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Organohalogen contaminants in blood of Kemp's ridley (*Lepidochelys kempii*) and green sea turtles (*Chelonia mydas*) from the Gulf of Mexico

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ABSTRACT

The threat that exposure to organohalogen (OH) contaminants poses to endangered populations of Kemp's ridley (*Lepidochelys kempii*) and green sea turtles (*Chelonia mydas*) is not well understood, partly because few OH data are available. Blood samples from live juvenile and sub-adult *L. kempii* ($n = 46$) and *C. mydas* ($n = 9$) from the Gulf of Mexico and from *L. kempii* from the southeastern US coast ($n = 3$) were extracted using microwave-assisted extraction, and analyzed by large volume injection gas chromatography–mass spectrometry for 85 polychlorinated biphenyls (PCBs), 25 organochlorine pesticides (OCPs) and 27 polybrominated diphenyl ethers (PBDEs). Plasma chemistries, hematology and immune responses were also assessed. Concentrations of ΣPCBs (geometric mean, range: 3190 pg g^{-1}, 227–21 590 pg g^{-1} blood), ΣDDTs (geometric mean, range: 541 pg g^{-1}, 161–4310 pg g^{-1} blood) and OCPs in *L. kempii* from the Gulf were comparable to those reported in *L. kempii* from the Atlantic. ΣPBDEs were detected in all samples (geometric mean, range: 146 pg g^{-1}, 19.5–1450 pg g^{-1} blood), with PBDE 47, 99, 100, 153 and 154 being the predominant congeners. ΣPCBs, ΣDDTs and Σchlordanes were one order of magnitude lower in green turtles, and ΣPBDE concentrations were lower by half due to trophic level differences. *L. kempii* from the southeast USA had higher percentages of highly chlorinated PCBs indicating exposure to Aroclor 1268. Blood urea nitrogen was positively correlated to Σchlordanes, and ΣPCBs were inversely correlated to creatine phosphokinase in *L. kempii*. These data help establish baseline contaminant concentrations in live *L. kempii* and *C. mydas*.

1. Introduction

Many toxic organohalogen (OH) compounds, such as polychlorinated biphenyls (PCBs), dichloro-diphenyl-trichloroethane (DDT) and other organochlorine pesticides (OCPs), have been phased out of use, but they persist in the environment. Flame retardant polybrominated diphenyl ethers (PBDEs) are currently being phased out of use. The deca-brominated PBDE formulation is still being used as policy makers balance its beneficial properties against emerging evidence that PBDEs are environmentally persistent, ubiquitously distributed, bioaccumulative and toxic (Darnerud, 2003; Hites, 2004). Relatively recent use of DDT in countries fighting malaria is another source of OHs to marine environments that sea turtles inhabit (Roberts et al., 2002).

All seven species of sea turtles are threatened or endangered, and the Kemp's ridley (*Lepidochelys kempii*) is the most critically endangered (Marquez, 1994). Populations of green sea turtles (*Chelonia mydas*) are threatened by fibropapillomatosis, a debilitating disease characterized by the growth of large benign tumors. Contaminants have been implicated or suspected as potential contributors to sea turtle diseases or altered health (Aguirre et al., 1994; Keller et al., 2006). Understanding the effects of OHs on health of Kemp's ridley and green turtles is important for effective management of these species.

Altered health of loggerhead sea turtles (*Caretta caretta)* has been linked to OH exposure (Keller et al., 2004c, 2006). Prior studies have shown that blood OH levels were correlated with increased immune B-cell and T-cell proliferation (Keller et al., 2006), and stimulation of immune responses could potentially result in autoimmune disorders or hypersensitivity (Krzystyniak et al., 1995). In addition, Keller et al. (2004b) observed correlations between blood or adipose OH concentrations and clinical plasma

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chemistry parameters and blood cell counts indicating possible hepatotoxic effects, immunomodulation, and anemia. These correlative findings offer evidence that sea turtles may be susceptible to the detrimental health effects of OHs at environmentally relevant concentrations.

The majority of sea turtle contaminant data has come from dead or cold-stunned, stranded animals or turtles incidentally captured by fisheries operations (Pugh and Becker, 2001; Innis et al., 2008). Decomposition of stranded carcasses can complicate contaminant analysis. Samples collected from stranded sea turtles may positively skew contaminant concentrations, because dead animals may have higher concentrations than do live animals, as has been observed in marine mammals (e.g. Hobbs et al., 2003). Recent studies have demonstrated the utility of blood as an alternative tissue for OH analysis in sea turtles (Keller et al., 2004b,c). OH concentrations in blood of Kemp's ridley and loggerhead sea turtles were shown to be correlated to concentrations in adipose tissue, indicating that contaminants in blood are in equilibrium with storage tissues and that blood could be used to monitor OH contaminants in free-ranging sea turtles (Keller et al., 2004a).

While legislation and international treaties have begun to address overt threats to sea turtle survival, the more subtle threat posed by exposure to OH contaminants is not well understood. The goal of this study was to determine baseline concentrations and patterns of OH contaminants in blood from live Kemp's ridley and green turtles from the Gulf of Mexico and to determine if these concentrations have an impact on their health.

2. Materials and methods

2.1. Sample collection

Juvenile and sub-adult Kemp's ridley ($n = 46$) and green turtles ($n = 9$) were captured in 2001–2002. Kemp's ridley straight carapace lengths (SCLs), measured from the nuchal notch to the most posterior marginal notch, were 24.3–46.7 cm (mean ± SD: 37.4 ± 6.8 cm) and green turtle SCLs ranged from 29.5 to 57 cm (mean ± SD: 46.1 ± 4.6 cm). Four sites along the US coast in the northern Gulf of Mexico, Calcasieu Pass (CP), Sabine Pass (SP), Lavaca Bay (LB), and the lower Laguna Madre near South Padre Island (LLM), were sampled using large-mesh entanglement nets that were checked for turtles every 30 min (Fig. 1). Whole blood samples were taken from the dorsocervical sinus using double-ended needles directly into 5 mL Vacutainer tubes containing lithium heparin (Beckton–Dickson Inc., Franklin Lakes, NJ). Three additional sub-adult Kemp's ridleys were captured using modified shrimp trawlers in 2001 in the Atlantic off the southeast coast of the United States (SE USA) for comparison with turtles from the Gulf (Fig. 1). Blood samples were stored at −20 °C until analyzed for OHs.

2.2. Morphometrics and health assessment

Morphometric data including SCL ($n = 26$), straight carapace width ($n = 18$), curved carapace length and width ($n = 18$), and body mass ($n = 18$) were recorded for turtles captured in the Gulf in 2002 and in the Atlantic. Body condition index (BCI) ($n = 18$) was calculated similarly to Keller et al. (2004c) using the formula:

$$\mathrm{BCI} = \frac{\text{Body mass (kg)}}{\mathrm{SCL}^3\ (\mathrm{m}^3)} \quad (1)$$

Plasma testosterone concentrations ($n = 22$) were measured using a radioimmunoassay for sex determination (Wibbels et al., 1990). Mitogen-induced lymphocyte proliferation assays were performed as described by Keller et al. (2005) using 5 days of culture with final well volumes of 0.1 mL. T-cell mitogens used were concanavalin A (ConA) type IV from Jack Bean (*Canavalia ensiformis*) (Sigma, St. Louis, MO) at a final culture mitogen concentration of 20 μg mL^{-1} and, phytohemagglutinin P (PHA; Amersham Pharmacia Biotech Inc., Pascataway, NJ) at 5 μg mL^{-1} and 10 μg mL^{-1} for PHA. B-cell mitogens used were lipopolysaccharide (LPS) from *Escherichia coli* serotype 0127:B8 (Sigma) at 10 μg mL^{-1} and phorbol 12,13-dibutyrate (PDB; Sigma) at 0.2 μg mL^{-1}. Clinical plasma chemistry and hematologic parameters ($n = 22$) were measured by a diagnostic laboratory (ANTECH, Memphis, TN).

