

1 **TITLE: PERSISTENT ORGANIC POLLUTANTS IN GREEN SEA TURTLES**
2 **(*CHELONIA MYDAS*) INHABITING TWO URBANIZED SOUTHERN CALIFORNIA**
3 **HABITATS**

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Abstract

37 Within Southern California, east Pacific green sea turtles (*Chelonia mydas*) forage
38 year-round, taking advantage of diverse food resources, including seagrass, marine algae,
39 and invertebrates. Assessing persistent organic pollutants (POP) in green turtle
40 aggregations in the Seal Beach National Wildlife Refuge (SBNWR, n = 17) and San
41 Diego Bay (SDB, n = 25) can help quantify contamination risks for these populations.
42 Blood plasma was analyzed for polychlorinated biphenyls (PCBs), organochlorinated
43 pesticides (OCPs), and polybrominated diphenyl ethers (PBDEs). PCBs and body size
44 explained much of the separation of turtles by foraging aggregation in a principal
45 component analysis. Turtles from SDB had significantly ($p < 0.001$) higher total PCBs
46 than SBNWR turtles. Most PCBs detected in turtles were non-dioxin-like PCB congeners
47 (153, 138, 99) that are associated with neurotoxicity. Recaptured turtles' POP levels
48 changed significantly over time indicating significant variation in POP levels through
49 time and space, even among adjacent foraging locations.

50

51 **Keywords:** *Chelonia mydas*, persistent organic pollutants, polychlorinated biphenyls,
52 organochlorinated pesticides, urbanized habitats, marine turtles

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Introduction

55 Marine species, such as sea turtles, have been negatively impacted by
56 anthropogenic activities, and as a result many sea turtle populations are considered
57 threatened or endangered (Gardner and Oberdorster, 2005; Hamann et al., 2010; Pugh
58 and Becker, 2001). Historically, threats such as overharvesting and habitat loss have

59 severely impacted sea turtle populations, but conservation efforts have initiated recovery
60 for some populations around the world (Hamann et al., 2010; Mazaris et al., 2017;
61 Seminoff et al., 2015). Additional threats, such as pollution, continue to impact marine
62 waterways that sea turtles inhabit (Hamann et al., 2010). Human activities, such as
63 manufacturing, agriculture, and electronic waste produce a variety of anthropogenic
64 contaminants that commonly enter the environment surrounding urban areas that are
65 known to cause cancer, reproductive and immune system impairments across a range of
66 wildlife (Carpenter, 2006; Darnerud et al., 2001; Lohmann et al., 2007; Sindermann,
67 2005). In particular, large coastal cities can heavily pollute surrounding coastal and
68 estuarine habitats (Sindermann, 2005). **Because some sea turtle species, such as green
69 sea turtles (*Chelonia mydas*), demonstrate high site fidelity and extended residency
70 in foraging habitats (Lutz et al., 2002), sea turtles that inhabit these urbanized
71 coastal foraging grounds have higher risk of exposure to anthropogenic pollutants
72 and can accumulate POPs over time serving as bioindicators for urban habitats
73 (Finlayson et al., 2016; Hamann et al., 2010; Keller, 2013).**

74 Persistent organic pollutants (POPs), one group of anthropogenic contaminants,
75 are made for many industrial and agricultural purposes and are introduced into the
76 environment via urban and agricultural runoff (Flynn and Kleiman, 1997; Gray, 1997;
77 Jones and de Voogt, 1999). POPs include compounds such as polychlorinated biphenyls
78 (PCBs), organochlorinated pesticides (OCPs), and polybrominated diphenyl ethers
79 (PBDEs). Due to their stability in the environment and accumulation in wildlife, PCBs
80 and OCPs are still found in the environment despite being banned in the 1970s (Jones and
81 de Voogt, 1999; Weber et al., 2008). Because of their prevalence and persistence in the

82 environment, there are still numerous questions about the exposure and potential health
83 impacts of POP accumulation in marine species.

84 Research into the potential effects of PCBs, OCPs, and PBDEs on sea turtle
85 health has increased in recent years (Finlayson et al., 2016; Gardner and Oberdorster,
86 2005; Keller, 2013; Pugh and Becker, 2001). For example, *in vitro* DDT exposure has
87 been shown to alter testosterone hormone binding in blood plasma obtained from nesting
88 green sea turtles in Malaysia, suggesting that DDT may disrupt sex steroid-protein
89 binding in green sea turtle blood (Ikonomopoulou et al., 2009). Research on nesting
90 turtles in Malaysia found that hatchlings with higher POP concentrations were smaller in
91 size, which may indicate reduced survival (van de Merwe et al., 2010b). Leatherback sea
92 turtle eggs in Costa Rica found that higher POP concentrations were correlated to lower
93 hatching success (De Andrés et al., 2016). Experiments with an immortal sea turtle testis
94 cell line was found to have increased cytochrome 450 aromatase activity following
95 exposure to POPs, indicating sex steroid alterations (Keller and McClellan-Green, 2004).
96 These studies indicate that POPs may competitively inhibit sex hormone binding, affect
97 hatchling survival, and impair immune function through depressed or increased white
98 blood cell count in sea turtles (De Andrés et al., 2016; Keller and McClellan-Green,
99 2004; Komoroske et al., 2011; Stewart et al., 2011; van de Merwe et al., 2010b). By
100 quantifying POP accumulation in urban green turtle foraging aggregations, wildlife and
101 endangered species managers can assess potential exposure risks and population recovery
102 strategies.

103 In California, USA, year-round foraging aggregations of green sea turtles are
104 found in several bays and estuaries from Los Angeles to San Diego (Crear et al., 2017,

105 2016; Eguchi et al., 2010; MacDonald et al., 2012). **These locations are highly-**
106 **urbanized areas known to have POPs present in sediment and wildlife through**
107 **previous research and sediment analysis** (Dodder et al., 2016; Lyons et al., 2014;
108 Lyons and Lowe, 2015; Schiff et al., 2011). Prior research has shown that green sea
109 turtles in San Diego Bay have detectable levels of POPs above the no effect threshold for
110 immunological impairment (e.g., altered lymphocyte production and cytokine gene
111 expression) (Komoroske et al., 2011; Lewison et al., 2011). A growing population of
112 green sea turtles has been identified in the Long Beach/Los Angeles area within the Seal
113 Beach National Wildlife Refuge (SBNWR) and San Gabriel River, which provides heat
114 effluent water allowing year around use of this foraging habitat (Crear et al., 2017, 2016).
115 These SDB and SBNWR green sea turtles provide an opportunity to compare these two
116 subpopulations that inhabit two distinct urban areas.

117 Considering distance and differences in exposure to pollutants between the two
118 locations, the current study hypothesizes that POPs will differ between green sea turtles
119 from SBNWR and SDB. To assess POP concentration patterns, blood plasma samples
120 will be used to measure POP concentrations in green sea turtles caught in SBNWR and
121 SDB. Previous studies have used lipid-rich tissues, such as liver and adipose, to quantify
122 body burden (Rozman and Klaassen, 2007) of POPs in green sea turtles. However, blood
123 samples have been shown to correlate with POP contamination in lipid-dense organs,
124 albeit at lower concentrations, and thus may provide a non-invasive sampling alternative
125 for measuring POP exposure and bioaccumulation (Finlayson et al., 2016; Gardner and
126 Oberdorster, 2005). This study aims to assess if location-based pollutant signatures are

127 present, how pollutant signatures differ between foraging aggregations, and compare the
128 results with other studies in green turtle POP accumulation.

129 **Methods**

130 *Study Sites*

131 San Diego Bay (32° 36' 54" N, 117° 6' 4" W; SDB) is a natural bay that contains
132 salt marsh, eelgrass bed, mud/tidal flat habitat. SDB's coastline is heavily urbanized with
133 homes, military bases, harbors, and shipyard activity throughout the bay (Figure 4).

134 Green sea turtles have inhabited these waters for a long period of time feeding on the
135 eelgrass beds within the bay (MacDonald et al., 2012; McDonald et al., 1994). SDB adult
136 greens nest and are from the genetic stock of the Revillagigedo Islands and Michoacan,
137 Mexico (Dutton et al., 2019). The SDB green turtle foraging aggregation has been studied
138 regularly with capture/recapture and monitoring taking place within the bay since 1990
139 (Eguchi et al., 2010).

140 The Seal Beach National Wildlife Refuge (33° 44' 07" N, 118° 03' 52" W;
141 SBNWR) is a wetland area within the Anaheim Bay estuary (Figure 4). SBNWR estuary
142 contains natural habitat as well as restored habitat that was constructed as part of a
143 restoration project. Within the restored habitat are a series of channels and basins that
144 house eelgrass beds, which green sea turtles are known to forage (Crear et al., 2017,
145 2016). Green sea turtle capture efforts were conducted within a pond that is fed by a
146 culvert which green sea turtles use to enter the pond and forage. This wetland is adjacent
147 to a naval weapons base with green sea turtles travelling between the ponds in the refuge
148 and the San Gabriel River (Crear et al., 2017, 2016).

149 The San Gabriel River (33° 45' 15" N, 118° 6' 13" W; SGR) is a concrete lined

150 river that ends in a 6 km stretch of estuarine habitat (Figure 4). The river acts as a flood
151 control channel with tributaries throughout the Greater Los Angeles area feeding into the
152 river. There are two power plants 3 km from the river's mouth that use once-through
153 cooling, drawing water from the San Gabriel River to cool steam generators (Crear et al.,
154 2016). As a result, the San Gabriel River's water temperatures are regularly altered via
155 heated water discharge from once-through cooling. Green sea turtles are increasingly
156 found within SGR in recent years and individuals are known to move regularly between
157 the SGR and adjacent SBNWR (Crear et al., 2016).

