ANALYSIS OF ANIMAL- AND PLANT-DERIVED FEED INGREDIENTS FOR DIOXIN-LIKE COMPOUNDS

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Introduction

During a national survey of polychlorinated dibenzo-*p*-dioxins (CDD), dibenzofurans (CDF), and dioxin-like coplanar PCBs (PCB) in poultry[1], elevated concentrations above 20 parts per trillion (ppt) toxic equivalents (TEQ) were found in the fat of 2 broilers. These TEQ values were driven by very high concentrations of CDD. A team comprised of individuals from the United States (US) Environmental Protection Agency (EPA), the US Food and Drug Administration (FDA), and the US Department of Agriculture (DA) traced the source of elevated CDD to a minor component in the poultry feed. This component was ball clay and it was used as an anti-caking agent in the soybean meal. The ball clay often comprised less than 0.2 % of the dry weight of the complete ration in contaminated poultry. The investigation traced the ball clay to a mine in Mississippi. After learning that other ball clay mines in Kentucky and Tennessee also contained elevated CDD levels, the FDA issued a letter to producers or users of clay products in animal feeds asking that they cease using ball clay in any animal feed or feed ingredient[2].

Subsequent contaminations of animal feed in Belgium[3] with PCB and of citrus pulp from Brazil[4] with CDD and CDF alerted countries worldwide that animal feeds can become contaminated with CDD/CDF/PCB (DFP) via contamination of minor feed components. This type of contamination can overshadow the normal air-to-leaf process that is thought to dominate the food chain for terrestrial food animals in background conditions. Following the ball clay incident, the US EPA, FDA and DA have coordinated their efforts to monitor for DFP in animal- and plant-derived food products, complete animal feeds, and animal feed ingredients. This has been done in order to better understand the background DFP status of the food supply and how that food supply comes to attain levels of DFP.[5,6,7]

In 1998, the Center for Veterinary Medicine (CVM) of FDA collected several anti-caking products used in animal feeds and oilseed meals. The samples from this survey were analyzed by the EPA Environmental Chemistry Laboratory (ECL) in Mississippi, USA, and the FDA Arkansas Regional Laboratory. Nine of 15 samples contained CDD/CDF (DF) at low levels, ranging from 0.4 to 22.5 ppt TEQ, with 1,2,3,7,8-pentachlorodibenzo-*p*-dioxin (PeCDD) contributing the most to the sample TEQ.[8] For comparison, the ball clay from the 1997 investigation contained TEQ concentrations over 1000 ppt and 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (TCDD) was the primary congener contributing to the sample TEQ. Together with EPA, CVM initiated a survey in 2000 where 47 feed samples were collected.[2] This survey is the subject of this paper, but analytical data will be presented on only 44 samples. In 2001, CVM continued its study of animal feeds by requesting that 50 samples of oilseed meals, fat-soluble vitamins, complete feeds, milk products, minerals, and wood products be collected for DFP analysis[2]. Results from the 2001 survey may be available during 2002.

Methods

Since dioxins are predominantly found in fats, most of the samples collected in the 2000 survey were fatty in nature and came from commonly used feed ingredients that were speculated to contain relatively high levels of DFP. Thus, beef fat, pork fat, fat and meat and bone meal from mixed animal species, poultry by-product meal, fish meals, and egg samples were taken. Some samples of plant origin speculated to contain meaningful levels of DFP were also collected. These included deodorizer distillates and molasses. Deodorizer distillates are by-products of one of the final steps in the commercial refining of vegetable oils. It has been demonstrated that this ingredient can contain toxic organic contaminants such as halogenated industrial contaminants (e.g., PCBs) and halogenated pesticides (e.g., aldrin, dieldrin)[9]. These contaminants are removed from the oil during the deodorizing (distillation) steps of refining and are concentrated in the deodorizer distillate fraction. Cane and beet molasses were selected because they are common feed ingredients, and although not fatty, may provide some insight into the relative importance of DFP contamination in soil (on beets) or from fire (in cane harvesting) on these feed ingredients.

The 44 samples from this survey are divided into 9 categories including:

1) Animal fat from slaughtered beef or slaughtered pork (AF-SL)

A total of six fat samples (three each for pork and beef) originated from six different integrated animal renderers that are associated with a slaughter facility. These renderers only process beef or pork or have segregated facilities for beef and pork.

2) Animal fat from mixed animal species (AF-MX)

Three samples from three independent animal renderers were collected. These renderers were not associated with a single slaughter facility and their products typically contain fat from several animal species.

