

Polychlorinated biphenyls in raptor and owl eggs in the Czech Republic

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ABSTRACT: In 2001, raptor and owl eggs were collected for the purpose of detection of contamination by polychlorinated biphenyls (PCBs) at 15 sites in the southeastern area of the Czech Republic. In total 31 unhatched eggs of 4 raptor species and 3 owl species were examined. PCB determination was carried out by high-resolution gas chromatography. PCB content was expressed in µg per 1 kg of lipid weight for the major PCB congeners 28, 52, 101, 118, 138, 153 and 180, serving as indicators. In all examined eggs the indicator PCB congeners were found. The ranges of total values of indicator PCB congeners found in different raptor and owl species were as follows: kestrel (*Falco tinnunculus*) 37.0 and 44.3 ($n = 2$), black kite (*Milvus migrans*) 28.3–40.5 ($n = 3$), imperial eagle (*Aquila heliaca*) 66.4 ($n = 1$), marsh harrier (*Circus aeruginosus*) 45.1 ($n = 1$), barn owl (*Tyto alba*) 20.6–46.1 ($n = 17$), long-eared owl (*Asio otus*) 32.0–41.9 ($n = 6$) and tawny owl (*Strix aluco*) 47.5 ($n = 1$) µg/kg of lipid weight. PCB congener 153 was the one most frequently found.

Keywords: eggs; raptors; owls; indicator PCB congeners; contamination

In the 70s to 90s of the last century, polychlorinated biphenyls (PCBs) were the most frequently occurring xenobiotic chemicals contaminating the environment. In the 1980s these substances were banned in many countries including the Czech Republic. PCBs are lipophilic substances and therefore they accumulate in animal tissues. They are present in all parts of food chains (Hoffman *et al.*, 1996). Due to the fact that the position of raptors and owls as predators is at the end of a food chain, high levels of these substances can be expected to be found in their tissues (Anthony *et al.*, 1999). Various authors described the role of raptors and owls with regard to PCBs in the environment. Gilbertson (1990) described the possibilities of utilisation of raptors and owls as biological indicators of contamination of the environment with PCBs. Negative impact of PCBs upon reproductive performance in raptors and owls was described by Johnstone *et al.* (1996).

Ban on use of PCBs and other chlorinated organic substances with long biological half-time resulted in an increase in natural populations of raptors since

the mid nineties (Henny, 1998). During recent years, low concentrations of organic pollutants in the raptor eggs were recorded by Henny *et al.* (1998) in the Volga basin of Russia. In raptors observed in the United Kingdom in 1992–1997 the numbers of deaths due to intoxications caused by organochlorine pollutants was decreased by 50% compared to the period of 1960–1992 (Newton *et al.*, 1999). During the period of 1974–1994, Donaldson *et al.* (1999) detected reduced contamination by PCBs in blood plasma and eggs of the bald eagle (*Haliaeetus leucocephalus*) living in the area of Lake Erie (USA, Canada). A considerably reduced PCB content in eggs of the osprey (*Pandion haliaetus*) was reported from New Jersey (USA); at the same the number of individuals in the population has increased by 200% since 1989 (Clark *et al.*, 2001).

In European wild birds PCB content in tissues was recently described in particular in water birds (Aurigi *et al.*, 2000; Berny *et al.*, 2002). The levels of PCBs in raptors and owls was studied in Iceland (Olafsdottir *et al.*, 1995), Spain (Negro *et al.*, 1993),

Germany (Disser *et al.*, 1992; Wiesmuller *et al.*, 1999) and Russia (Henny *et al.*, 1998). There have not been any recent studies done to cover the situation in the Czech Republic with respect to PCB content of wild raptors and owls. The aim of our work was to provide such information and to carry out a comparison with the data from other regions.

