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Cliff Swallows *Petrochelidon pyrrhonota* as Bioindicators of Environmental Mercury, Cache Creek Watershed, California

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Abstract To evaluate mercury (Hg) and other element exposure in cliff swallows (*Petrochelidon pyrrhonota*), eggs were collected from 16 sites within the mining-impacted Cache Creek watershed, Colusa, Lake, and Yolo counties, California, USA, in 1997–1998. Nestlings were collected from seven sites in 1998. Geometric mean total Hg (THg) concentrations ranged from 0.013 to 0.208 μg/g wet weight (ww) in cliff swallow eggs and from 0.047 to 0.347 μg/g ww in nestlings. Mercury detected in eggs generally followed the spatial distribution of Hg in the watershed based on proximity to both anthropogenic and natural sources. Mean Hg concentrations in samples of eggs and nestlings collected from sites near Hg sources were up to five and seven times higher, respectively, than in samples from reference sites within the watershed. Concentrations of other detected elements, including aluminum, beryllium, boron, calcium, manganese, strontium, and vanadium, were more frequently elevated at sites near Hg sources. Overall, Hg concentrations in eggs from Cache Creek were lower than those reported in eggs of tree swallows (*Tachycineta bicolor*) from highly contaminated locations in North America. Total Hg concentrations were lower in all Cache Creek egg samples than adverse effects levels established for other species. Total Hg concentrations in bullfrogs (*Rana catesbeiana*) and foothill yellow-legged frogs (*Rana boylii*) collected from 10 of the study sites were both positively correlated with THg concentrations in cliff swallow eggs. Our data suggest that cliff swallows are reliable bioindicators of environmental Hg.

The Cache Creek watershed, located in Colusa, Lake, and Yolo counties in California's Coast Range, USA, is an area with abundant geologic sources of mercury (Hg) and a long history of anthropogenic Hg contamination (Rytuba 2000). Sources of Hg in the Cache Creek watershed include agricultural runoff, erosion of naturally Hg-enriched soils, and atmospheric deposition. Most of the Hg exported from Cache Creek, however, originates from geothermal springs and abandoned and inactive Hg mines in the upper watershed (Domagalski et al. 2004). Active Hg mining occurred in the Cache Creek watershed during the latter part of the 19th century and continued until the 1950s (Domagalski et al. 2000). Tributaries affected by abandoned mining sites continue to be sources of Hg pollution to Cache Creek and downstream water bodies, including the Sacramento-San Joaquin River Delta and San Francisco Bay Estuary (Foe and Croyle 1999; Domagalski 2001; Domagalski et al. 2004).

Mercury, especially in its more bioavailable form, methylmercury (MeHg), is highly toxic and is readily bioaccumulated by both invertebrate and vertebrate biota. Benthic macroinvertebrates may serve as a link in the food chain for the transfer of Hg in sediments (Domagalski et al. 2004) to higher trophic levels (Steingraeber and Wiener 1995). The bioaccumulation of MeHg has been documented in aquatic invertebrates (Slotton et al. 1997) and fish (Slotton et al. 1995) from the Cache Creek watershed.

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Prior to this study, the bioaccumulation of Hg in insectivorous birds, namely, the cliff swallow (*Petrochelidon pyrrhonota*), had not been evaluated. Although tree swallows (*Tachycineta bicolor)* have been used more extensively as biomonitors of contamination (T. Custer et al. 2001; C. Custer 2003a, b, 2006, 2007; Gerrard and St. Louis 2001; Longcore et al. 2005), cliff swallows also have been used (Mora et al. 2005; Maruya et al. 2005).

Amphibians play a vital role in many aquatic food webs and may serve as indicators of metals contamination (Cooke 1981). The foothill yellow-legged frog (*Rana boylii*) is native to the Cache Creek watershed and is common in the upper reaches, while the bullfrog (*Rana catesbeiana*), an introduced species to California, is common in suitable habitat throughout the watershed (Kupferberg 1997).

The objectives of this study were (1) to quantify and compare the bioaccumulation of Hg and other elements detected in cliff swallow eggs and nestlings collected from the Cache Creek watershed, (2) to relate these accumulations to Hg sources, and (3) to evaluate the suitability of cliff swallows as biomonitors by comparing bioaccumulation of Hg by cliff swallows with that of amphibians collected from the same sites.

Methods

Study Area

The Cache Creek watershed (2950 km^2) is located about 130 km north of San Francisco and 30 km northwest of Sacramento (Fig. 1). Cache Creek flows from Clear Lake to the Cache Creek Settling Basin and ultimately the Yolo Bypass east of the city of Woodland. It has two major tributaries, the North Fork of Cache Creek and Bear Creek. Study sites (n = 16) were selected based on the presence of nesting cliff swallows and current knowledge of Hg contamination. For spatial comparisons, the study area was classified into five regions based on proximity to Hg mine sites and stream gradient (Table 1 and Fig. 1): reference, mine, canyon, upper valley, and lower valley. Three reference sites were located in the upper reaches of the watershed above known anthropogenic and natural sources of Hg (Domagalski et al. 2004). The mine region included four sites within 2 km downstream of mines and natural sources of Hg. The canyon region consisted of three study sites located 5 to 20 km downstream of mine sites along high-gradient streams. The upper valley region included three sites located approximately 25 to 65 km downstream of mine sites along the low-gradient main stem of Cache Creek. The lower valley region included two low-gradient sites within 10 km of the Sacramento River and one on the west bank of the Sacramento River (Table 1 and Fig. 1).