3) Meat and bone meal from mixed animal species (MBM-MX)

Three samples were obtained from two independent renderers. MBM is high in protein, calcium and phosphorus.

4) Poultry by-product meal and eggs (PBM&E)

Three poultry by-product meal samples were obtained from three integrated renderers associated with a poultry slaughter facility. In addition, two egg samples, one a dried egg sample and the other a broken egg sample were collected from two companies. These two ingredients were combined into one category because they had similar dioxin levels and were derived from poultry.

5, 6 & 7) Fish meal (FM-WHI), (FM-CAT), and (FM-MEN)

Eight fish meal samples including three from Pacific whiting, one from catfish, and four from menhaden (about 90% of fish meal production in the US is derived from menhaden) were collected from 8 separate facilities. The fish meal samples were divided by species into three categories because of the differences noted in dioxin levels.

8) Deodorizer distillates (DD)

Eleven deodorizer distillate samples were obtained from ten separate facilities. These samples were collected at oilseed processors that produce corn, soybean, peanut, cottonseed, and/or canola oils.

9) Molasses/Corn Oil/Canola Meal (MOL)

Three cane and three beet molasses samples from six companies were collected. In addition, one corn oil and one canola meal sample from two companies were collected. These eight samples were grouped together because they had similar dioxin levels and were derived from plants.

The 47 samples in this survey were collected by FDA investigators between June 19 and October 2, 2000. They were collected in dioxin-free containers using dioxin-free equipment. These samples came from 22 different states (n=6 for TX; n=5 for IL, n=3 for GA, LA, NE and WA; n=2 from AL, FL, MN,

MS, MT, NC, NY, and OH; and n=1 from AZ, HI, IA, MD, MI, ND, TN, and VA). Samples were shipped overnight to the EPA ECL.

Dry free-flowing samples were Soxlet extracted while liquid samples were liquid/liquid extracted. Samples were cleaned-up using acidified and basic silica, alumina and grafitized carbon. Analytical and QA/QC procedures were similar to those described in EPA's Method 1613 and Ferrario et al.[10] The detection limits using this high resolution gas chromatography/mass spectrometry method are expressed in total picograms for a one gram sample (pg/g) and the congener specific detection limit on a per sample basis was calculated by dividing the following amounts (pg) by the sample weight (sample weights ranged from 5.4-34.1 g): Tetra- CDD and CDF - 0.4; Penta- and Hexa-CDD and CDF - 1.0; Hepta- CDD and CDF - 3.0; OCDD - 30.0; OCDF.- 10.0; PCB 77 – 2.0; PCB 126 - 2.0; PCB 169 - 1.0. TEQ concentrations were calculated using the 1998 WHO TEFs.

Results

Table 1 shows the results of this survey, including average congener concentrations for groupings of samples, and TEQ results. The DFP-TEQ levels for the slaughtered animal fat samples (AF-SL; column 1) averaged about 0.5 ppt TEQ dry weight. The pork fat samples averaged about 0.2 ppt TEQ and were lower than the beef fat samples which averaged about 0.9 ppt TEQ. The DFP-TEQ concentrations for the mixed species animal fat (AF-MX; column 2) was dominated by one sample described as "animal fat beef/swine (bulk rendered)", whose concentration was 3.95. The DFP-TEQ levels for meat & bone meal from mixed animal species (MBM-MX; column 3) as well as the poultry by-product meal and eggs (PBM&E, column 4) were all below 0.3 ppt. The DFP-TEQ levels for the fish meal samples (FM-WHI; FM-CAT; FM-MEN; columns 5, 6 and 7) varied by species. The lowest values were consistently found from the Pacific whiting, all at less than 0.50 ppt DFP-TEQ, and the highest values were consistently found from the menhaden, averaging 2.9 ppt DFP-TEQ. The single catfish sample had DFP levels between the Pacific whiting and the menhaden (DFP-TEQ = 0.9 ppt) and had a different congener pattern. Specifically, the DFP-TEQ was dominated by dioxins for the catfish sample, with very little contribution by furans or PCBs. For the menhaden, the dioxin-like PCB made up 41% of the TEQ, the dioxins made up 40 %, and the furans made up 19 %. The deodorizer distillates (DD; column 8) showed the highest dry weight concentrations of DFP in this study. The average dry weight concentration of 4.4 ppt TEQ was not driven by any single sample, as the range for the 11 samples was narrow (1.4 to 7.1 ppt TEQ). An examination of the individual congeners suggests that both dioxin and furan congeners were elevated, but the dioxins more than the furans. This differs markedly from the ball clay profile, which was unambiguously elevated by the dioxin congeners only. The other vegetable-based samples (MOL; column 9) had very low DFP concentrations, mostly < 0.10 ppt TEQ.

