DIOXIN, PCB AND PBDE EXPOSURE IN GREY HERON (ardea cinerea)

Alwyn Fernandes[1], Helen Thompson[1], Kyle Dsilva[1], Shaun White[1], Martin Rose[1]

[1]Central Science Laboratory, York

Introduction

Grey herons (*Ardea cinerea*) are large, aquatic birds geographically distributed throughout Africa, Asia and Europe. They generally breed in colonies in woodland habitats in close proximity to water and are recognised as higher level predators with a varied range of prey from fish, small amphibians and molluscs to insects and even small rodents and birds (1). Given this variety in diet and the average lifespan of around two decades, there is considerable potential within the species for bio-accumulation of persistent environmental contaminants. The fact that established colonies do not tend to migrate coupled with the diet and bio-accumulation potential, makes herons a good indicator of local contamination by common pollutants.

There have not been many reports of contaminants in grey heron, but considerably more data exists on a related species – the great blue heron (ardea herodias) native to the north American continent. Here studies have established the validity of using these birds as indicators of local contamination, and levels of common contaminants such as dioxins and PCBs have been identified and measured at a number of locations in Canada and the United States (2,3). Studies have shown elevated levels of environmental contaminants in heron tissue and the related adverse effects on the colonies. In particular eggs show embryo deformities that have been linked to dioxin–like compounds and positive correlations between nest failure and TCDD concentrations have also been demonstrated (3,7).

In the United Kingdom recent investigations have detected elevated levels of mortality and bone disease in grey herons at an established colony in Nottinghamshire along the course of the river Trent (4). The causes of mortality are unclear but deformities recorded in the other birds include multiple fracture of the tarsus, tibia and metacarpal bones (4). These findings have prompted a pilot study into assessing the level of environmental contaminants in the tissue and eggs of these birds. Two classes of contaminants have the potential to cause the deformities observed in the birds – heavy metals such as selenium, cadmium, arsenic mercury and lead, and halogenated organic contaminants such as dioxins, and PCBs (5,6,7). This paper discusses levels of these contaminants (8) in the samples of eggs taken from the colony. Additional samples of eggs were also collected from a colony in Hertfordshire and from a site in the north of the country. The discussion will be limited to the halogenated organic contaminants as the levels of heavy metals were similar in all sites and were generally at or above background levels. Given the increased utilisation of

brominated flame retardant chemicals over the last decade and the similarities in structure and environmental persistence of some of these compounds to the dioxins and PCBs, polybrominated diphenylethers (PBDEs) were also measured in the samples.

Methodology

Samples of adipose tissue (fat) were collected in 2002 from nestlings and dead adult birds at the Nottinghamshire site (Site A). This site is located on a flood plain on the eastern bank of the River Trent, which flows through densely populated and industrialised areas, from the west midlands to the Humber estuary and into the North Sea. Samples of eggs taken from individual nests were collected under license, from this site and from nests at another site in the North of England (Site B). This site is located within rural woodlands in close proximity to urban centres and heavy industry. Additionally, egg samples were taken from a colony in Hertfordshire (Site C) that is semi-urban but without any heavy industry in close proximity.

Adipose tissue samples were pooled based on the state of the nestlings (dead, deformed, undeformed) and dead adults, and analysed as received. The egg samples were pooled based on location, homogenised and freeze-dried prior to analysis. Accredited and robustly validated methods used for the extraction and analysis of the samples have been reported previously (9). In brief, ^{13} Carbon labelled internal standardisation was used for all analytes coupled with either GC-HRMS or GC–LRMS for the ortho substituted PCBs. Frequent measurement of reagent blanks and reference materials (RMs and CRMs) form part of the analytical quality control. Data quality was ensured by continuous successful participation in international inter-calibration exercises (10,11,12) on the measurement of dioxins and PCBs in food and animal feed.

Results and Discussion

The results for the analyses of dioxins, PCBs and PBDEs are summarised in Table 1. Dioxin data is represented by WHO-TEQ values, as are non-ortho substituted PCBs and ortho substituted PCBs. For comparability with other data the sum of the ICES PCB congeners (PCBs 28, 52, 101, 118, 138, 153 and 180) is also reported, as are the sum of 16 PBDE congeners (BDE 17, 28, 47, 49, 66, 71, 77, 85, 99, 100, 119, 126, 138, 153, 154 and 183). Data for the eggs is reported as the average of pooled samples for the sites A (5 pools) and C (2 pools) and for the single pooled sample from Site B. Adipose tissue (fat) samples were all taken at site A from nestlings or adult birds.