Sample Collection

Eggs were collected from 14 colonies during 5–20 May 1997 (Table 1). Based on egg availability, samples of three pooled eggs or five individual eggs were collected from each location and analyzed for total Hg (THg) (Tables 1 and 2). At each of seven sites, an additional sample of three

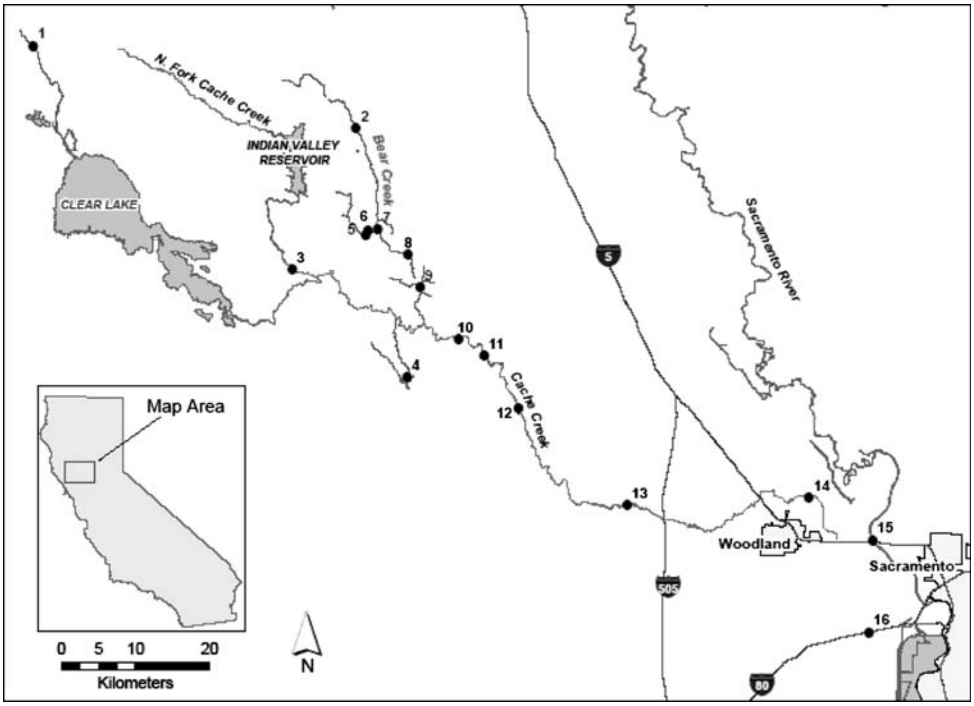

Fig. 1 Cliff swallow study sites, Cache Creek watershed, California, USA, 1997–1998

Table 1 Cache Creek study sites (see Fig. 1) and samples collected in 1997 and 1998

Site no.	Location	Region	Samples collected[a]		Latitude, longitude
			1997	1998	
1	W. Fork Middle Cr.	Reference	E*F	—	39.252°N, 122.954°W
2	Mill Cr. at Brim Rd.	Reference	E*BF	EN	39.163°N, 122.446°W
3	N. Fork Cache Cr.	Reference	E	—	38.988°N, 122.540°W
4	Davis Cr. Reservoir	Mine	E*BF	—	38.862°N, 122.357°W
5	Sulfur Cr. Barn	Mine	E*F	E	39.033°N, 122.427°W
6	Sulfur Cr. Bridge	Mine	—	EN	39.038°N, 122.423°W
7	Bear Cr. at Sulfur Cr.	Mine	E*BF	E	39.040°N, 122.408°W
8	Bear Cr. at Hwy 20	Canyon	E	EN	39.011°N, 122.361°W
9	Bear Cr., Thompson Canyon	Canyon	EF	—	38.971°N, 122.340°W
10	Cache Cr. at Camp Haswell	Canyon	EB	—	38.909°N, 122.278°W
11	Cache Cr. at Rumsey Bridge	Upper valley	E	—	38.890°N, 122.238°W
12	Cache Cr. at Guinda Bridge	Upper valley	E*B	—	38.828°N, 122.183°W
13	Cache Cr. at Esparto Bridge	Upper valley	EB	—	38.713°N, 122.011°W
14	Cache Cr. at Rd. 102 Bridge	Lower valley	E*B	EN	38.726°N, 121.729°W
15	Sacramento Riv. Pump Sta.	Lower valley	E	—	38.676°N, 121.629°W
16	Yolo Basin Wildlife Area	Lower valley	—	EN	38.564°N, 121.632°W

Note. W, west; N, north; Cr., Creek; Rd., Road; Riv., River; Sta., Station

[a] E*—eggs analyzed for metals, trace elements, and MeHg; E—eggs analyzed for THg only; N—nestling, B—bullfrog, F—foothill yellow-legged frog samples analyzed only for THg

pooled eggs was analyzed for THg and MeHg, and another sample of three to four pooled eggs was analyzed for trace elements (Tables 1 and 2). Based on the results in 1997, five study sites that appeared to be most heavily affected by Hg and two new sites thought to be affected by Hg (one in the mine area and one in the lower valley) were sampled between 29 April and 22 May 1998 (Table 1). Six eggs were collected from each site (with the exception of three eggs from site 5) and analyzed individually for THg. Prefledging nestlings (11 to 19 days old) were collected from five of the seven sites (Table 1) during May 26–July 1 and analyzed for THg.

