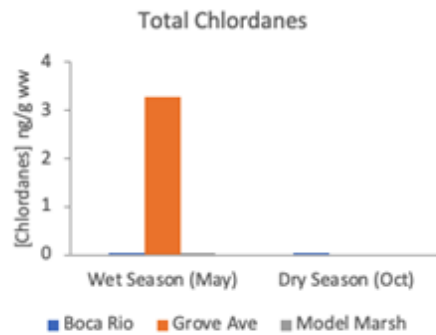
variability have been reported for other estuaries in North Carolina, U.S. and Portugal (Hyland et al., 2004; Mucha et al., 2005). Given these site and season differences, future studies might also include this sampling structure to better understand patterns and their implications.

References

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Inorganic Contaminant Concentrations in Horn Snails Across Tijuana River Estuary Sites (2021). Both Dietary. No Effect threshold for selenium and No Effect threshold for arsenic in invertebrates are shown in green. (U.S.D.O.I., 1998)



Organic Contaminant Concentrations in Horn Snails. Dietary screening levels for sensitive avian species developed for San Diego Bay bioaccumulation studies by Bay et al (2016) and Stransky (2016). Screening values are for species that rely on benthic invertebrates for food (from Zeeman 2016).

Appendix F

Reflection on Mussel Watch Program Sampling Protocol for Tijuana River Estuary

Although the Mussel Watch Program (MWP) is one of the most consistent, long-term environmental monitoring programs, having ran since 1986, it was found that the earliest records in the Tijuana River Estuary only had fish data. As mussel data started in 2007, the area would benefit from having continuous, long-term monitoring efforts from the Mussel Watch Program. The difference in sampling between mussels and fish was likely due to the differences in protocol that the State MWP (1977-2010) and the National MWP (1986 - present) had. It is recommended for the large-scale program to contact local experts to ensure the Tijuana River Estuary's data continues to be collected at the most beneficial location, specifically from the Boca Rio site as the location with the most long-term, consistent data. The site was revisited in 2018, but only the nearby coastal site of North Jetty at Imperial Beach was collected due to no mussels being found in the historic monitoring site. However, a great abundance was available for this study only a few years later. This may have been due to either not finding the correct location or shifts in the bivalves' availability. Invasive oysters have previously been reducing mussel populations but mussels are now abundant again.