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ScienceDirect
Chemosphere 70 (2008) 1563–1567
CHEMOSPHERE
www.elsevier.com/locate/chemosphere

Relationship of PCDD/Fs congener profiles between beef and raw milk in South Korea

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Received 11 April 2007; received in revised form 16 August 2007; accepted 17 August 2007
Available online 22 October 2007

Abstract

The relationship between profiles of residual PCDD/Fs in beef and raw milk was examined by measuring concentrations and detected frequencies. Unrelated samples of beef and raw milk were collected from nine regions in South Korea. Congener-specific profiles of PCDD/Fs in beef and raw milk were very similar. PCDFs, particularly 2,3,4,7,8-PeCDF, 1,2,3,4,7,8-HxCDF, 1,2,3,6,7,8-HxCDF, and 2,3,4,6,7,8-HxCDF, were dominant congeners in both beef and raw milk suggesting that sources of contamination may not significantly differ nationwide. The profiles of PCDD/Fs in domestic beef and raw milk in this study were closer to the profiles of emission from metal industries although Korea imports over 75% of feedingstuffs. The ratios of PCDF/PCDD in TEQ concentration were more than 5 and 15 in beef and raw milk, respectively. The mean concentrations of PCDD/Fs in 60 samples of beef and 60 samples of raw milk were 0.80 pg WHO-TEQ/g fat and 0.65 pg WHO-TEQ/g fat, respectively. The residual profiles of PCDD/Fs in raw milk resembled that in beef although the congener profiles might change throughout the food chain. This indicated that monitoring of dioxins in milk could provide information for contamination of milk itself or other associated food.

Keywords: PCDD/Fs; Congener; Residual profile; Beef; Raw milk

1. Introduction

Polychlorinated dibenzo-*p*-dioxins (PCDDs) and dibenzofurans (PCDFs) are found in a variety of foods. Although the animal metabolism and environmental fate could influence the congener profiles, congener-specific profiles of residual dioxins (PCDD/Fs) in food may reflect the sources of the contaminants. These environmental contaminants directly affect animals, agriculture, and crops used as animal feed, and indirectly affect foodstuffs of animal origin (McLachlan, 1997; Schuler et al., 1997; Wagrowski and Hites, 1998). Animal feed is a major source of dioxin contamination in food of animal origin (Covaci et al., 2002; Malisch, 2000; Hoogenboom et al., 2004). Contamination is difficult to detect before feed reaches livestock or food reaches market. Milk is probably one of the most convenient materials to examine for dioxin contamination, either in the milk itself or in the associated beef. Cattle have more time to accumulate dioxins in their bodies before slaughter because they live relatively longer than most other livestock animals. Profiles of residual PCDD/Fs in raw milk might help interpret the profiles of residual PCDD/Fs in beef. The concentration of dioxins excreted in milk divided by the concentration of dioxins taken from a cow will represent the carry-over rate. Carry-over rates and bioaccumulation are inversely related to the degree of chlorination (McLachlan and Richter, 1998; Huwe and Smith, 2005). Carry-over rates are usually independent of lactation rate, body fat weight, and feed mix of the animal (McLachlan and Richter, 1998), suggesting that the levels of PCDD/Fs in raw milk may help predict the levels of PCDD/Fs in beef.

In this study, beef and raw milk samples were not related to each other. Residual profiles in milk are likely to reflect the residual profiles in beef if the samples came

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0045-6535/$ - see front matter © 2007 Elsevier Ltd. All rights reserved.
doi:10.1016/j.chemosphere.2007.08.060

from the same dairy cow. Residual profiles observed in beef and milk from different cows should represent the general relationship between dioxin levels in beef and milk instead. It assumed that the samples used in this study were not incidentally contaminated with dioxins. Not all beef samples were from dairy cows, and samples were taken from different farms in the same region. A greater variety of samples might better reveal general trends in the background levels of contamination with PCDD/Fs. The purpose of this study was to evaluate congener profiles in beef and milk throughout South Korea to determine the main source of PCDD/Fs in animals and determine if a general correlation could be made between the matrices.