US Federal Agencies continue to investigate animal feed components as possible sources of contaminants to the human food supply.

Disclaimer

The views expressed in this article are those of the authors and do not necessarily reflect the views or policies of the U.S. Environmental Protection Agency and the U.S. Food and Drug Administration.

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Table 1. Summary data for feed ingredients (average analytical concentrations in ppt for 20 DFP congeners when non-detects (ND) = ½ detection limit; levels of 0.01-0.04 ppt indicate ND for all or most samples; average TEQ concentrations in ppt when ND = ½ detection limit and at ND=0).

Description	Animal-Based							Plant-Based	
	AF – SL	AF – MX	MBM – MX	PBM &E	FM – WHI	FM – CAT	FM – MEN	DD	MOL
Number of samples	6	3	3	5	3	1	4	11	8
2378-TCDD	0.04	0.08	0.01	0.01	0.01	0.09	0.37	0.23	0.02
12378-PeCDD	0.12	0.36	0.03	0.04	0.04	0.46	0.62	0.92	0.04
123478-HxCDD	0.13	0.53	0.03	0.04	0.03	0.61	0.17	1.31	0.04
123678-HxCDD	0.61	2.99	0.10	0.04	0.08	0.91	0.66	3.27	0.04
123789-HxCDD	0.19	0.66	0.06	0.04	0.03	0.64	0.46	3.45	0.04
1234678-HpCDD	3.17	16.26	1.66	0.61	0.29	6.62	3.11	62.55	1.12
OCDD	12.03	76.74	15.06	7.29	2.80	47.84	54.07	511.15	11.61
2378-TCDF	0.04	0.73	0.04	0.01	0.20	0.05	2.32	0.68	0.02
12378-PeCDF	0.06	0.11	0.02	0.04	0.03	0.02	0.31	0.40	0.04
23478-PeCDF	0.11	0.32	0.02	0.04	0.08	0.04	0.58	0.77	0.04
123478-HxCDF	0.13	0.63	0.04	0.04	0.03	0.02	0.12	0.70	0.04
123678-HxCDF	0.11	0.42	0.02	0.04	0.03	0.02	0.06	0.51	0.04
234678-HxCDF	0.06	0.22	0.04	0.04	0.04	0.05	0.12	0.65	0.04
123789-HxCDF	0.06	0.06	0.02	0.04	0.03	0.02	0.02	0.24	0.05
1234678-HpCDF	0.17	2.87	0.24	0.19	0.09	0.06	0.28	6.54	0.50
1234789-HpCDF	0.17	0.25	0.07	0.11	0.08	0.05	0.06	0.57	0.12
OCDF	0.50	2.25	0.42	0.27	0.25	0.02	0.33	10.55	1.63
PCB 77	2.06	22.15	1.68	119.06	2.01	2.37	82.10	133.70	0.71
PCB 126	1.62	6.60	0.32	0.50	1.42	0.37	11.63	9.73	0.02
PCB 169	0.54	1.29	0.04	0.13	0.40	0.08	1.97	0.44	0.04
TEQ – D	0.28	1.02	0.04	0.07	0.07	0.84	1.15	2.63	0.08
TEQ – F	0.10	0.40	0.03	0.04	0.07	0.04	0.57	0.76	0.05
TEQ – P	0.17	0.68	0.03	0.05	0.15	0.04	1.19	0.99	<0.01
TEQ – DF	0.38	1.43	0.11	0.11	0.15	0.87	1.72	3.38	0.13
TEQ – DF; ND=0	0.26	1.41	0.07	0.01	0.11	0.87	1.72	3.35	0.02
TEQ – DFP	0.54	2.10	0.14	0.16	0.29	0.91	2.91	4.39	0.13
TEQ – DFP; ND=0	0.43	2.09	0.10	0.06	0.26	0.91	2.91	4.34	0.02
Min TEQ – DFP	0.08	1.07	0.13	0.09	0.20	0.91	2.14	1.43	0.02
Max TEQ – DFP	1.49	3.95	0.16	0.30	0.44	0.91	3.33	7.08	0.18

Key: AF-SL = animal fat from slaughtered animals; AF-MX = animal fat from mixed animal species; MBM-MX = meat & bone meal from mixed animal species; PBM&E = poultry by-product meal and eggs; FM-WHI = Pacific whiting meal; FM-CAT = catfish meal; FM-MEN = menhaden meal; DD = deodorizer distillates; MOL = molasses, corn oil and canola meal.