

PCDD, PCDF, AND PCB LEVELS AND PROFILES IN MILK AND FEEDINGSTUFFS FROM FARMS NEAR TWO INCINERATION PLANTS IN TUSCANY, ITALY

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Introduction

Italy, as other European countries, has been involved in the debate on the impact of incineration emissions on human health. In the last decades, the release of PCDDs and PCDFs from incinerators has progressively decreased because of the use of abatement technologies and of the endorsement of the stricter emission limits envisaged by the EC normative. Nevertheless, the results of a large study recently carried out in France showed a correlation between an increase in human serum levels of PCDDs and PCDFs and the presence of an incinerator near the place of residence in people consuming important amounts of local produced food^{1,2}.

In the Province of Grosseto, Tuscany, concerns of people living around two old-generation incineration plants (Casone and Valpiana) prompted local sanitary authorities to carry out a study to characterize the impact of incinerator emissions on environmental and human exposure. The study included the measurement of serum levels of PCDDs and PCDFs in groups of individuals living in areas under impact with respect to groups of individuals living nearby areas at presumable background level of exposure³, as well as the assessment of the role of consumption of locally grown food in contributing to determine an eventual increase in serum levels of these contaminants. The present paper reports the results of the analysis of PCDDs, PCDFs, and PCBs in milk of grazing cows and sheep from farms located in the two type of areas, and in samples of locally grown feedingstuffs.

Materials and methods

Selection of study areas

Study areas were identified on the basis of the distance from the incineration plants and from the analysis of the atmospheric dispersion carried out by the use of the CALMET-CALPUFF model (CALPUFF Modelyng System⁴). The following farms were identified in the areas under the impact of the two incinerator emissions: V₁, V₂, and V₃ (Valpiana area), and C₁, C₂, and C₃ (Casone area). Farms located in the control areas at presumable background level of exposure were P (Prata) and G (Giuncarico).

Sampling and analysis

Bovine and ovine milk samples and samples of the feedingstuffs used to breed animals were collected in 2005, from the eight farms included in the study. All feedingstuffs were of local origin, with the exception of the sample "C₁, mixed feed", which was a mixed formulation of commercial origin.

Feedingstuff and milk samples were spiked before extraction with ¹³C-labelled PCDD, PCDF, and PCB internal standards and then liquid-liquid extracted with mixtures of acetone/n-hexane and diethyl ether/n-hexane (after addition of methyl alcohol), respectively. After treatment with concentrated sulphuric acid, clean up was performed by the automatic Power-Prep system. The moisture content of feedingstuff samples was determined on dedicated subsamples in drying oven at 105 °C. Lipid content of milk samples was gravimetrically determined.

Quantitative determination was performed by high resolution gas chromatography coupled with high-resolution mass spectrometry (HRGC-HRMS) used in the selected ion monitoring mode (SIM) at a resolution of 10000. The mass spectrometer was a VG Autospec. Quantification was accomplished by the isotope dilution technique.

Results and discussion

Cumulative results of the analysis of PCDDs, PCDFs, and PCBs in feedingstuff and milk samples are reported in Table 1.

Table 1. Cumulative concentrations (pgWHO-TE/g, upper bound approach) of PCDDs, PCDFs, dioxin-like, and non-dioxin-like-PCBs in feedingstuffs and milk samples from eight areas of the Province of Grosseto. For feedingstuffs all the values are referred to fresh weight, with a moisture content of 12 %. For milk samples all the values are expressed on a lipid base. Values rounded off to two figures.

Analytes	Areas under impact of the two incinerator emissions areas							Control areas		
	V ₁ , cereals	V ₂ , hay	V ₃ , grass	C ₁ , mixed feed	C ₁ , cereals	C ₂ , cereals	C ₂ , fodder	C ₃ , hay	P, hay	G, hay
Σ(PCDD+PCDF)	0.12	0.11	0.40	0.11	0.070	0.16	0.21	0.18	0.18	0.11
Σ(dl-PCB)	0.27	0.14	0.21	0.14	0.23	0.30	0.090	0.22	0.16	0.096
Total TEQ	0.39	0.25	0.61	0.25	0.30	0.46	0.30	0.40	0.34	0.21
Σ ₆ (PCB) ^a	0.14	1.2	1.1	1.6	0.16	9.3	0.13	0.20	1.9	1.2
	V ₁ , ovine milk	V ₂ , ovine milk	V ₃ , ovine milk	C ₁ , bovine milk		C ₂ , ovine milk		C ₃ , ovine milk	P, bovine milk	G, bovine milk
Lipid content %	3.1	6.9	5.9	3.4		6.1		5.9	2.9	4.1
Σ(PCDD+PCDF)	0.41	0.30	0.44	0.21		0.32		0.95	0.14	0.25
Σ(dl-PCB)	0.37	0.60	0.63	0.50		1.34		1.92	0.45	0.27
Total TEQ	0.78	0.90	1.1	0.71		1.7		2.9	0.59	0.52
Σ ₆ (PCB) ^a	1.4	2.3	3.0	1.5		3.9		8.2	1.6	0.90

(a) ng/g, sum of the six NDL indicator PCBs.