2. Materials and methods

2.1. Materials

PCDD/Fs and ^{13}C-labeled standards including a labeled compound spiking solution (LCS), cleanup standard solution (CSS), and internal standard solution (ISS) were purchased from Wellington Laboratories Co. (Canada). Calibration standard solutions (CS) were purchased from Cambridge Isotope Laboratories, Inc. (USA). Organic solvents used were HPLC-grade from J.T. Baker (USA).

Sixty beef samples from 60 cattle in 52 farms were collected from February to October 2004 at nine slaughter facilities in nine regions throughout South Korea. The cattle were from two to five years old and from 410 to 850 kg weight. Sixty raw milk samples from 21 gathering stations involving 129 farms were collected in April 2005 in the nine regions. Each farm provided a sample of collected raw milk from several dairy cows. The number of samples from each region depended upon production rates.

2.2. Preparation of beef samples

Fat was melted from the beef samples in an 80 °C oven before analysis (Covaci et al., 2002). LCS was spiked to 5 g of the liquefied fat in 200 ml of hexane. The isotope dilution method used was based on the US EPA protocol 1613B. Acidic silica gel (deactivated, 30%) was added and the mixture was shaken for 2 h followed by elution through an anhydrous sodium sulfate column. The sample was concentrated to 1–2 ml and a small amount of hexane was added. CSS was added to the sample and clean-up was performed using the Power-Prep™ (Fluid Management Systems, Waltham, MA, USA) system with a silica column, alumina column, and activated carbon column. ISS was added to the final extract, which was in nonane after concentration.

2.3. Preparation of raw milk samples

Extraction of PCDD/Fs from raw milk was carried out through several steps. First, 300 ml of methanol and 4 g of sodium oxalate were added to 200 ml of raw milk and shaken for 30 min by a mechanical shaker. Next, 200 ml of ethyl ether/petroleum ether (1:1, v/v) were added and the mixture was shaken for an additional 1 h. After centrifugation for 20 min at 3000 rpm, the upper solution was transferred to a separatory funnel. The bottom solution was extracted twice more with the above solutions and the upper solution was again transferred to the separatory funnel and then washed with NaCl-saturated distilled water. The organic layer was collected through anhydrous sodium sulfate and concentrated in a Turbovap II (Zymark, USA). The collected fatty part was dissolved in hexane and cleaned by silica, alumina, and carbon columns using the Power-Prep™ system as with the extraction of the beef samples.

2.4. HR-GC/MS Analysis

PCDD/Fs were analyzed by a gas chromatograph (HP 6890, Hewlett Packard, USA) with a DB5MS capillary column (50 m × 0.25 mm ID, 0.25 μm film thickness, J&W Scientific, USA) coupled to a mass spectrometer (Autospec Ultima, Micromass, UK). The two most abundant ions were measured. The GC oven temperature was held for 1 min at 160 °C, after which the temperature was increased to 220 °C at 5 °C/min and then held for 15 min, followed by an increase to 290 °C at 5 °C/min that was held for 10 min. The temperature was then increased to a final temperature of 300 °C at a rate of 10 °C/min and held for 12 min. Peak identifications were made by observation of retention time and masses for the two major ions. Recoveries of the internal standards were 67–138%.