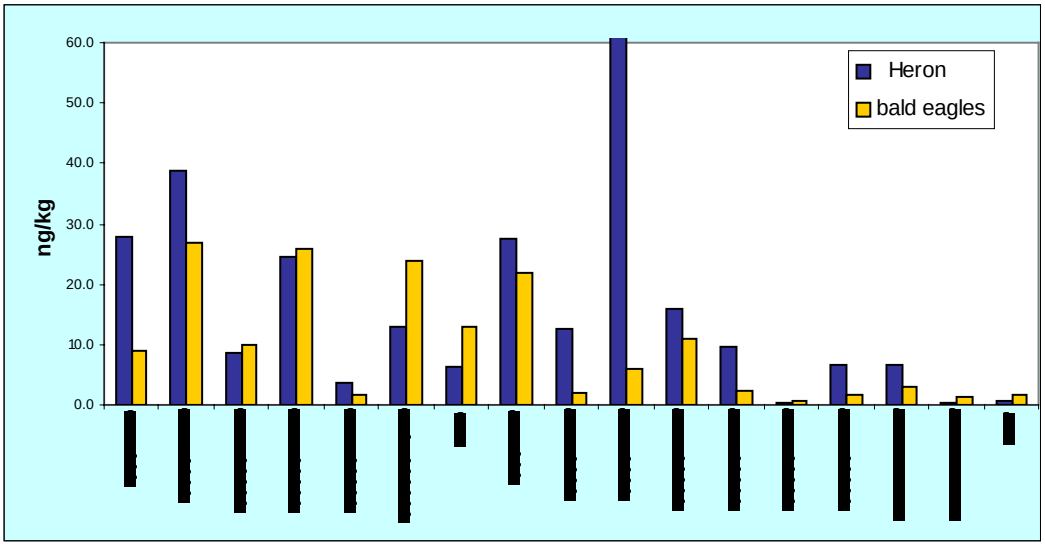
Table 1: Summary of contaminant levels (fat weight basis) in Heron eggs and Adipose tissue (fat).

Location=>	Site A (eggs)	Site B (eggs)	Site C (eggs)	Site A Undeformed (fat)	Site A Dead (fat)	Site A Deformed (fat)	Site A Adult (fat)
WHO-TEQs in ng/kg							
Dioxin	76	651	52	55	102	156	196
non ortho-PCB	281	992	191	137	217	304	341
ortho-PCB	152	590	88	69	51	159	118
Σ WHO TEQs	509	2233	331	260	370	619	655
Σ ICES 7PCBs, µg/kg	5769	14381	2091	1964	2004	5291	2718
Total PBDE, µg /kg	966	4118	376	2162	1361	2270	2906

The WHO-TEQ profiles for both eggs and adipose tissue samples are broadly similar, with non-ortho PCBs providing the largest contribution to the ΣWHO-TEQ. In general ortho PCBs contribute more than the dioxins except for the adipose tissue samples from dead nestlings and adult birds. PCBs therefore predominate the ΣWHO-TEQ as observed by the good correlation (correlation co-efficient – 0.96) between ΣWHO-TEQ and Σ ICES 7PCBs**.** No such correlation is observed between the PBDEs and the other pollutant groups. PCBs and dioxins have been declining in the environment and in other matrices such as human diet (13) and fish oils (14), but a slower decline has been observed for PCBs (13), particularly in matrices such as fish, some species of which form part of the herons diet. There is debate on the trends in PBDE concentrations, with levels in some countries declining, but showing increases in others.

The most striking feature of the data in Table 1 is the results for the egg sample from site B. This sample shows the highest levels of all contaminants with dioxin WHO-TEQ and PBDE concentrations occurring in excess of an order of magnitude higher than site C. The corresponding figure for PCB WHO-TEQ is more than 5 times higher. The levels observed for the eggs from site A are also higher at approximately 1.5 times the ΣWHO-TEQ values at site C, but more than 2.5 times the PBDE concentrations. The higher levels of WHO-TEQ observed in the adipose tissue samples from site A, for dead and deformed birds compared to those which had no deformities may indicate a relationship between the levels of WHO-TEQ and the deformities. Nestlings found dead in particular, showed WHO-TEQ levels approaching those of adult birds. Σ ICES 7PCBs were also highest in the fat from the deformed birds, but no similar correlation was observed for the PBDEs.