158 *Sea Turtle Capture and Sampling*

159 Whole blood samples were collected as described in Barraza et al. (2019).
160 Briefly, green sea turtles were captured, subadults were sexed via testosterone (T) levels
161 in the blood (Allen et al., 2015) and, adult-sized turtles via morphology (Caldwell, 1962),
162 weighed (± 0.1 kg), and measured for curved carapace length (CCL; ± 0.1 cm).
163 Methodology used is from previous research conducted within SDB (32° 36' 54" N, 117°
164 6' 4" W) and the SBNWR (33° 44' 06.8" N, 118° 03' 51.9" W)(Crear et al., 2016;
165 Eguchi et al., 2010). Whole blood (3–10 ml) samples were collected and prepared for
166 POP analysis (see below) following a modified National Institute of Standards and
167 Technology (NIST) protocol to reduce the possibility of sample contamination (Keller et
168 al., 2014b). Changes to NIST protocol include: using kilned glass containers with Teflon
169 lids (Thermo Scientific) instead of Teflon containers, and using kilned aluminum instead
170 of hexane rinse aluminum. Blood was collected with powder free nitrile gloves (Kimtech,
171 Roswell, Georgia) into glass sodium-heparinized tubes (Becton Dickson, San Jose,
172 California) and stored in a cooler with ice packs. At the end of each field day, blood

173 samples were centrifuged at 3000 rpm for 10 min to separate plasma. Plasma was
174 transferred into glass vials. Samples were placed at -20°C overnight then transferred to
175 -80°C freezers until POP analysis.

176 Sea turtles were held for approximately 1 hour for morphometric and blood
177 sample collection; afterward, they were released at their location of capture. Additional
178 plasma samples were provided for POP analyses from previous samples collected using
179 NIST protocols (2009–2014) of green sea turtles in SDB ($n = 4$) and the San Gabriel
180 River (SGR, $n = 6$), a river within 4 km of SNBWR. These supplementary samples
181 followed collection methods described in Keller et al. (2014).

182 *POP Analyses*

183 Blood plasma samples were analyzed for 14 PBDEs, 11 OCPs and 54 PCBs
184 (specific analytes listed in Table 2 and Table S4). POPs were extracted using a soxhlet
185 extraction with dichloromethane solvent. Blood plasma was placed directly into sodium
186 sulfate in a thimble to dry and placed through a soxhlet extraction overnight. Extracts
187 were column cleaned with an Alumina-B/Silica column to reduce lipid interference and
188 concentrated with a vacuum-sealed rotary-evaporator followed by nitrogen evaporation.
189 Extracts were analyzed using an Agilent Gas Chromatograph Mass Spectrometer
190 (7890A/5975C) equipped with a J&W 60 meter, 0.25 mm ID, 0.25 μm film thickness
191 DB-5 column via a splitless injection at a temperature of 285°C . The oven temperature
192 profile was programmed from 45°C to 150°C at $20^{\circ}\text{C}/\text{min}$, and then to 300°C at
193 $2.5^{\circ}\text{C}/\text{min}$. PCBs and OCPs were analyzed using a mass selective detector (MSD) in
194 Electron Ionization (EI) selected ion monitoring (SIM) mode to scan for PCB and OCP
195 specific ions at 1.67 times/sec. PBDEs were analyzed using the MSD in Negative Ion

196 Chemical Ionization (NCI) mode using methane. A standard curve for all pollutants was
197 based on a 6-point linear regression calibration curve with an R^2 value of 0.99. All
198 calibration standards were NIST traceable standards (AccuStandard©). For quality
199 assurance, each sample batch was analyzed with a standard reference material (SRM
200 1957, NIST), two laboratory blank spikes, and a laboratory blank. Limits of detection in
201 sea turtle plasma were determined using spiked sea turtle plasma with a 6-point linear
202 regression; the standard deviation of spikes was divided by the slope of the regression
203 and multiplied by 3.3. For a conservative approach, the congener with the highest limit of
204 detection for each POP type was used as the limit of detection for all congeners.
205 Recovery surrogates (TCMX, PCB 30, PCB 112, and PCB 198) were added to each
206 sample prior to extraction, and all samples were quantified using the internal standards
207 dibromobiphenyl and tetrabromobiphenyl. Blank spikes, SRM values, and recovery
208 surrogates were analyzed for percent recovery to assess the accuracy and precision of the
209 selected methods (supplementary Tables S2, S3, and S4). To account for lipid content
210 difference, percent lipid was determined via weighing a 25% lipid split of extract and
211 dividing by 25% volume of blood plasma used in the extract.

212 *Statistical Analyses*

213 Statistical analyses were done using R (version 3.3.3; R Core Team, 2018), with a
214 significance threshold of $\alpha = 0.05$. To assess current differences in POP levels in
215 SDB and SBNWR turtles, samples collected before 2015 were not included in summary
216 statistics of POPs. Samples collected before 2015 were used to provide data for key
217 habitat locations for which little other data exist, and to provide data for future
218 comparisons. Four blood plasma samples from SDB green sea turtles and six blood

219 plasma samples from individuals in the SGR collected during another study were
220 processes and analyzed separately. Since no PBDEs were detected below or above LOD
221 in any samples, they were not included in the current study's analyses.

222 Two methods were used to summarize POP data. First, all detected PCB and OCP
223 analytes were summed to calculate total POP (Σ POPs) concentration detected per turtle
224 and summed by pollutant group to calculate total PCB or total OCPs (Σ PCB or Σ OCP)
225 per turtle. Second, a non-parametric Kaplan-Meier model (as described in Helsel, 2012)
226 was used to account for detections below the limit of detection (LOD) and to provide an
227 average value for each PCB and OCP analyte. For a conservative approach to avoid
228 overestimation, and because of resource limitations, the highest congener LOD for PCBs
229 and OCPs, respectively, was used as the cut-off for categorizing values below LOD in the
230 Kaplan-Meier models (Table S1). Σ POPs/PCBs/OCPs for individuals were not calculated
231 using the Kaplan-Meier model because the data were over 70% censored, which previous
232 research has shown to not provide a good estimate of concentrations (Antweiler and
233 Taylor, 2008). As a result, to avoid problems added by substitution (Helsel, 2012) or
234 statistical treatment (Antweiler and Taylor, 2008), Σ PCB and Σ OCP included all detected
235 values (whether above or below LOD), and non-detected analytes (analytes not found at
236 all in scans) were treated as zero. To normalize data and account for individual lipid
237 differences, detected POPs were measured in ng/g blood plasma, converted to ng/g lipid
238 and natural log transformed for all statistical analyses. POPs that were not detected in any
239 plasma samples were omitted from summary tables (Tables 1 and 2) for clarity.

240 One SBNWR turtle and three SDB turtles were recaptured and had repeat blood
241 samples taken. Mean POP concentrations of recaptured turtles were used for analyses; no

242 green turtle was captured more than twice. Using the R package *vegan* (Oksanen et al.,
243 2015), principal component analyses (PCA) were conducted to assess location-based
244 pollutant patterns and included CCL, Σ PCB, and Σ OCP of each individual. Using the R
245 package *cluster* (Maechler et al., 2016), k-means cluster analyses of the PCA were
246 conducted to assess how individuals clustered by location. Regression analyses were used
247 to find relationships between POP types and CCL. A multivariate analysis of variance
248 (MANOVA) was used to compare Σ PCB and Σ OCP between locations and among size
249 classes. A second MANOVA was conducted that only included turtles of similar size for
250 comparison (Table 2; between 60 and 85 cm CCL), and therefore included similarly aged
251 turtles that are considered sub-adults (Eguchi et al., 2012; Figueroa et al., 1992; Juarez-
252 Ceron et al., 2003).

253 **Results**

254 From August 2015 to May 2017, whole blood samples were collected from 23
255 green sea turtles from SDB and 16 green sea turtles from SBNWR. Green sea turtles
256 captured in SDB were significantly larger than individuals from SBNWR ($p < 0.001$;
257 Figure 1). Of the 79 POPs assessed (14 PBDE congeners, 54 PCB congeners, and 11
258 OCP analytes), only 32 PCB congeners (LOD = 0.27 ng/g) and 6 OCP analytes (LOD =
259 0.18 ng/g) were detected at concentrations above the LOD (see Table 1). PCB congeners
260 not detected in any samples included: PCB 3, 8, 18, 28, 31, 33, 37, 44, 49, 52, 56 (60),
261 70, 74, 77, 81, 97, 119, 123, 126, 169, and 199 (200). OCP analytes not detected in any
262 samples included: 4,4'-DDMU, 2,4'-DDE, 4,4'-DDD, 2,4'-DDT, and 4,4'-DDT. No
263 PBDE analytes (PBDE 17, 28, 47, 66, 71, 85, 99, 100, 138, 153, 154, 183, 190, and 209)
264 were detected (LOD = 0.86 ng/g) in any green turtle blood plasma sample.