3. Results and discussion

Table 1 presents the concentrations by weight of PCDD/Fs, % frequency detected and the limit of detection for each congener in beef and raw milk. The concentrations were calculated using zero for non-detects. In samples of both beef and raw milk, OCDD, 2,3,4,7,8-PeCDF, 1,2,3,4,7,8-HxCDF, 1,2,3,6,7,8-HxCDF, 2,3,4,6,7,8-HxCDF, and 1,2,3,4,6,7,8-HpCDF were the major congeners in both concentration and frequency detected. It seems that the sources of contamination were very similar across the samples and the regions. However, 1,2,3,6,7,8-HxCDD with a detection frequency of 50% in beef, contributed 13% to the concentration of PCDDs, which was a significantly different profile from the raw milk, where the detection frequency of 2% contributed 0.7% to the concentration of PCDDs. 1,2,3,4,6,7,8-HpCDD was the only congener with higher concentrations and detection frequency in raw milk than in beef, although the carry-over rate was low (McLachlan and Richter, 1998; Fürst et al., 1992). 1,2,3,7,8-PeCDD had a concentration of 0.06 pg/g fat and represented 13% of detects in beef, which was 3–4 times more frequent than in raw milk. 1,2,3,4,7,8-HxCDD, 2,3,7,8-TCDF, 1,2,3,7,8-PeCDF, and 1,2,3,7,8,9-HxCDF were not detected in the 60 samples of raw milk. Overall,

Table 1
Mean, median, frequency detected (FD) and limit of detection (LOD) for PCDD/Fs analyzed in beef and raw milk (pg/g fat)

Congener	Beef ($n = 60$)			Raw milk ($n = 60$)			LOD
	Mean	Median (range)	% FD	Mean	Median (range)	% FD	
2,3,7,8-TCDD	0.01	0.00 (nd – 0.46)	3	0.01	0.00 (nd – 0.54)	3	0.024
1,2,3,7,8-PeCDD	0.06	0.00 (nd – 1.50)	13	0.01	0.00 (nd – 0.65)	3	0.013
1,2,3,4,7,8-HxCDD	0.03	0.00 (nd – 0.51)	10	nd	nd	0	0.027
1,2,3,6,7,8-HxCDD	0.31	0.06 (nd – 2.11)	50	0.01	0.00 (nd – 0.72)	2	0.024
1,2,3,7,8,9-HxCDD	0.06	0.00 (nd – 0.69)	18	0.02	0.00 (nd – 0.48)	5	0.024
1,2,3,4,6,7,8-HpCDD	0.52	0.38 (nd – 2.43)	58	0.96	0.97 (nd – 3.72)	85	0.029
OCDD	1.80	1.15 (nd – 8.73)	85	1.67	1.39 (nd – 9.57)	97	0.032
2,3,7,8-TCDF	nd	nd	0	nd	nd	0	0.015
1,2,3,7,8-PeCDF	0.07	0.00 (nd – 0.45)	2	nd	nd	0	0.013
2,3,4,7,8-PeCDF	0.88	0.73 (nd – 3.33)	88	0.86	0.73 (nd – 3.24)	90	0.010
1,2,3,4,7,8-HxCDF	0.82	0.71 (nd – 2.69)	88	0.65	0.53 (nd – 3.54)	78	0.023
1,2,3,6,7,8-HxCDF	0.70	0.61 (nd – 2.60)	82	0.57	0.47 (nd – 2.97)	78	0.021
2,3,4,6,7,8-HxCDF	0.69	0.52 (nd – 2.42)	87	0.49	0.44 (nd – 2.28)	70	0.029
1,2,3,7,8,9-HxCDF	0.004	0.00 (nd – 0.17)	3	nd	nd	0	0.029
1,2,3,4,6,7,8-HpCDF	0.62	0.55 (nd – 2.17)	75	0.57	0.47 (nd – 2.02)	83	0.019
1,2,3,4,7,8,9-HpCDF	0.01	0.00 (nd – 0.68)	2	0.006	0.00 (nd – 0.24)	3	0.028
OCDF	0.07	0.00 (nd – 0.80)	7	0.007	0.00 (nd – 0.11)	7	0.017
PCDDs	2.79	1.94 (nd – 12.1)		2.68	2.47 (nd – 9.84)		
PCDFs	3.86	3.09 (nd – 12.0)		3.15	2.73 (nd – 13.8)		
PCDD/Fs	6.66	5.08 (0.87 – 24.1)		5.83	5.14 (0.90 – 16.6)		
TEQ (PCDD/Fs)	0.80	0.60 (nd – 3.26)		0.65	0.53 (nd – 2.54)		

nd = not detected.

