PATTERNS OF PCBs AND PCDD/Fs IN CHICKEN AND PORK FOLLOWING A BELGIAN FOOD CONTAMINATION

Adrian Covaci, John Jake Ryan[1] and Paul Schepens

Toxicological Center, University of Antwerp (UIA), Universiteitsplein 1, 2610 Wilrijk, Belgium
[1] Health Canada, Bureau Chemical Safety, Ottawa K1A 0L2, Ontario, Canada

Introduction

In February 1999, a poisoning episode broke out in several farms in Belgium. Through the use of contaminated fat for production of approximately 500 tons of animal feed, about 50 kg of PCBs and 1 g of mostly furans were introduced into the food chain[1]. Analysis of contaminated feedstuff showed a pattern of PCBs closely matched with a mixture of Aroclors 1254 and 1260 and a consistent pattern of dioxin congeners, dominated by polychlorodibenzofurans (PCDFs). The outbreak of the Belgian PCB/dioxin contamination is similar with other past poultry poisoning[2,3] with PCBs. This study was undertaken to examine the accumulation patterns of PCBs and furans in chicken and pork resulting from the adulteration of the commercial food chain. The relative importance of PCB and PCDF congeners in these human foods is also assessed.

Methods and Materials

Sampling: Three samples of chicken fat (one low, medium and high) and two of pork fat (low and medium) along with a highly adulterated chicken feed sample obtained in the course of the Belgian dioxin crisis were chosen for detailed analyses.

A. Determination of PCBs. *Sample preparation:* A homogenised portion of animal fat was melted, weighed and solubilised in 10 ml n-hexane. Internal standards, PCB 46 and 143 were added to give a concentration of 50 ng/g fat. After ultrasonication, clean-up over 5 g acid silica (concentrated sulphuric acid : silica = 1 : 1, w/w), PCBs were eluted with 20 ml n-hexane. The eluate was concentrated to 0.5 ml and 5 ng 1,2,3,4 - tetrachloronaphthalene was added as recovery standard. To 0.5 g feedstuff (~6% fat content), 5 ml n-hexane were added together with internal standards to give a concentration of 50 ng/g fat. After ultrasonication, the supernatant was subjected to the same clean-up procedure as described above.

Analysis: A Hewlett Packard 6890 gas chromatograph with μ-ECD was equipped with a HT-8 (SGE) capillary column (30m x 0.22mm x 0.25μm). One μl was injected in pulsed splitless mode. For confirmatory purposes, a Hewlett Packard 6890 gas chromatograph was connected to a Hewlett Packard 5793 mass spectrometer (MS) and equipped with a DB-5ms (J&W Scientific) capillary column (30m x 0.25mm x 0.25μm). Peak identification was based on relative retention time (to TCN) and, for MS, on specific group ions. Recoveries of internals standards were 72±8 % for CB 46 and 78±10% for CB 143, respectively (n=3). Quality control was assured by the analysis of certified fish oil (CRM 349) and by successful participation to two interlaboratory tests organised by the Belgian Ministries of Health and Agriculture.

B. Determination of PCDD/Fs and non-ortho PCBs. To 1 to 5 g of chicken or pork fat or 0.1 g of chicken feed was added a mixture of seventeen ^{13}C-2,3,7,8-PCDD/Fs and three non-ortho PCBs. The samples were homogenized and extracted with acetone-hexane (a small aliquot was used for the lipid determination gravimetrically), defatted with concentrated sulphuric acid, adsorbed on acid/base silica and Florisil, and separated on activated carbon. Identification was by gas chromatography high resolution mass spectrometry and quantification by the isotope dilution internal standard method[4]. Detection limits varied depending on congener and sample size and were as low as 0.2 pg/g.

