CHAPTER 7 Invertebrate and Fish Trawls



Cover photos

Left: Pacific Electric Ray (Torpedo californica)

Left Center: Scientists from the Sanitation Districts' Marine Biology Laboratory setting the net for a trawl. Left to Right: Brent Haggin (Biologist II), Bill Power (Supervising Scientist, Retired), Percival Harper (Boat Deck Hand), Norbert Lee (Senior Lab Technician).

Right Center: Supervising Scientist of the Sanitation Districts' Marine Biology Laboratory Terra Petry, operating the crane during a trawl survey.

Right: Octopus californicus (North Pacific Bigeye Octopus)

INTRODUCTION

The Sanitation Districts of Los Angeles County (Sanitation Districts) own and operate the Joint Water Pollution Control Plant (JWPCP), which discharges secondary treated effluent into the Pacific Ocean pursuant to the Waste Discharge Requirements and National Pollutant Discharge Elimination System (NPDES) permit issued by the Los Angeles Regional Water Quality Control Board (LARWQCB Order No. R4-2017-0180; Appendix 1.1). The 2018-2019 trawl data for this report were collected semi-annually under the current NPDES permit. Monitoring and reporting requirements for the NPDES permit are specified in the Monitoring and Reporting Program (MRP) portion of the NPDES permit. The MRP has specified a number of monitoring elements for the JWPCP, including semi-annual trawls (winter/summer) since 2006. Prior to 2012, trawl data was collected quarterly (spring, summer, fall, and winter). From 2012-2019, due to take restrictions under the Marine Life Protection Act, only permit-required (winter/ summer) trawls were performed.

Epibenthic invertebrates and demersal fish live in close association with the sediments and are exposed, by direct contact and through ingestion, to contaminants associated with those sediments. As such, they can be valuable indicators of the integrated effects from the discharge of effluents and environmental factors on the soft-bottom ecosystem including changes in food resources, habitat, and oceanographic conditions. Therefore, monitoring the community structure and health of epibenthic invertebrates and demersal fishes is an integral part of the JWPCP NPDES monitoring program. However, distinguishing impacts due to the discharge of effluents from other anthropogenic stressors or natural oceanographic changes can be a considerable challenge.

During the early years of this monitoring program (1970s), effluent discharge-related impacts were observed in both demersal fishes and invertebrates. Community structure and organism health in areas closest to the discharge

were identified as altered based on community assemblages analyzed using multivariate statistics and the Fish Response Index (FRI, a measure of fish community response to pollution), and the prevalence of disease that included fin erosion, tumors, and lesions. While an individual stressor was not likely responsible for the observed responses, the combined stresses of organic over-supply, toxicants associated with discharged particulates, and the physiologic effects of hydrogen sulfide and depression of dissolved oxygen, were likely contributors. Subsequent JWPCP treatment plant upgrades and source control from the 1970s through the 1990s have resulted in the restoration of healthy epibenthic invertebrates and demersal fish communities. However, continued monitoring of the Palos Verdes shelf and slope is important to detect any potential future impacts associated with current discharges, other anthropogenic (e.g. human activities contributing to climate change), and natural shifts in community composition associated with general oceanographic conditions (e.g. El Niño).

Chapter overview

The objectives of this chapter are to assess the current (2018-2019) condition of demersal fish and epibenthic invertebrate health and communities off Palos Verdes in the vicinity of the JWPCP discharge, evaluate historical changes and trends, and compare current conditions off Palos Verdes to those observed across the Southern California Bight (SCB).

This chapter is one component of the JWPCP 2018-2019 Biennial Receiving Water Monitoring Report (LACSD 2020a). The complete report includes results and analyses for all MRP receiving water monitoring requirements as well as the associated appendices. Electronic copies of the JWPCP 2018-2019 Biennial Receiving Water Monitoring Report can be downloaded through the Sanitation Districts' website, <u>www.lacsd.org</u>.

MATERIALS AND METHODS

Field methods

Samples were collected at 16 stations (**Figure 7.1**), along four cross-shore transects (T0, T1, T4, and T5), and four isobaths (23 m, inner shelf; 61 m, mid-shelf; 137 m, outer shelf; and 305 m, upper slope). In 2018-2019, trawl surveys of the Palos Verdes shelf were conducted in the months of February and August. Temporal comparisons of the 23, 61, and 137 m isobaths incorporate data collected from monitoring surveys conducted from 1973-2019, and temporal comparisons of the 305 m isobaths utilized data collected from 1991 (the first year of sampling at this isobaths) through 2019.

As stipulated in the MRP, the same type of otter trawl used in Southern California Bight regional monitoring projects (SCCWRP 2013) was used to sample off Palos Verdes. All field and laboratory analyses for trawl samples were performed in accordance with the Sanitation Districts' Marine Biology Laboratory standard operating procedures (SOPs) for trawl monitoring (Appendix 7.1). Fishes and invertebrates captured during each trawl were identified to the lowest possible taxon and counted. The standard length of all fishes was measured and reported in size classes at one-centimeter increments. All invertebrates and fishes were examined for disease, external parasites, and other external anomalies, and weighed as species lots (i.e. a composite of all individuals of a species).

Debris collected by trawl was classified into natural and anthropogenic categories. Natural debris included marine vegetation, terrestrial vegetation and benthic debris such as dead gorgonian sea fans, rocks, and empty shells. Debris was reported following regional monitoring protocols, which was modified from previous regional surveys for compatibility with upstream debris sampling methods (SCCWRP 2013; Appendix 7.1).

Data analysis

All 2018-2019 trawl data are reported in the annual receiving water data summary reports (LACSD 2019, 2020b). Abundance, biomass, number of species, Shannon Diversity (diversity), Margalef's Species Richness (species richness), Simpson's Dominance Index (dominance), and Pielou's Evenness Index (evenness) were determined separately for invertebrate and fish catches in each sample. The diversity, species richness, and evenness indices measure the diversity of a sample as a function of the distribution of individuals among species. Size class ranges were determined for fish over the two -year period.

