- 1 **Title.** Plastics in estuarine fish and sediments at the mouth of an urban watershed.
- 2

# 3 Authors.

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# 12 Abstract.

The extent to which small plastics and potentially associated compounds are entering coastal 13 14 foodwebs, especially in estuarine systems, is only beginning to be realized. Using estuarine fish and sediment collected during June 2015 from urbanized Chollas Creek in San Diego, California, 15 16 we tested the hypotheses that small plastic composition in sediments would be reflected in fish 17 guts (non-selective consumption), and that semi-volatile organic compounds (SVOCs) would be 18 present in all fish. Sediments contained about 10,000 small plastic pieces per m<sup>2</sup>, consisting 19 mostly (90%) of fibers, and hard and soft pieces. Nearly 25% of fish contained small plastics, but 20 prevalence varied with size and between species. Of the 39 types of small plastics found in 21 sediment, fish preferred 10 types (distinct colors and forms). Several SVOCs, both water soluble 22 and sediment-associated compounds, were found in the two species of fish tested. We conclude that a species' natural history may influence contamination levels with consequences, and 23 24 lessons, for all consumers.

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26	Keywords. Chollas Creek, ichthyofauna, plastics, San Diego Bay, water contamination, wetland
27	fish
28	
29	Introduction. Microplastics (plastic particles <5mm; Rochman 2018) are pervasive in our
30	ocean and coastal ecosystems (Thompson 2015; Cole et al., 2011). These small plastics enter the
31	environment as either primary microplastics (those manufactured as tiny pieces, such as
32	microbeads) or secondary microplastics (those that form from the breakdown of larger plastic
33	items) (de Sá et al. 2018; Auta et al. 2017). Microplastics are of concern not just because of their
34	ubiquity, but because of the harmful effects they can have on biota. Microplastics can
35	accumulate in the gut or gills of organisms, interfering with important life history processes such
36	as feeding, growth, and reproduction (Chae and An 2017; Cole et al. 2015; Watts et al. 2014;
37	Watts et al. 2015; Sussarellu et al. 2016). The monomers and additives that compose
38	microplastics can be toxic to biota if they leach from their parent plastics into the environment
39	(Smith et al. 2018; Thaysen et al. 2018; Teuten, et al. 2009). Furthermore, microplastics can sorb
40	toxins such as metals, PCBs, PAHs, and DDT from the aquatic environment, and transmit these
41	toxins to organisms, causing stress to internal organs, disruptions in normal bodily functions
42	(e.g., enzyme inhibition, endocrine disruption), and reductions in organisms' abilities to defend
43	themselves against predators and other threats (Chae and An 2017; Smith et al. 2018; Rochman
44	et al. 2013a,b; Browne et al. 2013; Barboza et al. 2018b; Rochman et al. 2014). Microplastics
45	have been found to transfer between trophic levels (Welden et al. 2018; Farrell and Nelson 2013;
46	Setälä et al. 2014), and thus may pose health risks to humans via consumption of contaminated
47	seafood (e.g., Van Cauwenberghe and Janssen 2014; Rochman et al. 2015a; Barboza et al.

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48 2018a), though the impacts of microplastics on human health remain largely unknown (Wright

49 and Kelly 2017; Smith et al. 2018).

#### 50

One suite of contaminants of concern are semi-volatile organic compounds (SVOCs), which 51 52 include additives commonly used in plastics manufacturing, such as phthalates, bisphenol A, and PBDEs, as well as PCBs, PAHs, and DDT (Lucattini et al. 2018; Weschler and Nazaroff 2008; 53 Lusher et al. 2017b). SVOCs are susceptible not only to leaching out of plastics into the 54 55 environment, but to resorbing to microplastics once present in the environment, due to their 56 hydrophobic properties (Lusher et al. 2017b; Teuten et al. 2009; Cheng et al. 2013; Rochman et al. 2013a). SVOCs are a health concern for humans and wildlife because they are endocrine 57 disrupting chemicals (EDCs) that have been linked with neurological, reproductive, metabolic, 58 and behavioral abnormalities, as well as increased incidences of some forms of cancer (Koch and 59 60 Calafat 2009; Xu and Zhang 2011; Gore et al. 2015). Microplastics facilitate the accumulation of 61 SVOCs in organismal tissues (Rochman et al. 2013b; Besseling et al. 2013), however it is important to note that microplastics' role as a conduit for SVOCs into coastal foodwebs may be 62 63 relatively unimportant when compared with other vectors, such as contaminated water, prey, or 64 sediments (Lusher et al. 2017b; Koelmans et al. 2016). It is also important to note that, while 65 often associated with plastics, SVOCs found in the environment may stem from a number of 66 different sources, including household cleaning products, cosmetics, and pesticides (Luccattini et 67 al. 2018; Weschler and Nazaroff 2008). 68