MATERIAL AND METHODS

Unhatched eggs were collected in 2001 from nests of raptors and owls from 15 different sites in the south-eastern area of the Czech Republic. This lowland area is intensively agriculturally used with relative high settlement in villages and towns. However, no local sources of pollution by PCBs were identified. The whole area covers a few square kilometres. The nests where the eggs were collected were often a few tens km apart. This area is exploited by amateur ornithologists from The South Moravian Branch of the Czech Society for Ornithology, who are observing the nesting success of raptors and owls among others. During nest checking after the nesting season of 2001, they occasionally found abandoned, unhatched eggs. Following our request, the eggs were collected and preserved in the freezer at -18°C in microthene bags until our analyses at the end of 2001. The analysis was carried out in the eggs of the following species: kestrel (*Falco tinnunculus*, 2 eggs of 1 nest), the black kite (*Milvus migrans*, 3 eggs of 3 nests), imperial eagle (*Aquila heliaca*, 1 egg), marsh harrier (*Circus aeruginosus*, 1 egg), barn owl (*Tyto alba*, 17 eggs of 7 nests), long-eared owl (*Asio otus*, 6 eggs of 3 nests), and tawny owl (*Strix aluco*, 1 egg).

Raptor and owl eggs were usually unfertile. In some cases a dead embryo was found. Total content of the eggs was weighed and analysed. Egg content was ground with anhydrous sodium sulfate so as to obtain loose matter. PCBs were extracted together with fat by a continuous cold extraction with slightly polar organic solvent. The extraction was repeated three times, always using 100 ml of petroleum ether. The extracts were put together and the solvent was evaporated in a vacuum evaporator. The fat was weighed and total percentage of fat was determined. The quantities of 0.1–0.2 g of fat were used for further analysis. Lipids and other ballast substances were separated by column chromatography on Florisil. The elution was carried out with hexane-diethylether (94 : 6). The eluate

was concentrated by vacuum evaporator. Acid hydrolysis with concentrated sulfuric acid was used as needed for fine purification of the sample. The evaporation residue was dissolved in 1 ml of hexane and analysed by gas chromatography.

PCB analysis was carried out by high-resolution gas chromatography with electron capture detection as described by Vavrova *et al.* (2002). The conditions of the analysis were as follows: capillary column HP 5, column length 60 m, column inside diameter 0.25 μm ; film thickness 0.2 μm , split/splitless injector, injection temperature 250°C ; temperature mode 40°C for 40 min, increase to 180°C at 30°C per min and to 280°C at 2°C per min, and hold at 280°C for 10 min; detector ^{63}Ni ECD, detector temperature 300°C ; constant flow at 1.0 m per minute. Standards supplied by Ehrenstorfer (Germany) were used for congener identification. QA/QC conditions were assured with certified reference materials CRM of Ehrenstorfer (Germany) containing the studied substances in freeze-dried bovine liver tissue, freeze-dried beef, and bovine fat. Moreover, the materials of the National Reference Laboratory at the Institute of Chemical Technology in Prague were used. The detection limit for PCB congeners was 1 $\mu\text{g}/\text{kg}$. Arbitrary values of 0.5 $\mu\text{g}/\text{kg}$ were substituted for concentrations below the detection limit in the calculations of mean values. The content of major indicator congeners PCB 28, 52, 101, 118, 138, 153 and 180 in $\mu\text{g}/\text{kg}$ of fat was determined. These congeners were chosen by Community Bureau of Reference as the major components, which are almost present in the environment in high concentrations.

RESULTS

The content of indicator PCB congeners in raptor eggs, mean and median values for the total content of indicator PCB congeners are presented in Table 1. The highest PCB content was found in the eggs of the imperial eagle (total content of all indicator PCB congeners was 66.4 $\mu\text{g}/\text{kg}$ of fat). High-chlorinated congeners prevailed over low-chlorinated compounds in the eggs of all four raptor species included in the study. Highest levels were found for PCB 153, followed by PCB 138 and PCB 180.