the frequencies detected of PCDDs and PCDFs in beef and in raw milk were almost the same. The contributions of penta- and hexa-CDFs to the congener profile were 83% in beef and 93% in raw milk. The cattle sampled in the present study ranged in age from two to seven years old and the body weights were about 410–850 kg. Age, body weight, and type of cattle did not related to the residual levels of PCDD/Fs in this study.

Congener profiles of PCDD/Fs in beef and in raw milk from each region are shown in Figs. 1 and 2, respectively. The nine regions were labeled alphabetically from A to I. TEQ concentrations were determined by averaging the regional samples. The congener profiles were very similar between beef and raw milk. 2,3,4,7,8-PeCDF was the most highly concentrated in almost all samples of beef and raw milk. 1,2,3,7,8-PeCDD was the most concentrated in beef

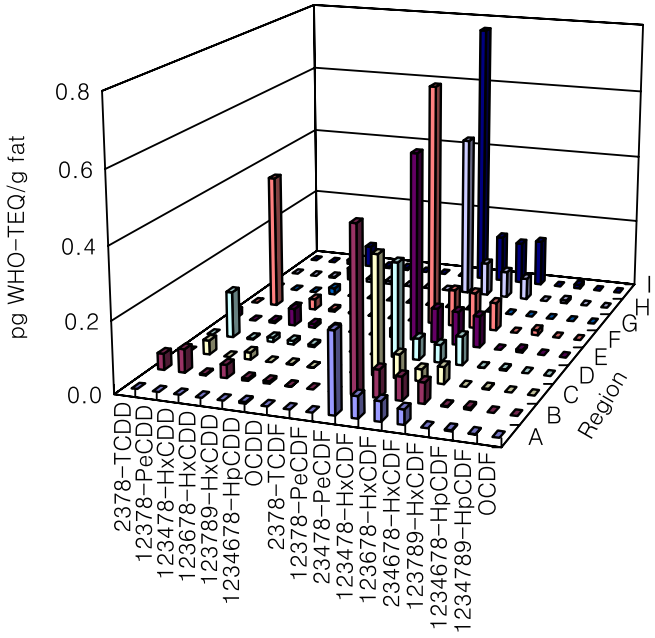

Fig. 1. Congener profiles of the average level of PCDD/Fs in beef from each region.

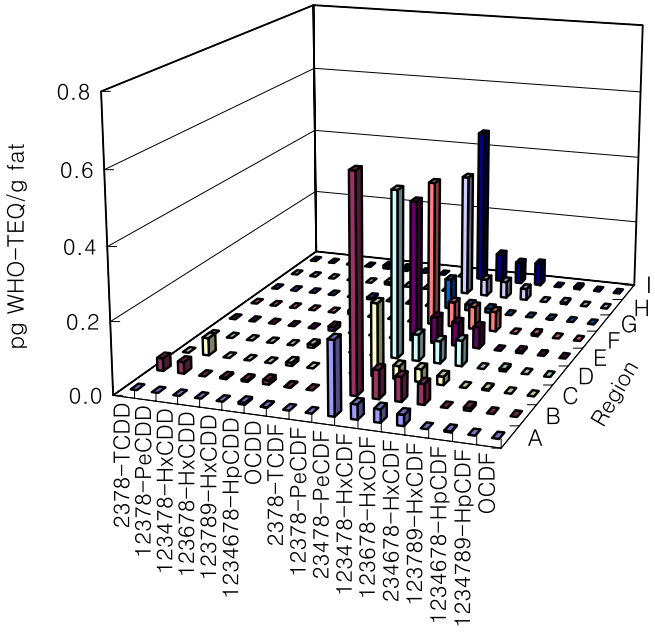

Fig. 2. Congener profiles of the average level of PCDD/Fs in raw milk from each region.