Only one egg was collected from each sampled nest. Nests were chosen based on egg availability and accessibility at each site. Eggs were kept on ice or refrigerated until processed within 7 days of collection. Eggs were measured and weighed, and then the contents were removed from the shell. Egg fertility was determined, and embryonic viability, normality, and stage of development were assessed. Total and sample mass (±0.1 g) for each egg were determined with an electronic balance. When nestlings were collected in 1998, the largest nestling, based on visual appearance, was collected from the same clutch previously sampled for an egg. Each nestling was euthanized by chest compression, placed on ice, and refrigerated until processed within 1 day of collection. To confirm nestling age, the wing, tarsus, gape, and selected feathers of each nestling were measured with calipers (±0.1 mm) (Stoner 1945; St. Louis and Barlow 1993). Using an electronic balance, the fresh mass (±0.1 g) of each nestling was measured, then the contents of the digestive tract were removed, and the sample mass was determined. Egg contents or nestling carcasses (including feathers) were placed individually in labeled chemically clean jars (VWR TraceClean), sealed with Parafilm, and frozen at −20°C pending chemical analysis.

In 1997, three individual bullfrogs were collected from each of seven cliff swallow colony sites, and three individual foothill yellow-legged frogs were collected from each of six swallow sites (Table 1). Both species of frogs were collected from three sites. Frogs were euthanized with MS-222 and kept frozen until they could be processed within 2 days of collection. The digestive tract was removed, and the carcass, including the stripped and rinsed digestive tract, was then analyzed for THg. Frog specimens were placed individually in labeled chemically clean jars (VWR TraceClean), sealed with Parafilm, and frozen at −20°C pending chemical analysis.

Chemical Analyses

The Trace Element Research Laboratory (TERL) in College Station, Texas, performed all chemical analyses. The mean lower levels of detection (µg/g dry weight [dw]) in 1997 were as follows: aluminum (Al), 0.52; arsenic (As),

Table 2 Geometric mean concentrations and 95% confidence intervals for total mercury (THg μg/g, wet wt) in cliff swallow eggs from the Cache Creek watershed, CA, 1997–1998

Region (site no.)	1997				1998			
	% moisture	Mean THg	95% CI	*n*[a]	% moisture	Mean THg	95% CI	*n*[b]
Reference (1)	74.1	0.023 BCD[c]	0.020–0.026	7	—	—	—	—
Reference (2)[d]	83.1	0.020 CD	0.018–0.023	7	79.7	0.044 C	0.041–0.047	6
Reference (3)	83.4	0.022 BCD	0.019–0.025	5	—	—	—	—
Mine (4)	79.9	0.059 ABCD	0.050–0.069	7	—	—	—	—
Mine (5)[d]	73.9	0.097 AB	0.076–0.122	7	68.1	0.142 ABC	0.075–0.269	3
Mine (6)	—	—	—	—	80.6	0.208 A	0.156–0.278	6
Mine (7)	81.7	0.081 AB	0.070–0.093	7	78.7	0.072 BC	0.062–0.084	6
Canyon (8)	86.2	0.070 ABC	0.055–0.089	5	80.0	0.085 ABC	0.069–0.105	6
Canyon (9)	86.1	0.118 A	0.091–0.152	5	—	—	—	—
Canyon (10)	86.3	0.026 ABCD	0.012–0.058	5	—	—	—	—
Upper valley (11)	78.0	0.024	—	1	—	—	—	—
Upper valley (12)	83.3	0.013 D	0.009–0.018	7	—	—	—	—
Upper valley (13)	81.5	0.019 CD	0.012–0.030	5	—	—	—	—
Lower valley (14)[d]	84.9	0.045 ABCD	0.038–0.052	7	79.9	0.088 ABC	0.071–0.109	6
Lower valley (15)	76.5	0.131	—	1	—	—	—	—
Lower valley (16)	—	—	—	—	82.1	0.173 AB	0.151–0.198	6

[a] When $n = 1$, one three-egg composite sample analyzed; when $n = 5$, five individual eggs analyzed; when $n = 7$, five individual eggs analyzed, one three-egg composite (also analyzed for MeHg) and one three- or four-egg composite (also analyzed for other elements)

[b] Means based on individual eggs; no composites were analyzed

[c] Based on Kruskal-Wallis one-way ANOVA on ranks and Dunn's method for all pairwise multiple comparisons, within years; site means not sharing a common letter differed ($p < 0.05$)

[d] Based on two-way ANOVA and Holm-Sidak method for all pairwise multiple comparisons; cliff swallow eggs at these sites had mean THg concentrations that were higher in 1998 than in 1997 ($p < 0.05$). Mercury concentrations at sites 7 and 8 did not differ between years

0.21; boron (B), 1.03; barium (Ba), 0.10; beryllium (Be), 0.10; calcium (Ca), 5.16; cadmium (Cd), 0.01; chromium (Cr), 1.03; copper (Cu), 0.52; iron (Fe), 1.03; potassium (K), 20.61; magnesium (Mg), 10.31; manganese (Mn), 0.10; molybdenum (Mo), 1.03; sodium (Na), 5.16; nickel (Ni), 0.26; phosphorus (P), 5.16; lead (Pb), 0.10; sulfur (S), 5.16; antimony (Sb), 1.03; selenium (Se), 0.21; strontium (Sr), 0.05; titanium (Ti), 1.03; vanadium (V), 1.03; zinc (Zn), 0.52; and MeHg, 0.006. The mean lower level of detection for egg THg was 0.002 μg/g ww in 1997 and 0.005 μg/g ww in 1998, and that for nestling THg in 1998 was 0.004 μg/g ww. The mean lower level of detection for THg in amphibians was 0.013 μg/g ww.