The values for indicator PCB congeners in the eggs of the barn owl and the long-eared owl are presented in Tables 2 and 3. The content of PCBs in the eggs of the tawny owl was as follows: 2.3 for PCB 28, 2.0 for PCB 52, 1.4 for PCB 101, > 1 for PCB

Table 1. Content of indicator PCB congeners in raptor eggs ($\mu\text{g}/\text{kg}$ of fat)

Species	Nest No.	Egg No.	Indicator congeners						Total	
			28	52	101	118	138	153		180
Kestrel	1	1	1.1	1.3	1.0	1.1	8.4	14.5	9.6	37.0
		2	2.3	1.2	1.0	1.1	12.6	17.1	9.0	44.3
Black kite	2	3 ^e	2.0	1.7	1.5	<1	6.9	17.2	10.7	40.5
		4	1.3	1.1	1.2	1.1	7.4	15.4	9.4	36.9
		5	1.3	1.2	1.0	<1	5.1	12.3	6.9	28.3
Imperial eagle	5	6 ^e	3.9	2.6	1.9	1.4	17.2	26.3	13.1	66.4
Marsh harrier	6	7	2.5	2.0	1.4	1.0	11.5	17.7	8.9	45.1
Mean	–	–	2.06	1.59	1.29	0.96	9.87	17.21	9.66	42.64
Median	–	–	2.00	1.30	1.20	1.10	9.87	17.10	9.40	–

^ethere was embryo in egg

Table 2. Content of indicator PCB congeners in eggs of the barn owl (*Tyto alba*; $\mu\text{g}/\text{kg}$ of fat)

Nest No.	Egg No.	Indicator congeners						Total	
		28	52	101	118	138	153		180
1	1	2.3	1.2	1.1	1.0	11.4	14.5	9.2	40.7
	2	2.2	1.1	1.0	<1	10.1	13.4	4.5	32.8
	3	1.4	<1	<1	1.3	9.2	12.4	9.3	34.6
	4	1.2	1.1	1.0	<1	12.2	16.4	10.0	42.4
	5	1.9	1.3	1.1	1.0	13.4	17.8	9.6	46.1
2	6	1.3	1.2	1.1	1.0	9.2	12.4	5.3	31.5
	7	1.4	1.1	<1	<1	8.3	11.4	2.9	26.6
	8	1.9	1.2	1.1	1.0	7.9	14.8	9.6	37.5
	9 ^e	1.7	1.2	<1	<1	8.4	15.2	10.7	38.2
3	10	1.6	1.2	1.0	<1	7.2	14.4	9.7	35.6
	11	<1	<1	<1	<1	6.1	8.2	4.3	20.6
	12	<1	<1	<1	<1	5.3	9.4	5.3	22.0
4	13	1.1	<1	<1	1.2	7.4	10.3	6.7	27.7
	14	1.2	<1	1.1	1.1	8.1	11.4	5.3	28.7
5	15 ^e	2.2	1.2	1.1	<1	13.6	15.5	10.3	44.4
6	16 ^e	1.4	<1	<1	1.0	7.3	9.5	7.0	27.2
7	17	1.2	1.1	1.1	1.0	9.3	12.5	7.4	33.6
	Mean	1.47	*	*	*	9.08	12.90	7.48	33.94
	Median	1.40	1.47	*	*	8.30	12.50	7.40	–

^ethere was embryo in egg

*not evaluated due to large proportion of values below detection limit

Table 3. Content of indicator PCB congeners in eggs of the long-eared owl (*Asio otus*; µg/kg of fat)

Nest No.	Egg No.	Indicator congeners							Total
		28	52	101	118	138	153	180	
1	1	1.9	1.3	1.1	1.0	6.2	13.3	10.0	34.8
2	2	2.1	1.2	< 1	< 1	8.6	19.8	8.9	41.6
	3	1.7	1.5	1.3	1.1	7.7	18.5	10.1	41.9
	4 ^e	2.3	1.9	1.0	1.1	6.5	11.9	7.3	32.0
	5	1.9	1.4	1.2	< 1	9.5	17.9	8.5	40.9
3	6	2.3	1.7	1.0	1.1	4.8	16.2	10.2	37.3
	Mean	2.03	1.50	1.02	0.88	7.22	16.27	9.17	38.08
	Median	2.00	1.45	1.05	1.05	7.10	17.05	9.45	–

^ethere was embryo in egg

118, 12.6 for PCB 138, 18.4 for PCB 153 and 10.3 for PCB 180. All values are given in µg/kg of fat. The highest content of PCBs was found in the tawny owl (total value for all indicator PCB congeners was 47.5 µg/kg of fat). Similarly to the findings in raptors, high-chlorinated congeners were detected in higher levels in all three owl species compared to low-chlorinated compounds. The ranking of different congeners according to the highest concentrations found was again the same: PCB 153, PCB 138 and PCB 180.