265 PCA k-means cluster analysis grouped green sea turtles from the same location
266 together with some overlap between the different locations (Figure 2). From the PCA,
267 PC1 (Σ PCBs) accounted for 58% of the variance, and PC2 (CCL) accounted for 30% of
268 the variance. There were more PCB congeners and OCP analytes detected (Table 1) in
269 SDB turtles (30 PCB congeners; 5 OCPs) than SBNWR turtles (19 PCB congeners; 4
270 OCPs). Several congeners that displayed differences in the frequency of detection
271 between populations included PCB 180, 187, 149, and 153 (see Table 1 and Table 2).
272 PCB 180 and 187 were detected in all SDB turtles, PCB 99 was only detected in one
273 SBNWR turtle but detected in nearly all SDB turtles, PCB 149 and 153 were detected in
274 all SBNWR turtles (and all but one SDB turtle), and PCB 138 was detected in every
275 turtle captured in the present study. Overall, the concentration of PCBs strongly varied
276 between SDB and SBNWR green turtle plasma samples; in particular, SDB turtles had
277 much higher concentrations of PCB 153, PCB 138, and PCB 99 in their blood plasma.
278 With the exception of moderate levels of PCB 118 in SDB green sea turtles, there were
279 low to non-detectable levels of dioxin-like PCBs (Table 1), such as PCB 77, 126, 169,
280 and 105 in the plasma of all green sea turtles from the current study. The Σ PCBs detected
281 in all turtles made up 95.1% by ng/g lipid of all POPs detected, and Σ OCPs made up
282 4.9% ng/g lipid of POPs detected. There were no significant differences in Σ PCB and
283 Σ OCP concentrations between male (n = 6) and female green sea turtles (n = 11)
284 regardless of capture location (data not shown). Not all turtles in the current study had
285 sex determined by testosterone measurements; all sex undetermined turtles were
286 juveniles. Two MANOVAs of PCB and OCP concentrations, one including all samples
287 and another including only sub-adults, both showed that location, not CCL or the

288 interaction of CCL and location, was a significant ($p < 0.001$) factor for PCB
289 concentrations (Table 3).

290 The MANOVA including all samples indicated that SDB green sea turtles had
291 significantly more Σ PCBs (7.16, 6.93 ± 0.22 ln ng/g lipid; median, mean \pm SE) in their
292 blood plasma than SBNWR (4.33, 4.49 ± 0.22 ln ng/g lipid) green sea turtles (Figure 3A).
293 MANOVA comparing similarly sized turtles (see methods) demonstrated that sub-adults
294 from SDB (6.45, 6.26 ± 0.29 ln ng/g lipid) had significantly more Σ PCBs (4.34, $4.36 \pm$
295 0.21 ln ng/g lipid) than SBNWR sub-adult turtles (Figure 3B).

296 There was no significant difference in Σ OCPs between turtles captured in SDB
297 (3.65, 2.90 ± 0.66 ln ng/g lipid) and SBNWR (3.80, 2.48 ± 0.94 ln ng/g lipid); and there
298 was no relationship between Σ OCPs and turtle CCL ($R^2 = 0.006$, $p = 0.52$). The most
299 commonly-occurring OCPs in green turtle plasma from both locations were chlordane-
300 gamma and trans-nonachlor. Most of the DDT metabolites were not detected or were
301 only present in blood plasma from one turtle at each location (Table 1).

302 Only four green sea turtles were recaptured for the duration of the study, three
303 from SDB and one from SBNWR. Three of the four turtles showed increased (+70.22%,
304 +248.30%, +367.55% ng/g lipid) Σ POP concentrations from their initial capture (Table
305 4). One turtle had decreased (-23.01%) Σ POPs from its initial capture (Table 4). Changes
306 in Σ POP concentrations were mainly driven by increases (+74.29%, +240.35%,
307 +383.71%) or decreases (-24.41%) in Σ PCB concentrations.

308 Samples from previous SDB green turtle blood collections had significantly ($p =$
309 0.002) higher Σ PCBs (8.95, 8.75 ± 0.30 ln ng/g lipid) and significantly ($p = 0.001$) higher
310 Σ OCPs (5.66, 5.63 ± 0.18 ln ng/g lipid) than SDB green sea turtles measured in the

311 current study. Green sea turtles from SGR had significantly ($p = 0.004$) higher Σ PCBs
312 (7.99, 7.52 ± 0.64 ln ng/g lipid) but similar Σ OCPs (5.09, 5.18 ± 0.49 ln ng/g lipid) than
313 green sea turtles from SBNWR. Results from additional samples are provided the
314 supplementary section for future comparisons (Table S4).

315 Percent recovery of SRM, plasma, blank, and blank spikes TCMX recovery
316 surrogate accuracy was 67.71–99.95% and precision was ± 1.79 –6.88% (see Table S2).
317 In all samples, accuracy of PCB 30 was 77.23–100.80%, and precision was 1.38–6.75%.
318 Detection of PCB 112 had an accuracy of 92.17–105.65% and a precision of 1.75–6.38%.
319 Accuracy of PCB 198 was 70.42–100.56% in all samples, and precision of PCB 198 in
320 all samples was ± 0.49 –8.16%. Detection of FPBDE had an accuracy of 76.55–88.01%
321 and precision of 3.44–8.77 %. Detection of DFPBDE had an accuracy of 52.35–74.75%
322 and precision of 2.26–6.70%. There was no outside contamination detected in any blanks
323 analyzed.

324 **Discussion**

325 *Location Pollutant Signatures*

326 As expected, there were unique location-specific pollutant signatures in green sea
327 turtles inhabiting SBNWR and SDB. Green sea turtles from SBNWR had similar PBDE
328 and OCP concentrations to SDB green sea turtles; however, the majority of POPs
329 detected in green turtle blood plasma were PCBs. As a result, PCBs constituted the
330 highest proportion of green turtle POP loads by ng/g lipid and were the strongest factor in
331 the separation of green sea turtles by location in the PCA loading plot (Figure 2). These
332 patterns are reflected in the higher concentration and variety of PCBs in SDB green turtle
333 blood plasma relative to SBNWR green sea turtles, which suggests that SDB green sea

334 turtles experience higher PCB exposure than SBNWR green sea turtles (Table 1; Table
335 2).

336 *Differences Between Sample Locations*

337 The current study measured PCB concentrations green sea turtles from two
338 locations, approximately 160 km apart. The two green sea turtle aggregations have some
339 biological factors (e.g., size and sex) that could influence their PCB accumulation
340 patterns. Previous research has shown that SDB green sea turtles' POPs correlate with
341 CCL (Komoroske et al., 2011); therefore, it could be expected that larger, older
342 individuals would have higher PCB concentrations as seen in other sea turtle species
343 (Finlayson et al., 2016; Pugh and Becker, 2001). SDB green sea turtles were larger than
344 green sea turtles caught in SBNWR (Figure 1), and PCB differences could be due to
345 longer exposure times. Although bioaccumulation with age/size has been found in turtles
346 from other locations (Finlayson et al., 2016; Gardner and Oberdorster, 2005) and the data
347 often trended with CCL (Figure 2, 3C), the current study did not find significant
348 relationships between size (CCL) and PCB levels (Table 3). However, the current study
349 captured only a small size range of SBNWR green sea turtles (50.8–82.6 cm CCL); thus,
350 future capture of an increased range in size of individuals from SBNWR would help
351 determine if there is a relationship between size and POP levels. It is possible that
352 SBNWR currently represents a relatively new year-round expansion of green turtle
353 foraging habitat, utilized primarily by smaller juvenile individuals that will take some
354 time to grow into adults. It is also possible that adult turtles could not fit into the culverts
355 that leads into the capture location within SBNWR as there were adult turtles caught

356 within the SGR in previous sampling years. However, one adult turtle was seen in
357 SBNWR, indicating some adults could access the ponds inside SBNWR.

358 Sex is another biological factor that may influence pollutant load (Keller, 2013).
359 Green sea turtles from SDB are mostly female (>75%) and of adult-size (>85cm CCL),
360 whereas turtles collected from SBNWR are also mostly female, but smaller than adult-
361 size (<85 cm CCL)(Allen et al., 2016, 2015). Female green sea turtles can maternally
362 offload POPs by metabolizing fat reserves that transfer lipids, and therefore POPs, to
363 their eggs (Munoz and Vermeiren, 2019; van de Merwe et al., 2010b). As a result, there
364 is potential for adult female green sea turtles to exhibit lower blood plasma POPs due to
365 maternal offloading, yet, the data did not support that conclusion as female and male
366 adult turtles in this study had similar POP concentrations. The current study had only six
367 adult males, and all but one was from SDB, which is insufficient to observed sex-based
368 POP differences.

369 Green sea turtles in SBNWR have access to anthropogenically-warmed waters
370 from power plants within SGR (Crear et al., 2017, 2016). In contrast, SDB green sea
371 turtles in the present study live under more natural conditions—although it should be
372 noted that data from Komoroske et al. (2011) in SDB were from a period when waters
373 were warmed from the south San Diego Bay power plant. Anthropogenically warmed
374 waters could explain why SDB green sea turtles had lower PCB levels when the power
375 plant was active (since monitoring began) than after power plant closure in December
376 2010. When SDB green sea turtles had access to anthropogenically warmed waters, their
377 growth rate was similar to green sea turtles that inhabit tropical waters (Eguchi et al.,
378 2012). While not confirmed, it is postulated that SBNWR/SGR green sea turtles have an

379 accelerated growth rate due to their access to anthropogenically-warmed waters.
380 Depending on pollutant availability, accelerated growth rates could be diluting SBNWR
381 green sea turtles' PCB concentrations through increased mass from growth and the
382 opposite for SDB green sea turtles currently inhabiting non-warmed waters. While it can
383 be argued that warmer waters can increase the rate at which green sea turtles feed on PCB
384 contaminated food, thereby increasing their potential PCB exposure, results indicate that
385 overall, SBNWR turtles have lower PCB concentrations than SDB green sea turtles.

