EXPOSURE OF CALIFORNIA PEREGRINE FALCON (*FALCO PEREGRINUS*) TO BROMINATED FLAME RETARDANTS (PBDES AND NEW ALTERNATIVE BFRS) AND PCBS: DIFFERENT PROFILES OF PBDES, PREY, AND ISOTOPE PATTERNS BETWEEN COASTAL AND BIG CITY NESTING BIRDS

Park J-S[1], Holden A[1], Chu V[1], Newsome S D[2], Henry R W[3], Wang M[1], Kim M[1], Rhee A[1], Patel P[1], Fong A[1], Linthicum J[3], Hooper K[1]

[1]California Department of Toxic Substances Control, California Environmental Protection Agency, Berkeley, CA 94710, USA; [2]Carnegie Institution for Science, Washington, DC 20015, USA; [3]Long Marine Laboratory, University of California-Santa Cruz, CA 95060, USA; [4]Division of Biostatistics, School of Public Health, University of California-Berkeley, Berkeley, CA, 94707, USA.

Abstracts

California peregrine falcon eggs (n = 90 eggs from 52 birds, 38 nest sites, collected 1986-2007) showed among highest levels of polybrominated diphenyl ethers (PBDEs) ranging 0.08-53.1 ug/g lipid with median 5.11. Over the 21-year period during which peregrine falcon eggs were collected, ∑PBDE levels in the California peregrine falcon population significantly doubled each decade ($r=0.55$, $p<0.01$), whereas PCB levels decreased slightly. For PBDEs, urban eggs had markedly different patterns from coastal eggs: BDE-209 and the higher brominated PBDEs (hexa-nona) were dominant congeners in urban eggs, while BDE-47 and -99 were dominant in coastal eggs. The patterns of peregrine prey and stable isotope values of eggs ($\delta^{13}C$, δD, $\delta^{15}N$) suggest that the differences in PBDE profiles seen in urban vs coastal eggs may arise from differences in peregrine prey and in turn from differences in diet of the prey birds. Levels of new BFR replacements for PBDEs were examined in a subset of eggs (n=19) that had the highest PBDE levels and/or were most recently collected (1999-2007). HBB and BTBPE were detected in most eggs. This indicates that the classic and new alternative BFRs are likely to escape from consumer products and become environmental contaminants, bioaccumulating in the aquatic and terrestrial food web.

Introduction

Polybrominated diphenyl ethers (PBDEs) have been widely used as a brominated flame retardant (BFR) for synthetic textiles in rugs, draperies, upholstery, and also in polyurethane foam and thermo-plastics which are found in a variety of consumer products and electronics. Presently, PBDEs are widely dispersed in the abiotic and biotic environments[1]. Penta and octa-BDE mixtures are now banned in the European Union (EU) and U.S. Continued production and use of deca-BDE, the major commercial PBDE formulation with 65-70% total production, in North America (mostly in the U.S.) poses a threat to wildlife and public health. This is because BDE-209, the major component (>97%) of deca-BDE[2], bioaccumulates in wildlife especially in the terrestrial food web[3-6]. BDE-209 also breaks down either via photolysis[7,8] or enzymatic transformation[9-11] to become lower brominated PBDEs, which are known to be more bioavailable, persistent, and toxicologically active than the parent compound. PBDE levels in California residents and aquatic wildlife were found to be among the highest in the world, with tetra-, penta-, and hexa-BDEs (BDE-47, -99, -100, -153, and -154) as the major congeners[12-16].

As a part of our Wildlife Early Warning System (WEWS), PBDE and PCB levels were measured in peregrine falcon (*Falco peregrinus*) eggs. Peregrine falcons are a useful sentinel species for monitoring environmental organic contaminants because, as bird-eating raptors, these top consumers occupy a high trophic niche in both aquatic/marine and terrestrial food chains. Peregrine falcons consume sedentary and migratory prey from both aquatic (waders and ducks) and terrestrial (pigeons, starlings and thrushes) environments. Peregrines can serve as indicators of regional differences in chemical pollution because they have: strong pair-bonds with a single mate; long life spans of 12-15 years; stable nest locations with long-term residency (7-9 years); and yearly clutches of 3-5 eggs[19]. Also, new other BFRs, introduced as replacements for PBDEs, such as tetrabromoethylcyclohexane (TBECH), pentabromoethylbenzene (PBEB), hexabromobenzene (HBB), pentabromotoluene (PBT), hexabromocyclododecane (HBCD), tetrabromobenzoate (TBB), bis(2-ethylhexyl) tetrabromophthalate (TBPH), and decabromodiphenylethane (DBDPE), are of concern due to their potential for bioaccumulation[17,18]. Scant data on environmental

presence of these compounds exists.