- 69 Much of the research conducted on the ecological impacts of microplastics over the last two
- 70 decades has focused on marine systems, only recently shifting to include terrestrial and

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<ol> <li>73</li> <li>74</li> <li>75</li> <li>76</li> <li>77</li> <li>78</li> <li>79</li> <li>80</li> <li>81</li> </ol>	important one, given the fact that significant portions of marine plastic pollution come from land-based sources, and rivers are major conduits of debris from land to sea (Rochman 2018;
<ol> <li>74</li> <li>75</li> <li>76</li> <li>77</li> <li>78</li> <li>79</li> <li>80</li> <li>81</li> </ol>	
75 76 77 78 79 80 81	
76 77 78 79 80 81	Jambeck et al. 2015; Lebreton et al. 2017). The effects of microplastics in urbanized riverine
77 78 79 80 81	ecosystems may be particularly acute (e.g., Peters and Bratton 2016), as rivers that flow through
78 79 80 81	densely populated, urban areas have been shown to carry higher loads of debris (Lebreton et al.
79 80 81	2017; SCCWRP 2016; Yonkos et al. 2014). This study contributes to the growing body of
80 81	research on microplastics upstream from marine ecosystems by investigating the presence of
81	microplastics and associated contaminants in estuarine sediments and fish in a brackish stretch of
	urbanized Chollas Creek, which empties into San Diego Bay. Understanding the types, fates, and
82	effects of small plastics in coastal watersheds is necessary to develop natural and social science-
	based solutions to marine debris and declining watershed health (Rochman 2018).
83	
84	Knowledge of seasonal precipitation patterns in Mediterranean climates, such as that found in
85	Southern California, result in accumulation and breakdown of photodegradable plastics during
86	the sunny, dry season, followed by wet season pulses of debris inputs and transport through
87	coastal watersheds (Anderson et al., 2012; Lee, 2011; Chandler, 2012; Moore et al., 2011). A
88	recent regional survey of the Southern California Bight, to which this study contributed, revealed
89	that 92% of the total stream length of urban watersheds in the Southern California Bight
90	contained debris, while 48% of undeveloped watersheds did (SCCWRP, 2016). More than 60%
91	of the debris found was plastic (SCCWRP, 2016). Many coastal wetland fishes, such as killifish,
92	of the debits found was prastic (See Witt, 2010). Many coustant workand rishes, such as minimus,
91	

93 contaminants accumulate, putting them at particularly high risk of contamination. These fish may

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94	in turn be important vectors in transferring small plastics and contaminants to the broader coastal		
95	food web given their abundance, their roles in connecting intertidal with both subtidal and		
96	terrestrial ecosystems, and their roles as forage fish for many species (Trexler et al., 1994; West		
97	et al., 2003; Able et al., 2012; Kang et al., 2015).		
98			
99	Project goals. The goals of this study were to: a) determine the extent and magnitude of		
100	microplastics pollution in estuarine sediments and fish, and of SVOC contamination in estuarine		
101	fish, at the mouth of an urban coastal watershed, and b) determine whether sampled fish		
102	preferentially ingested certain types of microplastics, when compared with types and abundances		
103	of microplastics in sampled sediments.		
104			
105	Materials and Methods.		
106	Study location. The Chollas Creek subwatershed (Fig. 1), considered one of the most		
107	impaired waterbodies in San Diego County, in part due to the large amounts of trash present in		
108	the creek (Anderson et al. 2012; State Water Board 2015), runs through a densely populated,		
109	urban section of the County and empties into San Diego Bay at one of the worst urban runoff		
110	sites of coastal San Diego (Pritchard 2014). In June 2015, we sampled sediments and fish along a		
111	250-m long reach of intertidal Chollas Creek, located about 1 km upstream of the mouth (Fig. 1).		
112	Sample collection and processing. Microplastics in sediment were sampled at low tide		
113	by collecting nine 10-cm diameter x 5-cm depth cores (393 cu cm) throughout the reach, placing		
114	cores in clean, airtight bags, and freezing cores until analysis in the lab. Plastics were sorted from		
115	sediments by placing a single layer of sediment at a time into a Petri dish (about 1 tablespoon or		
116	15 cu cm) along with a squeeze of milliQ water to slightly liquefy the moist sediment. The dish		

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**Commented [A4]:** It's hard to fine straightforward information about the pop density throughout SD County. This would be an okay reference to use if we need one here (tho not great): https://www.voiccofsandiego.org/topics/land-use/san-diego-the-eighth-largest-city-but-still-mostly-a-suburb/.