To gauge the condition of the Palos Verdes fish community relative to discharge impacts over time, a demersal fish biointegrity index, the Fish Response Index (FRI: Allen et al. 2001a) was also calculated. The FRI is a multivariate index that provides the abundanceweighted average pollution tolerance score of all the fish species in a sample. The FRI was designed to recognize that diverse fish communities are expected to occur at different depths and regions of the SCB. However, the FRI controls for this natural variability by utilizing a pollutant tolerance index to determine if observed populations are predominantly represented by pollution-tolerant (an indication of possible impact) or pollution-intolerant species. The pollution tolerances of the species were determined from ordination analysis of a large multi-year data set composed of trawl fish survey results across a range of shelf and slope depths, habitats, and sediment pollutant levels within the SCB. The FRI was calibrated and validated using almost 30 years of data around wastewater outfalls in depths between 20 and 215 m, and correlated with a well-documented pollution gradient (Allen et al. 2001a). This approach is similar to that used in the Benthic Response Index (Smith et al. 2001) to assess infaunal community condition in the SCB (Bergen et al. 2001). The reference condition threshold value of 45 was computed for the FRI as the 90% tolerance interval bound for observations from areas where the BRI was in reference condition. The FRI was developed for three depth zones; inner shelf (9-40 m), middle shelf (30-120 m), and outer shelf (100-215 m). An FRI score at or below 45 indicates that the fish community is in reference condition. A detailed presentation of analytic methodology is provided in Appendix 7.2.

Spatial and temporal trends were examined by comparisons of patterns of several parameters relative to sampling depth (inner shelf to upper slope), distance from the outfall, and wastewater treatment period. Wastewater treatment periods included baseline (1973), advanced primary (1974-1983), partial secondary (1984-2002), full secondary (2003-2017) and the current monitoring period (2018-2019).

PRIMER v7 (Plymouth Routines in Multivariate Ecological Research) multivariate statistical software was used to examine the spatial patterns of the invertebrate and fish assemblages on the Palos Verdes shelf (Clarke et al. 2014). Analyses included hierarchical clustering with group-average linking based on Bray-Curtis similarity indices, and ordination clustering of the data using non-metric multidimensional scaling (NMDS). Prior to the calculation of the Bray-Curtis indices, the abundances were square-root (fish) or fourth-root



Figure 7.1 Stations Sampled by Trawl

Map of semi-annual trawl monitoring surveys. The trawl was towed on the bottom along the isobath of each station for 10 minutes at approximately 1 m /sec, thus traversing about 0.6 km at each station. The Marine Protected Areas in Palos Verdes, Point Vicente and Abalone Cove State Marine Conservation Areas (SMCA), are outlined in gray.

(invertebrate) transformed. To visually depict the relationship of benthic infaunal communities to each other over time and space, a similarity profile (SIMPROF) analysis was used to confirm non-random structure of cluster groups (Clarke et al. 2008). The SIMPER ("similarity percentages") routine was also used to determine inter- and intra -group species differences (Clarke 1993). Analysis of similarity (ANOSIM), a nonparametric multivariate ANOVA using ranked distance or dissimilarity between pairs of samples or variables (Bray-Curtis), was used to test for differences in community composition between depths (shallow versus deep), distance from the outfall (near to far), and across time as the level of wastewater treatment increased. ANOSIM significance is based on the calculation of the R statistic, which measures the difference between sampling groups (Clarke 1993).

RESULTS

Invertebrates, 2018-2019

Megabenthic invertebrates were taken in abundance; a total of 118,400 individuals (an average of 1850 per trawl) representing 118 taxa from 6 phyla and weighing 2194 kg were collected in 2018-2019 (Figure 7.2, Appendix 7.3). The sea urchin Strongylocentrotus fragilis was the most abundant invertebrate overall, averaging 934 individuals per haul and accounting for 26% of the total abundance. S. fragilis, along with five additional species including the sea urchin Brisaster townsendi, the shrimp, Sicyonia ingentis, the pelagic red crab Pleuroncodes planipes, the brittle sea star, Ophiura luetkenii, and the painted urchin, Lytechinus pictus, comprised 95% of the total catch. Besides these abundant species, other frequently occurring species that occurred in at least 50% of the trawls included the cymothoid isopod Elthusa vulgaris, the sea star Astropecten californicus, the shrimp S. ingentis, the octopus Octopus rubescens, the sea slug Pleurobranchaea californica, and the spiny brittle star Ophiothrix spiculata (Appendix 7.3). Six species accounted for 95% of biomass including the urchins S. fragilis and B. townsendi, the shrimp S. ingentis, the pelagic red crab Pleuroncodes planipes, the brittle star O. luetkenii, and the sea cucumber Apostichopus californicus (formerly Parastichopus californicus).

Invertebrate metrics including abundance, biomass, number of species, diversity, species richness, and evenness for all individual trawl samples, and number of individuals per species are listed in Appendix 7.3. Megabenthic invertebrate community metrics varied among stations and between surveys during the sampling years (Figure 7.2, Appendix 7.3). Across all trawls for 2018-2019, summed species abundance ranged from 36 to 22,310 individuals, and the number of species ranged from 6 to 23. Shannon diversity values ranged from 0.04 to 2.17. The fewest species (6 or fewer) during 2018-2019 were recorded at station T0-23 during winter 2019, while the highest value (23 species) was recorded at station T1-137 also in winter 2019. Biomass values in 2018-2019 ranged from 0.27 kg to 223 kg. Evenness ranged from 0.02 to 0.84 (Figure 7.2, Appendix 7.3).