Samples were freeze-dried, and percentage moisture was determined by weight loss upon freeze-drying. Samples were wet-digested with nitric acid and converted into acidic digest solutions for analysis by atomic spectroscopy methods according to the standard protocol of TERL. Samples analyzed for Al, B, Ba, Be, Ca, Cr, Cu, Fe, K, Mg, Mn, Mo, Na, Ni, P, S, Sb, Sr, Ti, V, and Zn were determined by inductively coupled plasma optical emission spectroscopy (ICP). Arsenic, Cd, and Pb were analyzed by graphite furnace atomic absorption spectroscopy (GFAAS), and Se was analyzed by atomic fluorescence spectroscopy (AFS). Mercury was analyzed by cold vapor atomic absorption spectroscopy (CVAAS), and MeHg was analyzed by CVAAS and methods of Uthe et al. (1972). To assure the accuracy of the methods, blanks, spikes, duplicates, and certified reference material were all analyzed at a rate of 5%, with at least one per matrix per analytical run. Quality assurance values were within normal limits for all elements for both years. For THg, the recovery ranged from 81.2% to 96.0% in 1997 and from 81.8% to 101.0% in 1998. Recoveries of other elements in 1997 ranged from 67.0% for calcium to 116.0% for copper. Concentrations were not adjusted for percentage recovery.

Statistical Analyses

To satisfy the assumption of homogeneity of variance for analysis of variance (ANOVA), data were log-transformed. One-way ANOVA was used to analyze site effects within years. Where either the normality or the equal variance test failed ($p > 0.05$), a Kruskal-Wallis one-way ANOVA on ranks was performed. Multiple comparisons were conducted using Dunn's method. The significance level for all tests was $\alpha = 0.05$. Two-way ANOVA was utilized to test for differences in THg concentrations between eggs and nestlings collected at the same sites in 1998 and to compare differences in THg concentrations between years at the same sites. Pairwise multiple comparisons were conducted using the Holm-Sidak method. Linear regressions were used to evaluate the relationships between THg concentrations in cliff swallow eggs and those in amphibians collected from the same sites. Egg and nestling body burdens were calculated by multiplying the THg concentration (μg/g) by the sample mass (g). When an egg and a nestling were collected from the same clutch, the contribution of THg from the egg to the nestling was estimated by dividing the amount of THg (μg) detected in a nestling into the amount of THg (μg) detected in a sibling egg. All egg and nestling THg concentrations by site are reported as geometric means, unless otherwise stated.

Results

Mercury Concentrations in Cliff Swallow Eggs and Nestlings

Mercury levels detected in cliff swallow eggs are presented in Table 2, along with sample size and Kruskal-Wallis one-way ANOVA on ranks showing differences among sites. The natural occurrence of Hg in the Coast Range combined with potential atmospheric deposition resulted in detectable levels of Hg in all samples, including those collected at reference sites.

In 1997, geometric mean THg concentrations in eggs (n = 114) ranged from 0.013 to 0.118 μg/g ww. Mercury concentrations detected at mine sites 5 and 7 were higher than at one of the three reference sites ($p < 0.05$) (site 2). At site 9, located in the canyon region, the mean concentration was higher than at all reference sites ($p < 0.05$). The highest concentration (0.131 μg/g ww in one composite sample) was detected at site 15 in the lower valley region and was six times higher than the references. Intermediate concentrations of THg were detected in eggs from mine site 4 and canyon site 8, which were two to three times higher than, but not statistically different from ($p > 0.05$), those at the reference sites. Eggs collected from the upper valley sites on the main stem of Cache Creek had mean concentrations similar to (sites 11 and 13), or somewhat lower than (site 12), those at the reference sites.

In 1998, geometric mean THg concentrations in eggs (n = 39) ranged from 0.044 to 0.208 μg/g ww. As in 1997, concentrations were low above the point sources (site 2) and high at the mine sites (sites 5–7), just below the mines (site 8), and in the lower reaches of the watershed at the Yolo Basin Wildlife Area (site 16) (Fig. 2). The highest concentration, detected at mine site 6 (0.208 μg/g ww), was nearly five times higher than the reference ($p < 0.05$). The mean concentration in eggs from site 16 was significantly higher ($p < 0.05$) than that at the reference site, but was not significantly different ($p > 0.05$) from that at the three mine sites. Comparisons by two-way ANOVA of mean egg THg concentrations from the five sites sampled in both years indicate that site ($F_{4,50} = 46.9$), year ($F_{1,50} = 34.2$), and the interaction between site and year ($F_{4,50} = 6.8$) all contributed to differences ($p < 0.001$). Three of the sites had higher ($p < 0.05$) THg concentrations in eggs in 1998 than in 1997 (sites 2, 5, and 14; Table 2 and Fig. 2).