Feedingstuff samples

Total TEQ values in samples from the six areas under incineration impact ranged from 0.25 to 0.61 pgWHO-TE/g fresh weight (fw), and were similar to those found in the two areas at a presumed background level of exposure, in the range of 0.21–0.34 pgWHO-TE/g fw. PCDD+PCDF and DL-PCB contributions to the total WHO-TEQ were 0.070–0.40 pgWHO-TE/g fw and 0.090–0.30, respectively. All these levels were much lower than the limits and the action levels established by the European Commission⁵ and showed levels of contamination comparable to samples collected in other countries in areas under no direct exposure to incinerator emissions^{6,7}. Cumulative levels of the six NDL-PCBs ranged from 0.13 to 9.3 ng/g fw in potentially contaminated samples and from 1.2 to 1.9 ng/g fw in control samples. These values were below the mean value of occurrence (10.7 ng/g) reported for feeds or feed components of plant origin by the European Food Safety Agency⁸.

Milk samples

Total TEQ values were in the range of 0.71–2.9 pgWHO-TE/g fat in milk samples from the six farms under incineration impact and of 0.52–0.59 pgWHO-TE/g fat in samples from farms in the control areas. PCDD+PCDF and DL-PCB contributions to the total TEQ were respectively 0.21–0.95 and 0.37–1.92 pgWHO-TE/g fat in the areas near the incinerators and 0.14–0.25 and 0.27–0.45 pgWHO-TE/g fat in the control areas. In both kind of areas, contribution of DL-PCBs to total TEQ was predominant; this finding has been reported in literature data on ovine and bovine milk from other European countries^{6,9,10}. All TEQ values were below the maximum levels¹¹ and the action levels¹² established for raw milk, and similar to those observed in areas at background levels of exposure in other countries^{13,14,15}. NDL-PCB cumulative levels ranged from 1.4 to 8.2 ng/g

fat in samples collected in the six farms near the incineration plants and from 0.90 to 1.6 ng/g fat in control samples, values that are much lower than the mean value (10.7 ng/g fat) calculated by EFSA⁸.

Feedingstuffs-to-milk carry-over

Pattern of PCDDs and PCDFs, DL-PCBs, and NDL-PCBs of a milk sample (V₂, ovine milk) and of a feedingstuff sample (V₂, hay) from the same farm are shown in Figure 1. These patterns may be considered representative of the profiles observed in the two type of the analysed samples.

PCDD and PCDF congener-specific profiles of feedingstuffs were dominated by O₈CDD (D₇ in Figure 1), 1, 2, 3, 4, 6, 7, 8-H₇CDD (D₆), 2, 3, 7, 8-T₄CDF (F₁), 1, 2, 3, 4, 6, 7, 8-H₇CDF (F₈), and O₈CDF (F₁₀), while D₇, D₆ (with a relative abundance lower than in feedingstuffs), and PCDDs and PCDFs with a medium degree of chlorination were prevalent in milk samples. These differences in patterns can be partly explained by the different feed-to-milk carry over rates (CORs, the fractions of the ingested contaminants that are excreted in milk at steady state^{18,19}) that represent a measure of the transfer of a specific compound from feedingstuffs to milk. Within the 17 congeners of PCDDs and PCDFs tetra- to hexa-CDDs, 2,3,4,7,8-P₅CDF (F₃), 1,2,3,4,7,8-H₆CDF (F₄), and 1,2,3,6,7,8-H₆CDF (F₅) are the congeners with the highest CORs, while the other congeners are transferred from feed to milk to a lesser extent, because of a lower absorption or a greater metabolism (Costera et al., 2006); among these, in particular F₁ and 1, 2, 3, 7, 8-P₅CDF (F₂) are known to be strongly metabolized within the organism¹⁸. For these reasons, D₆, F₁, F₈, and F₁₀, which are the predominant congeners in feedingstuffs, in milk samples are present at a lower relative abundance, or even below the detection limit.