Fishes, 2018-2019

Results of fish catches in 2018-2019 are provided in Figure 7.3 and Appendix 7.3. Fish metrics including abundance, biomass, number of species, diversity, species richness, evenness, and FRI for all individual trawl samples, and number of individuals per species are listed in Appendix 7.3. A total of 29.351 fishes, representing an average of 458 individuals per trawl, and weighing 681 kg total, were taken on the Palos Verdes shelf and slope in 2018-2019. Seventynine species of fishes were collected, representing 37 families (Appendix 7.3). Six species of fishes occurred in at least 50% of the trawls, including Pacific Sanddab (Citharichthys sordidus), Dover Sole (Microstomus pacificus), California Tonguefish (Symphurus atricaudus), Slender Sole (Lyopsetta exilis), English Sole (Parophrys vetulus), and Hornyhead Turbot (Pleuronichthys verticalis) (Appendix 7.3).

Twenty fish species accounted for 95% of the abundance (Appendix 7.3). The top six of those species included Pacific Sanddab (family Paralichthyidae), Slender Sole and Dover Sole (family Pleuronectidae), Stripetail Rockfish and Halfbanded Rockfish (*Sebastes saxicola* and *Sebastes semicinctus*, family Scorpaenidae), and Speckled Sanddab (*Citharichthys stigmaeus*, family Paralichthyidae) (Appendix 7.3). Together, these six species accounted for over 70% of all fish captured during 2018-2019.



Figure 7.2 Invertebrate Catch Metrics, 2018-2019

Results from invertebrate catches of Winter and Summer trawl surveys in 2018-2019 at each station expressed as Abundance, Biomass, Shannon diversity (Diversity) and Pielou's evenness (Evenness).







Figure 7.3 Fish Catch Metrics, 2018-2019

Results from fish catches of Winter and Summer trawl surveys in 2018-2019 at each station expressed as Abundance, Biomass, Shannon diversity (Diversity) and Fish Response Index (FRI). Red line represents maximum FRI score (45) associated with reference condition. Twenty species of fishes accounted for 90% of the biomass. The top three of those species accounting for 50% of the biomass were Pacific Sanddab, Slender Sole, and Dover Sole.

Size class for all fish species ranged from 1 to 78 cm with a mode of 13 cm (**Appendix 7.4**). The modes for transects T0, T1, T4, and T5 ranged between 12 and 13 cm. The modes for each depth (23, 61, 137, 305 m) also ranged between 9 and 15 cm.

Fish abundance ranged from 1 to 8909 fishes, while biomass values ranged from 0.044 kg to 172.92 kg (Figure 7.3). Diversity ranged from 0.64 to 2.3. The number of fish species caught ranged from 6 to 24 species over the twoyear period (Appendix 7.3). Species richness ranged from 1.2 to 3.8. The FRI indicated reference condition for all 64 samples from 2018-2019 except for at station T1-23 in summer 2018, with FRI scores ranging from 14 to 46 (Figure 7.3, Appendix 7.3).

Organism health, 2018-2019

Organism health, as tracked by occurrences of anomalies, is depicted in **Table 7.1**, and further details of Palos Verdes anomaly data can be found in **Appendix 7.5**. There were few external anomalies in invertebrates and fish collected in 2018-2019. Anomalies occurred in <0.1 to 10.7% of the ten fish species affected. In total, there was <0.05% of total invertebrate catch and 0.4% of the fish catch found with anomalies in 2018-2019. Most anomalies observed occurred in flatfish.

The copepod eye parasite (*Phrixocephalus cincinnatus*), which primarily afflicts Pacific Sanddab, accounted for most of the anomalies in that species in 2018-2019. Eye parasites were only found on Pacific Sanddab, with an overall incidence rate of 0.6% for that species in Palos Verdes.

Color anomalies such as ambicoloration were observed most frequently amongst several species of flatfish; California Halibut (*Paralichthys californicus*) had the highest percentage of occurrences (10.7%), followed by Hornyhead Turbot (8.3%), and Slender Sole (0.2%).

Pseudotumors only occurred in Dover Sole, and were the most prevalent anomaly in that species, with 1.5% frequency. Skeletal deformities occurred in Dover Sole, which had an incident rate of <0.1% for this anomaly; the most common deformity was found on the caudal region.

Other types of anomalies found included one individual California Tonguefish with a black internal cyst (0.2% incidence), an individual Dover Sole with a healed bite mark (<0.1% incidence), and one individual Pacific Sanddab with a missing eye (<0.1% incidence). The missing eye was likely a result of the copepod eye parasite, *Phrixocephalus cincinnatus*.

External anomalies were rarely seen on invertebrates. There was one invertebrate species with an external anomaly. *Paralithodes californiensis* (California king crab), was occasionally parasitized with *Briarosaccus sp.*, a parasitic barnacle (Appendix 7.5).

Trawl debris, 2018-2019

A data summary of anthropogenic debris caught in trawls during 2018-2019 is provided in **Appendix 7.6**. Anthropogenic debris occurred across all stations during the two-year period. Plastic was the most common type of anthropogenic debris, constituting 65% of all anthropogenic debris pieces counted, and was found across 90% of all stations off Palos Verdes. Plastic included items such as plastic bags, monofilament fishing line, nets and plastic traps, polypropylene rope, and movie filmstrip. Other items found included glass (mostly beer bottles), and metal, including beer cans, and fishing gear including traps, hooks, lures, and metal line. Miscellaneous types of anthropogenic debris included items such as clothing, and scraps of cloth.

DISCUSSION

Current condition, 2018-2019

Fish community patterns, 2018-2019

Current fish community condition, as measured by average FRI scores for each station, was below the reference threshold, indicating the entire sampled area of the Palos Verdes shelf is in reference condition (**Figure 7.4**). All but one FRI score for each individual sampling event were indicative of reference condition. T1-23 taken during Summer 2018 did not meet the reference condition threshold. However, during the three

Table 7.1 Organism Health, 2018-2019

Percentage individuals of each species of fish and invertebrates with different anomaly types at each transect and percent anomalous trawl species collected at depths of 23-305 m on the Palos Verdes shelf and slope (PV), 2018-2019. For regional comparison, values from the 2013 SCB survey (Walther et al. 2017) are in **blue***.