Figure 1: Dioxin profile of adipose tissue in bald eagles (17) and heron

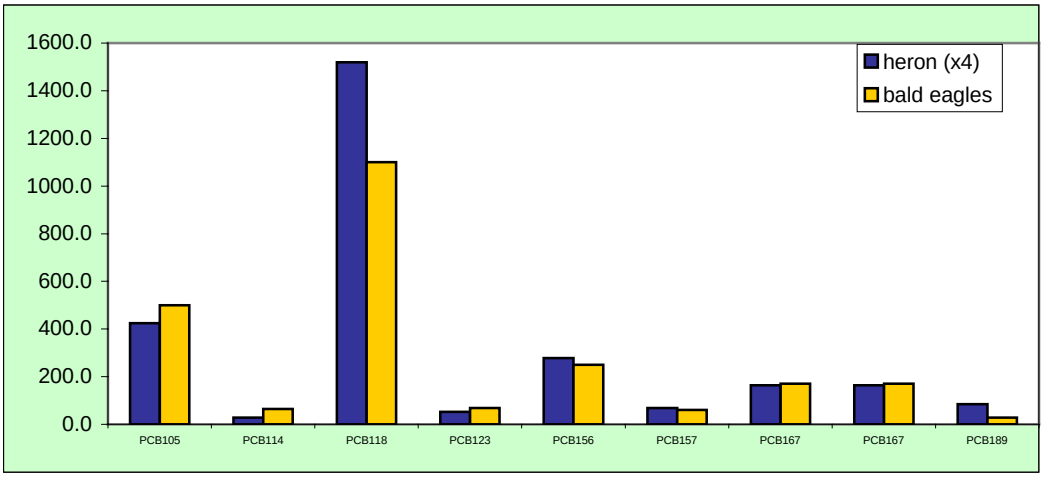


As mentioned earlier the site in the North of England (site B) is situated in close proximity to heavy industry, and densely populated areas. The site in Nottinghamshire (site A) is located within the flood plains of the river Trent that flows through a densely populated and industrialised course. There are also a number of coal-fired power stations in the area and historically, fly-ash from these was deposited in lagoons near the site. Both sites therefore have strong potential sources of contaminants in close proximity. Additionally, in an earlier study on the effects of contaminant deposition on floodplains arising from incidents of flooding, relatively high levels of PCBs and dioxins were observed in the milk of cows grazing on upstream floodplains on the river Trent (15).

There is very little species-specific data reported in the literature in order to make comparisons, particularly with WHO-TEQ contributing congener data. However data does exist for other fish eating avian species (16-18) and the profiles observed for bald eagle fat (17) collected from birds in Michigan, USA, bear a number of similarities to heron fat as shown in figure 1. PCDDs dominate to a larger extent than PCDFs in both species except for 23478 PCDF in the heron which makes the largest contribution to the WHO-TEQ

A greater degree of similarity is observed when WHO-TEQ contributing ortho PCB congeners are compared (Figure 2). Concentrations recorded for bald eagles are higher (the heron data in Figure 2 is scaled by a factor of 4), but in both species the largest WHO-TEQ contributions are observed for PCB 118 and PCB 156. The similarity is all the more remarkable given the differences in species, habitat, and geographical location, and may suggest a dominance of aquatic prey in the heron diet or a similar mechanism of metabolism in the two species. A full profile of all the contaminants measured here for herons is given in figure 3.

Figure 2 Ortho substituted PCB profile of adipose tissue in bald eagles (17) and heron (scaled).



