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M. A. Mora, J. L. Sericano & C. Baxter

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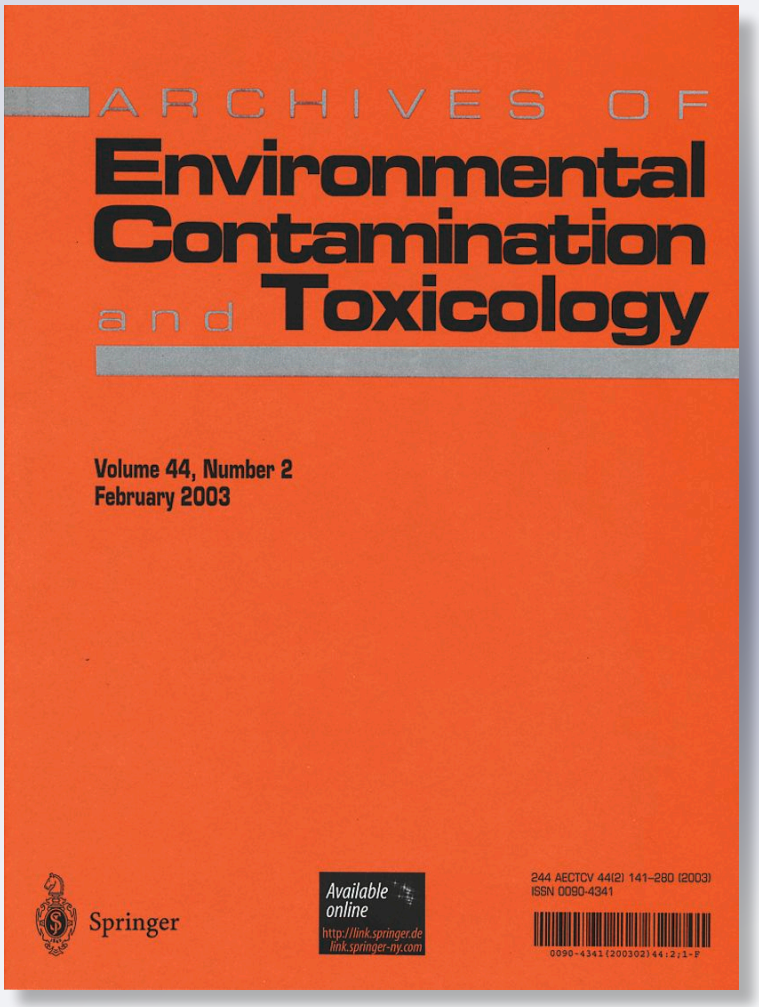

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Swallows as Indicators of Environmental Pollution of the Rio Grande/Rio Bravo Basin: Are Persistent Organic Pollutants a Concern?

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Abstract Swallows from two locations in the Rio Grande/Rio Bravo Basin and one reference site located 500 km away were analyzed for organochlorine pesticides (OCs), polychlorinated biphenyls (PCBs), and polybrominated diphenyl ethers (PBDEs). Of the OC pesticides, only p,p′-DDE was observed at levels of concern (carcass geometric mean range 642 to 8511 ng/g wet weight [ww]). DDE residues in carcass were significantly greater at El Paso than at other locations. PCBs were relatively low compared with locations in the northeastern United States and the Great Lakes. Geometric mean PBDE concentrations ranged from 18 to 258 ng/g ww (280 to 3395 ng/g lipid weight). PBDE congeners 47 and 99 comprised approximately 60% of total PBDEs. Concentrations of DDE measured in swallows from El Paso in 2000 and 2005 are among the highest observed in the last 20 years along the United States–Mexico border. The results from this study indicated that swallows are still being exposed to high concentrations of DDE, which could have adverse effects on reproduction or on predators that feed on swallows.