								PV	Bight'13
	-	By Transect		Total	Total	Anomaly	Anomaly		
Anomaly	Common Name	Т0	T1	T 4	Т5	Affe cte d	Catch	(%)	(%)
Color Anomaly	Bigmouth Sole	0	0	0	1	1	105	1.0	0.7
	California Halibut	3	0	0	0	3	28	10.7	3.4
	California Tonguefish	3	0	1	2	6	961	0.6	1.4
	Curlfin Sole	-	0	1	1	2	36	5.6	5.1
	Dover Sole	5	6	3	3	17	3,244	0.5	<0.1
	English Sole	1	0	0	0	1	166	0.6	0.1
	Homyhead Turbot	2	1	1	0	4	322	1.2	1.2
	Slender Sole	2	0	0	0	2	3,061	0.1	0.1
	Speckled Sanddab	0	0	0	1	1	2,521	0.0	<0.1
Skeletal Deformity	California Tonguefish	0	0	0	1	1	961	0.1	0
	Dover Sole	0	0	0	1	1	3,244	<0.1	0
	Pacific Sanddab	1	0	0	0	1	8,909	<0.1	<0.1
Parasite	Pacific Sanddab	5	17	4	8	34	8,909	0.4	1.1
	California King Crab	1	2	1	-	4	6	0.7	21.4
Tumor	Dover Sole	16	6	9	15	46	3,244	1.5	0.6
	Homyhead Turbot	0	0	0	1	1	322	0.3	0
Total for All Invertebrate Taxa		1	2	1	0	4	118,400	<0.1	<0.1
Total for All Fish Taxa		38	30	19	34	121	29,351	0.4	0.5

"-" Species not found in this transect

"0" Species found but no anomalies occurred

*Note: does not account for anomaly types in 2013 SCB survey that were not encountered

in PV durina 2018-19

other sampling events conducted over the twoyear period, the FRI at T1-23 was indicative of reference condition. Considering the relatively high amount of variability associated with field collected data, minor excursions in any one sampling event are to be expected. Averaging the four sampling events across the two-year period sufficiently addresses such variation. A more detailed discussion of anomalously high FRI values is provided under the Historical Trends section below. A closer examination of the T1-23 summer 2018 data revealed the reason for the increase in FRI resulted from a large number of medium to high-pollution tolerant species in the sample, primarily Pacific and Speckled Sanddabs (*Citharichthys sordidus* and *C. stigmaeus*, respectively), while lacking in abundance of any low pollution-tolerant species. The higher the pollution-tolerance of the species, the fewer individuals of that species it takes to indicate pollution impact at that station.

Legacy sediment contamination (Chapter 4) and effluent water quality (Chapters 1 and 2) can influence distributions of invertebrates and fishes. The dominant direction of the subthermocline current and effluent plume is to the northwest of the outfall. Distributions of both invertebrates and fishes must be interpreted with this water movement in mind due to the continuing effluent discharge plume and historic sediment contamination that could potentially impact biota. There were no discernable impacts to fish communities as measured by the FRI in 2018-2019 correlating to proximity to the JWPCP outfall or effluent plume. Depth remained the primary determinant of organism distribution on the shelf and slope off Palos Verdes in 2018-2019. The Palos Verdes FRI scores are consistent with those seen elsewhere in the SCB in 2013, where 11% of the inner shelf was non-reference as measured by the FRI (Walther et al. 2017).

Spatial patterns in fish assemblages are expected to occur across different depths throughout the region (Walther et al. 2017). These natural patterns in Palos Verdes fish communities were evaluated and confirmed by comparing diversity between depths and distance from outfall (Figure 7.3), as well as by comparing fish assemblage structure over depth and by transect in 2018-2019 using non-metric multidimensional scaling (nMDS) ordination of trawl fish assemblages (Appendix 7.7). NMDS ordination of trawl fish assemblages, based on the Bray-Curtis similarity index of square root transformed mean abundance, strongly grouped fish assemblages by depth (Appendix 7.7). An ANOSIM test confirmed the strong differences between fish assemblages at each depth, with assemblages at each depth being statistically unique to one another (R=0.97, p=0.0001; Appendix 7.7). Species that typified 137 m included Pacific Sanddab, Dover Sole, Slender Sole, and Stripetail Rockfish. ANOSIM also distinguished between fish assemblages by transect, except between T4 and T5 (R=0.295, p=0.0001), though to a much lesser degree than by depth. The small R-values indicated that there was an overlap in species that characterized most assemblages between transects. This was further demonstrated by the SIMPER results (Appendix 7.7) in which individual fish species such as

Pacific Sanddab and Dover Sole were large contributors to the assemblages of multiple transects.

Sites on the southern side of the peninsula, adjacent to the discharge (T4 and T5), differ in some physical characteristics from those on the western side. These sites have a much steeper and more gullied slope, and, on both the shelf and slope, have a greater legacy sediment contaminant burden from pre-1970s discharge deposits. Shallow sites (23 m isobath) located on the southern side of the peninsula have been subject to continuous sediment inputs from the Portuguese Bend landslide (Kayen et al. 2002). The physical structure of the discharge pipe itself and its associated armor rock also create a different habitat affecting organism distributions. These physical factors, along with exposure to historic sediment contamination, were likely more influential factors affecting organism distribution amongst transects, rather than exposure to the currently discharged effluent. A discussion about legacy outfall effects is contained in a later section.

Fish lengths were larger on the upper slope (305 m) compared to all other depths (Appendix 7.4). The median size class for upper slope was 16 cm, compared with that of the other three depths, which contained size classes that were more similar to one another, with averages ranging from 10-11 cm. Some of the largest fish species at 305 m included Longnose Skate (*Raja rhina*), Spotted Ratfish (*Hydrolagus colliei*), and Pacific Hagfish (*Eptatretus stoutii*). There was no difference detected in fish length when comparing transects (Appendix 7.4). The fish size classes are consistent with those seen elsewhere in the SCB in 2013 (Walther et al. 2017).

