Supplementary Material

Contribution of Polybrominated Dibenzo-*p*-dioxins and Dibenzofurans (PBDD/Fs) to the Toxic Equivalency of Dioxin-like Compounds in Archived Biosolids from the U.S. EPA's 2001 National Sewage Sludge Survey

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Tables (6)

Biosolids samples. Biosolids samples were collected by the U.S. EPA from 94 WWTPs in 32 U.S. states and the District of Columbia as part of the 2001 NSSS. Information on sampled facilities is available as supplementary material (Table S1). The facilities were selected by the U.S. EPA to obtain unbiased national estimates of chemical contaminants in U.S. sewage sludges that are disposed of primarily by land application. During the 1988 national sewage sludge survey, U.S. EPA collected information on facilities that had a minimum secondary biological treatment and a secondary clarifier for regulatory development efforts. About 11,400 facilities met these criteria, of which a statistical probability sample of 208 facilities comprised the analytical component of the 1988 survey. From this list, 101 facilities were statistically drawn for the 2001 NSSS to represent the population of 7,714 WWTPs across the following four strata based on their average daily flow of influent wastewater:

- Flow >100 million gallons per day (mgd: 1 mgd = 3.78 million liters per day)
- Flow >10 mgd but <100 mgd
- Flow > 1 mgd but < 10mgd
- Flow <1 mgd

The sampling fractions were derived using Bayes Theorem, the details of which could be found elsewhere[1]. Hence the samples analyzed in the present study are statistically 'representative' of the more than 16,000 WWTPs present in the U.S. The purpose of EPA's 2001 survey was to estimate levels of chlorinated dioxins, furans, and coplanar polychlorinated biphenyls in biosolids. Grab samples were collected between February and March 2001, according to an established protocol, exclusively targeting facilities featuring secondary treatment [1]. All samples were collected in 500 mL glass or polyethylene/polypropylene jars. Polyethylene gloves were used to prevent sample contamination. Solid biosolids samples were collected by using a polyethylene pail and polypropylene scoop. Samples were directly collected from the sludge discharge chute in the pail, thoroughly mixed using the scoop and then transferred to the containers. If the facility featured two or more dewatering units, equal amounts of samples were collected from each discharge point and mixed in the polyethylene pail. While collecting liquid samples from storage tanks, the pumps were ran for five minutes to clear stagnant sludge and then samples were collected in to the pail. The collected samples were thoroughly agitated in the pail using a polyethylene ladle and then transferred to the container. Samples were collected only from processed sewage sludge (biosolids) intended for disposal. Of the 94 WWTPs, 89 had single system (either aerobic or anaerobic digestion) and five of them had two systems for sludge treatment (both aerobic and anaerobic digestion). Samples were collected from each treatment system. In addition, duplicate samples were collected from 14 facilities, amounting to a total of 113 biosolids samples. Three samples were lost due to breaking of containers during transportation. The rest of the 110 biosolids samples were randomly grouped into five composite samples, each containing solids from between 21 and 24 individual samples. Composite samples were prepared by weighing out approximately 1 g of dry weight from each sample and pooling it to obtain five composites. The solids content of the MSS samples ranged between 1.5 and >90 %.

Quality assurance. Analysis batches consisted of a maximum of 20 samples, one procedural blank and one spiked matrix sample for ongoing precision and recovery (OPR) determination. Clean sand was used as the matrix for procedural blanks and OPR. A duplicate was analyzed for every analysis batch that had to agree to within ± 20 % of prior measurements on identical samples. Ion ratios (quantification-to-confirmation ion) had to fall within ±15% of the theoretical values for positive identification of all target analytes. The minimum signal-to-noise ratio was 3:1. Native analytes having an exact labeled surrogate had to elute 0-3 seconds after the labeled standard. Retention times for other congeners had to fall within 5 seconds of that in the daily calibration standard. In addition to these standard procedures, a duplicate of composite biosolids sample # 1 was prepared to serve as a blind (unknown) duplicate to the commercial laboratory and was shipped along with the other composites. This sample served to evaluate analysis precision of the method in addition to the laboratory's QA/QC protocol. Precision between samples and duplicates was expressed as relative percent difference (RPD), which was calculated using the following expression.