Recent studies addressing contaminant impacts on birds of the Rio Grande Basin are few. Previously, Mora et al. (2006) documented relatively high concentrations of p,p′-DDE [2,2-bis (*p*-chlorophenyl)-1,1-dichloroethylene; mean 12.4 μg/g wet weight (ww)] in swallows nesting on bridges along the Riverside Canal in the eastern part of El Paso. p,p′-DDE is a metabolite of DDT [1,1,1-trichloro-2,2-bis(*p*-chlorophenyl)ethane], a persistent OC pesticide that was banned in the United States in 1972 and of restricted use in Mexico for malaria control until 2000 (Chanon et al. 2003). The increased concentrations of DDE observed in swallow carcasses during 1999 and 2000 in El Paso were of concern because concentrations were great enough to evoke adverse effects on reproduction. Cliff swallows (*Petrochelidon pyrrhonota*) from El Paso had lower plasma thyroxine (T_4) concentration and increased spleen mass compared with swallows from other regions of the Rio Grande, which also had lower levels of DDE (Mora et al. 2006).

The use of industrial flame retardants in many commercial products has resulted in nearly ubiquitous distribution of polybrominated diphenyl ethers (PBDEs) in the environment (Chen and Hale 2010). PBDEs are a class of chemicals commonly manufactured as flame retardants for use in such products as upholstery, circuit boards, foams, and rubber (Chen and Hale 2010). There are several industrial mixtures composed of specific combinations of certain congeners. The three most common are the deca-, octa-, and penta- mixtures, which make up 80, 6, and 12%, respectively, of industrial production (Darnerud 2003). These flame retardant chemicals are known to accumulate in living organisms, sometimes causing significant biological and biochemical effects (Darnerud 2003). PBDEs have a tendency to migrate through ecosystems and accumulate in organisms at the top of the food chain, such as birds of prey (Voorspoels et al. 2006; Wan et al. 2008). Experiments with PBDEs have shown that exposure results in immediate and long-term problems, such as cancer, developmental delays, and organ damage (Darnerud 2003);

M. A. Mora (✉) · C. Baxter
Department of Wildlife and Fisheries Sciences, Texas A&M University, College Station, TX 77843-2258, USA
e-mail: mmora@tamu.edu

J. L. Sericano
Geochemical and Environmental Research Group, Texas A&M University, College Station, TX 77845, USA

hormonal, developmental, enzymatic abnormalities; and, in extreme cases, death (Fernie et al. 2005). The manufacture and import of penta-BDE and octa-BDE formulations was prohibited in the United States since January 1, 2005 (http://www.epa.gov/oppt/pbde/pubs/qanda.htm, consulted 9/07/2011). In addition, deca-BDE was proposed to be phased out by December 2013 (http://www.epa.gov/opptintr/existingchemicals/pubs/actionplans/deccadbe.html, consulted 9/07/2011).

For many years, swallows have been used as indicators of environmental pollution in the United States (Custer 2011). Swallows are good indicators of local pollution because of their insectivorous diet and use of sediments for nest building (Sitzlar et al. 2009). Cliff and cave (*P. fulva*) swallows nest all along the Rio Grande/Rio Bravo on the Texas–Mexico border and are likely exposed to agricultural and industrial contaminants, including pesticides, organic solvents, metals, and hazardous wastes, which are often washed or intentionally dumped into the Rio Grande. The concentrations of DDE reported by Mora et al. (2006) were the highest reported for the last 20 years for the Rio Grande Basin. The objectives of this study were to determine if concentrations of persistent organic pollutants, particularly DDE and PCBs, had decreased in birds of the Rio Grande (using swallows as indicator species), particularly at El Paso and Laredo, two of the most highly populated areas in the basin. In addition, we report concentrations of PBDEs, which, to our knowledge, have not been reported in avian species from the Rio Grande Basin.

Materials and Methods

Study Areas

We selected two locations in the Rio Grande and one reference location 500 km northeast of Laredo. The El Paso sampling site was located in the eastern part of the city of El Paso by the Riverside canal and downstream from the outfall effluent of the Roberto Bustamante wastewater treatment plant (31° 39′ N, 106° 19′ W; Fig. 1). The second site was in the city of Laredo, in Manadas Creek, a region with heavy vehicle traffic and industrial activity (27° 34′ N, 99° 28′ W). The reference location was near Somerville, Texas (mostly agriculture; 30° 20′ N, 96° 28′ W; Fig 1) and approximately 500 km northeast of the Rio Grande.