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117	with mud was systematically examined at 25-45x power using a dissecting microscope; sorting
118	of each dish took no more than 15 minutes. Particles that were clearly of anthropogenic origin, as
119	determined by the shape and/or color of each particle (e.g., spherical microbeads, fibers with
120	smooth surfaces and homogeneous thicknesses; often bright colors that stood out from the rest of
121	the sample) were sorted out of the sample, classified according to the item from which they
122	originated, when distinguishable (e.g., pieces of grocery bag, wrappers), or by hardness, shape
123	and color (e.g., film, hard or soft pieces, fiber), and then counted and measured for maximum
124	length. Particles that were not clearly of anthropogenic origin were examined using a compound
125	microscope to check for lack of cell structure. Any particles that remained of uncertain origin
126	were excluded from the analysis. It is important to note that because samples were only
127	examined using microscopy (and not run through a spectroscope to chemically verify polymer
128	types and particle counts), it is possible that the abundances of microplastics reported herein are
129	either over- or underestimates of actual numbers, as both false positives and failures to identify
130	very small plastic particles are relatively common when relying on microscopy to identify
131	microplastics (Song et al. 2015; Lenz et al. 2015; Lusher et al. 2017a).
132	
133	At the time this study was conducted, the risk of sample contamination from airborne
134	plastics was just beginning to be realized, and protocols to control for such contamination
135	followed soon after although such QA/QC protocols have not yet been standardized and debate
136	remains about how best to control and account for contamination of samples (NOAA, 2015;
137	REF??; but see Hidalgo-Ruz et al., 2012). We therefore attempted a post-hoc control of
138	environmental plastics contamination by conducting three trials separated in time on 20 June, 20
139	July and 19 August 2016 to determine average levels of contamination in the lab used during the

**Commented [A5]:** I wonder how confident we were in this – wonder whether we should reclassify the few pieces that we found in these categories as "clear film" etc...

**Commented [A6]:** Try to find a "history of microplastics research" paper, and look at the Norwegian research institute report's sections on QA/QC // Lusher's Sampling, isolating and identifying microplastics ingested by fish and invertebrates

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140	study. During each trial, six clean Petri dishes were set out for 15 minutes on the lab countertops.	
141	Three or four people were present in the lab each time (during sample sorting in 2015, two or	
142	three people were present at any one time). At 15 minutes, dishes were covered with clean, clear	
143	lids and examined for particle settlement using a dissecting microscope. Only fibers were found	
144	at an average of $0.5\pm0.3$ , $0.5\pm0.2$ and $0.5\pm0.3$ fibers per dish for the June, July and August trials,	
145	respectively (grand average = $0.5\pm0.0$ fibers per dish per 15 minute time period). Fiber	
146	contamination for each fiber color (type) was then calculated using the following steps: The	
147	average number of fibers per dish (0.5) was multiplied by the number of dishes likely sorted for	
148	each sample core (393 cu cm core / 15 cu cm spoonful per dish = $\sim$ 26 dishes per core) for an	
149	estimated total of 13.1 fibers contaminating each sample core. Since the color of fibers causing	
150	contamination in 2015 could not be determined after the fact, the estimate of 13.1 fibers per core	
151	was divided by the seven fiber color categories for an estimate of 1.87 fibers contaminating each	
152	fiber color category. This value (1.87 fibers) was then subtracted from each fiber color category	
153	of each core before analyses and summary statistics were calculated. If the result of the	
154	subtraction was a negative number, the value was assigned a 0.	
155	Common wetland fish (Boerger, et al. 2010; Rochman et al., 2013b) were trapped using	Commented [A7]: here
156	metal minnow traps baited with cat food placed in nylon sleeves (to prevent fish from consuming	
157	it), and set throughout the reach. Three species were captured: the native marsh residents	
158	California killifish (Fundulus parvipinnis; n = 68) and longjaw mudsucker (Gillichthys mirabilis;	
159	n = 4), and the introduced sailfin molly ( <i>Poecilia latipinna</i> ; $n = 82$ ). In the field, all fish collected	
160	for gut analysis were placed in ziplock bags (one bag per trap). Additionally, two composite	
161	samples (7 California killifish and 8 sailfin molly) were collected and immediately placed into	
162	clean glass jars for analysis of SVOCs. Only four longjaw mudsucker individuals were captured,	

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Check if we need these references