In this study, we measured levels of PCBs and PBDEs in peregrine falcon eggs that were collected and archived in California during 1986-2007, and compared levels and patterns of congeners in these eggs collected in the different California ecological regions (e.g., Big City and Coastal). We next examined the possible sources and pathways of exposure to PBDEs by comparing patterns of PBDE congeners with those of prey and carbon ($\delta^{13}C$), nitrogen, ($\delta^{15}N$), and hydrogen (δD) stable isotope values for each peregrine egg. In addition, the new BFRs were measured in a subset of eggs with the highest PBDE levels that were most recently collected (1999-2007).

Materials and Methods

Peregrine falcon eggs were collected and archived in California over a 21 year period (1986-2007) as part of the Peregrine Recovery Program in response to DDT-induced egg-shell thinning and population collapse in the 1960s[19]. Eggs were received frozen and were stored at -20°C until analysis. Sample analysis is described elsewhere[9]. In summary, lyophilized samples were extracted in Accelerated Solvent Extraction system (ASE200, Dionex, Sunnyvale, CA) and cleaned up using a glass column packed with acidic silica gel. The eluates were fractionated on an Envirogel GPC Cleanup Column (Waters, Milford, MA). PBDEs and new BFRs were measured by using isotope dilution/HRGC/MS (ThermoFinngan MAT95, Bremen, Germany) and external calibration/GC-NCI/MS (Varian 1200L Varian Inc., Walnut Creek, CA), respectively. New BFR congeners were also confirmed in HRGC/MS. PBDEs and new BFRs were analyzed on DB-5MS column (15 m × 0.25 mm i.d., 0.1 μm film thickness, J&W Scientific, USA) and GC temperature program; 175 °C for 2 min., ramped to 280 °C at 5.0 °C/min. and to 310 °C at 7.0 °C/min., and at 310 °C for 5 min. The accuracy and precision of analysis were measured by using duplicate samples, matrix spikes (corn oil), and in-house reference materials within 20%.

Peregrine prey species (n=185) were identified by their remains at 38 nest sites intermittently over a 14-year period (1974-1998). Prey were classified by diet and grouped into 15 categories; prey are expressed as the weight percentage consumed of their particular prey group. Peregrine eggs were lipid extracted using Accelerated Solvent Extraction system (ASE200, Dionex, Sunnyvale, CA). Carbon ($\delta^{13}C$), nitrogen ($\delta^{15}N$) and hydrogen (δD) isotope values were determined for the remaining solid fraction using a Carlo-Erba EA (NC 2500) for C and N or Finnegan TC/EA for H interfaced with a Thermo-Finnegan Delta Plus XL mass spectrometer (Carnegie Institution of Washington). Isotopic results are expressed as δ values and the standards are Vienna-Pee Dee Belemnite (V-PDB) limestone for carbon, atmospheric N_2 for nitrogen, and Vienna-Standard Mean Ocean Water (V-SMOW) for hydrogen. The units are expressed as parts per thousand, or per mil (‰). The within-run standard deviation of an acetalinide standard was ≤0.2‰ for $\delta^{13}C$ and $\delta^{15}N$.

The concentrations of PCBs and PBDEs were log-transformed to make the data more symmetrical. Generalized Estimating Equations (GEEs) were used to explore temporal trends in PBDE and PCB levels (n=90 eggs). All statistical analyses were performed using R. The PBDE levels of the time-series peregrine falcon eggs (multiple eggs from the same nest and the same bird) were averaged and treated as a single data point for the data summary.

Results and Discussion

High levels of PBDEs were measured in peregrine falcon eggs from California (n=90 eggs from 52 birds, 38 nest sites, collected 1986-2007, ΣPBDEs range 0.08-53.1 μg/g lipid, median 5.11). Significantly, over the 21 year period during which PFE samples were collected, ∑PBDE levels in the California peregrine falcon population doubled each decade ($r=0.55$, $p<0.01$), whereas PCB levels appeared to decrease slightly. PBDE levels were highest in eggs from urban nests, whereas PCBs were highest in eggs from coastal regions. For PBDEs, urban eggs had markedly different patterns from coastal eggs: BDE-209 and the higher brominated PBDEs (hexa-nona) were dominant congeners in urban eggs, while BDE-47 and -99 were dominant in coastal eggs (Figure 1). In contrast, PCB patterns showed no differences at urban vs coastal eggs. A diet may explain the urban PBDE pattern, as the pattern in urban pigeons and peregrine falcons were similar, with high proportions of BDE-209 and the higher-brominated PBDEs. Also, our prey data (bones from peregrine nests) showed urban peregrines having a higher proportion (2-fold) of garbage eating birds (e.g., 'introduced feral' pigeons, mourning doves, starlings) in their diet than coastal peregrines. In addition, $\delta^{13}C$, $\delta^{15}N$, and δD isotopic composition showed the distinct pattern between urban and coastal peregrines; respective values were -15.9±0.3 and -20.1±0.3 ($p<0.001$), 10.3±0.1 and 11.5±0.3 ($p<0.001$), and -50.4±1.4 and -28.0±2.1 ($p<0.001$). Those prey and isotope data suggest that the different