386 *Comparison to Previous Research*

387 Previous research by Komoroske et al. (2011) examined POPs in SDB green sea
388 turtles and captured a size range of green sea turtles similar to those captured in the
389 current study. In the current study, green sea turtles from SDB and SBNWR had lower
390 chlordane gamma and chlordane alpha than previously assessed in SDB, possibly
391 indicating a reduction in bioavailable pesticides or exposure (Komoroske et al., 2011;
392 Lewison et al., 2011). It is also important to note that Komoroske et al. (2011) had
393 different LODs for each of the POPs analyzed than the current study, with LODs lower in
394 the current study for PCBs and OCPs but higher for PBDEs. Unlike Komoroske et al
395 (2011), the current study did not find any PBDEs in blood plasma samples. These
396 findings could indicate a decrease in PBDE contamination at both sites samples were last
397 taken, especially since PBDEs concentrations are decreasing throughout the Southern
398 California Bight (Dodder et al., 2016, 2012). The current study had a higher LOD and
399 recovery surrogates were relatively low compared to other analytes, but SRM
400 measurements were accurate and precise. There is a possibility that the higher LOD
401 would miss low PBDE concentrations in green turtle blood plasma; however, there was

402 no indication of any PBDE concentrations at or below the LOD in gas chromatography
403 mass spectrometry scans.

404 SDB turtles have among the highest Σ PCBs detected in blood plasma compared to
405 previous SDB samples and other turtles worldwide, with studies detecting Σ PCBs as low
406 as 2.84 pg/g blood plasma and as high as 5.38 ng/g blood plasma compared to 30.11 ng/g
407 Σ PCBs found in an SDB turtle (Camacho et al., 2014; Keller et al., 2014a; Komoroske et
408 al., 2011; Lewison et al., 2011; Swarthout et al., 2010; van de Merwe et al., 2010b,
409 2010a). Conversely, SBNWR green sea turtles had similar or lower Σ PCB levels in their
410 blood than other studies worldwide (Keller, 2013). Additional samples collected in 2011–
411 2013 from green sea turtles inhabiting the SGR had higher PCB concentrations than
412 SBNWR turtles. Sediment samples from previous research found very low (0 – 1.77
413 ug/kg dw) PCB concentrations near the SGR and SBNWR (Dodder et al., 2016), possibly
414 due to differences in food items available between the locations (eelgrass in SBNWR and
415 algae in the SGR)(Crear et al., 2016). Green sea turtles sampled from the SGR were
416 similar in size to the current study's SBNWR green sea turtles, except for one adult turtle
417 (93 cm, CCL), which had PCB levels analogous to other adult turtles from SDB samples.
418 While the sample size was low for SGR turtles (n = 6) and the foraging locations are
419 within 8 km of each other, the current study's data indicate that turtles from SGR may
420 have higher PCB exposure and ensuing risk of PCB accumulation than turtles from
421 SBNWR. Overall, the current study's results indicate that PCB exposure risk in SDB
422 turtles may have increased since previous studies and that the SDB location may
423 represent one of the highest PCB-contaminated green turtle populations studied to date
424 (Finlayson et al., 2016; Gardner and Oberdorster, 2005; Keller, 2013). Nonetheless,

425 previous research has shown that blood plasma PCB concentrations can vary with time
426 and between samples (Lewison et al., 2011).

427 *Variance from Lipid Mobilization*

428 Previous research has shown that the mobilization of adipose or recent dietary
429 exposure could also temporarily increase POP concentrations in blood plasma, indicating
430 that POP concentrations can vary depending on whether green sea turtles are captured
431 before or after breeding migrations that require fasting or recent feeding events (Hamann
432 et al., 2002; Keller et al., 2004). The current study's Σ PCB, Σ OCPs, and Σ POPs
433 concentrations were lipid-normalized to account for these lipid mobilization events. Four
434 green sea turtles were recaptured, three from SDB and one from SBNWR, with three out
435 of the four turtles having increased Σ POPs (+74%, +240%, + 383% ng/g lipid) since their
436 initial capture (Table 4). Of note, three out for four recaptured green sea turtles had
437 higher Σ POPs in ng/g lipid in the fall/winter months than the spring/summer months,
438 suggesting that these green sea turtles POP concentrations continually change even when
439 accounting for increased lipid in blood plasma. Previous research that has repeated
440 sampling of individuals in SDB found that POP concentrations can vary with time of
441 sampling and even within samples of the same individual collected on the same day
442 (Lewison et al., 2011). The results are also supported by additional blood samples
443 collected from 2011–2013, which further indicate that SDB green turtle POP
444 concentrations continuously fluctuate over time. These samples had higher PCB and OCP
445 concentrations compared to current (2015–2017) SDB turtle samples and those found in
446 2009 (Komoroske et al., 2011). Only four SDB turtles were analyzed as recaptures,
447 which makes it difficult to suggest that the whole aggregation is reflected in the analysis.

448 However, these samples and trends may suggest that time of year and/or temperature may
449 be affecting Σ POP concentrations found in the blood of these green sea turtles.

450 *Future Directions*

451 Overall, both populations of green sea turtles in the current study had low to non-
452 detectable levels of dioxin-like PCBs, a heavily studied group of PCBs (Domingo and
453 Bocio, 2007; Srogi, 2008). Rather, the most abundant PCB congeners detected were non-
454 dioxin-like PCBs (Table 1), such as PCB 138, 153, 187, and 180. These PCBs are known
455 to activate the ryanodine receptor or alter dopaminergic-signaling pathways (Holland et
456 al., 2017; Kenet et al., 2007; Pessah et al., 2006; Wigstrand et al., 2013; Yang et al.,
457 2009). Non-dioxin-like PCB actions through these pathways have been related to induced
458 muscle impairment, altered neuronal growth, and impaired learning and memory (Pessah
459 et al., 2010; Wayman et al., 2012; Wigstrand et al., 2013; Yang et al., 2009). Although
460 these previous studies were conducted in mammalian species, their findings suggest that
461 green sea turtles could be at risk for the induction of neurotoxicity due to PCB burdens.
462 Juvenile/hatchling green sea turtles born from SDB green sea turtles may receive high
463 PCB burdens through maternal transfer of non-dioxin-like PCBs, potentially negatively
464 impacting their early life stages. Given that the majority of PCBs detected in SDB and
465 SBNWR turtles were non-dioxin-like PCBs, and the current lack of research into
466 neurotoxic non-dioxin-like PCBs in reptiles, it would be beneficial to investigate these
467 possible effects in the SBNWR and SDB green sea turtles (Finlayson et al., 2016;
468 Gardner and Oberdorster, 2005). The current study did not include many other pollutants
469 (e.g., PAHs, plastics) that could be accumulating within southern California green sea
470 turtles, which future research could help determine risk and possible mixture interactions.

471 New research into POPs include: health panels for investigating health effects (Banerjee
472 et al., 2019; Keller et al., 2014a; Komoroske et al., 2011), non-targeted extraction
473 methods to evaluate mixture effects and monitor multiple compound types (Dogruer et
474 al., 2018; Heffernan et al., 2017; Vijayasathy et al., 2019), and combining non-targeted
475 extractions with cell line bioassays as measurements of contamination and toxicity (Allan
476 et al., 2017; Finlayson et al., 2019b, 2019c, 2019a; Jin et al., 2015). These new methods
477 and tools help link POP concentrations to various health effects and assess the effects of
478 multiple pollutant types; and while the body of research is growing, pollutant
479 physiological tipping points and pollutant mixture interactions are still not well
480 understood (Cortes-Gomez et al., 2017; Finlayson et al., 2016). Until more research is
481 completed, it is uncertain whether the amount POPs detected are high enough to have a
482 detrimental effect on SDB and SBNWR green sea turtles' health.

483 **Conclusions**

484 Overall, evidence was found that green sea turtles from SDB accumulated higher
485 PCB levels than green sea turtles from SBNWR. While factors such as size, and lipid
486 mobilization events can change PCB levels in the short term (6 months to 1.5 years), the
487 results indicate greater PCB levels in green sea turtles in SDB than those found in other
488 parts of the world (Keller, 2013). The current study's results suggest that green sea turtles
489 foraging within SGR are at greater risk of PCB accumulation than SBNWR turtles and
490 similar PCB levels to SDB turtles. The most common PCBs accumulated were non-
491 dioxin-like PCBs, indicating opportunities for future research to investigate the possible
492 effects of non-dioxin-like PCBs impacts on green turtle physiology. Overall, considering
493 the disparity and fluctuations in PCB accumulation patterns found, additional monitoring

494 of turtles within the Los Angeles and San Diego areas may be necessary. Even green sea
495 turtles foraging within proximate habitats (SBNWR vs. SGR) exhibited different PCB
496 accumulation patterns. While the health effects of these PCBs are currently unknown,
497 green sea turtles will continue to inhabit these urban areas for the foreseeable future due
498 to increasingly warm waters and population recovery (Seminoff et al., 2015). Urban
499 populations, such as the green sea turtles inhabiting critical habitat within southern
500 California, provide clues and opportunities to elucidate how green sea turtles are affected
501 by anthropogenic pollutants.