$$RPD[\%] = \frac{\left|C_{sample} - C_{duplicate}\right| * 100}{\frac{C_{sample} + C_{duplicate}}{2}} \qquad (1)$$

Where, C_{sample} and $C_{duplicate}$ are the concentration detected in the original sample and in its duplicate, respectively. The blinded-RPD was calculated from the analysis of a blind-duplicate of composite sample #1. A duplicate of composite #3 was additionally analyzed by the laboratory as part of their QA/QC protocol to evaluate analysis precision (non-blinded RPD).

Modeling annual load of chemicals in biosolids. Annual load was estimated for all detected analytes based on the annual biosolids production of 5.1-6.4 million metric dry tonnes (5.6-7 million dry U.S. tons) estimated for the year 2001 in the U.S. [2-4].

Annual load = (mean analyte concentration in biosolids) ng/kg * (10^{-12} kg/ng) * (5.1-6.4 x 10^{9} kg of biosolids/year) (2)

Table S1. Facilities sampled in the 2001 national sewage sludge survey

Facility name	City	State	Facility name	City	State
Sacramento Regional WWTP	Elk Grove	CA	Metropolitan Council – Metro	Saint Paul	MN
Fallbrook Public Utility District	Fallbrook	CA	Crocker WWTP	Crocker	MO
Manteca WQCF	Manteca	CA	Mason Farm WTP	Carrboro	NC
Central Contra Costa Sanitary District	Martinez	CA	Whiteville WWTP	Whiteville	NC
Fairfield Suisun Sewer District	Suisun city	CA	Burwell WWTF	Burwell	NE
Boulder – 75th St WWTP	Boulder	CO	Middletown Sewerage Authority	Belford	NJ
Steamboat Springs	Steamboat Springs	CO	Joint Meeting Sewage Treatment	Elizabeth	NJ
Rocky Hill WPCP	Hartford	CT	Passaic Valley Sewerage Commision	Newark	NJ
Waterbury WPCF	Waterbury	CT	Bowery Bay WPC	Corona Queens	NY
DC WASA (Blue Plains)	Washington	DC	Hunt's Point WPC	Corona Queens	NY
Mulberry STP	Mulberry	FL	Cayuga Heights WWTP	Ithaca	NY
Escambia County – Main Street WTP	Pensacola	FL	Brewster WWTP	Mahopac	NY
St. Petersburg SW Treatment Plant	St. Petersburg	FL	NEORSD – Southerly	Cleveland	OH
Sunrise Sweage Treatment Plant No. 1	Sunrise	FL	Brentwood Estates STP #24	Cuyahoga Falls	OH
R.M. Clayton WPCP	Atlanta	GA	Delphos	Delphos	OH
Buford Westside WPCP	Buford	GA	Massillon	Massillon	OH
Cartersville WPCP	Cartersville	GA	North Olmsted	North Olmsted	OH
Dekalb Co – Snapfinger Cr WPCP	Decatur	GA	Port Clinton	Port Clinton	OH
Garden City WPCP	Garden City	GA	Twin Lakes WWTP	Ravenna	OH
Gwinnett Co Jackson Cr	Lilburn	GA	Thornville	Thornville	OH
Ocmulgee WPCP	Warner Robins	GA	West Carrollton	West Carrollton	OH
Boise	Boise	ID	Blackwell	Blackwell	OK
Belleville STP #1	Belleville	IL	Lebanon	Lebanon	OR
MWRDGC Stickney STP	Cicero	IL	Portland	Portland	OR
Jacksonville STP	Jacksonville	IL	Burnham STP	Burnham	PA
Morris STP	Morris	IL	Downingtown Area Regional Authority	Downingto wn	PA
Tolono STP	Westville	IL	Girard Boro	Girard	PA
Evansville STP – Westside	Evansville	IN	Kiski Valley Water Pollution Control	Leechburg	PA