163	so all were used for gut analysis. The protocol of the AVMA Panel on Euthanasia (American
164	Veterinary Medical Association, 2013) was followed, which recommends rapid chilling to
165	euthanize warm-water fish. All fish remained frozen until analysis. Composite samples for
166	SVOC analysis were analyzed for 67 SVOCs by a local analytical facility (Enviromatrix
167	Analytical, Inc.) using EPA Method 8270C (EPA, 1996). Fish used for gut analysis were thawed,
168	measured, weighed, and sexed in the lab. A ventral, longitudinal incision and two perpendicular
169	ventral incisions (anterior and posterior) were made in each fish to expose the intact guts, and
170	then the fish was placed in a clean glass dish under a dissecting microscope to complete the
171	dissection and removal of gut contents. All contents were removed from inside the fish gut
172	systematically as the gut was opened and analyzed a small section at a time for a total of ~15 min
173	of exposure time (i.e., low risk of contamination). Since the post-hoc estimated contamination
174	rates were $\leq 0.5$ fibers per sample (0.5 fibers per dish divided by the one to four fiber color
175	categories found in the fish samples equals 0.125-0.5 fibers contaminating each fiber color
176	category) we did not use a correction but acknowledge that our fiber counts may be slight
177	overestimates. Only materials drawn out of the gut were identified (or described) and counted.
178	For items not feasibly counted (e.g., sand grains, organic debris, filamentous algae), presence in
179	the gut was noted. Ten out of the 149 fish sampled had empty guts and were therefore excluded
180	from further analyses. As with sediment samples, plastics were categorized by color and type
181	(e.g., hard or soft pieces, fiber), then counted and measured for maximum length.
182	Data analyses. Descriptive statistics of sediment small plastics (average of all cores) and
183	fish gut contents (average of small plastics and prey items for each species) were calculated to
184	summarize findings. The concentrations of SVOCs, if present in at least one sample (at least one
185	of the species), are reported. Fish diet preference was explored using Manly's alpha (Chipps and

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186	Garvey, 2007), which compares the abundances of the types of plastic found in the environment		
187	and consumed by the fish. Differences in size and sex ratios of all fish sampled to those that had		
188	consumed plastics were tested using t-tests (size variables) and Chi Square (sex ratios) in JMP		
189	12.		
190			
191	Results		
192	Small plastics in sediment. All sediment cores collected contained small plastics; the		
193	average abundance ( $\pm 1$ SE) was 9,638 $\pm 1$ ,636 pieces m <sup>2</sup> and average lengths ( $\pm 1$ SE) of small		
194	plastics ranged from 1.8±0.3 to 4.6±1.1 mm. Common categories of plastics were plastic film		
195	pieces, such as film from bags and wrappers, polystyrene pieces, soft pieces, hard pieces,		
196	synthetic fibers, and a few miscellaneous items, such as strands of carpeting and synthetic turf		
197	(Fig. 2, Table 1). Synthetic fibers, hard pieces, and soft pieces were the most common types of		
198	plastics found across all sediment cores, together making up 90% of the fragments found (Fig.		
199	2).		
200			
201	Fish gut contents.		
202	Fish species present. Add short paragraph about ranges, and natural history characteristics of		
203	fundulus, sailfin molly, and mudsucker. Mention species captured here, put natural history in		
204	discussion.		
205			
206	Prey and non-plastics in guts. Half or more of these bottom feeding fish species had fed on sand		
207	or silt particles, and filamentous green algae (Table 2). Roughly 10% of both California killifish		
208	and sailfin molly also contained red algal filaments (Table 2). California killifish, primarily an		

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210	amphipod appendages), scales, unidentifiable fishes and insects, snails, tubificid oligochaetes,	
211	nematodes, and a sea cucumber (Table 2). The sailfin molly, although predominantly an	
212	herbivore, also commonly ingested tubificid oligochaetes and nematodes, and to a lesser extent,	
213	crustaceans and snails (Table 2). The predatory longjaw mudsucker contained scales, a digested	
214	fish, and nematodes (Table 2).	
215		
216	Characteristics of plastic eating fish. None of the longjaw mudsucker guts contained plastics,	
217	which may have been due to the small sample size of only four individuals. California killifish	
218	individuals that had plastics in their guts were, on average, 25% longer and 79% heavier than	
219	those free of plastics (p $\leq$ 0.03, Table 3). The ratio of males to females (to unknown sex) was	
220	similar between fish with and without gut plastics (Table 3). Neither size nor sex of sailfin molly	
221	individuals differed between those that contained plastics and those that did not (Table 3).	
222		
223	Small plastics in guts. Almost one quarter of fish examined contained small plastics, with 12% of	
224	California killifish (7 of 61) and 32% of sailfin molly (24 of 75) having consumed plastic (Table	
225	3). Of the 39 types of plastic available in the environment, the California killifish and sailfin	
226	molly each consumed 10-11 different types of plastic items, mostly consisting of fibers and hard	
227	pieces (Fig. 2, Table 1). California killifish also ingested clear microbeads. The consumed items	

invertebrate predator, contained remnants of small crustaceans (e.g., exoskeleton pieces,

228 ranged in average length from 0.05±0.0 to 3.25±2.75 mm for California killifish and from

229 0.10±0.01 to 2.58±1.10 mm for sailfin molly.