Results and Discussion

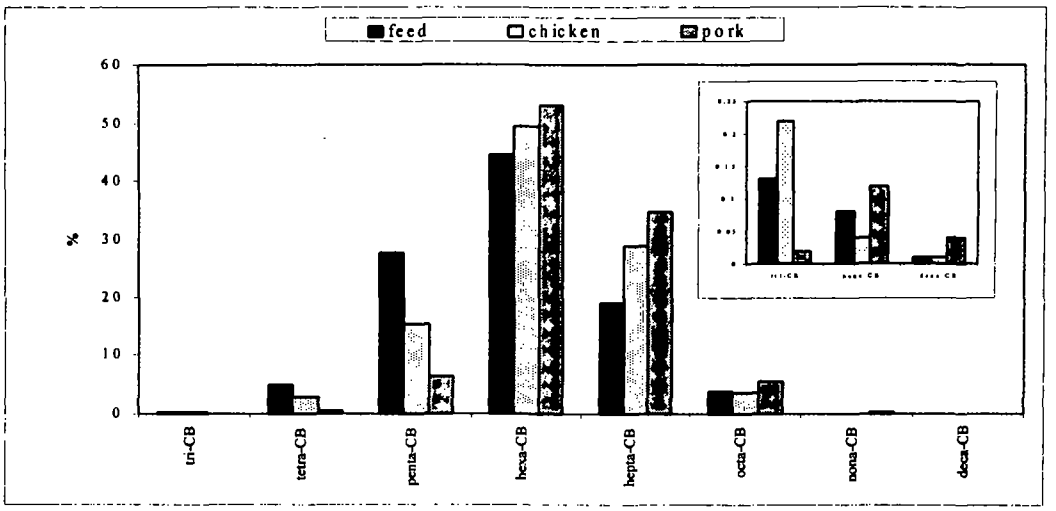
During the Belgian food contamination, the Toxicological Center performed over 2000 analyses on animal fat (mainly chicken and pork) for 7 marker PCBs (IUPAC no. 28, 52, 101, 118, 138, 153 and 180). The profiles varied between the two species and from the contaminated feedstuff, which necessitated data on a larger number of PCB congeners. Monitoring of more congeners gives a greater potential for estimation of congener profile changes and helps to resolve many of the ambiguities in the risk assessment. Three samples of chicken, two of pork and one the feedstuff were analysed for 56 PCB congeners, 3 non-ortho PCBs and 17 2,3,7,8-substituted PCDD/F. I-TEQ values calculated for the PCDD/F congeners, non- and mono-ortho PCBs are presented in Table 1. The patterns found (the individual number and/or relative proportion of each congener within a group) are approximately constant for each species analysed, which in turn are representative of other contaminated samples (same percentage of the 7 markers).

Table 1. I-TEQ values in chicken, pork and feedstuff.

	TEF	TEQ (pg/g fat)					
compound		feed-7057	ch-6133	ch-6134	ch-10078	po-7129	po-7133
105	0.0001	652.1	24.1	9.6	1.1	0.1	0.4
114	0.0005	151	3.0	1.2	0.2	0.1	0.5
118	0.0001	1224.4	63.4	25.3	2.9	0.9	4.5
123	0.0001	148.5	0.3	0.1	0.0	0.0	0.05
156	0.0005	1838	78.5	31.3	3.6	9.8	47.6
157	0.0005	277	13.0	5.2	0.6	1.7	8.1
167	0.00001	16.2	1.6	0.1	0.0	0.1	0.2
189	0.0001	10.6	1.8	0.7	0.1	0.3	1.6
77	0.000 1	31.7	0.1	0.1	0.0	0.0	0.0
126	0.1	5683.2	61.8	30.4	2.8	0.8	1.2
169	0.01	31.7	0.3	0.2	0.00	0.1	0.25
PCDF		13517	108.2	40.0	2.3	3.3	5.6
PCDD		1566	10.3	4.3	0.7	0.1	0.2
PCDD/F		15083	118.5	44.3	3.0	3.4	5.8
non-ortho PCB		5747	62.2	30.6	2.8	0.9	1.45
mono-ortho PCB		4317.8	185.7	73.5	8.5	13.0	63.0
total TEQ		25147	366.4	148.4	14.3	17.3	70.25
sum CB (ng/g fat)		316330	8540	3405	390	1065	5160
sum 7marker PCB (ng/g fat)		114463	4387	1750	200	600	2900

3.1. Patterns of PCBs in chicken and pork. Differences in metabolism are remarked in the region of lower chlorinated PCBs (tri- to penta-PCB congeners). Levels of tri- to penta-CBs are lower in chicken or pork than in the feedstuff, indicating that both species are able to metabolise to some extent these congeners. However, higher levels of lower chlorinated congeners can be found in chicken when compared with pork (Figure 1). Levels of higher chlorinated PCBs (hexa to octa-CB) are higher in animals than in the feedstuff, suggesting an accumulation for these congeners especially for the persistent ones (PCB 138, 153 and 180) in the following sequence: feedstuff – chicken – pork.