2.3. OH measurements

2.3.1. Standard solutions

A seven point external calibration curve was prepared from National Institute of Standards and Technology (NIST) Standard Reference Materials (SRM) 2261 (chlorinated pesticides in hexane), 2262 (chlorinated biphenyls in *iso*-octane), 2274 (chlorinated biphenyl congeners in *iso*-octane II), 2275 (chlorinated pesticides in hexane II) and solutions containing 46 additional PCB congeners, 28 PBDE congeners and octachlorostyrene, with target concentration ranges from 60 to 0.05 ng g^{-1} for PCBs and pesticides and 170 to 0.1 ng g^{-1} for PBDEs. A primary internal standard (IS) solution containing 13 isotopically-labeled compounds (^{13}C-PCB 28, 52, 118, 153, 180, 194 and 206, endosulfan I-d_4, 4,4′-DDE-d_8, 4,4′-DDD-d_8 and 4,4′-DDT-d_8, ^{13}C-*trans*-chlordane and ^{13}C-PBDE 99) purchased from Cambridge Isotope Laboratories (Andover, MA) and Wellington Laboratories (Guelph, Ontario, Canada) was prepared by combining individual compound solutions and diluting with acetone. A secondary IS solution containing ^{13}C-PCB 47, ^{13}C-PCB 155, ^{13}C-dieldrin and ^{13}C-hexachlorobenzene (Cambridge Isotope Laboratories) was prepared in *iso*-octane. Calibration solutions were spiked with the primary and secondary IS solutions (~5 ng of each compound), and concentrated under a stream of purified nitrogen to a final volume of approximately 0.1 mL.

2.3.2. Sample extraction

Whole blood samples (1.8–5.2 g) were thawed at 4 °C, homogenized by vortex and gravimetrically transferred to Mars Xpress Teflon extraction vessels (CEM Corp., Matthews, NC). Reconstituted NIST SRM 1589a (PCBs, pesticides, PBDEs, and dioxins/furans in human serum), an aliquot of an in-house loggerhead sea turtle plasma control material, and an analytical blank of hexane-rinsed Millipore (Billerica, MA) filtered water were analyzed with each batch of 10–20 samples. The primary IS solution (250 μL) was added to each extraction vessel. Vessels were sonicated for 15 min and refrigerated overnight to equilibrate. Extraction, lipid determination, and clean-up methods are described elsewhere (Keller et al., 2009). Briefly, samples were extracted using cavity dispersed microwave-assisted extraction with formic acid and 20% dichloromethane in hexane (v:v). Lipid or total extractable organic (TEO) content was determined gravimetrically from a subsample of extract. Extracts were then cleaned up using an alumina column followed by a size exclusion gel permeation column.

2.3.3. Determination of PCBs, PBDEs and OCPs

Samples were analyzed with three different injections on an Agilent Technologies 6890/5973 GC/MS (Palo Alto, CA). A programmable temperature vaporization inlet (Gerstel Inc., Baltimore, MD) was operated in the solvent vent mode. This allowed for the large, 20 μL (4 × 5 μL) injection needed to measure low levels in blood. GC parameters, DB-5 ms column lengths (Agilent Technologies), and MS parameters are listed in Supplemental Table 1. The transfer line was maintained at 300 °C. To further increase sensitivity, the

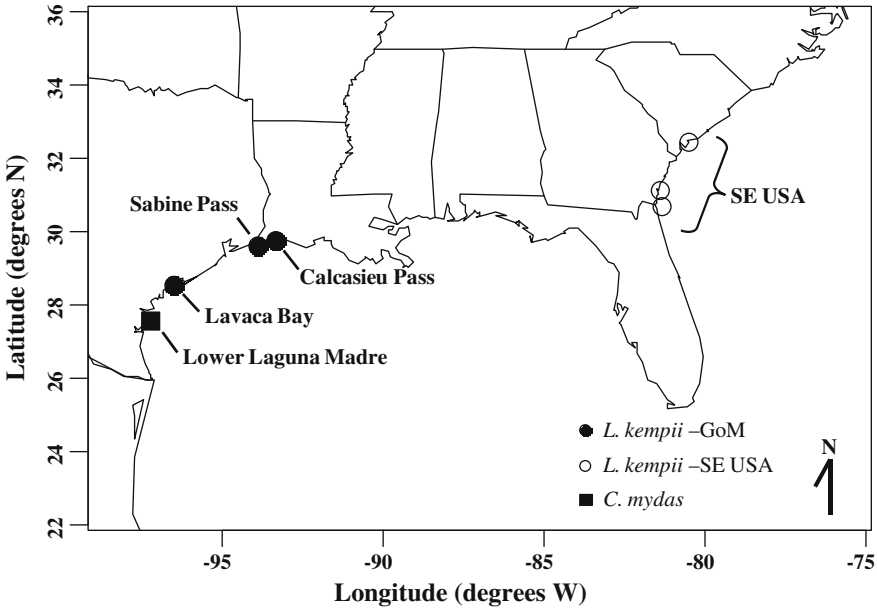

Fig. 1. Map of sea turtle capture locations. Kemp's ridley turtles were sampled at CP (n = 28), SP (n = 8), LB (n = 10) and SE USA (n = 3). Green sea turtles were caught at LB (n = 2) and LLM (n = 7).

MS source was operated in selected ion monitoring mode with the chromatogram separated into as many windows as feasible.

Analyte concentrations were calculated using the linear regression equation of three or four out of seven external calibration curve points which bracketed the analyte: primary IS peak area ratio in the sample. Limit of detection (LOD) was defined as the mean analyte concentration plus three times the standard deviation calculated in the five blank samples. Reported concentrations were not corrected for analyte recovery or for blank concentrations.

2.4. Statistical analysis

Statistical analyses were performed at the 95% confidence level on wet mass normalized concentrations using the software package R v2.1.0 (R Institute, Vienna, Austria). Statistically significant differences discussed below indicate a p-value less than 0.05. Data were reduced by summing compounds above the LOD in each group (ΣPCB, Σdioxin-like PCBs, ΣPBDEs, ΣDDTs, Σchlordanes). Individual compounds included in each group are listed in the Supplemental material. Differences in ΣPCBs, ΣDDTs, ΣPBDEs, Σchlordanes, dieldrin and mirex between species were examined by Student's t-test or the Wilcoxon test if the data could not be transformed to produce normally distributed or homoschedastic groups. Differences in mean contaminant concentrations between capture locations and across collection years were examined by MANOVA with interaction terms (location × year). Tukey's multiple comparisons of means was used as a post hoc test to determine which locations were significantly different.

Contaminant pattern differences were examined by principal component analysis (PCA). Compounds detected in over 80% of the samples and constituting more than 1% of the ΣOH concentration were included as variables. Variables were scaled to a variance of 1 and singular value decomposition was used to calculate the principal component (PC) values. Concentrations below the LOD were estimated as half the LOD for the sample. Concentrations were converted to percentage of ΣOH. To eliminate the loading bias created by compounds present at higher percentages, percent of ΣOH concentrations was scaled using the equation:

$$C_s = \frac{c_i - c_\mu}{\sigma} \quad (2)$$

where c_s is the scaled concentration, c_i is the percent concentration in the ith sample, c_μ is the mean percent concentration of the compound in all the samples and σ is the standard deviation of the percent concentrations in all the samples.

Relationships between OH concentrations and morphometric and health parameters were assessed by Pearson or Spearman correlations.

2.5. Quality control

Concentrations of all compounds in SRM 1589a were within 35% of the certified value. Concentrations of all compounds measured in the loggerhead turtle plasma control material were within 30% of the consensus values.

Mean coefficient of variation (CV) was 11% for all compounds quantified in four replicates of the loggerhead plasma control material, and it was 11% for all compounds quantified in five replicates of SRM 1589a, indicating acceptable reproducibility between sample batches.

Recoveries of primary IS compounds were similar to those obtained with other extraction methods, except for some lower recoveries of ^{13}C-PBDE 99 in a small number of samples (see Keller et al., 2009). One green and nine Kemp's ridley samples had recoveries of <30% for ^{13}C-PBDE 99. Concentrations of PBDEs in these samples were excluded from statistical analyses.