Frankton Municipal STP	Frankton	IN	Philadelphia Water Dept (SW)	Philadelphia	PA
Hammond Municipal STP	Hammond	IN	Philadelphia Water Dept (NE)	Philadelphia	PA
Muncie Sanitary District	Muncie	IN	Allengheny County Sanitary Authority	Pittsburgh	PA
Terre Haute Municipal STP	Terre Haute	IN	Narragansett Bay Commission – Bucklin	Providence	RI
Union city Municipal STP	Union City	IN	Florence – Pee Dee River Plant	Florence	SC
Oakland STP	Topeka	KS	WCRSA/Pelham WWTF	Greenville	SC
Shepherdsville STP	Shepherdsville	KY	Brooking	Brookings	SD
Billerica WWTP	Billerica	MA	Sioux Falls	Sioux Falls	SD
Fall River WWTF	Fall River	MA	Andrews STP	Andrews	TX
Medfield WWTP	Medfield	MA	Del Rio – San Felipe	Del Rio	TX
Pittsfield WWTP	Pittsfield	MA	Navasota, Grimes Co. STP	Navasota	TX
Patapsco WWTP	Baltimore	MD	Orange, Jackson St WWTP	Orange	TX
South Portland WPCF	South Portland	ME	Brazos River Authority (Waco)	Waco	TX
Dowagiac WWTP	Dowagiac	MI	Fredericksburg City STP	Fredericksburg	VA
Iron Mountain – Kingsford WWTP	Kingsford	MI	Augusta County Service Authority	Verona	VA
Genesee County – Ragnone WWTP	Montrose	MI	HRSD – James River STP	Virginia Beach	VA
Port Huron WWTP	Port Huron	MI	HRSD – Chesapeake/Elizabeth STP	Virginia Beach	VA
Wyandotte WWTP	Wyandotte	MI	Metropolitan King County	Renton	WA
Western Lake SSD	Duluth	MN	Greenbrier County PSD No 2	Rainelle	WV

Table S2. PBDD/Fs ions monitored, retention time, and surrogates used.

Analytes	Quantified Against	Retention Time (min:sec)	Quantification Ion (Qt)	Confirmation Ion (Cf)	Qt/Cf ratio
Dioxins					
2,3,7,8-Tetrabromo dibenzo-*p*-dioxin (TBDD)	$^{13}C_{12}$-2,3,7,8-TBDD	22:59	497.7	499.7	0.69
1,2,3,7,8-Pentabromo dibenzo-*p*-dioxin (PeBDD)	$^{13}C_{12}$-1,2,3,7,8-PeBDD	37:27	577.6	579.6	1.03
1,2,3,4,7,8-/1,2,3,6,7,8-Hexabromo dibenzo-*p*-dioxin (HxBDD)	$^{13}C_{12}$-1,2,3,4,7,8-HxBDD	44:17	655.5	657.5	0.77
1,2,3,7,8,9-HxBDD	$^{13}C_{12}$-1,2,3,4,7,8-HxBDD	44:39	655.5	657.5	0.77
Octbromo dibenzo-*p*-dioxin (OBDD)	$^{13}C_{12}$-1,2,3,4,7,8-HxBDD	53:19	813.3	815.3	0.82
Furans					
2,3,7,8-Tetrabromo dibenzofuran (TBDF)	$^{13}C_{12}$-2,3,7,8-TBDF	21:50	481.7	483.7	0.69
1,2,3,7,8-Pentabromo dibenzofuran (PeBDF)	$^{13}C_{12}$-2,3,4,7,8-PeBDF	35:27	561.6	563.6	1.03
2,3,4,7,8-PeBDF	$^{13}C_{12}$-2,3,4,7,8-PeBDF	36:58	561.6	563.6	1.03
1,2,3,4,7,8-Hexabromo dibenzofuran (HxBDF)	$^{13}C_{12}$-1,2,3,4,7,8-HxBDD	43:33	639.5	641.5	0.77
1,2,3,4,6,7,8-Heptabromo dibenzofuran (HpBDF)	$^{13}C_{12}$-1,2,3,4,7,8-HxBDD	48:18	719.4	721.4	1.03
Octabromo dibenzofuran (OBDF)	$^{13}C_{12}$-1,2,3,4,7,8-HxBDD	53:23	797.3	799.3	0.82
Recovery Standard					
$^{13}C_{12}$-1,2,3,7,8,9-HxCDD		20:54	401.856	403.853	1.24
Interfering Brominated Diphenylethane (BDPE) Ions and Lock Masses					
Hexa-BDPE			643.5		
Hepta-BDPE			721.4		
Octa-BDPE			799.4		
Nona-BDPE			879.3		
Deca-BDPE			957.2		
Lock Mass			492.9		
Lock Mass			566.9		
Lock Mass			654.9		
Lock Mass			730.9		
Lock Mass			804.9		