Congener profiles of DL-PCBs are dominated by congeners 118, 105, and 156 in both feed and milk samples. All these congeners have a high COR value (about 80 %¹⁹) and are easily

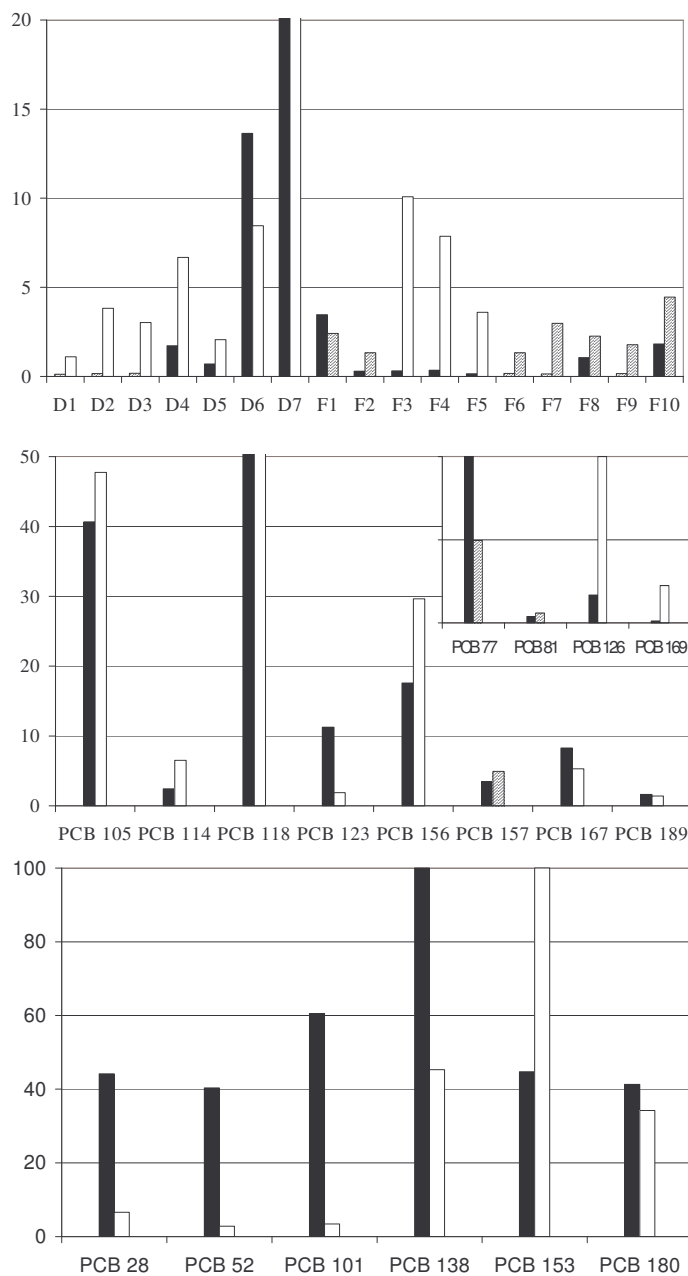


Figure 1. PCDD, PCDF, DL-PCB, and NDL-PCB congener profile comparison between feedingstuff (hay, black bars) and milk (ovine milk, white bars) samples collected in the same farm (V₂, in Valpiana area). Congeners under the limit of detection are shown as grey bars.

transferred from feedingstuffs to milk. The major differences observed between DL-PCB profiles of feed and milk are the relative abundances of PCBs 77 and 123, the DL-PCB congeners with the lowest transfer rates (values of COR < 20 %¹⁹). These congeners have a higher relative abundance in feed than in milk, and PCB 77 in particular is the most abundant non-*ortho* congener in feedingstuffs, often found below the limit of detection in milk samples. The most abundant non-*ortho* congener in milk is PCB 126 that has the highest COR¹⁹ among these PCB congeners.

Indicator PCB congeners 138, 153 and 180 are predominant in feedingstuffs and milk samples, and are the indicator PCBs with the highest transfer rates values (COR > 40 %¹⁹). The other three indicator congeners (PCBs 28, 52 and 101) are present in feedingstuff samples with relative abundances highest than in milk. These findings are in agreement with the indication that PCBs 28, 52 and 101 are easily metabolized by the organisms, while PCBs 138, 153 and 180 are more resistant to biotransformation²⁰.

In conclusion, results of the analysis of feedingstuffs and milk samples did not reveal any statistically significant difference in levels ($p > 0.05$, Mann Whitney U test) and profiles of PCDDs, PCDFs, DL-PCBs, and NDL-PCBs between samples from areas under impact from the incineration plants and from areas at background exposure.

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