3. Results

3.1. Species and geographic comparison

3.1.1. Contaminant concentrations

OH compounds detected and TEO content are displayed in Table 1 by species and capture location. Compounds below the detection limit were hexachlorocyclohexanes, endosulfans, aldrin, endrin, pentachlorobenzene, hexachlorobenzene, octachlorostyrene, tri-brominated PBDEs, and hepta- to deca-brominated PBDEs. Correlations of TEO content with the summed contaminant classes showed that lipid content explained only 15%, 0.01%, 11%, 20%, 6.8% and 1.1% of the variation in ΣPCB, ΣPBDE, ΣDDT, Σchlordane, dieldrin and mirex concentrations, respectively. Because these

Table 1
Organohalogen concentrations (pg g^{-1} wet mass) and percent TEO content in Kemp's ridley *(L. kempii)* and green turtle (C. *mydas)* blood.

Location	Gulf of Mexico			Combined Gulf of Mexico sites			CP[a]			LB[a]			SP[a]			SEUSA		
Species	*C. mydas*			*L. kempii*			*L. kempii*			*L. kempii*			*L. kempii*			*L. kempii*		
n	9			46			28			10			8			3		
Analyte	Geomean (SD)	Range	*n*	Geomean (SD)	Range	*n*	Geomean (SD)	Range	*n*	Geomean (SD)	Range	*n*	Geomean (SD)	Range	*n*	Geomean (SD)	Range	*n*
∑PCB	331 (701)[d]	117–2330	9	3190 (3620)[d]	227–21 590	46	4010 (4130)[f]	895–21 590	28	1720 (1930)[f]	227–6910	10	3130 (2390)	924–7870	8	6610 (12 200)	2400–24 700	3
3C1	10.5 (7.9)	<3.5–27.7	8	15.8 (14.7)	<3.5–68.8	18	18.8 (19.1)	<3.5–68.8	9	12.9 (2.5)	<3.5–15.6	3	13.5 (8.7)	<3.5–28	6	12.9 (NA)	<3.5–12.9	1
4C1	23.6 (57.4)	<1.7–111	3	41.0 (50.0)	<1.7–205	43	58.5 (52.6)	<1.7–205	27	15.6 (13.8)	<1.7–45.4	8	32.6 (38.9)	6.76–97.9	8	55.8 (NA)	<1.7–131	2
5C1	61.0 (131)	13–425	9	461 (502)	40.8–2890	46	591 (566)	91.6–2890	28	244 (267)	40.8–908	10	429 (331)	64.1–1050	8	747 (1800)	236–3470	3
6C1	149 (355)	56.5–1160	9	1790 (2080)	101–12 500	46	2250 (2380)	510–12 500	28	946 (1110)	101–3980	10	1810 (1370)	570–4350	8	2930 (6910)	845–13 400	3
7C1	58.2 (135)	21.7–440	9	680 (762)	46.8–4560	46	838 (872)	198–4560	28	388 (449)	46.8–1560	10	657 (507)	231–1750	8	1490 (2470)	573–5100	3
8C1	28.7 (51.7)	<0.4–167	8	150 (184)	21.8–1060	46	187 (209)	50.7–1060	28	91.3 (96.5)	21.8–352	10	127 (142)	34.3–472	8	897 (670)	539–1790	3
9C1	30.1 (NA)	<0.5–30.1	1	28.0 (48.3)	<0.5–237	38	34.0 (55.4)	<0.5–237	25	14.6 (13.4)	<0.5–45.3	7	26.9 (29.2)	<0.5–88.3	6	295 (258)	187–647	3
10Cl	7.3 (NA)	<0.6–18.2	2	21.0 (34.3)	<0.6–168	34	25.6 (38.8)	<0.6–168	23	13.1 (10.5)	<0.6–31.8	5	14.5 (23.7)	<0.6–68.7	6	45.5 (65.4)	19.4–140	3
Dioxin-like	13.2 (37.1)	4.03–120	9	155 (196)	11.7–1170	46	191 (220)	50.1–1170	28	83.7 (123)	11.7–431	10	162 (164)	51.7–481	8	322 (989)	78.8–1860	3
∑PBDE	80.6 (217)	24.4–623	9	146 (273)	19.5–1450	46	148 (109)	19.5–535	28	160 (453)	19.6–1450	10	125 (371)	39.1–1140	8	105 (141)	38–307	3
PBDE 47	58.7 (139)	<1.4–370	8	54.7 (100)	<1.4–521	37	54.5 (41.3)	<1.4–162	21	65.2 (180)	<1.4–521	8	46.6 (102)	13.4–324	8	38.7 (27.0)	16.3–69.4	3
PBDE 49	15.7 (NA)	<2.7–15.7	1	5.0 (4.6)	<2.7–21	19	4.4 (2.8)	<2.7–10	15	9.1 (8.9)	<2.7–21.2	3	4.6 (NA)	<2.7–4.6	1	2.6 (NA)	<2.7–3.1	2
PBDE 99	28.7 (70)	<2.0–208	7	35.9 (153)	<2.0–657	30	27.2 (22.8)	<2.0–75.6	17	82.8 (250)	<2.0–657	6	34.2 (208)	<2.0–574	7	17.7 (21.9)	6.8–48.7	3
PBDE 100	32.1 (NA)	<2.3–70	2	34.1 (36.9)	<2.3–167	35	37.9 (36.3)	<2.3–167	19	29.3 (42)	<2.3–135	8	31 (37.8)	7.4–128	8	26.2 (19.4)	12.6–51.1	3
PBDE 153	19.3 (NA)	<0.5–19.3	1	21.1 (17.4)	<0.5–59.2	13	15.2 (19.6)	<0.5–59.2	6	27.5 (13.8)	<0.5–48.8	5	29 (NA)	<0.5–49.8	2	39.9 (NA)	<0.5–39.9	1
PBDE 154	33.1 (NA)	<0.6–33.1	1	21.0 (17.2)	<0.6–67.8	27	21.1 (13.9)	<0.6–64.9	16	21.3 (21.5)	<0.6–67.8	6	20.1 (24.5)	<0.6–66.6	5	67.8 (NA)	<0.6–67.8	1
PBDE 155	<0.5	<0.5	0	9.4 (NA)	<0.5–9.9	2	9.4 (NA)	<0.5–9.9	2	<0.5	<0.5	0	<0.5	<0.5	0	30.2 (NA)	<0.5–30.2	1
∑DDT	80.0 (114)[d]	6.31–348	9	541 (656)[d]	161–4310	46	602 (788)	175–4310	28	490 (330)	169–1170	10	422 (385)	161–1370	8	794 (1790)	188–3530	3
4,4′-DDE	66.4 (110)	<10.1–348	8	472 (633)	43.9–4110	46	540 (754)	140–4110	28	378 (368)	43.9–1170	10	386 (367)	104–1290	8	773 (1800)	188–3530	3
2,4′-DDT + 4,4′-DDD	<5.7	<5.7	0	38.5 (25.6)	<5.7–90.1	12	38.1 (29.3)	<5.7–90.1	8	50.3 (11.1)	<5.7–58.1	3	18.7 (NA)	<5.7–18.7	1	<5.7	<5.7	0
4,4′-DDT	56.4 (13.5)	<8.7–68.9	4	33.4 (22.1)	<8.7–95.6	28	36.7 (24.3)	<8.7–95.6	16	29.8 (22.2)	<8.7–67.1	8	29.0 (7.2)	<8.7–39.6	4	26.7 (NA)	<8.7–26.7	1
4,4′-DDT:4,4′-DDE[b]	0.266 (0.634)[e]	0.019–1.80		0.032 (0.238)[e]	0.003–1.53		0.028 (0.107)	0.006–0.485		0.054 (0.469)	0.024–1.53		0.028 (0.085)	0.006–0.256		0.013 (0.021)	0.002–0.039	
∑chlordane	16.4 (25.7)[d]	<2.2–64.3	6	163 (249)[d]	10.9–1190	46	199 (250)	17.9–1190	28	128 (220)[h]	10.9–700	10	111 (300)[h]	30.0–809	8	1140 (1850)[h]	353–3870	3
Cis-chlordane	<4.3	<4.3	0	13.6 (12.5)	<4.3–43.7	24	12.9 (10.6)	<4.3–41.7	17	15.3 (20.0)	<4.3–43.7	3	15.7 (16.6)	<4.3–37.4	4	82.3 (NA)	<4.3–973	2
Trans-chlordane	3.05 (2.19)	<1.4–6.71	4	16.4 (47.9)	<1.4–213	43	17.7 (44.5)	3.06–213	28	14.0 (49.5)	<1.4–150	8	14.5 (64.7)	4.07–166	7	26.3 (48.5)	9.98–97.9	3
Cis-nonachlor	6.9 (NA)	<2.0–12.5	2	16.1 (17.1)	<2.0–101	30	17.7 (20.1)	<2.0–101	19	14.7 (12.1)	<2.0–39	6	12.5 (7.05)	<2.0–22	5	29.2 (37.5)	12.3–82.6	3
Trans-nonachlor	9.33 (17.4)	<1.6–45.1	5	41.8 (51.3)	<1.6–264	30	46.3 (60.6)	<1.6–264	19	40.2 (32.6)	<1.6–97	6	29.5 (18.3)	<1.6–60.1	5	115 (224)	30.1–453	3
Oxychlordane	<1.8	<1.8	0	33.5 (40.7)	<1.8–188	26	31.9 (28.6)	<1.8–112	18	62.6 (66.9)	<1.8–188	5	16.0 (9.91)	<1.8–28.8	3	387 (625)	103–1310	3
Heptachlor epoxide	51.7 (NA)	<7.7–51.**7**	1	103 (204)	<7.7–933	37	107 (205)	<7.7–933	25	82.0 (159)	<7.7–506	8	122 (294)	<7.7–605	4	453 (378)	198–949	3
Dieldrin	96.0 (NA)	<74.8–96	1	199 (119)	<74.8–609	38	226 (123/	<74.8–609	25	172 (117)[h]	<74.8–395	8	132 (59.3)[g,h]	<74.8–203	5	500 (409)[h]	206–1020	3
Mirex	11.1 (11.4)	<0.6–37.7	7	10.0 (4.2)	<0.6–19	18	10.6 (4.53)	O.6–19	7	9.6 (4.8)	<0.6–18.5	7	9.94 (3.54)	<0.6–15.4	4	9.17 (NA)	<0.6–9.52	2
SOH	655 (846)	287–2820	9	4500 (4510)	567–27 700	46	5360 (5190)	1640–27 700	28	2960 (2520)	567–9120	10	4110 (3080)	1280–10 600	8	9330 (16 400)	3240–33 500	3
% TEO (w:w)[c]	0.296 (0.096)	0.17–0.436	9	0.227 (0.153)	0.061–0.996	46	0.210 (0.177)	0.061–0.996	28	0.245 (0.114)	0.123–0.504	10	0.276 (0.107)	0.168–0.437	8	0.518 (0.182)	0.377–0.736	3