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540 **Table 1**

541 Summary persistent organic pollutants (PCB and OCP) detected in blood plasma samples
 542 collected from green sea turtles from the Seal Beach National Wildlife Refuge (n = 16) or
 543 San Diego Bay (n = 23). Analytes that were not detected in any green sea turtles sampled
 544 are not displayed for clarity, including PBDEs. Non-detects are listed as zeros. Mean \pm
 545 SE and median (range) are ng/g blood plasma. Limit of detection (LOD) for PCB
 546 congeners and OCP analytes is 0.27 and 0.18 ng/g, respectively.

<i>Congeners</i>	<i>San Diego Bay</i>			<i>Seal Beach National Wildlife Refuge</i>		
	n > LOD	Median (Range)	Mean \pm SE	n > LOD	Median (Range)	Mean \pm SE
PCB066	1	0 (0 – 0.27)	0.01 \pm 0.05	0	0 (0)	0
PCB087	1	0 (0 – 0.31)	0.01 \pm 0.06	0	0 (0)	0
PCB095	11	0 (0 – 0.22)	0.05 \pm 0.06	1	0 (0 – 0.11)	0.01 \pm 0.007
PCB099	21	0.73 (0 – 3.31)	0.86 \pm 0.68	1	0 (0 – 0.66)	0.05 \pm 0.04
PCB101	1	0 (0 – 0.13)	0.006 \pm 0.02	1	0 (0 – 0.14)	0.008 \pm 0.008
PCB105	1	0 (0 – 0.22)	0.01 \pm 0.04	0	0 (0)	0
PCB110	0	0 (0)	0	1	0 (0 – 0.08)	0.005 \pm 0.004
PCB114	1	0 (0 – 0.44)	0.02 \pm 0.08	0	0 (0)	0
PCB118	18	0.175 (0 – 0.94)	0.21 \pm 0.19	0	0 (0 – 0.17)	0.01 \pm 0.01
PCB128	11	0 (0 – 0.70)	0.12 \pm 0.15	0	0 (0 – 0.12)	0.007 \pm 0.007
PCB138	23	2.07 (0.13 – 11.82)	2.92 \pm 2.61	16	0.15 (0.07 – 2.06)	0.32 \pm 0.12
PCB141	0	0 (0)	0	1	0 (0 – 0.13)	0.008 \pm 0.008
PCB149	22	0.095 (0 – 0.33)	0.12 \pm 0.06	16	0.05 (0.02 – 0.28)	0.07 \pm 0.02
PCB151	17	0.03 (0 – 0.11)	0.03 \pm 0.03	6	0 (0 – 0.09)	0.01 \pm 0.006
PCB153	22	1.345 (0 – 11.92)	2.79 \pm 3.01	16	0.08 (0.03 – 1.32)	0.18 \pm 0.08
PCB156	3	0 (0 – 0.10)	0.01 \pm 0.02	0	0 (0)	0
PCB157	3	0 (0 – 0.09)	0.01 \pm 0.02	0	0 (0)	0
PCB158	1	0 (0 – 0.20)	0.01 \pm 0.04	0	0 (0)	0
PCB167	9	0 (0 – 0.22)	0.06 \pm 0.07	0	0 (0)	0
PCB168+132	1	0 (0 – 0.12)	0.01 \pm 0.02	1	0 (0 – 0.13)	0.008 \pm 0.008
PCB170	12	0.015 (0 – 0.31)	0.07 \pm 0.08	1	0 (0 – 0.07)	0.007 \pm 0.005
PCB174	9	0 (0 – 0.10)	0.02 \pm 0.02	2	0 (0 – 0.14)	0.01 \pm 0.008
PCB177	9	0 (0 – 0.10)	0.02 \pm 0.03	1	0 (0 – 0.08)	0.007 \pm 0.005
PCB180	23	0.21 (0.05 – 2.02)	0.38 \pm 0.41	11	0.04 (0 – 0.21)	0.05 \pm 0.01
PCB183	20	0.075 (0 – 0.57)	0.15 \pm 0.14	1	0 (0 – 0.10)	0.01 \pm 0.006
PCB187	23	0.26 (0.05 – 0.66)	0.30 \pm 0.15	13	0.03 (0 – 0.35)	0.06 \pm 0.02
PCB189	1	0 (0 – 0.02)	0.001 \pm 0.003	0	0 (0)	0
PCB194	5	0 (0 – 0.15)	0.02 \pm 0.04	0	0 (0)	0
PCB195	1	0 (0 – 0.04)	0.002 \pm 0.007	0	0 (0)	0
PCB201	9	0 (0 – 0.38)	0.08 \pm 0.10	0	0 (0 – 0.08)	0.005 \pm 0.005

PCB206	4	0 (0 – 0.09)	0.01 ± 0.02	0	0 (0)	0
PCB209	1	0 (0 – 0.04)	0.002 ± 0.007	0	0 (0)	0
Σ PCB	23	5.07 (0.36 – 30.11)	8.31 ± 7.25	16	0.32 (0.12 – 5.46)	0.85 ± 0.33
Chlordane-gamma	19	0.095 (0 – 0.19)	0.09 ± 0.05	14	0.1 (0 – 0.2)	0.10 ± 0.01
Chlordane-alpha	1	0 (0 – 0.09)	0.004 ± 0.02	1	0 (0 – 0.08)	0.005 ± 0.005
Trans-Nonachlor	19	0.09 (0 – 0.51)	0.12 ± 0.11	11	0.05 (0 – 0.21)	0.05 ± 0.01
4,4'-DDE	1	0 (0 – 0.40)	0.02 ± 0.07	0	0 (0)	0
2,4'-DDD	0	0 (0)	0	1	0 (0 – 0.89)	0.05 ± 0.05
Cis-Nonachlor	3	0 (0 – 0.14)	0.01 ± 0.03	0	0 (0)	0
Σ OCP	21	0.23 (0 – 0.75)	0.25 ± 0.16	14	0.15 (0 – 0.95)	0.20 ± 0.05
Σ POP	23	5.275 (0.36 – 30.79)	8.57 ± 7.36	16	0.44 (0.25 – 5.61)	1.05 ± 0.34
% Lipid	23	0.5 (0.13 – 2.61)	0.68 ± 0.10	16	0.41 (0.16 – 1.06)	0.47 ± 0.05

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581 **Table 2**

582 Summary persistent organic pollutants (PCB and OCP) estimated in blood plasma
 583 samples collected from green sea turtles from the Seal Beach National Wildlife Refuge (n
 584 = 16) or San Diego Bay (n = 23) using Kalpan-Meier model estimates. NAs are present
 585 where the model could not provide an estimate. Mean \pm SEM and 95% confidence
 586 interval (Conf. Int.) are ng/g blood plasma.

<i>Congener</i>	<i>San Diego Bay</i>		<i>Seal Beach National Wildlife Refuge</i>	
	Mean \pm SEM	95% Conf. Int.	Mean \pm SEM	95% Conf. Int.
PCB 3	0	0	0	0
PCB 8	0	0	0	0
PCB 18	0	0	0	0
PCB 28	0	0	0	0
PCB 31	0	0	0	0
PCB 33	0	0	0	0
PCB 37	0	0	0	0
PCB 44	0	0	0	0
PCB 49	0	0	0	0
PCB 52	0	0	0	0
PCB 56+60	0	0	0	0
PCB 66	0.27 \pm NA	NA	0	0
PCB 70	0	0	0	0
PCB 74	0	0	0	0
PCB 77	0	0	0	0
PCB 81	0	0	0	0
PCB 87	0.31 \pm NA	NA	0	0
PCB 95	0	0	0	0
PCB 97	0	0	0	0
PCB 99	0.88 \pm 0.16	0.57 - 1.19	0	0
PCB 101	0	0	0	0
PCB 105	0	0	0	0
PCB 110	0	0	0	0
PCB 114	0.44 \pm NA	NA	0	0
PCB 118	0.33 \pm 0.035	0.26 - 0.40	0	0
PCB 119	0	0	0	0
PCB 123	0	0	0	0
PCB 126	0	0	0	0
PCB 128	0.29 \pm 0.02	0.25 - 0.34	0	0
PCB 138	2.91 \pm 0.62	1.69 - 4.12	0.33 \pm 0.02	0.28 - 0.40
PCB 141	0	0	0	0
PCB 149	0.33 \pm NA	NA	0.28 \pm NA	NA
PCB 151	0	0	0	0

PCB 153	2.75 ± 0.72	1.35 - 4.16	0.54 ± NA	NA
PCB 156	0	0	0	0
PCB 157	0	0	0	0
PCB 158	0	0	0	0
PCB 167	0	0	0	0
PCB 168+132	0	0	0	0
PCB 169	0	0	0	0
PCB 170	0.28 ± 0.002	0.28 - 0.28	0	0
PCB 174	0	0	0	0
PCB 177	0	0	0	0
PCB 180	0.49 ± 0.09	0.31 - 0.66	0	0
PCB 183	0.32 ± 0.02	0.28 - 0.35	0	0
PCB 187	0.35 ± 0.03	0.29 - 0.40	0	0
PCB 189	0	0	0	0
PCB 194	0	0	0	0
PCB 195	0	0	0	0
PCB 199+200	0	0	0	0
PCB 201	0.34 ± 0.002	0.34 - 0.35	0	0
PCB 206	0	0	0	0
PCB 209	0	0	0	0
4,4'-DDMU	0	0	0	0
Chlordane-gamma	0	0	0	0
2,4'-DDE	0	0	0	0
Chlordane-alpha	0	0	0	0
trans-Nonachlor	0.43 ± 0.005	0.42 - 0.44	0	0
4,4'-DDE	0.40 ± NA	NA	0	0
2,4'-DDD	0	0	0.89 ± NA	NA
4,4'-DDD	0	0	0	0
2,4'-DDT	0	0	0	0
cis-Nonachlor	0	0	0	0
4,4'-DDT	0	0	0	0

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598 **Table 3**
599 Multivariate Analysis of Variance (MANOVA) results of persistent organic pollutant
600 concentrations in green sea turtles inhabiting the Seal Beach National Wildlife Refuge (n
601 = 16) or San Diego Bay (n = 23). “PCBs” are polychlorinated biphenyls, “OCPs” are
602 organochlorinated pesticides, and “CCL” refers to curved carapace length. “All” refers to
603 comparisons that include all turtles captured and analyzed in the current study (n = 39);
604 and “Sub-Adult” refers to comparisons which only include similar sized subadult turtles
605 (n = 23). “Comparison” refers to the dependent and independent variables in the
606 MANOVA.