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232	eaten (Manly's alpha $\geq 0.025$ ; Table 1), meaning the fishes' guts contained higher proportions of
233	these items than were found in the environment (Fig. 2). The items the fishes selected included
234	blue, yellow, orange and/or red hard plastic pieces, and all colors of synthetic fibers (Table 1).
235	
236	SVOCs in fish.
237	Three SVOCs of 67 tested were found in the tissues of these species. Both the California killifish
238	and sailfin molly contained diethyl phthalate and benzyl alcohol, and the sailfin molly
239	additionally contained 4-(3-) methylphenol (Fig. 3). The sources of these compounds in this
240	creek are uncertain. The phthalate is a synthetic compound, while benzyl alcohol and 4-(3-)
241	methylphenol have natural, albeit small, localized sources (PubChem, 2019 a,b,c). All three have
242	common industrial applications, including additives in plastics, solvents, antiseptics,
243	preservatives, pesticides, and/or additives in cosmetics and perfumes (WHO, 2003; Wade, 2019;
244	Wiki, 2019; PubChem, 2019a,b,c).
245	
246	Discussion.
247	Small debris in sediment and fish. This study, along with other recent studies (Moore et al.,
248	2011; Rochman et al., 2015a; Rochman et al., 2016), illustrate the risk of plastic and plasticizer
249	(SVOC) pollution to coastal food webs. A concurrent regional study of debris in the Southern
250	California Bight found that sediments in embayments exhibited the highest abundance of small

Of the 10-11 types of small plastics that were consumed by the fishes, 7-8 types were selectively

- 251 plastics, while continental shelf sediments exhibited the lowest abundance of small plastics
- 252 (SCCWRP, 2016). Bay sediments, for example, were found to have a mean of 140 pieces/m<sup>2</sup>,
- and continental shelf sediments were found to have ≤20 pieces/m<sup>2</sup> (SCCWRP, 2016). By

**Commented [A9]:** Could it also be that these items just remain in the gut/get stuck in the gut, as opposed to other items?

**Commented [A10]:** Hit harder the discussion about pathways through which plastics become integrated into foodwebs (still big knowledge gaps)//about connections between presence of plastics in the sediment and presence in fish guts...

While there is much knowledge about the presence of small plastics in coastal ecosystems, the extent to which/pathways through which small plastics and potentially associated compounds are being integrated into coastal foodwebs remains an emerging area of research.

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254	contrast, our study of the debris at the mouth of an urban stream emptying into San Diego Bay
255	revealed a much denser concentration of small plastics, with almost 10,000 pieces/m <sup>2</sup> . This may
256	be because the mouth of Chollas Creek is in a highly urbanized area, in close proximity to roads,
257	which are responsible for large inputs of debris (SCCWRP, 2016). Streams like Chollas Creek
258	are conduits for plastics, carrying them into embayments and eventually to the ocean, where they
259	accumulate at high densities in marine sediment (SCCWRP, 2016).

While we found a greater variety of small plastics in wetland fish than were observed in select species of pelagic and demersal fish sold in California for human consumption (Rochman et al., 2015a), both sets of fish predominantly ingested fibers (~60% of the plastics ingested by wetland fish, ~80% of market fish). Further, all of the small debris found in California-grown Pacific oysters were fibers (Rochman et al., 2015a). It is important to note that the fibers found within aquatic organisms, while undoubtedly anthropogenic, could be either synthetic or organic. We were unable to perform spectroscopic analysis to identify their composition.

Natural history and contamination risk. The extent that contaminants impact food webs
depends upon the types of plastics and contaminants present, the environment (Renick et al.
2015), and the natural history of the organisms present (e.g., Renick et al. 2016), including
feeding behavior, and changes associated with ontogeny and sex (Temming and Hammer, 1994;
Smith et al., 2000; Talley, 2000; Borg et al., 2014).

- 274
- 275 The marsh fish in this area selectively fed on most of the small plastics found in their guts
- 276 including all colors of fibers, blue and warm colors of hard pieces, and, in the case of California

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277	killifish, microbeads. Anecdotally, these items often resembled prey, especially a similar
278	morphology between fish eggs and microbeads, and between synthetic fibers and filamentous
279	algae, oligochaetes and nematodes (e.g., Fig. 4), further raising the concern that fish may mistake
280	small plastics as food (Corley, 2014). The likelihood of plastics ingestion or the ability to pass
281	plastics may change throughout the life of an organism, as revealed by the higher incidence of
282	plastics ingestion in larger (older) California killifish individuals during this study. This is
283	consistent with other ontogenetic dietary shifts observed in California killifish, including changes
284	in prey type, prey size and different microhabitat use with time (Smith et al., 2000; Talley,
285	2000).
286	
287	As with the patterns of plastics ingestion, contamination by SVOCs varied with fish species.
288	Diethyl phthalate, a water-insoluble, sediment-penetrating compound (PubChem 2019a), was
289	found in both species but was almost three times higher in the California killifish. Both species
290	ingest sediment while feeding (e.g., Table 3), so an explanation for the higher phthalate
291	concentration in California killifish is uncertain but may be linked to diet, with higher
292	abundances of benthic deposit feeders observed in the guts of killifish in this study (i.e.,
293	potentially more diethyl phthalate-laden sediment), or an artifact of small sample size (n=1). The
294	reasons underlying the presence of 4-(3-) methylphenol and the 3.5 fold greater benzyl alcohol
295	concentration in the sailfin molly compared with the California killifish are also uncertain. These
296	compounds, which are used as solvents, pesticides, antiseptics, anesthetics, and additives in
297	cosmetics and fragrances, are water soluble, have fairly rapid degradation rates in water, and do
298	not tend to accumulate in tissues (PubChem 2019b,c). Again, stomach contents (i.e., water
299	content) or small sample size could explain this observed difference. Although the sources and