Table S3. Method performance of PBDD/Fs analysis in U.S. biosolids

Compound	Method Detection Limit (pg/g dw)	Matrix Spike Recovery (%)	Relative Percentage Difference (%)	
			Non-blinded	**Blinded**
123478/123678-HxBDD	10	93.2	-	-
1,2,3,7,8,9-HxBDD	10	64.4	-	-
2,3,7,8-TBDF	2	207	4	-
1,2,3,7,8-PeBDF	10	88.1	9	-
2,3,4,7,8-PeBDF	10	83.4	10	43
1,2,3,4,7,8-HxBDF	10	67.3	12	14
1,2,3,4,6,7,8-HpBDF	10	50.1	55	16

- Represent non-detects in samples

Table S4. Detection of OBDD and OBDF in biosolids composites (for information only)

Compound	Concentration (pg/g dw) Avg. (Min, Max)	Detection Frequency (%)
OBDD	231 (47, 574)	60
OBDF	14,980 (3590, 29400)	40

Table S5. TEQ of PBDD/Fs estimated from World Health Organization (WHO)-TEFs of PCDD/Fs

Compound	WHO-TEF for PCDD/Fs	TEQ of PBDD/Fs in biosolids (ng/kg) Avg (Min, Max)
Dioxins		
1,2,3,4,7,8/1,2,3,6,7,8-HxBDD	0.1	4.8 (0.5, 20)
1,2,3,7,8,9-HxBDD	0.01	2.7 (0.5, 12)
OBDD	0.0001	0.0005*
Furans		
2,3,7,8-TBDF	0.1	0.7 (0.1, 1.7)
1,2,3,7,8-PeBDF	0.05	0.7 (0.3, 2.2)
2,3,4,7,8-PeBDF	0.5	25 (2.5, 100)
1,2,3,4,7,8-HxBDD	0.1	33 (3.9, 129)
1,2,3,4,6,7,8-HpBDF	0.01	95 (5.8, 409)

Table S6. TEQ of polybrominated diphenylether (PBDEs) in U.S. biosolids from the 2001 NSSS

Compound	**Concentration in biosolids (µg/kg-dw)[a] Avg (Min, Max)**	**Relative Potency (REP)[b]**	**TEQ (ng/kg dw) Avg (Min, Max)**
BDE-47	789 (314, 1120)	7.1E-7	0.56 (0.22, 0.80)
BDE-77	0.06 (0.05, 0.08)	3.2E-3	0.19 (0.16, 0.26)
BDE-99	1004 (402, 1510)	5.9E-6	5.9 (2.4, 8.9)
BDE-100	179 (75.3, 229)	2.4E-5	4.3 (1.8, 5.5)
BDE-119	1.7 (1.2, 2.4)	3.5E-5	0.06 (0.04, 0.08)
BDE-153	103 (43.9, 138)	4.3E-6	0.44 (0.19, 0.59)

[a]Concentrations source: [5]
[b]REP compiled and reported elsewhere [6]

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