PBDE profiles of peregrine eggs between two ecological regions arise from the prey eating different food sources. A subset of eggs (n=19) was analyzed for the new replacement BFRs such as TBECH, PBEB, HBB, PBT, HBCD, BTBPE, TBB, TBPH, and DBDPE. These eggs had most recent collection years (1999-2007) and the highest PBDE levels. Some of the alternative BFRs, including HBB and BTBPE, were detected, but at much lower levels than PBDEs, respectively. However, results from recovery studies in chicken eggs of new BFRs indicated that the conventional extraction/cleanup method for the PBDE analysis used in this study is not suitable for some of these new alternative BFRs. For example, a major portion of TBPH was lost during the acidic silica column clean up and possibly more was lost in other steps (e.g., evaporation, GPC). Thus further optimization for the sample extraction/clean up method was need for the new alternative BFR analysis.

We report that the classic and new alternative BFRs are likely to escape from consumer products and become environmental contaminants, which bioaccumuate in the aquatic/terrestrial food web, and that the differences in PBDE profiles seen in urban vs coastal eggs may arise from differences in peregrine prey and in turn, from differences in diet of the prey birds. For the new BFRs, we continue to develop data sets on the sources and pathways of exposure in support of measures to characterize and reduce environmental contamination.

Acknowledgements and Disclaimer
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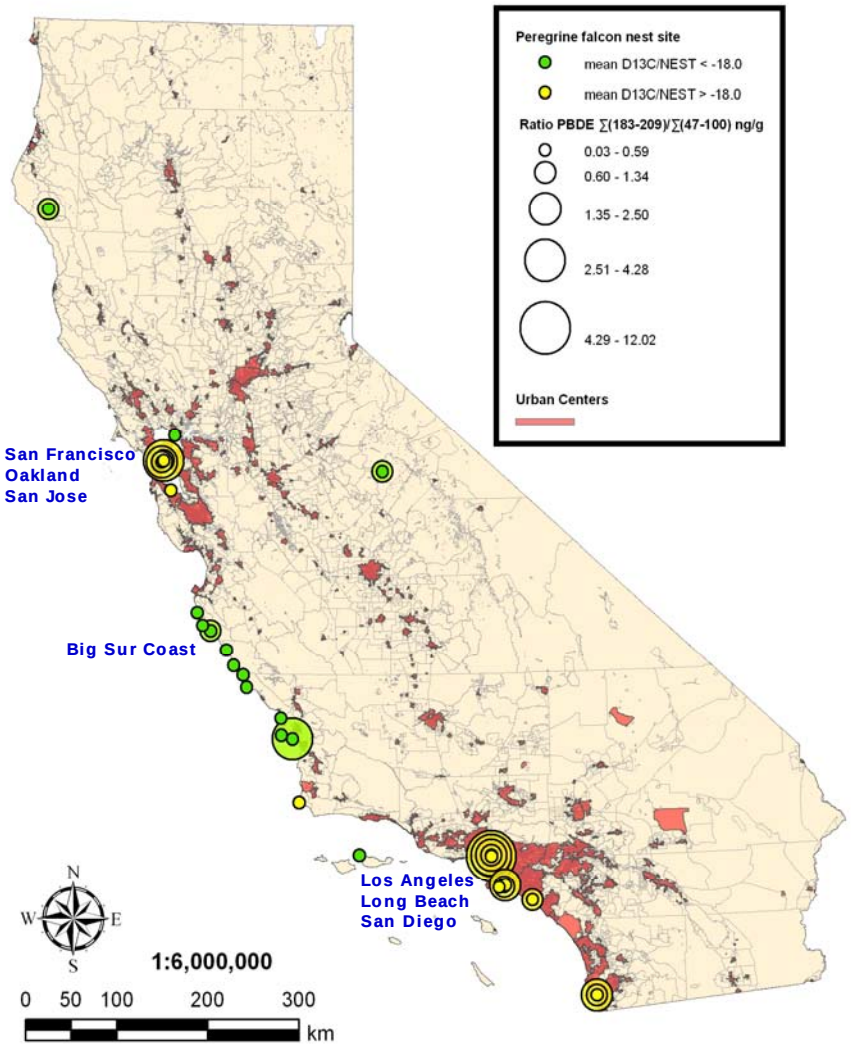

Figure 1. Profiles of PBDE pattern (high to low brominated PBDEs) and stable isotope ratios ($\delta^{13}C$) in California peregrine falcon eggs in relation to the ecological regions**.**