**Commented [A11]:** Do these fish use vision to help them feed? Or do they rely more on scent? Might be worth it to look at papers about feeding behavior (Savoca's thesis noted that sea birds often ingested plastics because they smelled like food)

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300	pathways of exposure are uncertain (Weschler and Nazaroff, 2008), the presence of these	
301	compounds in our two samples reveals that transfer of contaminants, even those that are	
302	relatively transient, from the environment to food webs is a real risk. Further study is needed to	
303	understand the interactions among these contaminants, the species' natural history, and other	
304	environmental stresses (e.g., predation, parasitism; Renick, et al., 2015; Renick et al. 2016).	
305		
306	Health effects of small plastics and SVOCs. The effects of small plastics and SVOC	
307	contamination on organisms may be complex (e.g., Renick et al., 2015) and remain largely	
308	uncertain, but knowledge of these effects is needed to understand the consequences of exposure	
309	to the organisms themselves and those that eat them. The acute and chronic effects of the three	
310	SVOCs found in this study have been observed on the growth, reproduction, enzyme activity,	
311	metabolic activity, respiration, kidney function and/or liver function in animals, while the effects	
312	on humans are less well known and are of concern (NIOSH, 1997; Lithner et al., 2011; Groshart	
313	and Okkerman, 2000; Ghorpade et al., 2002; Okkerman and van der Putte, 2002; Gore et al.,	
314	2015; NIH, 2018; PubChem 2019a,b,c). Dietary guidelines and warnings about the risks of	
315	contamination in higher trophic level and longer-lived seafood species due to bioaccumulation	
316	are common, but studies like this illustrate that smaller and/or lower trophic level fish may have	
317	hazards of their own. There is increasing realization that plastics (Murray and Cowie, 2011; Van	Commented [A12]: Super good – highlight more?
318	Cauwenberghe and Janssen, 2014; Rochman et al., 2015a; Sussarellu et al., 2016) and SVOCs	
319	(Windward Environmental, 2010) occur in species consumed by humans. Improved knowledge	
320	of the types and distributions of contaminants in an area, as well as of the biology and natural	
321	history of organisms, are needed to improve predictions of contamination risks of fish, shellfish	
322	and dependent higher trophic levels, including humans.	

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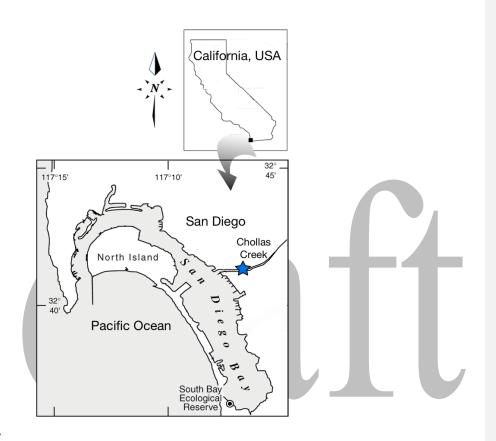
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668 Fig. 1. Study site located in lower Chollas Creek near the mouth with San Diego Bay, California,