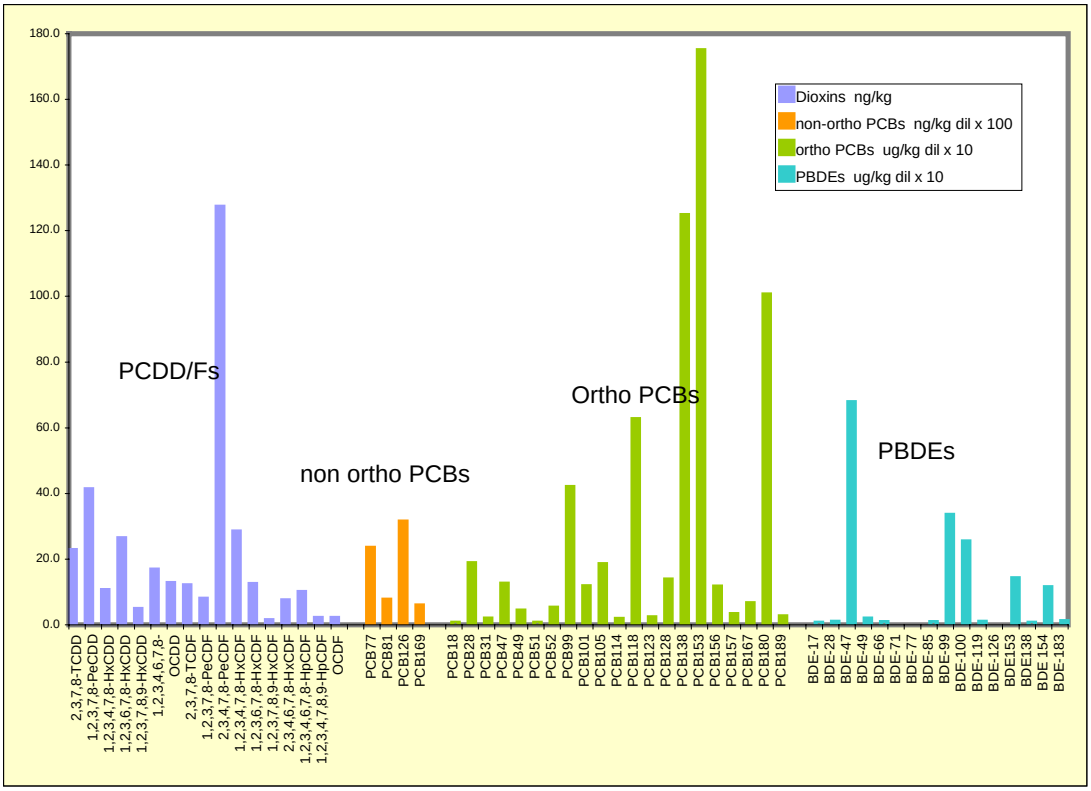
The levels of WHO-TEQ and Σ ICES 7PCBs observed in some of the individual egg pools are similar to those reported (Hoffman) to show reproductive effects such as embryo mortality in other avian species such as pheasants, chickens and bluebirds. However, given the dearth of similar data on ardea cinera, in the literature, it is advisable for any extrapolation of this data to these or other similar effects to consider the following points:

- The number of observations reported here are small – further structured measurements at all sites, especially site B, are essential. It is important to identify control sites far removed from potential dioxin/PCB sources and determine contaminant levels in birds and eggs at these sites in order to establish a baseline level.
- The measurements so far refer to WHO-TEQs arising from dioxins and PCBs only. Dioxin-like toxicity can be attributed to other chemical contaminants such as PCNs, and brominated and mixed bromo-chloro dioxins and furans; thus the recorded WHO-TEQs may be partial.
- WHO-TEFs used here to calculate dioxin-like toxicity are derived from biological effects and observations on mammals (19). Avian response and susceptibility may well be different as observed elsewhere (18). Additionally, it is widely accepted that dioxin-like toxicity varies considerably among and sometimes even within species, eg, differences in AhR proteins causes the DBA mouse to be much more resistant to the toxic effects of dioxins than the C57Bl/6 mouse (20).

Notwithstanding these arguments, the data presented is certainly indicative of a relationship between the levels of WHO-TEQ, Σ ICES 7PCBs and the deformities observed at site A. Additional work has already been commissioned for further investigations to record more data points and clarify this relationship. Such work may also facilitate a fuller understanding of the exposure pathways. In particular, as ingestion of food is likely to be the main route of exposure, accurate data on contaminant levels in (a), the species that the herons prey on and (b), local

environmental matrices (sediment, biota) may provide confirmation of the heron's role as indicator of local contamination.

Figure 3 Profile of chlorinated and brominated contaminants in heron (ardea cinera)



Conclusions

The individual WHO-TEQ profiles for all eggs and adipose tissue samples are broadly similar and show similarities to other fish eating avian species.

Levels of contaminants are higher at sites that have proximity to urban and industrial areas, which is consistent with the observation that herons are good indicators of local contamination.

The levels of dioxin and PCB WHO-TEQ recorded in dead and deformed nestling tissue at the Nottinghamshire site, compared to unaffected nestlings are indicative of a relationship between the levels of these contaminants and the deformities observed in the nestlings

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