<i>Comparison</i>	Mean Squares	F Value	P Value
All PCBs : Location	55.61	60.77	p < 0.001
All PCBs : CCL	2.02	2.21	p = 0.14
All PCBs : CCL*Location	2.32	2.53	p = 0.12
All OCPs : Location	0.23	0.12	p = 0.73
All OCPs : CCL	0.39	0.2	p = 0.65
All OCPs : CCL*Location	0.90	0.48	p = 0.50
Sub-Adult PCBs : Location	19.66	29.93	p < 0.001
Sub-Adult PCBs : CCL	1.32	2.01	p = 0.17
Sub-Adult PCBs : CCL*Location	0.01	0.02	p = 0.89
Sub-Adult OCPs : Location	0.10	0.04	p = 0.84
Sub-Adult OCPs : CCL	0.01	0.004	p = 0.94
Sub-Adult OCPs : CCL*Location	0.24	0.10	p = 0.75

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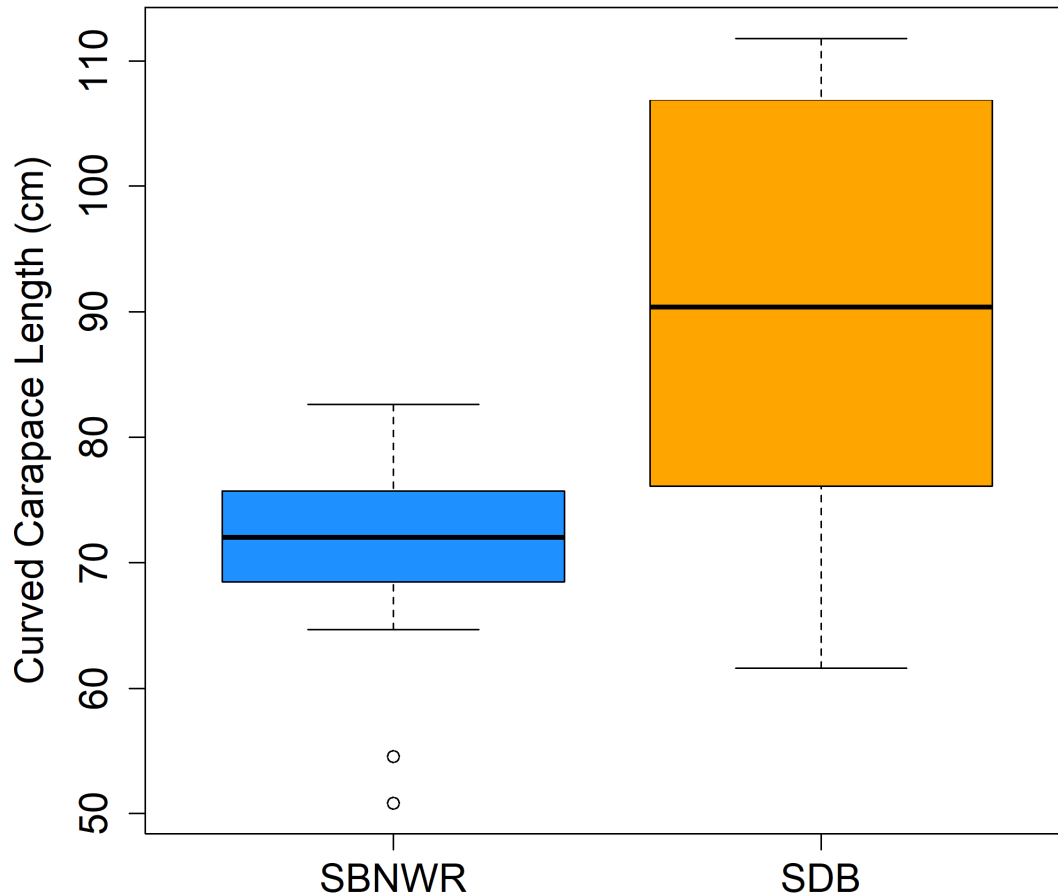
629 Summary of persistent organic pollutants (Σ POPs, Σ PCB, Σ OCP) concentrations in
 630 recaptured turtles, as well as their change in POP concentrations since their first capture.
 631 No turtle was caught more than twice. N.D. signifies that no analytes were detected.
 632 Σ POPs, Σ PCB, and Σ OCP concentrations are in ng/g blood plasma lipid. (% Δ) Refers to
 633 the percent change in analyte concentrations from first capture to recapture.

ID	Location	Date Captured	Σ PCBs	Σ OCPs	Σ POPs	% Δ PCBs	% Δ OCPs	% Δ POPs	Δ Time
GK-33	SBNWR	25/08/2015	37.31	16.42	53.73				
GK-33	SBNWR	7/12/2016	180.49	70.73	251.22	383.71	330.82	367.55	1 year, 4 months
GK-23	SDB	1/06/2016	635.56	37.78	673.33				
GK-23	SDB	10/11/2016	1107.69	38.46	1146.15	74.29	1.81	70.22	5 months
LB-326	SDB	15/12/2015	3645.21	38.36	3683.56				
LB-326	SDB	15/06/2016	2755.56	80.56	2836.11	-24.41	110.02	-23.01	6 months
LB-337	SDB	27/08/2015	173.58	N.D.	173.58				
LB-337	SDB	2/11/2016	590.80	13.79	604.60	240.35	NA	248.30	1 year, 3 months

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640 **Figure 1:** Boxplot of curved carapace length (CCL; cm) of green sea turtles captured

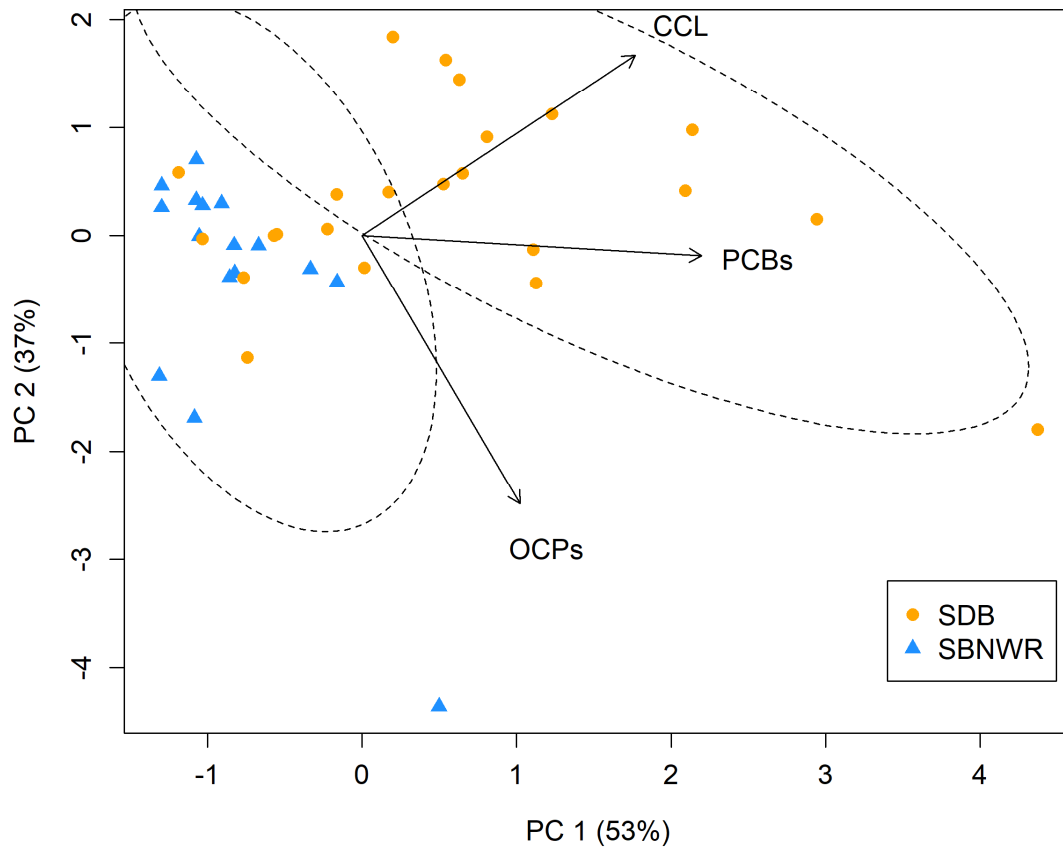
641 from Seal Beach National Wildlife Refuge (SBNWR, n = 16 turtles) and San Diego Bay