Sample Collection

As part of a monitoring program we collected cave and cliff swallows during 1999 and 2005 in El Paso, during 2003 in Laredo, and during 2004 in Somerville. Some of the swallows were analyzed previously for contaminants (Mora et al. 2006; Sitzlar et al. 2009), but others were not; thus, we report only new data in this article. The birds were captured with mist nets dropped under a bridge where the swallows were nesting. The swallows were taken out of the net within 15 min of capture and then killed by cervical dislocation, wrapped separately in aluminum foil, placed in plastic bags, and stored in dry ice until taken to the laboratory where they were stored at −80°C until chemical analyses.

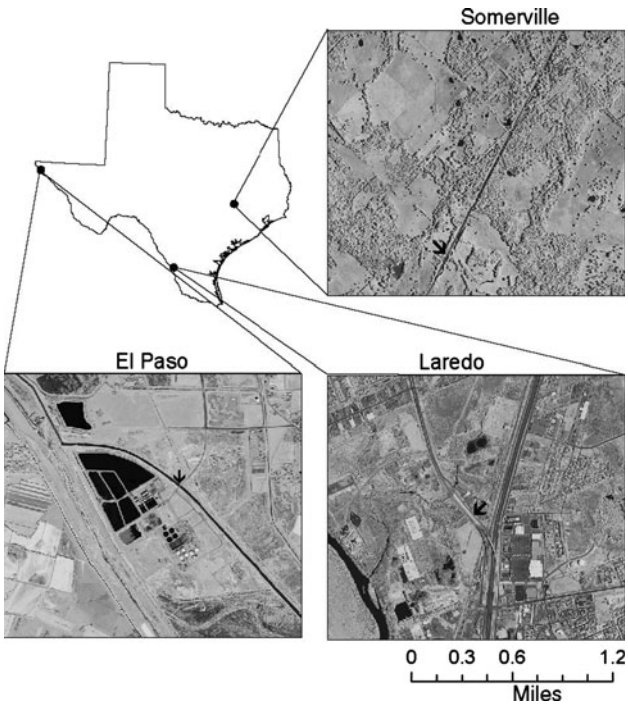

Fig. 1 Map of Texas showing the sampling locations along the Rio Grande and the reference location near Somerville

Chemical Analysis

All of the samples were analyzed for chlorinated pesticides and PCBs. A subset of the cliff swallow samples (eight from El Paso, eight from Laredo, and four from Somerville) also were analyzed for PBDEs. Carcasses were prepared by removing head, beak, wings, feet, and stomach contents. The remaining carcass was ground and homogenated with a Hobart meat grinder. The analytical procedures used for the extraction, fractionation, and cleanup of samples in the analyses of PBDEs, PCBs, and chlorinated pesticides in avian carcasses followed GERG (Geochemical and Environmental Research Group, Texas A&M University) standard operating procedures. Approximately 5 g of carcass homogenate was mixed with anhydrous Na_2SO_4 and extracted with methylene chloride using a homogenizer. 4,4′ dibromooctafluorobiphenyl, PCB 103,

and PCB 198 were added as internal standards. Extracts were fractionated by partially deactivated silica- and alumina-column chromatography and a 1:1 mixture of pentane and methylene chloride. Fractions were purified by high-performance liquid chromatography and concentrated to a volume of 1 ml in hexane for gas chromatographic analysis. PBDEs, PCBs, and chlorinated pesticides were analyzed by gas chromatography-mass spectrometry (6890 N GC system coupled to a 5975C Inert MSD) in selected ion mode. All of the sample extracts were injected in the splitless mode into a 30 m × 0.25 mm i.d. (0.25 μm film thickness) DB-5MS fused silica capillary column (J&W Scientific) and temperature programmed for optimum resolution of analytes. In all cases, the instrument was calibrated by injection of standard mixtures at four different concentrations before the analysis of the samples. 2,4,5,6-tetrachloro-*meta*-xylene was used as internal standard.