DISCUSSION

Indicator PCB congeners were detected in all raptor and owl eggs included in the study, apparently because of the history of use of commercial products containing PCBs in the past. These xenobiotic substances had contaminated the environment in the Czech Republic and even many years after their ban it was possible to detect them in soil (Korinek, 1999), water and water sediments (Zlamalova-Gargosova *et al.*, 2002), and tissues of game and small mammals (Vavrova *et al.*, 2003). As regards the levels of indicator PCB congeners identical to those in our study, Vavrova *et al.* (2003) reported the following values found in skin, muscles and liver of small ground mammals in the southeastern area of the Czech Republic (all values are given in µg/kg of fat): low-chlorinated PCBs < 1–10 in all types of samples; PCB 138 – 11.8–19.6 (skin), 19.1 to 32.3 (liver), 6.9–13.6 (muscles); PCB 153 – 7.8–10.9 (skin), 10.9–24.5 (liver), 4.8–9.3 (muscles); PCB 180

– 5.0–7.9 (skin), 8.2–19.7 (liver), 2.9–8.3 (muscles). Small ground mammals and small game animals are the most important components of prey of raptors and owls. This may be the cause of occurrence of PCBs in raptor and owl eggs from the given area. Our conclusions are in conformity with the study containing analysis of PCB in eggs of 20 avian species with regard to their position in the food chain (Disser *et al.*, 1992). Markedly highest content of PCBs was found in raptors and owls.

In the second half of the eighties Kredl and Kren (1986), Hudec *et al.* (1988), and Kredl *et al.* (1988) studied the contamination of raptor and owl eggs by organochlorine derivatives in the Czech Republic. A contamination with PCBs was on a relatively high level in the Czech Republic in the second half of eighties. The values presented in these works cannot be compared with our results because the quantitative evaluation was different – separate congeners were not determined. Kredl *et al.* (1988) reported the following values in raptor and owl eggs (all values are given in mg/kg of dry matter): the kestrel (*Falco tinnunculus*) – 0.6 ($n = 29$), the long-eared owl (*Asio otus*) – 13.7 ($n = 4$), the barn owl (*Tyto alba*) – 2.9 ($n = 18$), and the tawny owl (*Strix aluco*) – 0.4 ($n = 1$).

By far the highest content of PCBs was found in the egg of the imperial eagle (*Aquila heliaca*), which can be explained by the following facts: A pair of young birds nested for the first time in 2001. The female was 4 years old (P. Horak, personal communication). It can be therefore concluded that the time of accumulation of PCBs in the body of the female was considerably longer than in other raptors and owls. The largest proportion of PCBs

is transferred from the body of the female into the first laid egg (Disser *et al.*, 1992). In our case a single egg only was laid.

The major PCB indicator congeners, in particular the high-chlorinated compounds, such as PCB 153, PCB 180 and PCB 138, were found in the examined eggs of raptors and owls. These congeners are present in the environment because of the use of commercial products with higher content of high-chlorinated PCBs (hexa- and heptachlorobiphenyls). Out of such products in particular Delor 106 was used in the Czech Republic (Zlamalova-Gargosova *et al.*, 2002). Similarly to our results, Disser *et al.* (1992) detected in raptor and owl eggs in Germany highest levels of congeners PCB 153, PCB 180 and PCB 138. In the barn owl in Germany congeners PCB 138, PCB 153 and PCB 180 accounted for the highest PCB levels (Wiesmuller *et al.*, 1999). The higher levels of high-chlorinated congeners compared to low-chlorinated compounds are explained by the tendency of the former to accumulate in the body. Similar results were also described by Elliott and Norstrom (1998) and Elliott *et al.* (2001) in Canada. Although the proportions of different congeners varied in different sites, the highest content in Canada was always found for PCB 153.

CONCLUSION

Indicator PCB congeners were found in all the eggs of raptors and owls. The range of the sum of indicator PCB congeners in the eggs of all species was between 20–50 µg/kg lipid weight. Congener PCB 153 and others high-chlorinated congeners are present in the surroundings of the Czech Republic as the result of former using commercial mixture Delor 106.

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