642 (SDB, n = 23 turtles). Boxes are the middle 50% quartile with the line representing the

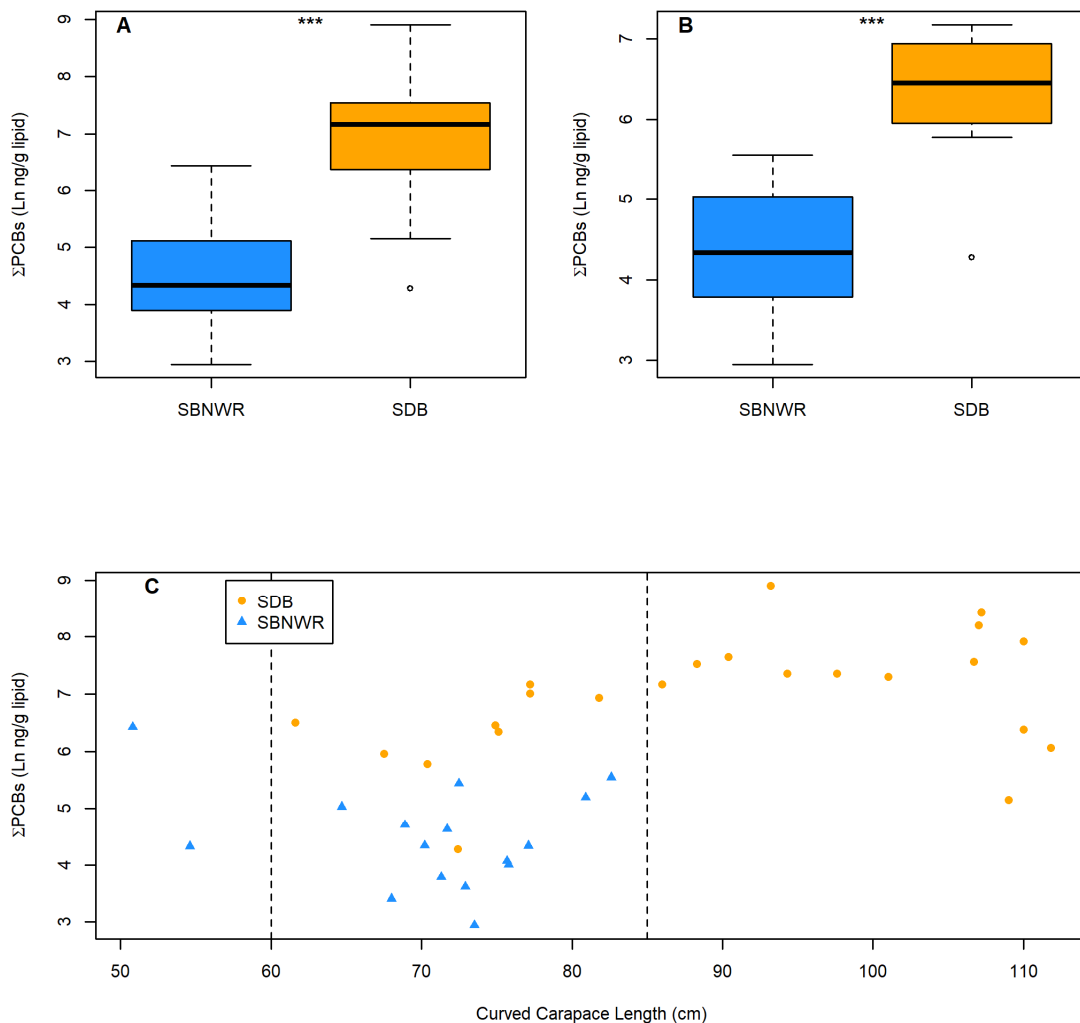
643 median, whiskers are top and bottom 25% quartile. X-axis represents CCL and y-axis

644 capture location of green sea turtles. SDB turtles are significantly ($p \leq 0.001$) larger than

645 SBNWR.

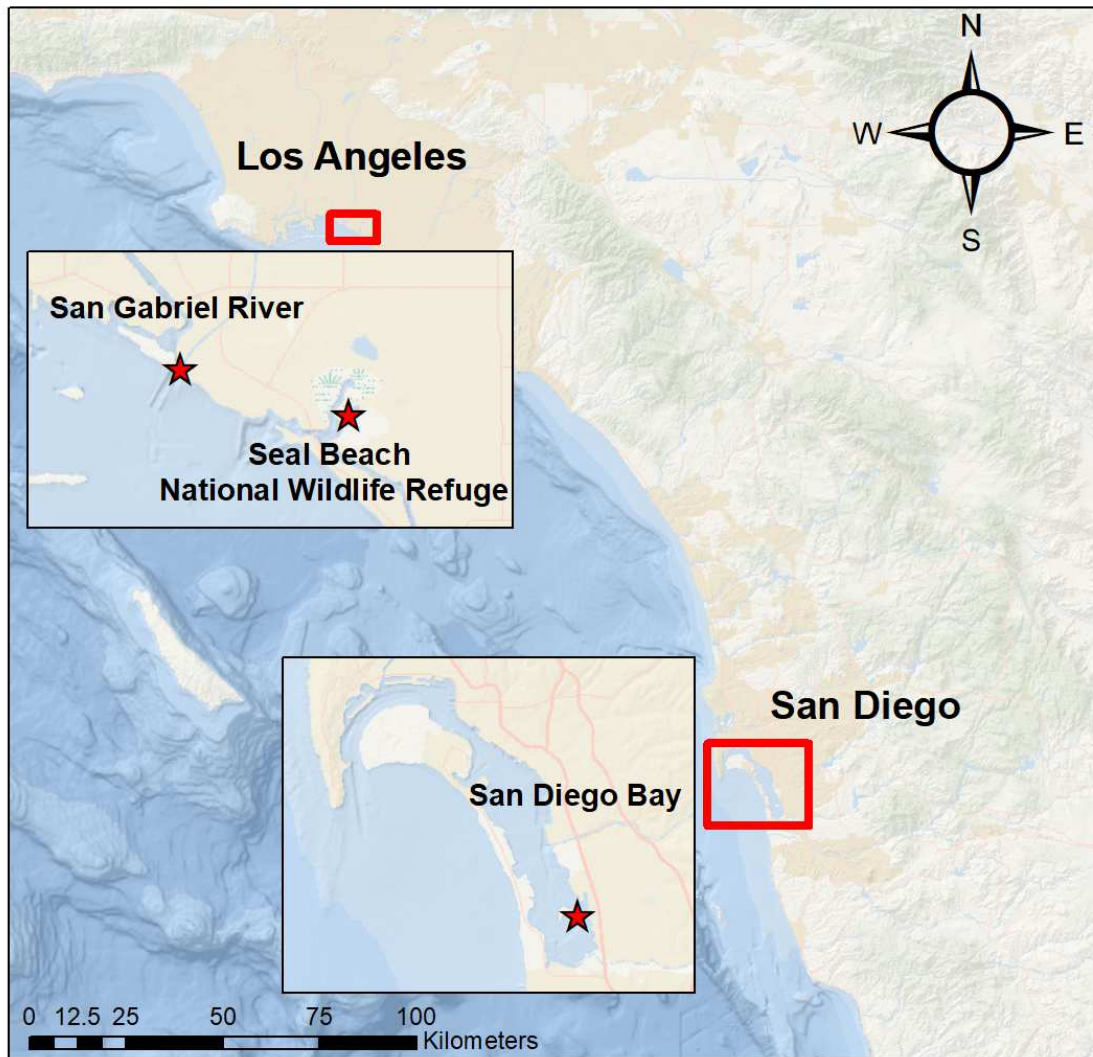


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 647 **Figure 2:** Principal component analysis of Σ PCBs and Σ OCPs in blood plasma lipid and
 648 curved carapace length (CCL) of green sea turtles from the Seal Beach National Wildlife
 649 Refuge (SBNWR; n = 16 green sea turtles) and San Diego Bay (SDB; n = 23 green sea
 650 turtles). Vectors indicate the direction that each factor affects principal component scores
 651 for each turtle (point). PC1 (x-axis) refers to Σ PCBs in ng/g blood plasma lipid, and PC2
 652 refers to CCL (cm).
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Figure 3: (A) Natural log transformed ΣPCBs in blood plasma lipid of green sea turtles from Seal Beach National Wildlife Refuge (SBNWR; n = 16 turtles; SBNWR) and San Diego Bay (SDB; n = 23 turtles; SDB). (B) Natural log transformed ΣPCBs in blood plasma lipid of sub-adult (between 60 and 85 cm CCL) green sea turtles from Seal Beach National Wildlife Refuge (SBNWR; n = 14 turtles) and San Diego Bay (SDB; n = 9 turtles). Y-axis represents ng/g lipid ΣPCBs natural log transformed in green turtle plasma samples (corrected for lipid content). X-axis represents location of sea turtles captured. Asterisks indicate significant differences via one-way ANOVA (*** ≤ 0.001). (C) Relationship of natural log ΣPCBs in blood plasma lipid and curved carapace length of green sea turtles from Seal Beach National Wildlife Refuge (SBNWR; n = 16 turtles) and San Diego Bay (SDB; n = 20 turtles). X-axis represents curved carapace length in centimeters of sea turtles captured. Y-axis represents natural log transformed ng/g ΣPCBs in sea turtle blood plasma lipid.