Figure 1. Patterns of PCBs in feed, chicken and pork samples



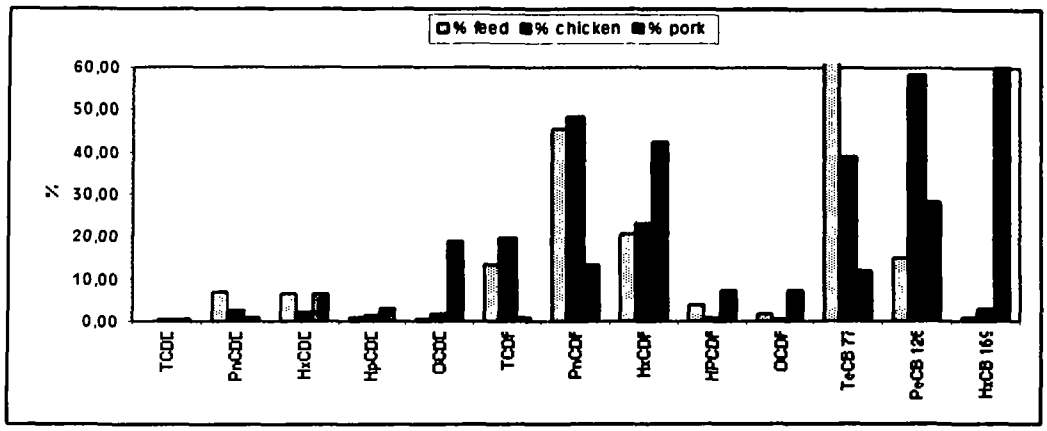
The following PCB congeners were found to accumulate more in chicken than in pork: 28, 49, 56/60, 74, 105, 114, 118, 126, 135, 141, 149, 174, 176 and 179. It seems that there is an ability of the mammals to metabolise those PCB congeners with 2 vicinal hydrogen atoms inmeta/para or ortho/para positions

It is interesting to mention the relatively high percentage of some congeners (PCB 101 and 110) in the contaminated feed and Aroclor mixtures, while their concentration in animal body fat is low. Congeners 132, 141, 149 and 151 are cleared in both investigated species and as expected, some congeners (PCB 99, 138, 153, 156, 170, 177, 180 and 187) are accumulating in both species. These congeners are usually analysed in routine work (highest concentration in the residues) and have not assigned TEF values.

3.2. Patterns of PCDD/PCDFs in chicken and pork. Because of high incidence of PCDFs, the dioxin/furan contamination could not have arisen from either PCP waste (predominant presence of higher chlorinated congeners), or from incineration processes (PCDDs and PCDFs are in similar amounts). This is consistent with the known presence of PCDF congeners in Aroclor mixtures in higher proportion than PCDDs, which is typical for PCB type of contamination as already seen in Yusho oil[5]. Chicken tissues contain more PCDD/F congeners than pork tissues, including non-2378 substituted ones such as 12478-PnCDF and 124689-HxCDF. This is probably related to the different metabolic capabilities of the mammalian and bird classes. The same congeners are present in the feedstuff, but to a higher extent. In both animal species, the 2378 substituted isomers are accumulating at a higher rate than the non-2378 substituted ones. In chicken, 2378-TCDF and 23478-PnCDF isomers account for 80 % of the PCDD/F TEQ value, while in pork only for 60 %.

3.3. TEQ from dioxins and from PCBs. The preferential accumulation of some PCB/dioxin congeners in birds (e.g. PCB 105, 118 and 126, 2378-TCDF and 23478-PnCDF) is partially responsible for their increased TEQ values. Contribution of PCBs to the total TEQ is higher in pork (mainly due to PCB 156 and 157) although the difference in overall concentration between pork and chicken may be a factor. Because of the ability of pork to metabolise tetra and penta-PCBs and PCDD/Fs (Figure 2), the total TEQ are lower than from chicken. For chicken, samples with levels of PCBs higher than the tolerance level (200 ng/g fat) contained higher TEQ values than the tolerance level for dioxins (5 pg I-TEQ/g fat), while for pork the situation is not the same, because of the metabolism of congeners with high TEFs (PCB 126, 2378-TCDF, 23478-PeCDF).

Figure 2. Patterns of PCDD/Fs



3.4. Remarks on the metabolism of different congeners in other species. As already observed, PCB patterns in fish and birds are different than those in mammals including humans. The presence of more tri, tetra and penta chlorinated PCBs and non 2378- PCDFs are observed in fish and birds. The appearance of these compounds in fish and birds indicates the inability for metabolism that leads to a selective accumulation. In the real life situation of the Belgium incident, we have shown that birds are better markers of the source of contamination of persistent organic pollutants than mammals since the residue pattern changes less.

References

1. A. Bernard, C. Hermans, F. Broeckaert, G. Depoorter, A. De Cock, G. Houins. *Nature* 1999, 401, 231-232.
2. D.P. Drotman, P.J. Baxter, J. A. Liddle, C.D. Brokopp, M.D. Skinner. *Am. J. Pub. Health* 1983, 73, 290-292.
3. M. Gilbertson, T. Kubiak, J. Ludwig, G.J. Fox. *J. Toxicol. Environ. Chem.* 1991, 33, 455-520.
4. J.J. Ryan, C. Shewchuk, B.P.Y. Lau, W.F. Sun. *J. Agr. Food Chem.* 1992, 40,919-923.
5. L.G. Hansen. *Environ. Health Perspect.* 1998, 106 (suppl.1), 171-189.
6. S.Tanabe, N.Kannan, T.Wakimoto, R.Tatsukawa. *Toxicol. Environ. Chem.* 1989, 24, 215-231.