Notes: geomean = geometric mean; SD = standard deviation; *n* = number of samples with concentrations above the limit of detection; NA = not applicable; 3 Cl through 10 Cl = sum of PCB homolog groups.

[a] Capture locations within the Gulf of Mexico.

[b] 4,4′-DDT:4,4′-DDE ratio was calculated for all individuals in each group and concentrations below the LOD were defined as one-half of the LOD. The value reported is the arithmetic mean (SD).

[c] TEO is used as an estimate of lipid content.

[d] *L. kempii* statistically greater than *C. mydas* ($p < 0.05$).

[e] *C. mydas* statistically greater than *L. kempii* ($p < 0.05$).

[f] CP statistically greater than LB ($p < 0.05$).

[g] CP statistically greater than SP ($p < 0.05$).

[h] SE USA statistically greater than LB and SP ($p < 0.05$).

correlations were not significant ($p > 0.05$), wet mass normalized analyte concentrations were used for all statistical analyses.

Concentrations of ΣPCBs, ΣDDTs and Σchlordanes were significantly higher (9.6, 6.8, and 9.9 times higher, respectively) in Kemp's ridley than in green turtles from the Gulf (Table 1). The ΣPBDE and mirex concentrations in these two species were not significantly different. Concentrations of 4,4′-DDT were detected in 44% of the green turtles and 59% of the Kemp's ridley turtles. The mean ratio of 4,4'-DDT to 4,4′-DDE concentrations was higher in green turtles (0.979) than in Kemp's ridleys (0.070) primarily due to the nearly 10-fold greater concentration of 4,4′-DDE in Kemp's ridleys.

Contaminant concentrations were also compared among the Kemp's ridley capture locations (Table 1). ΣPCBs were significantly higher in turtles from CP than LB, and dieldrin was significantly higher in turtles from CP than SP. SE USA turtles had significantly higher Σchlordanes and dieldrin concentrations than turtles from LB or SP. The mean ratio of 4,4′-DDT to 4,4′-DDE in the blood of Kemp's ridleys was not significantly different among capture locations (see Table 1). Green turtles were captured at LLM and LB, but small sample sizes prohibited statistical geographical comparisons.

3.1.2. Contaminant patterns

ΣPCBs constituted the largest proportion of ΣOHs in both species followed by ΣDDT, ΣPBDE and Σchlordane in Kemp's ridleys and ΣPBDE, ΣDDT and Σchlordane in green turtles. Differences in the overall contaminant pattern were investigated by PCA (Fig. 2). The first three PCs accounted for 88% of the variation in the contaminant data. With the exception of one green turtle captured at LB, green turtles were separated from Kemp's ridleys on PC1. Examination of the loadings showed that PC1 primarily explained the difference in the ΣPBDE to ΣPCB ratio. The three turtles from the SE USA were separated from Gulf turtles on PC2. High loading values for PC2 were observed for PCBs 183, 196 + 203 and 199.

PCB congener patterns were compared between species within the Gulf and among Kemp's ridley capture locations (see Table 1). Green turtles had significantly higher percentages of tri- and octa-PCBs than did Kemp's ridleys, while the latter had significantly higher percentages of hexa- and nona-PCBs. SE USA Kemp's ridleys had significantly higher percentages of octa- and nona-PCBs and significantly lower percentages of hexa-PCBs than did Gulf conspecifics. Individual PCB congener profiles of the two species were similar with congeners 99, 118, 138, 146, 149, 151, 153, 163, 170, 180 + 193, 183, 187, 196 + 203, 199 being the predominant congeners (Fig. 3a). These congeners comprised, on average, 82% of the ΣPCB concentration in each sample. However, there were a few notable species differences within the Gulf. Congeners 138, 158 and 187 were more prevalent in Kemp's ridleys, while congeners 28 + 31 and 149 constituted a greater percentage of ΣPCBs in green turtles (Fig. 3a). The pattern of predominant PCB congeners in blood of Kemp's ridleys did not differ among capture locations within the Gulf, but the turtles captured in SE USA had higher percentages of PCBs 194, 196 + 203, 199, 202 and 206 (Fig. 3b). Turtles from the SE USA also had a lower percentage of PCB congeners 138, 146, 149 and 158 (Fig. 3b).

PBDE congener patterns were also compared between species and among Kemp's ridley capture locations. Only tetra-, penta- and