Epibenthic invertebrate community patterns, 2018-2019

There is no currently accepted multivariate index for epibenthic invertebrate community condition such as the FRI (Walther et al. 2017). Therefore, evaluation of epibenthic invertebrate community condition was conducted using nonmetric multidimensional scaling (nMDS) ordination of trawl invertebrate assemblages, based on the Bray-Curtis similarity index of fourth root transformed mean abundance. In this analysis, depth was by far the largest factor in determining differences between assemblages



Figure 7.4 Fish Response Index (FRI) 1973-2019

FRI scores by wastewater treatment level at transect (T0, T1, T4, T5) at shelf depths (23, 61 and 137 m). Scores below red reference threshold value line (FRI =45) are in reference condition.

(Appendix 7.5). The stations clearly grouped together mostly by depth rather than by transect/ proximity to the JWPCP outfall. Groups that were statistically similar, as measured by SIMPROF, are depicted within a circle. An apriori ANOSIM test detected differences between invertebrate species and their abundances. The two-way ANOSIM indicated statistically significant differences between depths (global R=0.9, p=0.0001), and some smaller but still statistically significant differences in community assemblages between transects (R=0.4, p=0.0001). These statistical results are further evidence that depth was likely a major contributor to invertebrate assemblage patterns during 2018-2019 (Appendix 7.7). Species contributing most to the differences in assemblages between depths included Sicyonia *ingentis* (most characteristic of outer shelf), Brisaster townsendi (characteristic of upper slope), Strongylocentrotus fragilis (characteristic of upper slope, and, to a lesser extent, outer shelf), *Ophiura luetkenii* (characteristic of mid-shelf), and Astropecten californicus (characteristic of inner shelf). A complete list of species that contributed most to each depth and transect assemblage as determined by SIMPER is given in Appendix 7.7. When pairwise comparisons were made by transect to determine any effects of distance from outfall, there were some patterns. The transect farthest from the outfall (T0) had an invertebrate assemblage, across all depths, that was different from all of the other transects, and was most different from the assemblage of the transect closest to the outfall, T4. Species that most contributed to the differences between these two transects were greater abundances of Strongylocentrotus fragilis and Ophiura luetkenii at T4, and greater abundances of Brisaster townsendi at T0 (Appendix 7.7). The differences seen between assemblages at T0 and T4 could be due to differences in the topography and sediment grain size at T0 and are not likely due to a legacy outfall effect at T4 (a discussion on legacy outfall effects is contained in the following sections).

Invertebrate and fish community metrics, 2018-2019

Most trends in invertebrate and fish catch metrics across Palos Verdes were similar to spatial patterns seen in recent years (Appendix 7.7). There was no general pattern across transects. Transect T4 had the highest invertebrate abundance and Transect T0 had the highest abundance of fishes. Diversity at individual stations was consistently lower when there were large catches of one or two species of fishes and/ or invertebrates. Patterns of total invertebrate abundance mirrored variation in populations of *Ophiura luetkenii* due to the overwhelming dominance of this brittle star (Appendix 7.3). For example, high invertebrate abundances with 2,766 to 14,568 individuals reflected large hauls of O. luetkenii (2,573 to 11,886), recorded at station T5-137 in the winter and station T4-61 during the both winter and summer surveys. In addition, large numbers of the urchin Brisaster townsendi (2,574 to 3,181) contributed to high total abundances at T0-305. The low diversity (≤ 0.5) and evenness (<0.2) values observed at T4-61 and T0-305 were caused by the numerical dominance of the echinoderms, O. luetkenii, B. townsendi and S. fragilis.

High abundances of Pacific Sanddab at T1 -137 contributed to the pattern of high abundance and low diversity for fishes (Appendix 7.3). The outer shelf (137 m) produced the largest catches while the inner shelf (23 m) had the lowest invertebrate and fish catches (Figures 7.2 and 7.3).

Spatial trends in invertebrate biointegrity were measured by comparing diversity between depths and distance from outfall (by transect) (Figure 7.2) as well as by comparing the assemblage structure over depth and by transect in 2018-2019 (Appendix 7.7). No trends in diversity were apparent in fish in Palos Verdes, but invertebrates had the lowest diversity on the upper slope both in Palos Verdes and throughout the SCB in 2013. (Appendix 7.3; Walther et al. 2017). Though invertebrate diversity and evenness were both lowest for on the Palos Verdes upper slope (305 m) compared to other depths, these metrics increased moving east across the transects, with the lowest at far-field stations (T0) to the highest at nearfield stations (T5). The reason for this pattern is uncertain but could be due to differences in sediment grain size or slope.

Organism health, 2018-2019

Several categories of external anomalies are monitored off Palos Verdes; an anomaly is a departure from the regular arrangement and does not necessarily bear a negative connotation. There are several subgroups of anomalies. Some anomalies, such as ambicoloration, are frequently seen in bottom fish, and are not necessarily signs of impaired organism health or other undesirable conditions. Eye parasites and epidermal pseudotumors are examples of another category in which the anomaly is widely prevalent throughout the fish's range, and is not related to local environmental conditions, but may have significant cost to the organisms. A third category has been related to environmental conditions and can impact fish health; an example is fin erosion. Many anomalies show species specificity and can co-vary with the abundance of the species.

There were rare incidents of fish and invertebrate disease observed off Palos Verdes in 2018 and 2019 (Table 7.1). Anomalies of six categories were seen: pseudotumor, parasite, skeletal deformity, color anomaly, black cyst, and "other" anomaly. "Other" anomalies were likely not associated with pollution effects but were rather caused by other animals, parasites, or as a result of sampling. There was no difference in anomalies with relation to distance to the Palos Verdes outfall (Appendix 7.5).