Geometric mean THg concentrations in nestlings (n = 24) ranged from 0.047 to 0.347 μg/g ww (Table 3). Mean concentrations for all sampled nestling sites (6, 8, 14, and 16) were significantly higher than at the reference site (one-

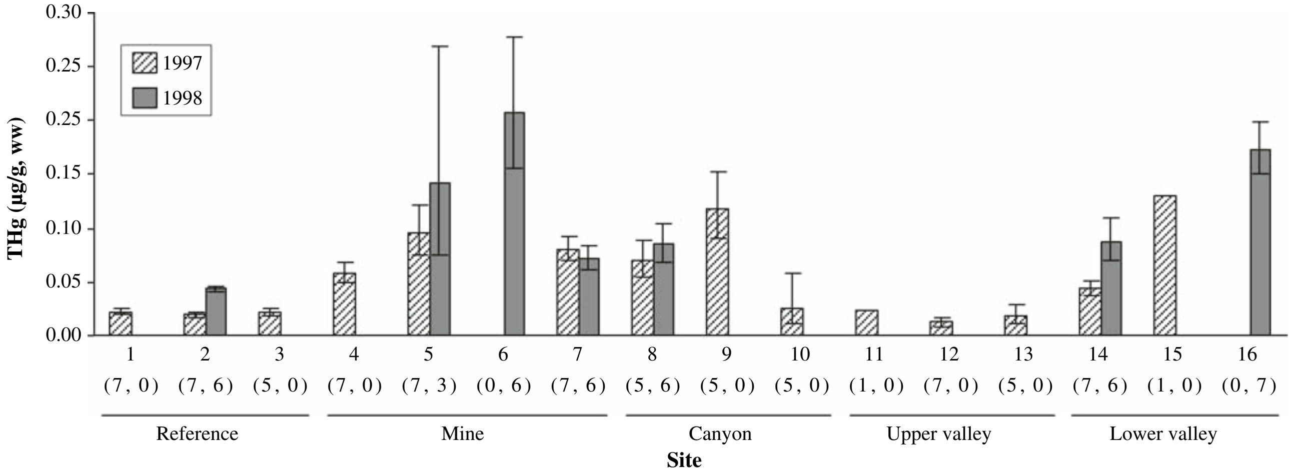

Fig. 2 Geometric mean concentrations of THg (μg/g wet wt) ± 95% confidence intervals in cliff swallow eggs collected from the Cache Creek watershed in 1997 and 1998 (sample size in parentheses)

Table 3 Geometric mean concentrations, burdens, and 95% confidence intervals (in parentheses) for total mercury (THg, wet wt.) in nestlings and sibling eggs of cliff swallows from sites in the Cache Creek watershed, CA, 1998

Site no. (region)	Egg			Nestling carcass				
	n % moisture	THg (µg/g)	Hg burden (µg)	*n* % moisture	THg (µg/g)	Hg burden (µg)	Mean age (days)	Mean % burden from egg
2 (reference)	6	0.044 C[a]	0.058 C[a]	6	0.047 C[a]	1.06 B[b]	13	5.7
	79.7	(0.041–0.047)	(0.052–0.065)	68.1	(0.041–0.054)	(0.880–1.27)		(4.40–7.04)
6 (mine)	3	0.193 A[c]	0.268 A	3	0.116 B	2.25 AB	16	13.4
	80.6	(0.120–0.313)	(0.165–0.435)	66.5	(0.067–0.201)	(0.800–6.35)		(4.36–22.5)
8 (canyon)	6	0.085 B	0.116 B	6	0.347 A	6.41 A	14	1.9
	80.0	(0.069–0.105)	(0.100–0.136)	66.6	(0.290–0.416)	(4. 56–9.01)		(1.47–2.27)
14 (lower valley)	6	0.088 B	0.118 B	6	0.167 B	4.05 AB	16	3.0
	79.9	(0.071–0.110)	(0.097–0.145)	67.5	(0.157–0.178)	(3.77–4.35)		(2.52–3.42)
16 (lower valley)	3	0.175 A[c]	0.268 A	3	0.157 B	3.36 AB	18	9.2
	82.1	(0.138–0.223)	(0.166–.0433)	63.8	(0.117–0.210)	(2.63–4.30)		(1.95–16.5)

[a] Based on one-way ANOVA and all pairwise multiple-comparison procedure (Holm Sidak); site means within tissue type not sharing a common letter differed ($p < 0.05$)

[b] Based on Kruskal-Wallis one-way ANOVA on ranks and all pairwise multiple comparison procedure (Dunn's method); site means within tissue type not sharing a common letter differed ($p < 0.05$)

[c] Means different from those in Table 2 because only sibling eggs to collected nestlings included in the calculation of means in this table

way ANOVA, $p < 0.001$). The highest mean concentration was detected in nestlings collected from the canyon region at site 8, which was seven times higher than that at the reference site. Based on two-way ANOVA, the geometric mean concentrations of THg in eggs and nestlings (Table 3) from five sites sampled in 1998 were different (*p*

Table 4 Concentrations of elements (µg/g, dry wt) from composites of cliff swallow eggs from the Cache Creek watershed in 1997