Method blanks, sample duplicates, and matrix spikes were processed with each sample preparation batch for quality control. The average percent recoveries of the internal standard used for quantitation (PCB 103) were 73.6 ± 8.2, 85.9 ± 13.3, and 71.1 ± 11.8 for PCBs, chlorinated hydrocarbons, and PBDEs, respectively. The average recoveries of analytes in the spiked matrices were 93.7 ± 8.3 for PBDEs and the relative percent differences between the matrix duplicates were <10% for all compounds. The most predominant PCB congeners in the mixture were 128, 153, 138, 146, 160, 170, 180, 187, and 196/203. The most commonly detected PBDE congeners were BDE 47, 99, 100, 118, 153, 154, and 183; however, BDE 28, 49, 66, 85, 138, and 155 also were detected in a few samples. The lowest limits of detection for p,p′-DDE, total PCBs, and total PBDEs were 1 ng/g ww.

Statistical Analysis

The contaminant data were log-transformed to satisfy the assumptions of normality and homogeneity of variance. There were only two locations (Laredo in 2003 and El Paso in 2005) with enough samples for statistical comparisons by sex. In both instances, Student *t* test indicated no significant differences in any of the contaminants detected greater than detection limits in 50% of the samples. Therefore, the data from both sexes were combined by location and year for further statistical analysis. We tested for differences in contaminant concentrations among species and locations by analysis of variance (PROC GLM ANOVA) with year as a covariate. The Tukey multiple comparisons procedure was further used to determine which means were significantly different. Concentrations of PBDE congeners were also compared among locations by analysis of variance (PROC GLM ANOVA) and the Tukey multiple comparisons procedure. All statistical comparisons were performed with SAS software (Cary, NC). The level of significance was set *a priori* at $P < 0.05$.

Results

There were no significant differences in body mass between males and females of the same species for the same location and year of collection. However, body mass of cave swallows collected during 2005 and cliff swallows collected in 1999 was significantly greater ($F_{4,31} = 7.13$, $P = 0.0003$) than that of cliff swallows collected also during 2005 and those collected in Laredo in 2003 (Table 1).

Concentrations of OC pesticides, including hexachlorobenze (HCB), heptachlor epoxide, oxychlordane, mirex,

Table 1 Geometric mean concentrations and ranges (ng/g ww) of OC pesticides and PCBs in cave and cliff swallows from El Paso, Laredo, and Somerville, TX

Bird	Location	Year	*n*	Body Mass (g)	HCB	Heptachlor Epoxide	Oxychlordane	Mirex	DDE	PCBs
Cave swallow	El Paso	2005	4	20.9 ± 1.3[a]	13[a]	7[b]	8[a]	3[b]	8.308[a]	114[b]
					(11–16)	(2–13)	(4–21)	(2–5)	(2185–26.218)	(41–261)
Cliff swallow	El Paso	1999	4	20.1 ± 2.6[a]	7[a]	50[a]	8[a]	21[ab]	8511[a]	698[a]
					(3–20)	(37–64)	(2–16)	(2–795)	(5475–13.256)	(576–957)
		2005	16	17.4 ± 1.2[b]	12[a]	11[b]	5[a]	39[a]	6182[a]	128[b]
					(3–51)	(1–55)	(2–15)	(6–333)	(3945–10.750)	(42–659)
	Laredo	2003	8	18.0 ± 1.2[b]	2[b]	51[a]	5[a]	52[a]	642[b]	193[b]
					(1–3)	(31–152)	(3–8)	(20–262)	(425–869)	(112–417)
	Somerville	2004	4	19.1 ± 1.2[ab]	1[b]	61[a]	5[a]	104[a]	987[b]	175[ab]
					(1–2)	(45–86)	(4–10)	(57–186)	(508–2158)	(139–205)

Within columns, data not sharing the same superscript lower-case letter are significantly different

Table 2 Geometric means and ranges (ng/g ww) for PBDEs in cliff swallows from El Paso and Laredo, TX, along the United States–Mexico border and a reference site