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Figure 4: Capture locations (red stars) for green sea turtles inhabiting the Los Angeles area (top square) and the San Diego area (bottom square) within Southern California, USA. Top square shows the San Gabriel River (left star) and the Seal Beach National Wildlife Refuge (right star); bottom square shows San Diego Bay (bottom star).

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689 **Supplementary Figures and Tables**

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691 **Table S1**692 Summary percent recovery of six persistent organic pollutant recovery surrogates in
693 standard reference material (SRM) and green turtle blood plasma samples (Plasma).694 Numbers are percent recovery \pm SE.

Surrogate	SRM	Plasma	Blanks	Blank Spikes
TCMX	91.11 \pm 6.88	99.95 \pm 1.79	67.71 \pm 2.46	98.11 \pm 3.44
PCB 30	91.31 \pm 6.75	100.8 \pm 1.38	77.32 \pm 1.75	100.13 \pm 2.7
PCB 112	105.65 \pm 6.38	110.03 \pm 1.75	92.17 \pm 4.62	98.97 \pm 2.14
PCB 198	70.42 \pm 8.16	72.69 \pm 0.49	84.83 \pm 6.58	100.56 \pm 0.57

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697 **Table S2**698 Summary of standard reference material (SRM) concentrations expected (Reference), the
699 mean \pm SE SRM concentrations detected (Detected), and the percent recovery of those
700 reference concentrations.

Analytes	Reference (ng/g)	Detected (ng/g)	Percent (% Recovery)
PCB 118	18.9 \pm 1.2	16.92 \pm 3.3	89.51 \pm 0.72
PCB 138	36.9 \pm 9.0	59.65 \pm 2.9	161.65 \pm 0.62
PCB 153	58.2 \pm 0.9	66.52 \pm 4.8	114.30 \pm 1.04
PCB 170	16.2 \pm 2.0	21.66 \pm 2.4	133.69 \pm 0.51
PCB 180/193	54.5 \pm 0.5	54.39 \pm 3.7	99.80 \pm 0.80
PCB 187	15.5 \pm 0.5	25.09 \pm 1.4	161.87 \pm 0.30
PCB 194	11.9 \pm 0.3	9.49 \pm 1.2	79.78 \pm 0.27
Trans-Nonachlor	58.3 \pm 1.9	77.19 \pm 7.9	132.40 \pm 1.71
4,4'-DDE	921 \pm 76	923.99 \pm 17.9	100.32 \pm 3.88
PBDE 47	268 \pm 14	271.46 \pm 3.9	101.29 \pm 1.43
PBDE 99	76 \pm 3.8	74.93 \pm 1.7	98.59 \pm 2.21
PBDE 100	49.7 \pm 2.7	49.63 \pm 3.7	99.87 \pm 7.53
PBDE 153	61.0 \pm 3.2	30.17 \pm 0.9	49.46 \pm 0.24

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702 **Table S3**

703 Summary percent recovery of blank spike analytes.

Congener Spiked	% Recovery
PCB 3	117.12 \pm 5.66
PCB 8	109.50 \pm 3.93
PCB 18	102.55 \pm 2.62
PCB 31	105.30 \pm 2.76
PCB 28	104.56 \pm 2.11
PCB 33	104.95 \pm 2.92

PCB 52	99.07 ± 2.16
PCB 49	99.27 ± 1.46
PCB 44	100.15 ± 0.99
PCB 37	102.57 ± 1.12
PCB 74	99.66 ± 0.88
PCB 70	97.91 ± 1.12
PCB 66	97.39 ± 1.23
PCB 95	93.09 ± 1.43
PCB 56(60)	101.06 ± 1.00
PCB 101	95.23 ± 1.26
PCB 99	93.76 ± 1.47
PCB 119	93.26 ± 1.20
PCB 97	95.23 ± 1.31
PCB 87	92.99 ± 1.53
PCB 81	94.35 ± 1.40
PCB 110	92.60 ± 1.46
PCB 77	94.76 ± 1.13
PCB 151	93.75 ± 2.09
PCB 149	93.48 ± 2.92
PCB 123	95.71 ± 1.89
PCB 118	92.02 ± 2.47
PCB 114	94.77 ± 2.23
PCB 153	97.42 ± 0.96
PCB 168+132	79.57 ± 8.60
PCB 105	93.52 ± 0.53
PCB 141	97.24 ± 0.58
PCB 138	100.54 ± 1.33
PCB 158	98.29 ± 1.34
PCB 126	97.08 ± 0.84
PCB 187	99.42 ± 0.77
PCB 183	100.31 ± 0.68
PCB 128	99.87 ± 0.73
PCB 167	94.56 ± 1.37
PCB 174	97.88 ± 0.79
PCB 177	96.52 ± 1.01
PCB 156	91.37 ± 1.15
PCB 199(200)	100.03 ± 2.49
PCB 157	94.21 ± 1.15
PCB 180	94.98 ± 1.91
PCB 169	90.18 ± 3.33
PCB 170	96.44 ± 3.11

PCB 201	106.63 ± 3.34
PCB 189	90.62 ± 3.26
PCB 195	97.46 ± 1.73
PCB 194	101.86 ± 4.83
PCB 206	105.09 ± 5.42
PCB 209	108.46 ± 5.27
4,4'-DDMU	104.31 ± 1.96
Chlordane-gamma	103.14 ± 2.25
2,4'-DDE	106.90 ± 3.78
Chlordane-alpha	101.86 ± 1.29
trans-Nonachlor	107.00 ± 3.30
4,4'-DDE	108.72 ± 3.59
2,4'-DDD	109.19 ± 4.81
4,4'-DDD	105.03 ± 4.64
2,4'-DDT	119.14 ± 2.50
cis-Nonachlor	100.86 ± 2.35
4,4'-DDT	118.55 ± 1.52

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Table S4

Summary persistent organic pollutants (PCB and OCP) detected in additional blood plasma samples collected in 2011–2013 from green sea turtles inhabiting the San Gabriel River (n = 6) or San Diego Bay (n = 4). ND means there was no detection for that congener in any blood plasma sample analyzed. Values are ng/g blood plasma. ¹Values are percent lipid in blood plasma. ²Values are ng/g blood plasma lipid.

<i>Congener</i>	<i>San Diego Bay</i>	<i>San Gabriel River</i>
	Mean ± SE	Mean ± SE
PCB 3	ND	ND
PCB 8	ND	ND
PCB 18	ND	ND
PCB 28	ND	ND
PCB 31	ND	ND
PCB 33	ND	ND
PCB 37	ND	ND
PCB 44	ND	ND
PCB 49	ND	ND
PCB 52	ND	ND
PCB 56(60)	ND	ND
PCB 66	ND	ND
PCB 70	ND	ND
PCB 74	ND	ND
PCB 77	ND	0.04 ± 0.04

PCB 81	ND	ND
PCB 87	ND	ND
PCB 95	ND	ND
PCB 97	ND	ND
PCB 99	0.72 ± 0.36	0.66 ± 0.32
PCB 101	ND	ND
PCB 105	ND	ND
PCB 110	ND	ND
PCB 114	0.47 ± 0.2	0.29 ± 0.11
PCB 118	ND	ND
PCB 119	ND	ND
PCB 123	ND	ND
PCB 126	ND	ND
PCB 128	ND	0.18 ± 0.16
PCB 138	2.27 ± 0.47	3.71 ± 2.41
PCB 141	ND	ND
PCB 149	0.18 ± 0.06	0.09 ± 0.03
PCB 151	0.03 ± 0.03	0.01 ± 0.01
PCB 153	ND	ND
PCB 156	ND	0.04 ± 0.04
PCB 157	ND	0.03 ± 0.03
PCB 158	0.61 ± 0.19	0.33 ± 0.07
PCB 167	0.02 ± 0.02	0.08 ± 0.04
PCB 168+132	1.85 ± 0.41	1.9 ± 1.18
PCB 169	ND	0 ± 0
PCB 170	ND	0.07 ± 0.07
PCB 174	ND	ND
PCB 177	ND	ND
PCB 180	0.21 ± 0.13	0.49 ± 0.4
PCB 183	0.13 ± 0.07	0.1 ± 0.09
PCB 187	1.6 ± 0.61	0.78 ± 0.17
PCB 189	ND	ND
PCB 194	ND	ND
PCB 195	ND	ND
PCB 199(200)	ND	ND
PCB 201	ND	ND
PCB 206	ND	ND
PCB 209	ND	ND
Σ PCB	8.09 ± 1.26	8.81 ± 4.66
4,4'-DDMU	ND	ND
Chlordane-gamma	0.31 ± 0.07	0.14 ± 0.07

2,4'-DDE	ND	0.33 ± 0.2
Chlordane-alpha	ND	ND
trans-Nonachlor	0.04 ± 0.04	0.21 ± 0.07
4,4'-DDE	ND	0.03 ± 0.03
2,4'-DDD	ND	ND
4,4'-DDD	ND	ND
2,4'-DDT	ND	0.18 ± 0.18
cis-Nonachlor	ND	ND
4,4'-DDT	ND	ND
Σ OCP	0.35 ± 0.06	0.88 ± 0.32
Σ POP	8.44 ± 1.27	9.69 ± 4.77
Percent Lipid ¹	0.13% ± 0.02%	1.48% ± 1.30%
Σ PCB Lipid ²	7122.26 ± 1631.53	3631.23 ± 1528.20
Σ OCP Lipid ²	292.38 ± 50.60	310.56 ± 149.31
Σ POP Lipid ²	7414.64 ± 1660.87	3941.79 ± 1566.12

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