Analyte	Reference		Mine			Upper valley	Lower valley
	Site 1	Site 2	Site 4	Site 5	Site 7	Site 12	Site 14
N	3	4	4	4	4	4	4
% moisture	79.6	81.8	79.8	78.5	80.4	76.0	82.0
Al	<0.510	<0.498	2.65	10.5	1.02	<0.522	0.589
B	<1.02	1.59	2.54	5.84	7.18	<1.04	2.89
Ba	11.6	8.38	6.09	10.6	11.4	17.9	5.73
Be	<0.102	0.192	0.157	0.398	0.167	<0.104	<0.103
Ca	2540	3110	23000	19300	3010	2610	12200
Cu	2.14	1.10	3.59	2.30	1.44	1.78	2.37
Fe	123	108	153	130	87.4	103	132
THg	0.085	0.100	0.422	0.393	0.470	0.064	0.293
MeHg	0.113	0.098	0.402	0.406	0.535	0.087	0.282
K	6930	6320	8480	7920	6900	6530	7720
Mg	347	369	647	555	376	346	498
Mn	3.80	2.66	4.13	5.37	3.50	2.91	2.95
Na	7330	6940	9220	8710	7220	6970	8580
P	7230	7090	7820	6840	6740	7030	6990
S	5020	4870	6170	5660	5090	4880	5800
Se	2.11	2.30	2.94	2.09	1.68	1.65	2.37
Sr	11.3	11.1	46.8	55.9	15.8	9.27	24.9
V	<1.02	<0.996	1.59	1.77	<1.03	<1.04	<1.03
Zn	59.9	48.9	54.7	52.6	54.1	51.3	54.1

Note. < indicates element below listed detection limit

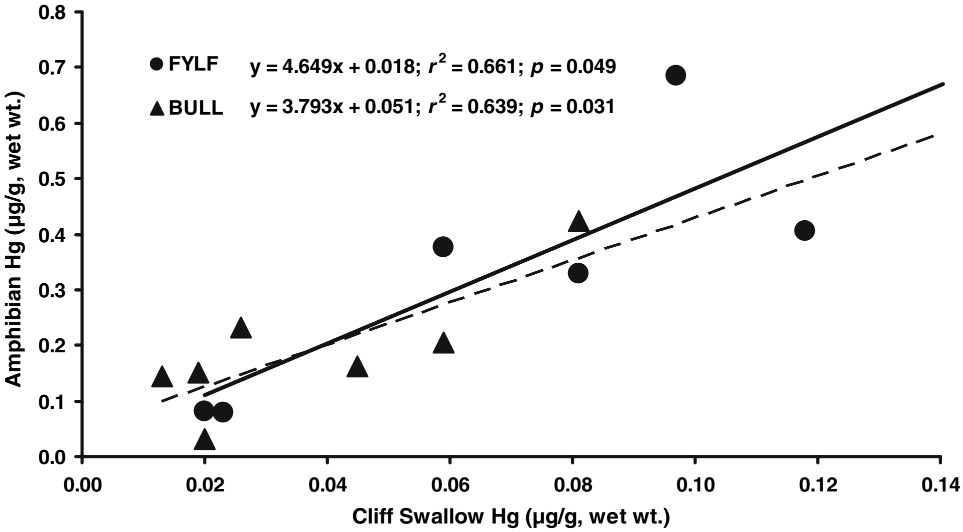

Fig. 3 Linear regression of geometric mean THg (µg/g wet wt) in bullfrogs (triangles and dashed trend line) and foothill yellow-legged frogs (circles and solid trend line) on geometric mean THg in cliff swallow eggs from the Cache Creek watershed, California, 1997

< 0.001) by site ($F_{4,44} = 59.8$), for sample matrix (i.e., egg or nestling) ($F_{1,44} = 17.6$), and for the interaction between site and sample matrix ($F_{4,44} = 25.3$). Mean concentrations differed between eggs and nestlings at three of the five sites, with nestlings having a higher mean THg concentration at sites 8 and 14 but a lower THg at site 6. The mean percentage burden from the egg, in other words, the percentage of THg transferred from the egg to the nestling, ranged from 1.9% to 13.4%. Gross abnormalities were not observed in advanced embryos or nestlings either year.

Other Elements in Eggs

Total Hg and MeHg were highly correlated ($r^2 = 0.97$, $p < 0.001$); nearly all THg was in the methylated form (Table 4). Eight other metals, Cd, Cr, Ti, Mo, Pb, As, Ni, and Sb, were not detected in the composite samples of eggs from any of the seven sampled sites. Three metals, Al, Be, and B, were detected at the mine sites but were low or not detected at the other sites (Table 4). Except for Zn and Ba, all of the elements detected at one or more sites had the highest concentration at one of the mine sites (site 4, 5, or 7; Table 4). Essential elements, K, Na, P, and S, were generally similar among all sites, but the concentrations of Ca in eggs from mine sites 4 and 5 were seven to nine times higher than at the reference sites.

Correlations with Amphibians

In 1997, geometric mean THg concentrations in bullfrogs from seven sites ($n = 21$) ranged from 0.032 to 0.423 µg/g ww, with the highest concentration at mine site 7. In foothill yellow-legged frogs collected from six sites ($n = 18$), THg ranged from 0.080 to 0.686 µg/g ww; the highest concentration detected was at mine site 5. Based on linear regressions, geometric mean THg concentrations in swallow eggs and in bullfrogs ($r^2 = 0.63$, $p = 0.0316$) and foothill yellow-legged frogs ($r^2 = 0.66$, $p = 0.0496$) from the same sites were positively correlated (Fig. 3).