Location	Year	*n*	% Moisture	% Lipid	BDE 47	BDE 99	BDE 100	BDE 118	BDE 153	BDE 154	Total PBDEs	Total PBDEs (lw)
El Paso	1999	4	65.9 ± 2.8	7.6 ± 3.9	77[a] (48–123)	92[a] (55–131)	33[a] (21–51)	5[a] (0–17)	29[a] (20–42)	9[a] (5–13)	258[a] (154–385)	3.395
El Paso	2005	4	67.9 ± 1.6	5.2 ± 0.7	64[a] (42–87)	67[a] (39–102)	23[a] (17–32)	9[a] (5–12)	20[a] (16–25)	6[a] (4–8)	196[a] (128–272)	3.769
Laredo	2003	8	67.8 ± 0.9	6.3 ± 1.0	6[b] (3–11)	8[b] (4–13)	3[b] (1–6)	1[b] (0–2)	4[b] (2–9)	1[b] (0–1)	25[b] (13–44)	388
Somerville	2004	4	67.6 ± 2.4	6.6 ± 2.4	7[b] (6–8)	5[b] (4–7)	3[b] (2–3)	1[b] (0–1)	2[c] (1–2)	1[b] (0–1)	18[b] (15-21)	280

Total PBDE values are also provided on a lipid-weight basis for comparisons with the literature. Within columns, data not sharing the same superscript lowercase letter are significantly different

lw Lipid weight

and p,p′-DDE, were detected in >50% of the samples at levels greater than the detection limits (Table 1). However, except for DDE, all other OC pesticide concentrations were relatively low and near detection limits. HCB concentrations were significantly greater ($F_{4,31} = 14.2$, $P < 0.0001$) in swallows from El Paso (both species, both years) than in those from Laredo in the Rio Grande and the reference site in Somerville. Mean heptachlor epoxide concentrations were significantly greater ($F_{4,31} = 8.5$, $P < 0.0001$) in swallows from Laredo, Somerville, and El Paso (1999) than those collected in 2005 at El Paso. Oxychlordane levels were the lowest of all OCs and were not different among locations. Mirex mean concentrations were significantly greater in cliff swallows from Somerville, Laredo, and El Paso (2005) ($F_{4,31} = 4.4$, $P = 0.0064$), than in cave swallows collected in El Paso in 2005. Geometric mean DDE concentrations were significantly greater ($F_{4,31} = 47.8$, $P < 0.0001$) in all swallows collected at El Paso in 1999 and 2005 than in swallows from Laredo and Somerville. Geometric mean DDE concentrations in cliff swallows collected in 2005 at El Paso were approximately 30% lower than those observed in 1999; however, DDE residues in cave swallows were similar in 2005 to those observed in all swallows in 1999. Mean total PCBs were significantly greater in cliff swallows from El Paso collected in 1999 ($F_{4,31} = 5.0$, $P = 0.0033$) than in swallows collected in 2005 in the same location and swallows from Laredo (Table 1).

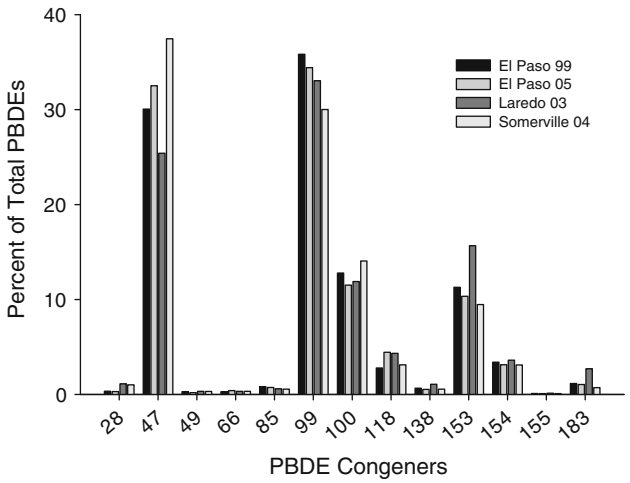

Fig. 2 Percent contribution of PBDE congeners to total PBDEs

The most predominant PBDE congeners in swallow carcasses were 47, 99, 100, 118, 153, and 154. Other BDEs, 28, 49, 66, 85, 138, and 155, were detected at concentrations near detection limits, i.e., <2 ng/g ww. Concentrations of BDE congeners 47, 99, 100, 118, 153, and 154 were significantly greater in swallows from El Paso collected in 1999 and 2005 ($F_{3,16} = 8.8$ to 61.7, $P < 0.0001$) than in swallows from Laredo and Somerville (Table 2). Concentrations of PBDES were mostly not significantly different between the Laredo location and Somerville, except for PBDE 153, which was significantly greater in swallows from Laredo than Somerville ($F_{3,16} = 51.1$, $P < 0.0001$; Table 2). The major BDE congeners contributing to total PBDEs were, in order of significance, PBDE 99, 47, 100, 153, 118, and 154. BDE congeners 47 and 99 comprised approximately 60% of total PBDEs (Fig. 2).