collected from the region F. From a toxicological point of view, 2,3,7,8-TCDF and OCDF were found in few of the 60 samples of beef and 1,2,3,4,7,8-HxCDD, OCDD, 2,3,7,8-TCDF, 1,2,3,7,8-PeCDF, 1,2,3,7,8,9-HxCDF, 1,2,3,4,7,8,9-HpCDF, and OCDF were found in few of the 60 samples of raw milk. The samples containing 2,3,7,8-TCDD and/or 1,2,3,7,8-PeCDD showed the highest TEQ levels in both beef and raw milk.

The number of samples and mean concentrations of PCDDs and PCDFs in beef and raw milk from each region were presented in Table 2. PCDFs were found at much higher concentrations than PCDDs in beef and raw milk from almost all the regions. The levels of PCDFs in beef were about five times higher than PCDDs in samples from the five regions. PCDDs alone were found in 3% of the total samples of beef, and PCDFs alone were found in 20% of the total samples of beef. The mean concentrations of PCDDs and PCDFs in raw milk were 0.04 and 0.61 pg WHO-TEQ/g fat, respectively. The levels of PCDFs in milk were about 15 times higher than PCDDs in samples from all nine regions. PCDD/Fs were not found in 5% of the total samples of raw milk. PCDDs and PCDFs were not found in 13% and 5%, respectively, of the total samples of raw milk. Overall, the residual TEQ in raw milk was mostly from PCDFs, as in beef.

The residual profiles of PCDDs and PCDFs in domestic beef and raw milk were very different from products in other countries. Beef in the USA as reported by Ferrario et al. (1996) and ground beef and milk from southern Mississippi as reported by Fiedler et al. (1997) showed similar congener profiles. The total contributions of PCDDs were higher than PCDFs in the USA beef fat and milk. Pentachlorophenol-treated wood was one of the sources of contamination in the USA (Fries et al., 2002; Huwe et al., 2004). Belgium beef showed higher levels of PCDFs than PCDDs, which was in line with the results of this study (Focant et al., 2002). The TEQ concentration of PCDFs was slightly higher than PCDDs in Belgium milk although the concentration by weight of PCDDs was higher than PCDFs (Focant et al., 2002). A comparison of different congener-specific profiles between domestic beef in Korea and imported beef from four non-European countries (Australia, Canada, New Zealand, and USA) in 2001–2002 indicated that the sources of dioxins were different (Kim et al., 2003a). However, the levels and detects of 1,2,3,6,7,8-HxCDD had a similar profile to the USA beef fat compared to other congeners.

The residual profiles of PCDD/Fs in beef and raw milk were similar to that in feed of animal origin, especially fish stuffs (Kim et al., 2005). Food and feed production is very complex. For example, dioxins enter the atmosphere from incinerators and settle onto the ground and crops by dry and wet deposition. PCDD/Fs are then directly transferred to animals through inhalation and indirectly transferred to animals through crops as animal feed (Schuler et al., 1997). Animal proteins and fats that contain PCDD/Fs are also used as feed (Eljarrat et al., 2002). A small amount of PCDD/Fs in a component of feed can accumulate to make a larger amount in an animal consuming the feed. At the same time, a large amount of PCDD/Fs in a component of feed can be diluted to a small amount by the many ingredients in the production of animal feed. The environment and livestock feed both influence the profiles of residual PCDD/Fs in cattle (McLachlan, 1997). Food of animal origin from non-European countries has been related more to contamination from minerals and chemical impurities. For the last few years, animal feed has been the major contamination source of food of animal origin. Mineral origin or anthropogenic sources were major contributors of PCDD/Fs in accidental contaminations of animal feed (Ferrario et al., 1999; Abad et al., 2002; Llerena et al., 2003). However, accidents involving citrus pulp or PCBs with animal fat might have different origins from those of PCDD/Fs (Malisch, 2000; Covaci et al., 2002). The residual profiles of PCDD/Fs in domestic beef and raw milk in this study were closer to the profiles of emission from the metal industry (Kim et al., 2003b, 2003c; Yu et al., 2003). The residual profiles of PCDD/Fs in fish oil, shell powder, and animal fat were also similar to that in this study (Kim et al., 2005). It is difficult to investigate the sources of contamination because South Korea is importing 75% of consumed feed and feed additives. In this