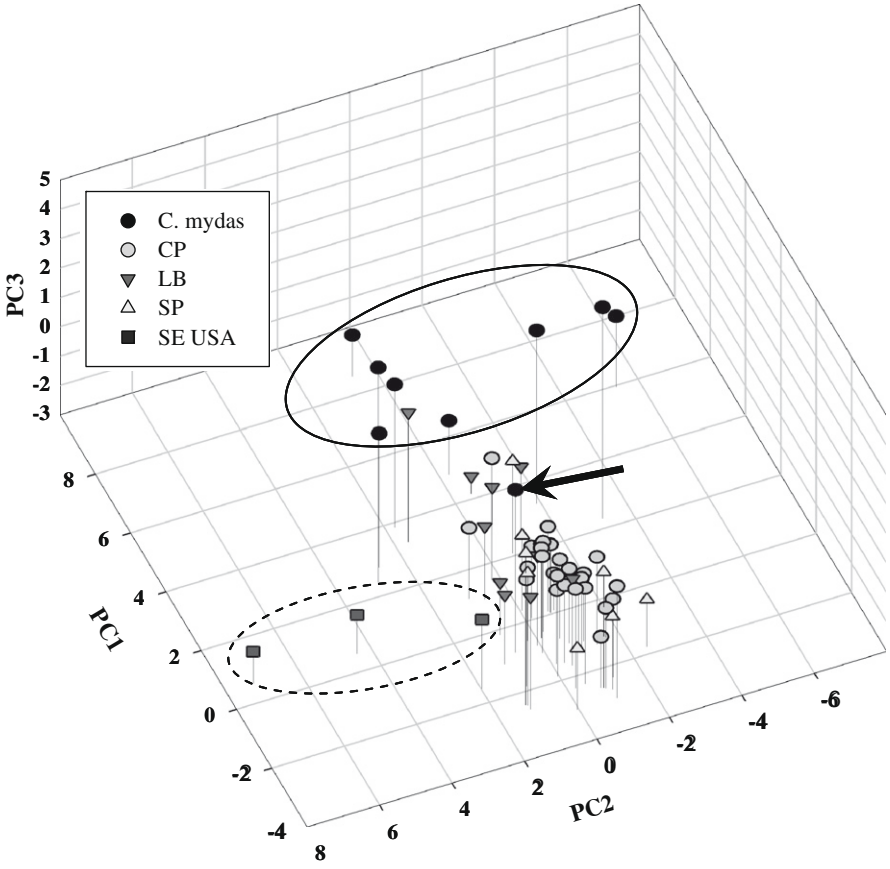

Fig. 2. Three-dimensional scatterplot of the first three principal component (PC) scores of organohalogen contaminant patterns in Kemp's ridley and green sea turtles. The solid ellipse indicates green turtles that were separated from Kemp's ridley turtles, and the dashed ellipse groups the three Kemp's ridleys captured off the SE USA. The arrow highlights one green turtle captured at LB that did not group with the other conspecifics. Kemp's ridleys were captured at CP, LB, SP, and SE USA.

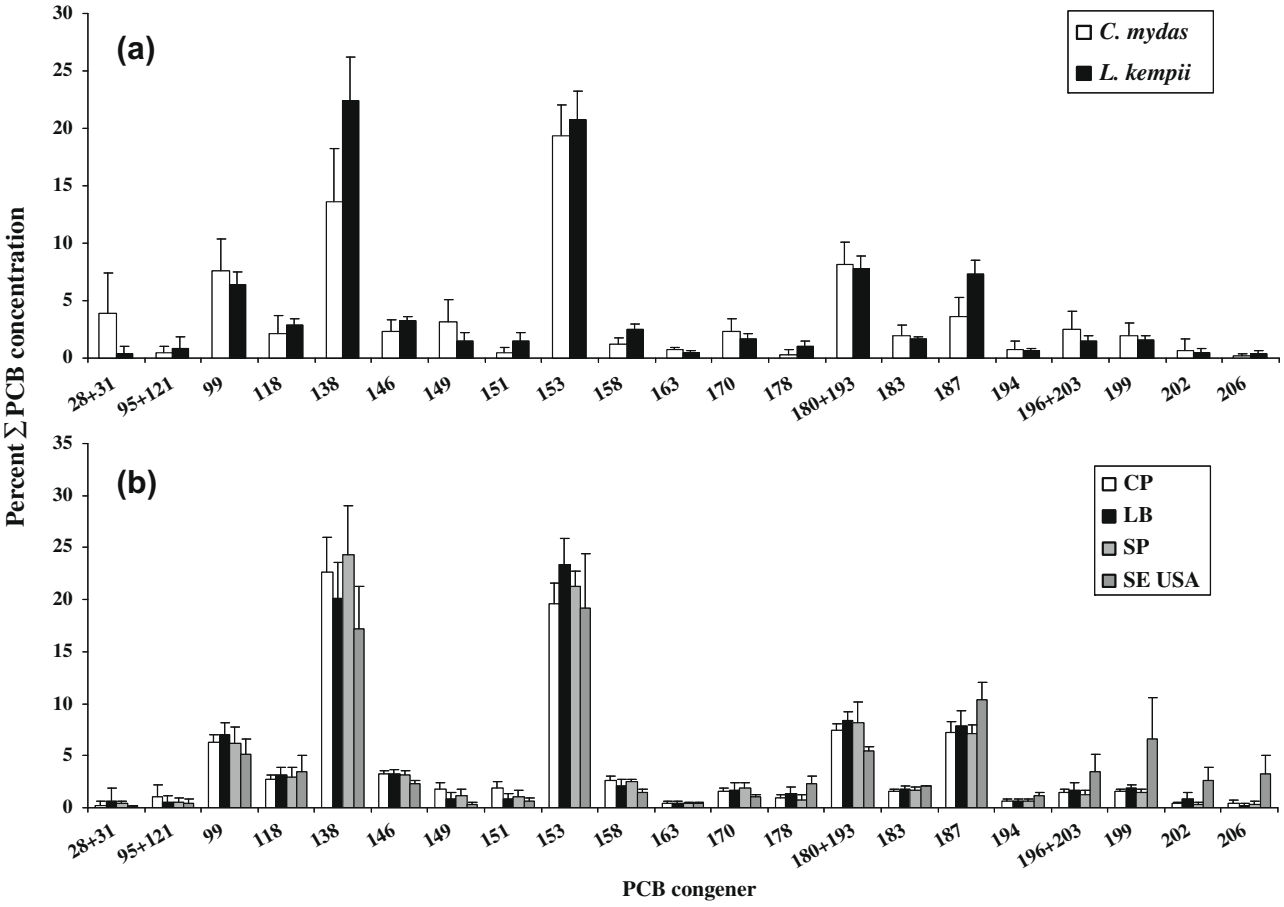

Fig. 3. Comparison of PCB congener profiles between sea turtle species (a) and among Kemp's ridley capture locations (b). Error bars represent one standard deviation.

hexa-PBDEs were detected in the blood samples, with PBDEs 47, 49, 99, 100, 153 and 154 being the predominant congeners (Fig. 4a). Kemp's ridley PBDE congener profiles were dominated by PBDEs 47 and 100, while PBDEs 47 and 99 dominated the green turtle profile (Fig. 4a). The green turtle PBDE profile, however, could be skewed because concentrations of PBDE 49, PBDE 153 and PBDE 154 were <LOD in all but one green turtle sample, but were quantified in 41%, 28% and 59%, respectively, of the Kemp's ridley samples. PBDE congener patterns in Kemp's ridley blood did not significantly differ between capture locations (Fig. 4b).

3.2. Correlations with morphological, health and immune function parameters

Mean values of all morphological and health parameters measured and correlation coefficients with contaminants are given in Table 2. Only five significant correlations between summed contaminant concentrations and morphological, health or immune function parameters were observed. Correlations between Σdioxin-like PCBs and morphological, health and immune function parameters were not investigated because Σdioxin-like PCBs was highly correlated to ΣPCBs ($r^2 = 0.96$). The significant correlation between ΣDDT and potassium concentrations was highly influenced by a single high potassium concentration. Significant negative correlations were observed between ΣPCBs and creatine phosphokinase (CPK) concentrations, between ΣDDTs and the proliferative response of T-cells to PHA (5 mg mL^{-1}), and between dieldrin and testosterone concentrations in female turtles. Blood urea nitrogen concentrations were positively correlated to Σchlordane concentrations. Morphological parameters were not significantly correlated with any contaminant concentrations.

4. Discussion

4.1. Species and geographical comparison

4.1.1. Concentrations

Higher concentrations of ΣPCBs, ΣDDTs and Σchlordanes in the blood of Kemp's ridleys compared to those of green turtles found in this study are likely due to differences in dietary preferences and biomagnifications potential. Green turtles are primarily herbivorous, feeding mostly on seagrasses and algae (Lopez-Mendilaharsu et al., 2005). As generalist predators, Kemp's ridleys primarily consume benthic invertebrates, including crabs and mollusks (Witzell and Schmid, 2005). Concentrations of OHs in Kemp's ridley turtle blood were similar to concentrations reported in the blood of loggerheads (Keller et al., 2004c; Carlson, 2006) (Table 3), and both species feed at similar trophic levels (Tomas et al., 2002). The higher ratio of 4,4′-DDT to 4,4′-DDE in green turtles relative to that in Kemp's ridleys may also be due to differences in dietary exposure; however, differences in the ability to metabolize the parent compound 4,4′-DDT to 4,4′-DDE may also explain this observation.