Discussion

Results from this study indicated that DDE residues decreased slightly in cliff swallows from El Paso; however, concentrations in cave swallows remained increased and close to values reported in earlier studies. Mora and

Wainwright (1998) reviewed DDE concentrations reported in water, sediments, and biota of the Rio Grande from 1965 to 1995. Within this period, the greatest mean DDE concentrations were reported in bird carcasses of western kingbirds (*Tyrannus verticalis*: 60,900 ng/g ww) collected in 1982 in Reeves and Hudspeth counties east of El Paso (Hunt et al. 1986). Applegate (1970) also reported high DDE residues in breast muscle of cliff swallows (10,800 ng/g ww) collected in the same counties during 1968. In addition, Mora et al. (2006) reported mean DDE concentrations of 12,400 ng/g ww in swallow carcasses during 1999 and 2000 at El Paso. However, four samples from the same batch collected in 1999 had lower mean values and were not different from those in 2005. The mean DDE values observed in swallows from El Paso in 2005 were approximately five times lower than the values reported in kingbirds from Reeves and Hudspeth counties. The DDE values reported in swallows from El Paso during 1999 to 2000 were among the highest reported in passerine birds from Texas in the last 20 years (Mora et al. 2006). The most recent data collected in 2005 suggest that DDE levels in swallows from El Paso are still high and are of potential concern for endrocrine-disrupting effects on birds. Cliff swallows collected in 1999 to 2000 at El Paso had increased spleen mass, lower plasma thyroxine (T_4) concentrations, and greater half-peak coefficient of variation of DNA (measured by flow cytometry) than swallows from other locations in the Rio Grande (Mora et al. 2006). We hypothesized that these effects could be associated with high concentrations of DDE; however, swallows from El Paso also had high concentrations of copper and selenium (Mora et al. 2006).

Swallows are known to be prey of different raptor species (Enderson et al. 1982; Mora et al. 2007); thus, they could be a significant source of contaminants for their predators. Potential avian prey (insects and passerine birds) of the northern aplomado falcon (*Falco femoralis septentrionalis*) collected in the Laguna Atascosa National Wildlife Refuge during 1994 had low levels of DDE (mean range 10 to 250 ng/g ww) (Mora et al. 1997). Similarly, mean DDE residues in potential avian prey of the aplomado falcon in Matagorda Bay, TX, were rather low and ranged from 0.4 to 24 ng/g ww during 2004 (Mora et al. 2008). Northern rough-winged swallows (*Stelgidopteryx serripennis*) from the Big Bend region in 1997 had mean DDE concentrations in carcass of 5140 ng/g ww (Mora et al. 2002). Repeated sampling during 2001 also provided low DDE concentrations in potential prey (mostly passerine birds, mean 2 to 216 ng/g ww) of the peregrine falcon in Big Bend National Park (Mora et al. 2007).

The concentrations of DDE in swallows from El Paso in 2000 and 2005 were among the highest observed in the last 20 years in swallows from along the United States–Mexico border. Therefore, it is likely that birds from El Paso are at a greater risk of impacts from DDE given that this region has shown the highest concentrations in birds since the late 1960 s (Mora and Wainwright 1998). Raptors that feed on prey with DDE concentrations between 1 and 3 μg/g ww may produce thin-shelled eggs (Enderson et al. 1982; Mendenhall et al. 1983). Therefore, swallows and likely other potential avian prey in the El Paso region should be of concern for reproductive effects on raptor species. The potential sources of DDE are most likely heavy use of DDT in the past, although dicofol impurities and potential illegal recent use of DDT cannot be ignored. Concentrations of other OC pesticides, for the most part, were rather low and of no concern for impacts on birds. However, it is important to notice that mirex, which was heavily used in agriculture in the past, is still detected in birds in Texas, particularly in agricultural environments.