Discussion

Bioaccumulation of Hg in Cliff Swallow Eggs and Nestlings

Although Hg bioaccumulation is a dynamic process influenced by multiple variables, the relationship between Hg in cliff swallow eggs and nestlings in the Cache Creek watershed and the sources (abandoned mines and geothermal sources) is strong. One site-specific variable is the degree of Hg methylation, which is primarily controlled by hydrological and biogeochemical factors and the presence of methylating bacteria. As expected, egg THg concentrations were low at reference sites and higher near mine sites. It was surprising, however, that THg concentrations in both eggs and amphibians from the upper valley sites were relatively low and were similar to those found at the reference sites. These upper valley sites were likely not significant methylation areas. However, farther downstream, at the lower valley sites (Fig. 2), intermediate to high egg THg concentrations were detected (Table 2). Water flow rates at these sites were visibly reduced compared to those at the upper valley sites. The Yolo Bypass Wildlife Area (site 16) receives floodwaters from upstream, including Cache Creek, and is on the west side of the Yolo Bypass, where flows originating in the Cache Creek watershed are most likely to drop sediments contaminated with Hg. Under the lentic conditions present at the lower valley sites (especially 14 and 16), it is assumed that THg was more likely to be deposited and then transformed to MeHg, thus increasing its bioavailability to the swallows.

Mercury in avian eggs reflects short-term exposure (Evers et al. 2005), and caution is necessary for the interpretation of temporal trends because we collected only 2 years of data (Burger and Gochfeld 1995). Between-year differences were significant at two of the five compared sites, including a lower valley site with moderate contamination (site 14) and a reference site (site 2). The reasons for higher Hg concentrations in 1998 were not clear, but they could be related to increased precipitation in 1998, atmospheric deposition of Hg, or other unknown factors. Annual variation of this magnitude is common in swallow eggs (C. Custer et al. 2006).

In migratory species, such as the cliff swallow, females may accumulate contaminants from wintering grounds and along migration routes, confounding egg residue interpretation. Once a bird arrives at the breeding grounds, however, dietary uptake and transfer of MeHg to the egg can occur rapidly (Kambamanoli-Dimou et al. 1991). Additionally, barn swallows (*Hirundo rustica*) have been shown to form eggs mainly from current food intake and not from reserves (Ward and Bryant 2006). Cliff swallows return to California's central valley as early as February (Small 1994) and were observed in both 1997 and 1998 building nests and presumably foraging 10 to 40 days before egg laying. Thus, we conclude that concentrations of THg in eggs represent primarily local exposure. Egg burdens represent the transfer of contaminants from the female to her offspring. Our results indicate that no more than 13.4% of the nestlings' body burden was derived from the egg. Therefore, the source of the majority of the THg in the nestlings was contaminated food from the local environment.

Geometric mean THg concentrations in cliff swallow eggs ranged from 0.013 to 0.208 μg/g ww (or, for comparison purposes, 0.08 to 1.07 μg/g dw). The mean egg THg concentration for Cache Creek mine sites (0.6 μg/g dw, years combined) was two to three times higher than in tree swallow eggs from the Arkansas River, CO (0.2 μg/g dw [C. Custer et al. 2003a]), and the North Platte River, WY (0.3 μg/g dw [T. Custer et al. 2001]), and nearly twice that in eggs from Ontario, Canada (0.365 μg/g dw [Gerrard and St. Louis 2001]). Mean mine site THg concentrations were similar to those in eggs from the Housatonic River, MA (0.6 μg/g dw [C. Custer et al. 2003b]). However, the mean THg concentrations in tree swallow eggs from the Hg-contaminated Carson River, NV (7.35 μg/g dw [C. Custer et al. 2007]), were 12 times higher than the mean concentrations from the Cache Creek watershed mine sites and nearly 7 times higher than the highest detected value in cliff swallow eggs. The range of egg THg concentrations detected in cliff swallows were also lower than the concentrations detected in eggs collected from Acadia National Park, ME (range = 0.097 to 1.313 μg/g ww), and a USEPA superfund site, Ayer, MA (range = 0.231 to 1.075 μg/g ww [Longcore et al. 2005]). Mercury concentrations in Cache Creek cliff swallow eggs were below observed adverse effects levels of 0.5 μg/g ww in ring-necked pheasants (*Phasianus colchicus* [Fimreite 1971]) eggs and ~1.0 μg/g ww in mallard (*Anas platyrhynchous*) eggs (Heinz and Hoffman 2003).

Mean cliff swallow nestling THg body burdens (range = 1.06 to 6.41 μg ww) were lower than the range (4.47 to 13.6 μg ww) of Hg in nestling tree swallows from the Longcore et al. (2005) study. The estimated average percentage contribution of the egg to the body burden was 1.9% to 13.4% in cliff swallows in the present study, compared with 2.4% to 23.9% in Maine (Longcore et al. 2005) and 1.2% to 10.7% in Ontario (Gerrard and St. Louis 2001). Longcore et al. (2005) found that most (81% to 92%) of the Hg that tree swallow nestlings ingest was deposited and retained in feathers, leaving less Hg in the nestling body to affect physiological processes. Feathers were not analyzed separately in this study.