The lack of a species difference in ΣPBDE concentrations suggests that turtles may be exposed to PBDEs through a different route, such as consumption of non-food items containing PBDEs. Both Kemp's ridley (Shaver, 1991) and green turtles (Bugoni et al., 2001) have been shown to consume plastic and foam marine debris that could potentially contain PBDEs. Additionally, ΣPBDEs

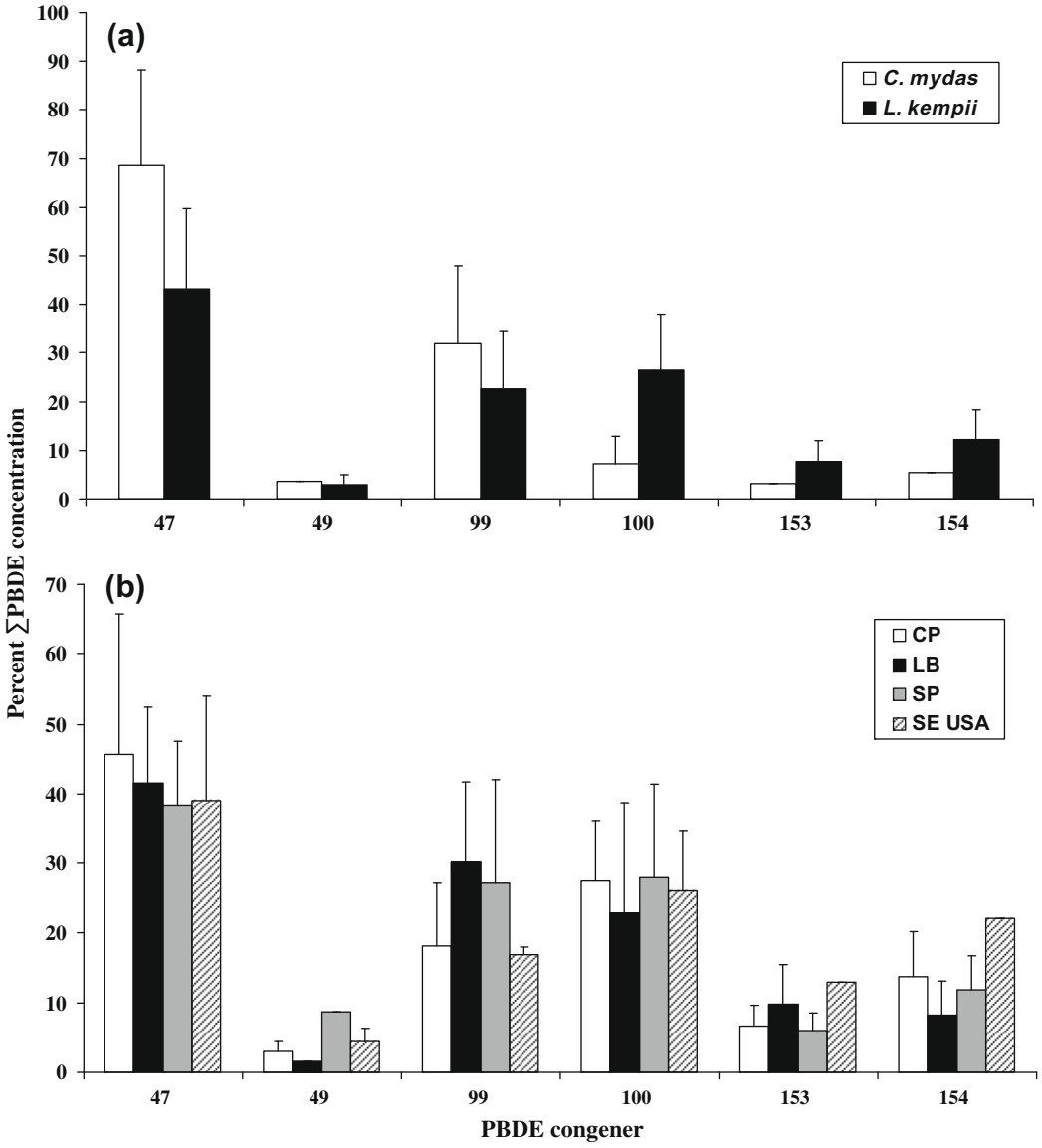

Fig. 4. Comparison of PBDE congener profiles between sea turtle species (a) and among Kemp's ridley capture locations (b). Error bars represent one standard deviation. Lack of an error bar indicates that the congener was above the limit of detection in only one sample in the group.

in green turtles exceeded ΣDDTs. With the exception of low level use as a malarial control agent, DDT use has ceased, while PBDEs have been shown to be increasing in selected biota with a doubling time in the range of 2–7 years (Hites, 2004; Elliott et al., 2005). Analysis of archived turtle samples from different years could be used to examine temporal trends of PBDEs in sea turtles in order to determine how the rate of accumulation will change as PBDE use decreases.

The lack of contaminant concentration differences across capture locations for Kemp's ridleys within the Gulf suggests that exposure is similar across this water body. Turtles captured closer to Mexico were expected to have a higher ratio of the parent compound, 4,4′-DDT, to the persistent metabolite, 4,4′-DDE, because of the relatively recent use of DDT to battle malaria in Central America (Roberts et al., 2002). While the ratio of 4,4′-DDT to 4,4′-DDE was higher in the blood of Kemp's ridleys captured closest to Mexico (Table 1), these differences were not statistically significant. A larger sample size or sampling further south may reveal significant differences.

4.1.2. Contaminant patterns

Differences in contaminant patterns were evident between species and among capture locations for Kemp's ridleys. Green turtles were separated from Kemp's ridleys on PC1 of the PCA (Fig. 2) driven by higher concentrations of PBDEs relative to PCBs in green turtles (Table 1). As stated above, this difference could be due to a different dietary exposure route for PBDEs or differences in the current use of PCBs and PBDEs. Kemp's ridleys from the SE USA were separated from turtles from the Gulf on PC2, because they had higher proportions of PCBs 183, 196 + 203 and 199.

The overall PCB congener profile in green and Kemp's ridley turtle blood was similar to patterns reported in other biota (Kucklick et al., 2002; Vander Pol et al., 2004). The PCB homolog pattern was slightly different between green and Kemp's ridley turtles. Reasons for this species difference could be differences in dietary preferences, migratory pathways, or ability to biotransform and excrete different congeners. While reptilian biotransformation of PCBs has been demonstrated (Schlezinger et al., 2000), insufficient data are available to even moderately describe the metabolic capacity of

Table 2
Morphometric and clinical plasma chemistry parameter values, and correlations with blood contaminant concentrations in Kemp's ridley sea turtles.