PCBs were notably low in swallows from all locations, including the Ciudad Juarez-El Paso region, which is highly industrialized. Mean total PCBs reported during the last 15 years in eggs and carcasses of birds from the Rio Grande/Rio Bravo ranged from 68 to 1265 ng/g ww (Mora 1996a, b; Wainwright et al. 2001; Mora et al. 2006). The results from the present and former studies confirm the presence of low concentrations of PCBs in avian wildlife of the Rio Grande/Rio Bravo region relative to concentrations observed in more industrialized regions around the Great Lakes and the northeastern United States (Rice et al. 2003).

To our knowledge, this is one of the first studies to provide PBDE data in birds from the Rio Grande Basin and in passerine birds from the United States. Not surprisingly, we found that birds from El Paso had the highest concentrations of PBDEs both in 1999 and 2005. Swallows from El Paso were nesting downstream from areas with significant industrial development: the city of El Paso and Ciudad Juarez. It is conceivable that the swallows accumulated some PBDEs from the plant's discharge effluent and from water of the Rio Grande. It is known that heavier PBDE congeners in wastewater accumulate in the particulate runoff from treatment plants (Hale et al. 2006). Water flowing through or near urban areas is likely to carry greater loads of PBDEs through direct dumping and runoff from contaminated land. The PBDE data in swallows from El Paso were approximately 8 to 13 times greater than those from Laredo and Somerville and approximately 30 times greater than those reported in eggs of passerine birds (great tits [*Parus major*]) from 14 European countries (Van den Steen et al. 2009). The PBDE congener patterns, however, were similar in swallows to those reported in great tits from Belgium and Europe (Van den Steen et al. 2008, 2009; Dauwe et al. 2009; Chen and Hale 2010).

Analysis of the relative concentrations of congeners at all sites suggests the industrially produced penta-mixture as

a potential source. The composition of the penta-mixture is approximately 37% BDE-47, 35% BDE-99, 6.8% BDE-100, 3.9% BDE-153, 2.5% BDE-154, and 1.6% BDE-85 (along with traces of other BDEs) (Sjodin et al. 1998). The relative percentages found in our samples are close to the previously mentioned pattern, although discrepancies can be explained by the tendency of some PBDE congeners, such as BDE-209, to break down into other congeners (Van den Steen et al. 2007; McKernan et al. 2010). In addition, some other mixtures that may be present contain the same congeners in different percentages. Our findings show that all sample sites have some sort of industrial (likely penta-mixture) PBDE contamination, with El Paso having the highest concentrations. Noticeable, the manufacture and import of penta-BDE and octa-BDE formulations has been prohibited in the United States since 2005 (http://www.epa.gov/oppt/pbde/pubs/qanda.htm, consulted 9/07/2011). In addition, a commitment to phase out deca-BDE in the United States by 2013 is currently in place (http://www.epa.gov/opptintr/existingchemicals/pubs/actionplans/deccadbe.html, consulted 9/07/2011).

Concentrations of PBDEs in swallows also were relatively lower than those reported in greater trophic–level birds from the northeastern United States and other industrialized countries (Chen et al. 2008; Johansson 2009; Chen and Hale 2010; Chen et al. 2010). However, concentrations of PBDEs in swallows from El Paso were greater than those observed in aplomado falcon eggs from two regions in Mexico (Mora et al. 2011). The swallows were collected from locations much closer to industrial sources than the falcons. Overall, the concentrations of PBDEs in swallows from El Paso were much lower than those associated with negative effects in American kestrels (*Falco sparverius*) and other birds (Fernie et al. 2006; McKernan et al. 2009). In addition, a recent study suggested that passerine birds may not be as sensitive to the effects of PBDEs as other birds. Exposure to a penta-PBDE mixture (1740 μg/kg body weight) for 6 months did not result in any endocrine-disruptor, hematological, or biochemical alterations in European starlings (*Sturnus vulgaris*) (Van den Steen et al. 2010).

Conclusion

The results from this study indicated that swallows in some portions of the Rio Grande were still being exposed to DDE at concentrations that could have adverse effects on their health and reproduction or on avian predators that make these species part of their diet. In addition, swallows were exposed to concentrations of PBDEs that could further exacerbate problems in predator species feeding on swallows and other passerine birds. Other OCs, including PCBs, were either not detected or were present at low concentrations.

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