Other Elements in Eggs

Comparisons of THg and MeHg in composite egg samples confirmed that most of the Hg in the eggs was in the methylated form (Gerrard and St. Louis 2001). Where the percentage MeHg exceeded 100%, it is likely that aliquots from the same composite sample were not uniformly homogenized at the laboratory (Bloom 1992).

Several elements that could potentially adversely affect swallow reproduction (Cd, Cr, Mo, Pb, As, and Ni) were either at background levels or not detected. In general, higher concentrations of other metals were most frequently associated with mine sites. With the exception of Ca, which was elevated in eggs from two of the mine sites (Table 4), the essential elements were similar among all sites. Concentrations of Se from Cache Creek swallow eggs were higher than the means reported for barn swallows from reference sites in the San Joaquin Valley (Ohlendorf et al. 1987). However, all values from Cache Creek were lower than the mean concentration observed at Kesterson Reservoir (4.37 μg/g), a site with significant embryo mortality and deformities in other species (Ohlendorf et al. 1989).

Concentrations of Ba and Sr were elevated in cliff swallow eggs from Cache Creek compared to concentrations in tree swallows from other North American locations (T. Custer et al. 2001; C. Custer 2003b, 2007). Concentrations of Ba (5.73–17.9 μg/g dw) in swallow eggs from Cache Creek were higher than the range reported in three deformed clapper rail late-stage embryos (2.16 to 4.13 µg/g dw [Schwarzbach et al. 2006]), while the range of Sr concentrations (9.27–55.9 µg/g dw) was lower than the mean in the same clapper rail embryos (121.4 µg/g [Schwarzbach et al. 2006]). Strontium concentrations in eggs from mine sites 4 and 5 were higher than mean Sr concentrations found in eggs of two passerine species from Arizona (means, 23.9 and 35.1 µg/g [Mora 2003]). The concentrations of Sr in the eggshells of the Arizona birds were considered to be elevated sufficiently to reduce hatching success by adversely affecting eggshell integrity. Further investigations would be required to determine if Ba or Sr are potentially affecting cliff swallow reproduction.

Correlations with Amphibians

Mercury residues in amphibians from the Cache Creek watershed provided a useful comparison for the cliff swallow results. Based on the 1997 data, trends in THg concentrations in frogs generally followed those in swallows. Mean THg concentrations (µg/g, ww) in all frogs were low at sites above known contamination sources and higher at or just below those locations. In 1997, only foothill yellow-legged frogs were found at three sites in the upper reaches of Cache Creek (sites 1, 5, and 9). Foothill yellow-legged frogs were sympatric with bullfrogs at certain sites (sites 2, 4, and 7), but only bullfrogs were present in the lower reaches of Cache Creek (sites 10 and 12–14) (Hothem et al., in preparation). The diets of the swallows were assumed to be primarily emergent aquatic insects (Brown and Brown 1995). Both species of frogs were observed to feed on insects, but the bullfrogs also fed on vertebrates, including fish, snakes, and other frogs (Hothem et al., in preparation). Although we did not observe frogs feeding on birds, Longcore (personal communication) reported bullfrogs with nestling tree swallows in their stomachs in Maine. We found that bioaccumulation in swallows, as reflected by their eggs, was closely correlated with the carcasses of the resident amphibians. This indicates that prey consumed by both swallows and amphibians were similarly contaminated with Hg at individual sites and that bioaccumulation by both taxa was reflective of the level of contamination at those sites.

Cliff Swallow Suitability for Biomonitoring

Preliminary data indicate that colonially nesting cliff swallows may serve as good biomonitors of Hg in the ecosystem. Cliff swallow breeding distribution covers most of temperate North America (Brown and Brown 1995), allowing comparable studies at multiple locations. They are commonly found in close association with aquatic habitats, where contaminants frequently accumulate in sediments and aquatic invertebrates. The food web for swallows is generally uncomplicated during the breeding season because they feed almost exclusively on aerial insects (Brown and Brown 1995) within a relatively short distance (0.4 km) of their nesting sites (Brown et al. 1992). The feeding habits of tree swallows are similar (McCarty and Winkler 1991; Robertson et al. 1992), and although comparative studies have not been done, it is likely that differences in Hg exposure for these two species would be minimal. Although cliff swallow eggs are easy to collect, assessment of their reproductive success is more challenging than for tree swallows, which will use nest boxes.

This study demonstrates the transfer of Hg from the aquatic food web to the terrestrial food web. We conclude that THg concentrations in cliff swallow eggs collected from the Cache Creek watershed in 1997 and 1998 generally followed the assumed spatial distribution of Hg in the watershed, based on proximity to both anthropogenic and natural sources. Additionally, depositional areas for Hg, such as the lower valley sites, may have elevated THg even though they are farther removed from Hg sources. Furthermore, THg concentrations in cliff swallow eggs were correlated with concentrations in bullfrogs and foothill yellow-legged frogs collected from the same sites. Overall, the general patterns of THg concentrations in cliff swallows and amphibians confirm the findings of other studies (Slotton et al. 2004) in that a portion of the Hg transported from abandoned mine sites and geothermal sources (particularly in Sulfur and Bear creeks) is bioavailable and continues to contaminate biota in the watershed. We recommend the use of cliff swallow eggs and nestlings to test the effectiveness of future remedial measures.

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