Parameter			Mean (SD)	ΣPCB	ΣDDT	ΣPBDE	Σchlordane	dieldrin	mirex	n^c
Morphometrics		SCL (cm)	34.6 (5.3)	0.070 (0.172)	0.078 (0.151)	0.036 (0.333)	0.003 (0.799)	0.028 (0.397)	0.036 (0.333)	28 (19)
		Body mass (kg)	5.0 (2.8)	0.004 (0.792)	0.003 (0.822)	−0.084 (0.258)	0.004 (0.794)	0.001 (0.894)	−0.084 (0.258)	21 (14)
		BCI (kg m^{-3})	11.4 (3.0)	0.104 (0.155)	0.125 (0.116)	−0.003 (0.837)	0.392 (0.079)	0.001 (0.893)	−0.003 (0.837)	21 (14)
Immune function	Cell counts	WBC (10^3 L^{-1})	10.7 (2.6)	0.100 (0.151)	0.069 (0.237)	−0.195 (0.051)	0.313 (0.155)	−0.020 (0.526)	0.195 (0.051)	22 (13)
		Neutrophils (10 μL^{-1})	2.7 (1.5)	0.095 (0.164)	0.007 (0.708)	−0.015 (0.608)	0.302 (0.171)	−0.013 (0.617)	0.015 (0.608)	22 (13)
		Lymphocytes (10^3 μL^{-1})	7.9 (2.1)	0.030 (0.440)	0.082 (0.197)	0.014 (0.619)	0.011 (0.643)	−0.012 (0.627)	−0.014 (0.619)	22 (13)
		Neut:Lymph ratio	0.128 (0.191)	0.014 (0.596)	<0.001 (1.00)	−0.123 (0.142)	0.004 (0.782)	<0.001 (0.989)	−0.123 (0.142)	22 (13)
	Lymphocyte Proliferation	Con A (20 μg L^{-1})		0.011 (0.680)	−0.372 (0.129)[b]	0.133 (0.244)[a]	<0.001 (0.973)	0.005 (0.773)	0.133 (0.244)[a]	18 (10)
		LPS (10 μg mL^{-1})		−0.016 (0.622)	−0.058 (0.337)[a]	0.001 (0.941)[a]	−0.007 (0.737)	0.033 (0.473)	0.001 (0.941)[a]	18 (10)
		PDB (0.2 μg mL^{-1})		−0.040 (0.427)	−0.025 (0.532)[a]	0.079 (0.376)[a]	−0.006 (0.753)	0.075 (0.271)[a]	0.079 (0.376)[a]	18 (10)
		PHA (5 μg mL^{-1})		−0.075 (0.271)	−0.568 (0.016)[b,*]	0.015 (0.705)[a]	−0.062 (0.321)	0.005 (0.775)	0.015 (0.705)[a]	18 (10)
		PHA (10 μg L^{-1})		−0.030 (0.505)	0.221 (0.393)[b]	−0.011 (0.743)[a]	−0.017 (0.414)	−0.022 (0.569)	0.011 (0.743)[a]	17 (10)
Clinical plasma chemistry	Overall health indicators	Glucose (mg dL^{-1})	128 (38)	0.096 (0.151)	−0.069 (0.228)	−0.019 (0.563)	0.303 (0.160)	0.124 (0.100)	−0.019 (0.563)	23 (14)
		Total protein (g dL^{-1})	2.9 (0.5)	−0.023 (0.492)	0.018 (0.536)	0.010 (0.677)	0.255 (0.239)	−0.067 (0.232)	−0.010 (0.677)	23 (14)
		Albumin (g dL^{-1})	1.2 (0.2)	0.123 (0.101)	0.024 (0.479)	0.086 (0.21)	0.296 (0.169)	0.037 (0.377)	0.086 (0.210)	23 (14)
		Globulin (g dL^{-1})	1.8 (0.4)	−0.046 (0.326)	0.035 (0.395)	0.001 (0.908)	0.001 (0.923)	−0.108 (0.126)	−0.001 (0.908)	23 (14)
		Alb: glob ratio	0.69 (0.12)	0.102 (0.137)	0.130 (0.090)	−0.037 (0.419)	0.001 (0.893)	0.136 (0.084)	−0.037 (0.419)	23 (14)
		BUN (mg dL^{-1})	47 (25)	0.166 (0.054)	0.012 (0.613)	0.083 (0.218)	0.564 (0.006) ·	0.002 (0.825)	−0.083 (0.218)	23 (14)
		Uric acid (mg dL^{-1})	1.1 (0.5)	0.002 (0.844)	−0.003 (0.793)	0.001 (0.914)	−0.027 (0.452)	0.029 (0.434)	−0.001 (0.914)	23 (14)
	Tissue damage	AST (U L^{-1})	121 (35)	−0.001 (0.909)	−0.001 (0.862)[a]	0.197 (0.050)[a]	−0.011 (0.635)	−0.001 (0.878)	0.197 (0.050)[a]	23 (14)
		CPK (U L^{-1})	2362 (1792)	−0.179 (0.044)*	<0.001 (0.946)[a]	0.033 (0.443)[a]	−0.048 (0.313)	−0.031 (0.422)	0.033 (0.443)[a]	23 (14)
	Ion regulation	Na (mM)	156 (5)	0.002 (0.843)	0.122 (0.103)	0.065 (0.279)	−0.001 (0.864)	<0.001 (0.931)	−0.065 (0.279)	23 (14)
		K (mM)	6.7 (2.7)	0.001 (0.913)	0.264 (0.012) ·	−0.008 (0.702)	−0.002 (0.854)	0.056 (0.277)	−0.008 (0.702)	23 (14)
		Ca (mg dL^{-1})	6.8 (1.8)	<0.001 (0.999)	−0.001 (0.882)	0.071 (0.257)	−0.010 (0.651)	0.001 (0.897)	−0.071 (0.257)	23 (14)
		P (mM)	9.2 (2.5)	0.009 (0.667)	−0.122 (0.102)	−0.029 (0.472)	−0.001 (0.871)	0.028 (0.442)	−0.029 (0.472)	23 (14)
		Cl (mM)	118 (6)	<0.001 (0.956)	−0.135 (0.085)	0.192 (0.053)	<0.001 (0.988)	−0.003 (0.813)	0.192 (0.053)	23 (14)
	Testosterone	Female (pg mL^{-1})	180 (200)	−0.003 (0.832)	−0.083 (0.246)[a]	−0.074 (0.308)[a]	<0.001 (0.997)	−0.290 (0.026)*	−0.074 (0.308)[a]	17 (11)
		Male (pg mL^{-1})	4130 (2520)	−0.570 (0.140)	0.092 (0.621)	−0.478 (0.196)	0.071 (0.666)	−0.259 (0.381)	0.478 (0.196)	5 (3)

Notes: All values are the r^2 (p-value) from Pearson product-moment correlations unless indicated otherwise. The sign in front of each correlation coefficient indicates whether the correlation was in the positive or negative direction.

[a] Contaminant concentrations were log-transformed.

[b] Spearman rank correlation, r_S (p-value).

[c] n = number of samples and the numbers in parentheses are sample numbers for the correlations with ΣPBDEs.

* Significant correlation ($p < 0.05$).

** Significant correlation ($p < 0.05$) strongly influenced by an outlier in the clinical plasma chemistry parameter; BCI = body condition index; WBC = white blood cell count; HP:lymph ratio = heterophil:lymphocyte ratio; alb:glob ratio = albumin:globulin ratio; AST = aspartate amino(s)transferase.

Table 3
Mean concentration (SD) (pg g^{-1} wet mass) of major contaminants and TEO content in blood of different sea turtle species.

Sea turtle species	Location	*n*	ΣPCB	ΣPBDE	ΣDDT	Σchlordane	Dieldrin	Mirex	% TEO
Green[a]	Gulf of Mexico	9	534 (701)	158 (217)	128 (114)	11.2 (20.5)	96.0	14.7 (11.4)	0.310 (0.096)
Kemp's ridley[a]	Gulf of Mexico	46	4270 (3620)	230 (273)	686 (656)	113 (100)	225 (119)	15.2 (14.5)	0.262 (0.153)
Kemp's ridley[a]	SE coast USA	3	10 700 (12 200)	148 (141)	1490 (1790)	1220 (1490)	608 (409)	9.18 (0.484)	0.538 (0.180)
Kemp's ridley[b]	MA, USA	8	4540 (5760)	NA	793 (678)	356 (376)	82.5 (75.1)	32.8 (62.2)	0.461 (0.313)
Loggerhead[b]	NC, USA	44	5560 (5280)	NA	649 (685)	225 (201)	60.8 (141)	44.5 (70.8)	0.262 (0.080)
Loggerhead[c]	NC, USA	45	3780 (5810)	66.0 (71.9)	2400 (3610)	30.8 (42.3)	NA	NA	NA
Green[d]	Queensland, Australia	7 pooled	NA	4.44	NA	NA	NA	NA	0.12
Flatback[d]	Queensland, Australia	1	NA	6.09	NA	NA	NA	NA	0.07
Hawksbill[d]	Queensland, Australia	1	NA	1.30	NA	NA	NA	NA	0.01

Notes: NA = not available.
[a] This study.
[b] Keller et al. (2004b).
[c] Carlson (2006).
[d] Hermanussen et al. (2008).

sea turtles for different contaminants. Overall profiles of individual PBDE congeners were also similar to those found in many other species, including humans (Hites, 2004).