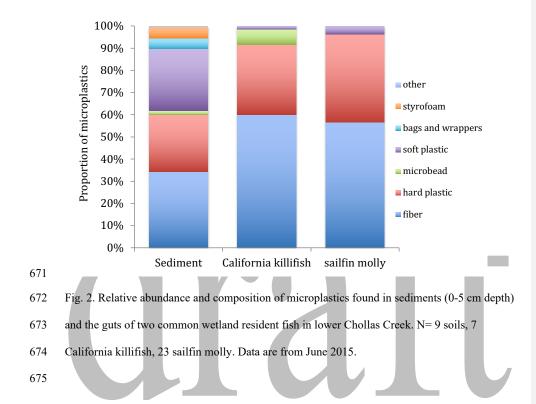
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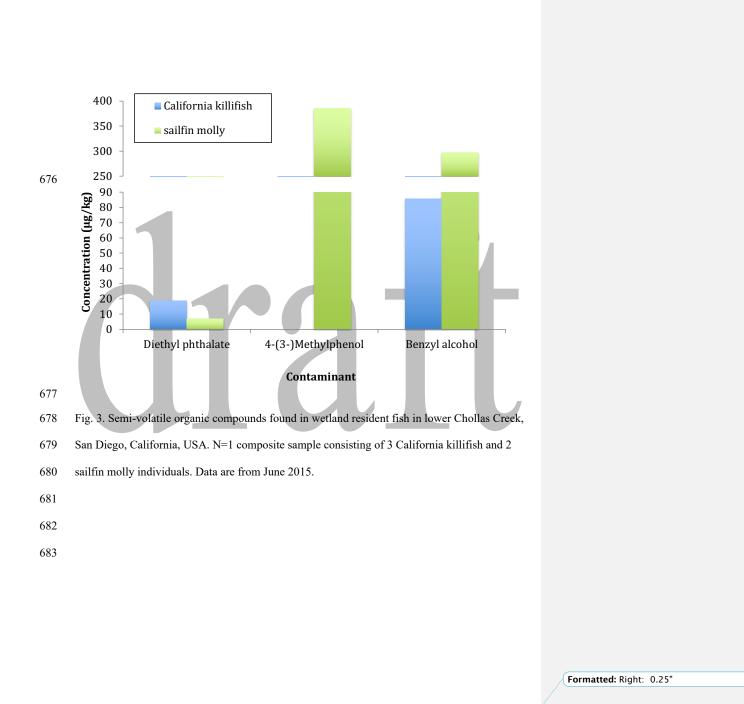
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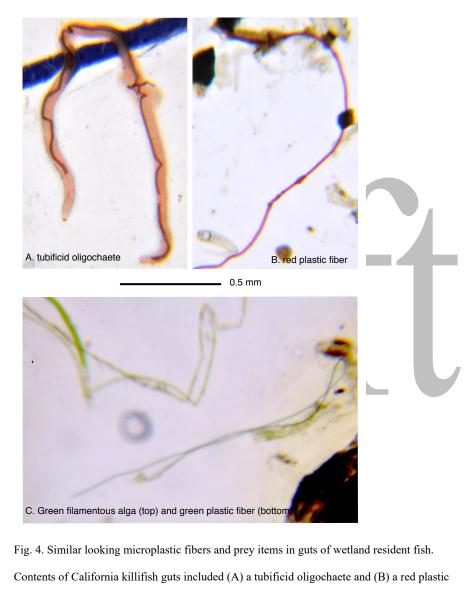


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687 fiber. Contents of a sailfin molly guts included (C) a filament of green algae (top) and a green

688 plastic fiber (bottom). Scale shown applies to all photos.

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684 685

- 689 Table 1. Abundance of microplastics in surface sediments and the guts of common marsh
- 690 resident fish, and Manly's alpha where a ≥0.025 (in bold) indicates a dietary selective preference
- 691 for microplastics compared to what were available in the environment. Only individuals with
- 692 microplastics present in the gut were included in this summary. Samples were collected from
- 693 lower Chollas Creek, San Diego, California, USA during June 2015.

[					
		California		California	
Type of microplastic	Sediment	killifsh	sailfin molly	killifsh	sailfin molly
n=	9 cores	7 individuals	23 individuals	7 individuals	23 individuals
	Abundance				
	(no. m <sup>-2</sup> )	Abundance	e (no. gut <sup>-1</sup> )	Manly's alph	na (α ≥ 0.026)
	Avg ± 1 SE	Avg ± 1 SE	Avg ± 1 SE	Avg ± 1 SE	Avg ± 1 SE
Bag and packaging pieces					
nylar wrapper	28 ± 19	0	0	0	0
lear or opaque wrapper	127 ± 60	0	0	0	0
rocery bag	85 ± 37_	0	0	0	0
hick, opaque (retail bag)	14 ± 14	0	0	0	0
hin translucent (produce bag)	14 ± 14	0	0	0	0
hick, clear, soft (bread or zip bag)	28 ± 28	0	0	0	0
lear, thin crinkly wrapper	42 ± 30	0	0	0	0
hick, clear, crinkly (nut, pasta bag)	14 ± 14	0	0	0	0
strapping or packaging tape	42 ± 30	0	0	0	0
lear hard plastic shell packaging	14 ± 14	0	0	0	0
styrofoam piece	467 ± 116	0	0	0	0
ar or bottle label	28 ± 28	0	0	0	0
ubber or foam piece	14 ± 14	0	0	0	0
Soft plastic pieces					
lear or white	467 ± 108	0	0	0	0
reen	241 ± 109	0	0	0	0
blue	467 ± 192	$0.14 \pm 0.14$	0.13 ± 0.13	$0.004 \pm 0.004$	0.012 ± 0.009
red	1089 ± 399	0	0	0	0
vellow	410 ± 137	0	0	0	0
range	99 ± 59	0	0	0	0
Hard plastic pieces					
lear or white	552 ± 209	0	$0.04 \pm 0.08$	0	0.005 ± 0.005
reen	368 ± 157	$0.14 \pm 0.14$	$0.04 \pm 0.08$	$0.005 \pm 0.005$	$0.020 \pm 0.020$
bink	28 ± 28	0	0	0	0
blue	311 ± 150	0.29 ± 0.18	0.65 ± 0.52	0.027 ± 0.021	0.220 ± 0.080
red	424 ± 133	$0.14 \pm 0.14$	0.17 ± 0.19	0.005 ± 0.005	
vellow	1019 ± 376	0.14 ± 0.14	0.13 ± 0.13	0.143 ± 0.143	0.046 ± 0.043
olack	14 ± 14	0	0	0	0
silver	14 ± 14	0	0	0	0
orange	14 ± 14	0	0.04 ± 0.08	0	0.042 ± 0.042
nicrobead	212 ± 124	$0.29 \pm 0.18$	0	0.066 ± 0.043	0
Fibers			-		
lear or white	2716 ± 1008	0.57 ± 0.43	0	0.145 ± 0.145	0
black	85 ± 56	0	0.39 ± 0.36	0	0.108 ± 0.054
blue	57 ± 43	0.43 ± 0.30	0.43 ± 0.25	0.213 ± 0.148	
green	14 ± 14	0.14 ± 0.14	0.13 ± 0.13		0.130 ± 0.072
red	71 ± 37	0.57 ± 0.30	0.22 ± 0.20	0.254 ± 0.127	0.121 ± 0.061
net or thick line	396 ± 312	0	0	0	0
Pieces of other items		-		-	
ponge	14 ± 14	0	0	0	0
oys: Lego, beads	28 ± 28	0	0	0	0
artificial grass, astro turf	28 ± 28	0	0	0	0
synthetic carpet	42 ± 30	0	0	0	0
Total per core or gut:	9638 ± 1638	2.86 ± 1.22	2.43 ± 0.68		