External parasites on invertebrates were rare, and there was no spatial pattern of occurrence. They occurred at transects where *Paralithodes californiensis* was found, including T0, T1, and T4, all at 305 m depth. (Table 7.1). Six individuals of *Paralithodes californiensis* were found at T0, T1, and T4, four of which were parasitized with *Briarosaccus sp.*, a parasitic barnacle. The anomaly incidence rate for *Paralithodes californiensis* (66%) was higher than that found during the SCB survey in 2013 (21.4%) (Walther et al. 2017).

Anomaly incidences for the species observed in Palos Verdes were higher than for the same species observed in the SCB 2013 survey, but the overall incident rate for all species was the same in Palos Verdes as in the SCB in 2013 (<0.1% for invertebrates and 0.5% for fish). There were several species with anomalies observed in the SCB 2013 survey (Walther et al. 2017) that were not observed in Palos Verdes in 2018-2019 and were not assessed in this report. Likewise, there were no organisms found with "other" category anomalies during the SCB 2013 survey. Comparisons of local data with regional data must be made cautiously, since some participant groups did not report anomalies regularly (Walther et al. 2017). The 2018-2019 measures in Palos Verdes remain at or below recent SCB background levels, and as demonstrated by spatial patterns, the anomalies observed are not likely associated with the effluent discharge.

Marine debris, 2018-2019

The Sanitation Districts' treated effluent is not a current source of trawl-caught marine debris. Patterns of debris occurrence and abundance follow stormwater and boat use patterns. The 137 m depth zone contained the most occurrences of debris at any depth. Transect T5 had more occurrences of debris than other transects (Appendix 7.6). Proximity to the Los Angeles Harbor and the gullied slope associated with T5 are likely contributors to this pattern. Marine debris was also prevalent across the rest of the SCB in 2013 (Moore et al. 2016).

Historical trends, 1973-2019

Demersal fish biointegrity, 1973-2019

Results of the FRI analysis for 23-137 meters at each transect during various treatment periods are presented in Figure 7.4, and additional analyses are included in Appendix 7.3. Impacts have declined over time across all transects. During the baseline (1973) wastewater treatment level period, all trawl locations were impacted except for T0-61. After advanced primary wastewater treatment improved effluent quality (1974-1983), recovery was seen in all areas, although recovery at transect T4, immediately down current from the outfalls, lagged compared to other transects. Palos Verdes trawl-caught fish communities have been in reference condition since the partial secondary treatment level period (1984-2002) and continue through the current period (2018-2019), as indicated by the median FRI scores (Appendix 7.3).

Since 2003, several individual trawls at 23 meters scored above the reference threshold of 45, including the single value, discussed above, that occurred during the current treatment period (2018-2019) at Station T1-23 during summer 2018. However, FRI scores above reference were not observed during this period at discharge depth (61 m) or at other depths. The elevated FRI scores in the 23 m samples were due to a higher abundance of C-O Sole (*Pleuronichthys coenosus*), Curlfin Sole, Stripetail Rockfish, Pacific Sanddab and Dover Sole. These species have higher pollution tolerance scores, and relatively small increases in the number of individuals of these species can significantly elevate the overall FRI score for the station. Changes in the abundance of these species may have been driven by largescale changes in ocean temperature, or by the persistent influence of the Portuguese Bend landslide on the shallow stations. These inputs likely played some part to increased FRI scores which indicate less than complete recovery at 23 m as compared to deeper stations. In addition, it has been suggested that some fishes with higher p-codes may not be reliable indicators of outfallrelated environmental stress, thereby skewing the FRI values, but further research is needed before a definitive statement can be made with regard to these species (Walther et al. 2017). Although FRI index scores for individual trawl samples were occasionally above reference condition, the median score for those 23 m stations within a sampling period was within reference condition.

Epibenthic invertebrate biointegrity, 1973-2019

Diversity, dominance, and species richness were amongst the metrics used to assess the general condition of invertebrate communities over the Sanitation Districts' wastewater treatment periods between 1973 and 2019. There was no observed change in diversity associated with distance from outfall, but some significant differences in diversity occurred after implementation of additional wastewater treatment, and between the shelf (23-137 m) and upper slope (305 m), with diversity decreasing with an increase in depth (Appendix 7.3). Unlike the FRI, which was designed to measure biointegrity of an area using abundance-weighted pollution tolerance scores of fishes, no similar predictor of biointegrity that is specific to a pollution gradient has proven to be accurate for invertebrates. In Appendix 7.7, distinct invertebrate assemblage structures are depicted using nMDS ordination. Three factors were tested: depth (23, 61, 137, and 305 m); wastewater treatment level: baseline (1973), advanced primary (1974-1983), partial secondary (1984-2002), full secondary (2003-2017), and current treatment period (2018-2019); and distance from outfall (transects T0-T5). ANOSIM tests confirmed that depth was the major factor associated with invertebrate assemblage patterns.

All transects grouped into the ordination space by depth, forming a clear pattern (Appendix 7.7). In a two-way crossed ANOSIM of water treatment level and distance from outfall, baseline was different from treatment periods occurring from 1984 on, including partial secondary, full secondary, and the current treatment period (R=0.431, p=.001). There was no significant difference between the current treatment period and full secondary, or between current and partial secondary. There was also no difference between transects (Appendix 7.7).