The greater prevalence of highly chlorinated PCB congeners in the three SE USA Kemp's ridleys compared to that of Gulf turtles indicates that SE USA turtles were exposed to a more highly chlorinated technical formulation of PCBs. In fact, a chloralkali plant in coastal Georgia manufactured and released a highly chlorinated PCB mixture, Aroclor 1268, leading to its designation as a Superfund site. The top ten PCB congeners in Aroclor 1268 are PCB 206, 199, 208, 196, 202, 209, 194, 187, 207 and 201 (Maruya and Lee, 1998). A highly conserved Aroclor 1268 PCB profile has been seen in local sediments (Kannan et al., 1997) and in local and regional fauna (Pulster and Maruya, 2008). SE USA Kemp's ridleys were caught near this source of Aroclor 1268 and had higher percentages of PCBs 187, 194, 196 + 203, 199, 202 and 206, indicating that these turtles were feeding on prey exposed to Aroclor 1268. Although only three turtles from this region were sampled and the findings should be viewed as highly tentative, the PCB pattern in these turtles indicates that Kemp's ridleys may integrate regional contamination.

4.2. Correlations with morphological, health and immune function parameters

No correlations were observed between morphometric characteristics and OH concentrations, indicating that length and body condition do not influence OH accumulation in this age range of Kemp's ridleys. However, the sample size for this correlation ($n = 26$) and range of values (e.g. SCL range: 29.1–43.0 cm; mean: 35.2 cm) may have limited the ability to properly investigate the relationship. A study of younger, smaller Kemp's ridleys (mean SCL of 26 cm) was unable to quantify any of the 20 OCPs investigated in blood, including DDTs, chlordanes, dieldrin and mirex (Innis et al., 2008). Unfortunately, no conclusions about the relationship between age and contaminants can be drawn from these results because the limits of quantification (10 ng g^{-1} wet mass) of the methods employed by Innis et al. (2008) were two to four orders of magnitude greater than those of the methods used in this study, and were inadequate for the measurement of trace levels of OHs in blood. In a study similar to ours, no correlation between blood contaminant concentrations and turtle length was observed for immature loggerheads (Keller et al., 2004c).

The majority of the clinical blood chemistry values reported in Table 2 fell within the range of values reported in other studies of cold-stunned Kemp's ridleys (Innis et al., 2008, 2009) and free-ranging loggerheads (Keller et al., 2004c). While Keller et al. (2004c) found numerous correlations between OH contaminant concentrations and health and clinical blood chemistry parameters in loggerhead turtles, only five of the 168 examined correlations between summed contaminant classes and health and immune function parameters analyzed in the current study were statistically significant. Concentrations of OHs in the blood of Kemp's ridleys investigated in this study were generally one to three orders of magnitude lower than concentrations that have been associated with health effects in other reptile species from contaminated sites. Plasma concentrations of PCBs, 4,4′-DDE, oxychlordane, trans-nonachlor, and mirex that were approximately two or more orders of magnitude higher have been associated with altered sexual morphology in adult male snapping turtles (*Chelydra serpentina*) (De Solla et al., 1998). Guillette et al. (1999) reported altered sexual morphology in male juvenile American alligators (*Alligator mississipiensis*) where mean serum concentrations of 4,4′-DDE, dieldrin, mirex, oxychlordane and trans-nonachlor were one or more orders of magnitude higher than those reported in juvenile Kemp's ridleys in this study. While the effects observed in both of these studies were not correlated with any one class of contaminants, studies involving loggerheads reported that similar blood concentrations of OHs were correlated to altered health parameters (Keller et al., 2004c).

Turtles with higher Σchlordane concentrations had higher concentrations of blood urea nitrogen (BUN). The same correlation was reported for loggerheads (Keller et al., 2004c). The authors of that paper suggest that increased BUN in turtles is less of an indicator of renal disease (as it is in mammals) and more of an indicator of recent feeding. Debilitated and injured sea turtles followed through rehabilitation and recovery show increased BUN, indicating that their nutritional status was improving (Harms et al., 2000). It is plausible that a turtle in good nutrition, which has fed recently, would have higher blood lipid content and concomitantly higher chlordanes because of the larger number of available binding sites in the blood. Interestingly, Σchlordane concentrations measured in the current study were the most closely related contaminant class to percent lipid, which would support the above reasoning.

A negative association was observed between ΣPCBs and CPK. Increased CPK indicates cellular damage to muscle (Evans, 1996); therefore, the negative correlation suggesting that turtles with higher blood contaminant concentrations may have less organ damage was unexpected. PCBs are known to be hepatotoxic (Kutlu et al., 2007), and significant positive correlations have been observed in loggerheads between several OHs and aspartate aminotransferase (AST), another indicator of damage to organs, including the liver (Keller et al., 2004c). Little is known, however, about which organs produce these enzymes in sea turtles and which stressors lead to increased enzyme concentrations in blood.

One correlation was observed between OH concentrations and the hematological and immune function parameters. Concentrations of ΣDDTs were correlated to decreased proliferation of T-cells stimulated by 5 mg mL^{-1} of PHA. Similar effects have been seen in Caspian terns (*Sterna caspia*) from the Great Lakes (Grasman et al., 1996) and in beluga whale (*Delphinapterus leucas*) *in vitro* exposure assays (De Guise et al., 1998). Loggerheads from North Carolina showed the opposite, a positive correlation between OH concentrations and lymphocyte proliferation (Keller et al., 2006). The relationship observed in the present study between T-cell proliferation and ΣDDT concentrations adds to the evidence that environmental contaminants may modify the immune response of sea turtles.

The inverse association between dieldrin and plasma testosterone levels in females is consistent with the evidence that this compound may interfere with hormone receptors and hormone production. While complex contaminant mixtures containing dieldrin were shown to override temperature-dependant sex determination signals in red eared slider (*Trachemys scripta elegans*) embryos, dieldrin assayed alone had no effect on sex determination (Willingham and Crews, 1999). Although this correlation was statistically significant, the biological significance is difficult to determine. Female sub-adult testosterone concentrations observed in this study (mean ± SD: 17.1 ± 20.1 pg mL^{-1} blood) were in the range of values observed for non-mating and post-nesting adult females (Rostal et al., 1998). The authors of this study suggested that increased levels of testosterone in female Kemp's ridleys shortly prior to mating might play a role in pre-mating behavior or receptivity, but the role that testosterone plays in sub-adult turtles is unknown.

Correlations between organic contaminants and numerous other health and immune function parameters have been reported for juvenile loggerheads at similar contaminant concentrations (Keller et al., 2004c, 2006). Only one of the correlations observed in Kemp's ridleys, a positive correlation with BUN and Σchlordanes, has been previously observed in loggerheads (Keller et al., 2004c). Differences in correlations observed in the current study and studies involving loggerheads demonstrate the difficulty in examining health effects of contaminants on wildlife using a weight of evidence approach. Future monitoring studies of free-ranging sea turtles would benefit from the development of more sensitive biomarkers of exposure to PCBs, DDTs and PBDEs.

5. Conclusions

Persistent OH contaminants, including PCBs, PBDEs, DDTs, chlordanes, and other OCPs were present in the blood of free-ranging Kemp's ridley and green sea turtles. Concentrations of PCBs, DDTs and dieldrin in the blood of Kemp's ridleys were similar to those reported previously in Kemp's ridley and loggerhead sea turtles. Concentrations of all contaminants were lower in herbivorous green sea turtles than in omnivorous Kemp's ridleys. Contaminant concentrations differed little among capture sites, but the PCB congener patterns differed spatially. A distinctive Aroclor 1268 PCB profile observed in Kemp's ridleys caught off the southeastern coast of the US was indicative of exposure from a nearby Superfund site; however, the small number of turtles sampled here limits the interpretation of this finding.

Concentrations of OHs in Kemp's ridleys were not correlated to any morphological proxies for age. Few correlations between OH contaminant concentrations and health or immune function parameters were observed. While this suggests these contaminants may not be an important impediment to the recovery of this critically endangered species, further studies with larger sample sizes are needed to determine this conclusively. Additionally, sensitive endocrine system endpoints, such as thyroid hormone levels, were not tested and assays should be developed for future studies. The suite of organic compounds analyzed should also be expanded to include perfluorinated compounds and additional brominated flame retardants.

Disclaimer

Certain commercial equipment, instruments, or materials are identified in this paper to specify adequately the experimental procedure. Such identification does not imply recommendation or endorsement by the National Institute of Standards and Technology, nor does it imply that the materials or equipment identified are necessarily the best available for the purpose.

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Appendix A. Supplementary material

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.chemosphere.2009.10.059.

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