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## 695 Table 2. Abundance of prey and other non-plastics found in the guts of common marsh resident

696 fish from lower Chollas Creek, San Diego, California, USA. Data are from June 2015.

697

	California		longjawed	
Gut content items	killifsh	sailfin molly	mudsucker	
n (number of individuals)=	61	74	4	
Items that could not be counted	Avg ± 1 SE	Avg ± 1 SE	Avg ± 1 SE	
	% of fi	sh with items pr		
sand or silt	48%	99%	50%	
scales	5%	0%	25%	
unknown exoskeleton pieces	53%	1%	0%	
unknown amphipod or shrimp pieces	2%	0%	0%	
unknown decapod pieces	0%	1%	0%	
unknown organics or digested pieces	18%	0%	0%	
green filamentous algae	75%	85%	50%	
red filamentous algae	10%	11%	0%	
Enumerated items	Avg ± 1SE	Avg ± 1SE	Avg ± 1SE	
	(no. gut-1)	(no. gut-1)	(no. gut-1)	
snails (Barleeia californica, Assiminea				
californica)	$0.49 \pm 0.40$	0.04 ± 0.03	0	
tubificid oligochaetes, nematodes	1.66 ± 0.28	0.12 ± 0.05	1.75 ± 0.75	
unnown whole digested-fish	$0.02 \pm 0.02$	$0.01 \pm 0.01$	$0.25 \pm 0.25$	
unknown fish eggs or larvae	$0.05 \pm 0.05$	0	0	
unknown insect larvae or adult parts	$0.05 \pm 0.03$	0	0	
sea cucumber (Leptosynapta sp.)	$0.05 \pm 0.03$	0	0	

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Table 3. Comparison of fish that had and did not have small plastics in their guts with all fish

700 analyzed in this study. Results of t-tests (fish morphological variables) and Chi square (sex

701 ratios) are shown.

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	California killifsh									sailfin molly									
	Fish without plastics			Fish with t plastics			t-test/Chi square results			Fish without plastics			Fish with plastics			t-test/Chi square results			
n=		61			7					74		23							
								t/									t/		
Variable	Avg	±	1 SE	Avg	±	1 SE	Р	Chi sq	df	Avg	±	1 SE	Avg	±	1 SE	Р	Chi sq	df	
standard length (cm)	4.34	±	0.16	5.50	±	0.32	0.003	2.52	59	4.05	±	0.12	4.15	±	0.29	0.655	0.55	72	
total length (cm)	5.12	±	0.18	6.40	±	0.40	0.005	2.41	59	4.96	±	0.14	5.08	±	0.34	0.624	0.58	72	
weight (g)	2.35	±	0.27	4.20	±	0.32	0.005	2.25	59	2.05	±	0.19	2.39	±	0.38	0.466	1.04	72	
sex: female / male / unknown	24	/23	3/6	3	/3/	1	0.571	1.13	2	28	/2:	1/2	15	5/7	/1	0.677	0.78	2	

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