Shelf stations (23-137 m) grouped closely by treatment period and did not differ greatly by distance from the outfall. On the inner shelf (23 m), all stations during the current treatment period (2018-2019) significantly grouped together using the SIMPROF test, independent of distance from outfall. Assemblages changed as wastewater treatment improved. Several invertebrates' assemblage patterns correlated with treatment level improvements. Increases in wastewater treatment were correlated with Astropecten *californicus* on the inner shelf (23 m), and with *Luidia foliolata* on the mid-shelf (61 m). On the outer shelf (137 m), Platymera gaudichaudii (armed box crab) was associated with earlier treatment periods, and Octopus rubescens was associated with improvements in wastewater treatment. In addition, as wastewater treatment level increased, the communities at T0 (a far-field station) on the inner and mid-shelf (23 and 61 m) indicated recovery as the assemblages near the outfall changed from impacted to far-field condition. Upper slope assemblages (305 m), which were first sampled beginning in the partial secondary treatment period, showed a similar pattern of assemblage change as treatment level increased but had higher similarity amongst transects than assemblages at shelf depths. These trends, from impacted toward a state of recovery, follow a similar pattern of recovery seen in the FRI (Figure 7.4). Appendix 7.7 provides detailed results from SIMPER tests, which revealed the invertebrate species that most contributed to each assemblage by depth, wastewater treatment period, and distance from outfall.

The ANOSIM tests of depth, treatment period, and distance from outfall confirmed the patterns (Appendix 7.7). R-values and pair-wise comparisons indicated that the strongest differences were caused by changes in depth (Appendix 7.7), followed by changes in treatment period. Within each depth, there were distinct assemblages detected. The degree that these assemblages overlapped was dependent upon the treatment period and distance from outfall. For instance, there were some differences in assemblages by treatment period, the strongest of which were always between the least and most treated samples (e.g. between baseline and current treatment periods). Some assemblages could be distinguished by transect, but only when they were first isolated by the factors of depth or treatment period, prior to testing, to remove the strong influence of those factors. In general, assemblages furthest from the outfall (T0) had less in common with the assemblages found at other transects.

Outfall impacts on trawl-caught organisms

Effluent discharge has not appeared to be a major factor influencing demersal fish and epibenthic invertebrate communities for over 20 years. Outfall-related gradients were apparent in the 1970s during advanced primary treatment levels, decreasing over time and becoming insignificant beginning in the 1990s when treatment had progressed to partial secondary. Over this period, effluent quality improvements resulted in decreased mass emissions of organic matter and trace contaminants and surface sediment contamination. Water clarity increased, kelp beds expanded and food resources diversified (Conversi and McGowan 1994, Stull 1995).

These observations are consistent with those of Thompson et al. (1993), who analyzed trawl-caught mega-invertebrate data from 1,168 SCB trawls at 224 sites in depths ranging from 10 -915 m collected between 1971 and 1984. These scientists found that the nature of this community on the Palos Verdes shelf had changed over time, from a distinctive low diversity assemblage typifying Palos Verdes prior to 1980, transforming to a normal shelf assemblage after 1980.

Reports of regional surveys from 1994 (Allen et al. 1998, Allen et al. 2001b), 1998 (Allen et al. 2002), 2003 (Allen et al. 2007), 2008 (Allen et al. 2011), and 2013 (Walther et al. 2017) also found that the community of trawlcaught organisms near major treated wastewater outfalls were comparable to areas far removed from such discharges, providing additional support for the health of this community off Palos Verdes.

Fish health, 1973-2019

Since the early 1970s, all fish from Palos Verdes NPDES monitoring have been examined for external anomalies, as described above in the Current Condition section. Spatial and temporal trends of fish anomalies can be seen in Figure **7.5**. The baseline and advanced primary treatment periods had higher levels of anomalies than other treatment periods, with transects nearest and adjacent to the outfall (T4 and T5) having higher incidence rates. Fewer anomalies have been observed since partial secondary treatment began, and no longer vary by distance from the outfall (Appendix 7.5). Appendix 7.5 provides additional information regarding prevalent fish anomaly types over the history of the Districts' treatment periods.

Fin erosion is an environmentally and historically relevant disease that was not present during the current wastewater treatment period (2018-2019). Numerous studies on the syndrome, its etiology, and connection to discharge impacts have been conducted, and ultimately attributed the condition to contact with contaminated sediment and possibly exposure to high concentrations of PCBs (Sherwood 1976, 1978). The rapid decline in individuals with fin erosion in the late 1970s coincided with treatment plant upgrades and reductions in sediment contamination (Sherwood and Mearns 1977). The disease was rarely seen by the mid-1980s and was last seen in four juvenile California Tonguefish (a flatfish) during the full secondary treatment period (two in 2010 and two in 2011) at T4-61, the station in closest proximity to the outfall. These were the first occurrences of fin erosion in California Tonguefish since 1993 and the first occurrence of fin erosion in any fish species collected during Palos Verdes trawl surveys since 1999, when the condition was last detected in Dover Sole. Histological evaluation of the two affected California Tonguefish caught in 2011 was performed by the National Oceanic and Atmospheric Administration (NOAA), who reported a lack of definitive findings in the samples (M. Myers, e-mail message, August 27, 2012). NOAA found that the damage to the fins could have been, at least partially, a reaction to a subcutaneous infection of the fish muscle tissue by a microsporidan parasite, but NOAA was not certain that the parasite was the only factor contributing to the fin damage. NOAA also reported that, in general, histopathologic

diagnosis of fin erosion in fish is very uncertain and can sometimes be due to healing of traumatic wounds (e.g. bite wounds), net trauma, or healing of areas previously or currently infected with parasitic organisms, all influenced by exposure to pollutants. The final conclusion by NOAA was that it is almost impossible to prove that pollutant exposure was the triggering event in this process.

Of 798 California Tonguefish collected from 2010-2011, there were only four with signs of fin erosion, an incidence rate of 0.5% for this species over the two-year period. This suggests that these four specimens likely represented isolated occurrences, not a return to previously observed impacts to fish resulting from degraded sediment conditions. The limited cases of fin erosion did not appear to represent a negative impact on fish communities related to current epibenthic/sediment conditions or the JWPCP discharge as demonstrated by the catch, diversity, and the FRI at T4-61, during the same time period, all of which indicated the station was not impacted. Further evidence of lack of impact is that during the full secondary and current wastewater treatment period, 24,559 Dover Sole were collected, all without signs of fin erosion. Although the detection of fin erosion must be acknowledged and investigated as a potential impact from contaminated sediments near the outfall, these incidents alone did not signify a trend towards impacted fish health.