Table 2
Number of samples (*N*) and mean concentrations (pg TEQ/g fat) of PCDD/Fs in samples from each region

Region	Beef				Raw milk			
	N	Concentration			*N*	Concentration		
		PCDDs	PCDFs	PCDD/Fs		PCDDs	PCDFs	PCDD/Fs
A	6	0.00	0.39	0.39	4	0.01	0.31	0.32
B	10	0.17	0.67	0.84	20	0.09	0.81	0.90
C	9	0.06	0.49	0.55	5	0.06	0.30	0.36
D	7	0.16	0.47	0.63	5	0.01	0.69	0.70
E	9	0.09	0.85	0.94	5	0.01	0.61	0.62
F	5	0.42	1.00	1.42	5	0.01	0.62	0.63
G	2	0.02	0.00	0.02	2	0.00	0.12	0.12
H	7	0.06	0.71	0.77	10	0.01	0.49	0.50
I	5	0.09	1.17	1.26	4	0.02	0.68	0.70
Total	60	0.12	0.68	0.80	60	0.04	0.61	0.65

present study, the residual profiles of PCDD/Fs in raw milk resembled that in beef, although the congener profiles might change through the food chain. The results should be considered that milk is probably a useful indicator to test for PCDD/Fs contamination of milk and the associate food.

References

Abad, E., Llerena, J.J., Sauló, J., Caixach, J., Rivera, J., 2002. Comprehensive study on dioxin contents in binder and anti-caking agent feed additives. Chemosphere 46, 1417–1421.

Covaci, A., Ryan, J.J., Schepens, P., 2002. Patterns of PCBs and PCDD/PCDFs in chicken and pork fat following a Belgian food contamination incident. Chemosphere 47, 207–217.

Eljarrat, E., Caixach, J., Rivera, J., 2002. Determination of PCDDs and PCDFs in different animal feed ingredients. Chemosphere 46, 1403–1407.

Ferrario, J., Byrne, C., McDaniel, D., Dupuy Jr., A., 1996. Determination of 2,3,7,8-chlorine-substituted dibenzo- *p*-dioxins and furans at the part per trillion level in United States beef fat using high-resolution gas chromatography/high-resolution mass spectrometry. Anal. Chem. 68, 647–652.

Ferrario, J., McDaniel, D., Byrne, C., 1999. The isomer distribution and congener profile of polychlorinated dibenzo-p-dioxins (PCDDs) in ball clay from the Mississippi embayment (Sledge, Mississippi). Organohalogen Compd. 40, 1–5.

Fiedler, H., Cooper, K.R., Bergek, S., Hjelt, M., Rappe, C., 1997. Polychlorinated dibenzo- *p*-dioxins and polychlorinated dibenzofurans (PCDD/PCDF) in food samples collected in Southern Mississippi, USA. Chemosphere 34, 1411–1419.

Focant, J-F., Eppe, G., Pirard, C., Massart, A-C., Andre, J-E., De Pauw, E., 2002. Levels and congener distributions of PCDDs, PCDFs and non-ortho PCBs in Belgian foodstuffs: Assessment of dietary intake. Chemosphere 48, 167–179.

Fries, G.F., Feil, V.J., Zaylskie, R.G., Bialek, K.M., Rice, C.P., 2002. Treated wood in livestock facilities: Relationships among residues of pentachlorophenol, dioxins, and furans in wood and beef. Environ. Pollut. 116, 301–307.