Fish pseudotumors (smoothly furrowed, epidermal papillomas) were most commonly observed in Dover Sole (Appendix 7.5). Historically, significantly fewer Dover Sole had epidermal pseudotumors than exhibited fin erosion. However, there have not been the same relative reductions in pseudotumors as in fin erosion. Over the past 47 years of monitoring Palos Verdes, pseudotumors occurred most frequently in small Dover Sole (7-14 cm standard length) and rarely in smaller or larger fish. Incidence was historically highest at transects T4 and T5. Relationships between pseudotumors and survival are not known. However, stations with the highest abundances of larger Dover Sole (transects T4 and T5) had a larger incidence of pseudotumors among the smaller sole. No consistent temporal trend is apparent.

The copepod eye parasite, *Phrixocephalus cincinnatus*, was typically hosted by sanddabs, especially Pacific Sanddab. Other external parasites, such as isopods and leeches, have also been observed over the years. No consistent spatial or temporal pattern is apparent.

Skeletal deformities include bent fin ray, spinal and caudal fin deformities, bent nose, and other similar skeletal anomalies. Historically, bent fin rays (believed to be related to fin erosion) were the typical anomaly, mostly on Dover sole. During the partial secondary treatment period, when sampling at 305 m began, clubbed pectoral fins were often observed in Shortspine Thornyhead. The distribution of skeletal deformities was not correlated with outfall proximity.

Black lesions, which are of unknown origin, have appeared only in California Tonguefish. This anomaly was not observed prior to 1985. In the years when it was detected during partial secondary treatment, annual incidence rates vacillated between 0.2% (1994) and 1.7% (2002). It occurred in fish captured at mid-shelf depth (61 m isobath) and was most prevalent near outfall stations (T4-61 and T5-61). Since implementation of full secondary treatment, incidence rates have generally decreased, and black lesions have not been observed since 2010.

Ambicoloration is the only color anomaly historically seen in flatfishes on Palos Verdes although other forms of color anomalies have been seen in recent years. Higher frequencies of ambicoloration, along with the detection of albinism and diffuse pigmentation (other forms of ambicoloration), have been reported since 1994, when a more sensitive definition was adopted. Historically, it has been most prevalent near outfall transect T4, although currently it is higher at both T0 and T1 (Table 7.1). Ambicoloration is believed to be an example of natural variability; there is no relationship to fish size.

Debris, 1996-2019

Debris has been monitored in Palos Verdes trawls for the past 23 years (1996-2019). Although the outfalls are not a current source of debris, it is possible that during early periods of treatment (baseline and advanced primary), small debris items could have been discharged from the Palos Verdes outfalls. Legacy debris of this type has never been detected in the trawl samples but has been found in sediment samples. Debris from boating and fishing activities (e.g. fishing



Figure 7.5 Spatial and Temporal Trends in Fish Anomalies, 1973-2019

Percent of anomalies in all fish species for each transect by wastewater treatment level.

gear, plastic food wrappers) and stormwater events (e.g. terrestrial vegetation) have been the major components of debris seen in the trawl nets.

Historically, depth has been the sole significant factor determining amount of debris, with the 137 m depth zone containing the greatest amounts and most occurrences of debris at any depth. Proximity to shipping lanes and topography of the slope are likely contributors to this pattern.

CONCLUSIONS

There was a marked relationship between effluent discharge, sediment contamination, and

the structure and function of the epibenthic invertebrate and demersal fish communities off Palos Verdes during baseline (1973) and advanced primary treatment periods (1974-1983). Impacts were most noticeable at sites closest to the discharge, and along the discharge isobath. Subsequent improvements to effluent quality, decreases in particulate quantity, and reduction in surface sediment contaminants have dramatically improved conditions for these communities over the past 47 years. This is evident in most data gathered, but is clearest in the FRI. Examined over time, the FRI demonstrates the recovery of the community, station by station, from its impacted state. Improvements were not simultaneous over the Palos Verdes shelf, and were initiated first at

stations most distant from the outfall, proceeding towards the discharge after lags of several years.

While the fish community can be characterized by the FRI, independent observations of the epibenthic invertebrate community are provided by Thompson et al. (1993), for the first two decades of this 47-year period. These scientists reported that by 1980, a normal epibenthic invertebrate community had been reestablished on Palos Verdes. The few persistent differences in epibenthic invertebrate populations on the mid and outer shelf near the outfall may be due to topography (e.g. outfall structure, slope gullies, proximity to the Portuguese Bend landslide), food resources (less diverse infauna), or legacy sediments from wastewater discharge in the 1970s (Lee 1994, Stull et al. 1996).

Historically discharged contaminants such as DDT and PCBs, as well as organicallyenriched sediment found in a partly buried sediment reservoir off Palos Verdes, are still bioavailable and may influence some species directly through contact with contaminated sediments or indirectly through effects on recruitment. As long as the legacy of pre-1970s sediment contamination persists on the Palos Verdes Shelf, some faint echo of its effects may be detected despite the extensive and well documented recovery of both invertebrate and fish communities.

The health of individuals in both the epibenthic invertebrate and demersal fish populations remains excellent, with rare disease and anomalies evident in invertebrates, and rare disease in fish. The overall incidences of anomalies remain higher than those found in the SCB. As demonstrated by spatial patterns within Palos Verdes, the anomalies observed are not likely associated with current effluent discharge.

Anthropogenic marine debris is prevalent on the Palos Verdes shelf and slope and appears to be related to stormwater and boat use patterns. Spatial patterns exist, indicating areas near harbors and certain topographical features may serve as a source and/or sink for the debris.

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