Fürst, P., Fürst, C., Wilmers, K., 1992. Survey of dairy products for PCDDs, PCDFs, PCBs and HCB. Chemosphere 25, 1039–1048.

Hoogenboom, R., Bovee, T., Portier, L., Bor, G., van der Weg, G., Onstenk, C., Traag, W., 2004. The German bakery waste incident; use of a combined approach of screening and confirmation for dioxins in feed and food. Talanta 63, 1249–1253.

Huwe, J.K., Davison, K., Feil, V.J., Larsen, G., Lorentzsen, M., Zaylskie, R., Tiernan, T.O., 2004. Levels of polychlorinated dibenzo- *p*-dioxins and dibenzofurans in cattle raised at agricultural research facilities across the USA and the influence of pentachlorophenol-treated wood. Food Addit. and Contam. 21, 182–194.

Huwe, J.K., Smith, D., 2005. Laboratory and on-farm studies on the bioaccumulation and elimination of dioxins from a contaminated mineral supplement fed to dairy cows. J. Agric. Food Chem. 53, 2362–2370.

Kim, M., Kim, S.Y., Kwon, J.W., Kim, D.G., Son, S.W., 2005. Survey of PCDD/Fs in animal feed and feed additives. Organohalogen Compd. 67, 1492–1494.

Kim, M., Kim, S.Y., Yun, S.J., Cho, B.H., Lee, M.H., Son, S.W., Park, J.M., 2003a. Congener-specific profile of PCDDs/PCDFs in beef, pork, and chicken. Organohalogen Compd. 64, 9–12.

Kim, S.-C., Na, J.-G., Choe, S.H., Lee, J.-H., Kim, Y.-H., Hwang, S.-R., Joo, C.-H., Jung, D.-H., You, J.-C., Lee, S.-W., Jeon, S.-E., 2003b. PCDDs/PCDFs emission from nonferrous metal industry. Organohalogen Compd. 63, 81–85.

Kim, S.-C., Na, J.-G., Choe, S.-H., Lee, J.-H., Kim, Y.-H., Hwang, S.-R., Joo, C.-H., Moon, D.-H., You, J.-C., Jung, E.-K., Lee, C.-J., Choi, K.S., 2003c. PCDDs/PCDFs emission from ferrous metal industry. Organohalogen Compd. 63, 77–80.

Llerena, J.J., Abad, E., Caixach, J., Rivera, J., 2003. An episode of dioxin contamination in feedingstuff: the choline chloride case. Chemosphere 53, 679–683.

Malisch, R., 2000. Increase of the PCDD/F-contamination of milk, butter and meat samples by use of contaminated citrus pulp. Chemosphere 40, 1041–1053.

McLachlan, M.S., 1997. A simple model to predict accumulation of PCDD/Fs in an agricultural food chain. Chemosphere 34, 1263–1276.

McLachlan, M., Richter, W., 1998. Uptake and transfer of PCDD/Fs by cattle fed naturally contaminated feedstuffs and feed contaminated as a results of sewage sludge application. 1. Lactating cows. J. Agric. Food Chem. 46, 1166–1172.

Schuler, F., Schmid, P., Schlatter, C., 1997. Transfer of airborne polychlorinated dibenzo- *p*-dioxins and dibenzofurans into dairy milk. J. Agric. Food Chem. 45, 4162–4167.

Wagrowski, D.M., Hites, R.A., 1998. Partitioning of polychlorinated dibenzo- *p*-dioxins and dibenzofurans between the atmosphere and corn. Environ. Sci. Technol. 32, 2389–2393.

Yu, B.-W., Moon, Y.-H., Kim, M.-K., Kyoung, J.-D., Chang, Y.-S., 2003. Inventory study of PCDD/Fs for metal industries in South Korea. Organohalogen Compd. 63, 94–97.