THE UNIVERSITY OF MICHIGAN DIOXIN EXPOSURE STUDY: A FOLLOW-UP INVESTIGATION OF A CASE WITH HIGH SERUM CONCENTRATION OF 2,3,4,7,8-PENTACHLORODIBENZOFURAN

Franzblau Alfred[1], Hedgeman Elizabeth[1], Knutson Kristine[1], Towey Tim[2], Chen Qixuan[3], Hong Biling[1], Adriaens Peter[4], Demond Avery[4], Garabrant David Hay[1], Gillespie Brenda Wilson[3], Lepkowski James[5]

[1]Dept of Environmental Health Sciences, University of Michigan School of Public Health, Ann Arbor, Michigan 48109-2029 USA; [2]LimnoTech, Ann Arbor, Michigan 48108-9799 USA; [3]Dept of Biostatistics, University of Michigan School of Public Health, Ann Arbor, Michigan 48109-2029 USA; [4]Dept of Civil and Environmental Engineering, University of Michigan College of Engineering, Ann Arbor, Michigan 48109-2135 USA; [5]Institute for Social Research, University of Michigan, Ann Arbor, Michigan 48109-1248 USA.

Introduction

As part of the University of Michigan Dioxin Exposure Study (UMDES) the 29 congeners of polychlorinated dibenzo-*p*-dioxins (PCDDs), polychlorinated dibenzofurans (PCDFs) and dioxin-like polychlorinated biphenyls (PCBs) that have consensus toxic equivalency factors (TEFs) were measured in serum of 946 subjects who were a representative sample of the general population in five Michigan counties. The study was motivated because of concerns about possible human exposure to dioxin-contaminated sediments in the Tittabawassee River (TR). Most of the total toxic equivalency (TEQ) in TR floodplain sediments is due to two furan congeners, 2,3,7,8-TCDF and 2,3,4,7,8-penta-CDF.[1,2] The half life of 2,3,7,8-TCDF is short and thus it does not accumulate in humans; the penta-CDF congener has a prolonged serum half life in humans (~7.8-10 years) and therefore can serve as a biomarker of exposure to contaminated sediment.[3] The individual with the highest adjusted serum level of 2,3,4,7,8-penta-CDF in the UMDES [42.5 parts per trillion, or 4.29 studentized residuals above the log-normalized mean of the control population after adjustment for age, age^2, and body mass index (BMI)] reported a unique exposure history involving consumption of beef and vegetables raised in the floodplain (FP) of the TR. This report describes this person's results and results of a follow-up investigation of friends and family members who also reported eating the beef and vegetables.

Materials and Methods

The UMDES involved a two-stage clustered random sampling design to recruit subjects from five counties in the State of Michigan, USA. Eligible subjects were required to be at least 18 years old, and to have lived in their homes for at least 5 years. The main study involved an hour-long interview and obtaining blood, house dust and soil samples for chemical analyses from eligible subjects. As noted, 946 subjects provided blood samples that were analyzed for PCDDs, PCDFs and PCBs by Vista Analytical Laboratory (El Dorado Hills, California) using modified United States Environmental Protection Agency methods 8290 and 1668, Revision A.[4,5] Serum results are reported in parts per trillion (ppt) on a lipid adjusted basis and soil results are reported in ppt on a dry weight basis. TEQ values are calculated with all 29 dioxin-like congeners, using 2005 TEFs.[6]

As noted above, the index case was the person found to have the highest adjusted serum level of 2,3,4,7,8-penta-CDF in the UMDES. As part of an effort to better understand why his serum level of 2,3,4,7,8-penta-CDF was elevated, he underwent a follow-up interview, at which time his unique dietary history involving consumption of beef and vegetables from the TR FP was discovered (described in detail below). Subsequently, friends and family members of the index case who were most likely to have consumed beef and/or vegetables were invited to participate in this follow-up investigation. Subjects were interviewed about diet (particularly consumption of beef and/or vegetables from the TR flood plain), occupation, residential history, personal habits (e.g., smoking), height, weight and change in weight, breast feeding, hobbies and recreational activities in or near the TR. Subjects were also invited to undergo phlebotomy for measurement of PCDDs, PCDFs and PCBs in serum. One subject had already undergone blood testing in 2004 as part of the UMDES and was not retested. All subjects provided written consent that had been reviewed and approved by the University of Michigan Health IRB.

Results and Discussion

The index case identified 15 friends and family members who were most likely to have consumed beef and/or vegetables from the TR FP; all 15 were invited and agreed to participate in the follow-up investigation.

The index case owned property in the TR FP that fronted on the river and in about 1984 or 1985 he began to raise cows on his land. Each year in the spring he bought 2-3 calves from local farms (not in the FP) and typically raised them for ~18 months. He had 4-6 cows on the property at a time, and the cows routinely roamed and grazed in areas that flooded annually. They ate FP grass, grain not grown in the FP, and hay from a nearby field that was near the FP but did not flood. The cattle were slaughtered at a local commercial abattoir. The meat always passed federal inspection, but the livers never passed, and no organ meats were consumed. The meat was never sold, but was distributed to friends and family members. He stopped raising beef in about 1996. At the time of this follow-up investigation (2008) no meat samples were available for chemical analyses. Beginning in the early 1980's he started a vegetable garden in an area of the TR FP that regularly flooded. Vegetables included: asparagus, tomatoes, cucumbers, green beans, corn, radishes, potatoes, beets, green peppers, onions, Swiss chard, watermelon and pumpkins. The vegetables were shared with the same friends and family members. Use of the vegetable garden was largely discontinued in about 1997, except for occasional tomato plants and asparagus.

Among properties in the UMDES study that fronted on the TR, 30% had a near-river or FP soil sample with a TEQ above 1,000 ppt, and 90% had a near river sample above 90 ppt. The mean and median of near-river samples among such properties were 975 ppt and 674 ppt, respectively. The percentage of such properties that had garden soil samples with a TEQ above 1,000 ppt and 90 ppt were 6% and 37%, respectively. The mean and median of garden samples among such properties were 227 ppt and 23.1 ppt, respectively.

The index case had the highest serum concentration of 2,3,4,7,8-penta-CDF among all subjects in this follow-up study (42.5 ppt, or 4.29 studentized residuals above the adjusted mean of the UMDES referent population). One other subject (case #2) had a serum concentration of 2,3,4,7,8-penta-CDF that was more than 3 studentized residuals above the mean (14.7 ppt, or 3.48 studentized residuals above the mean); all remaining subjects' serum concentrations of 2,3,4,7,8-penta-CDF were less than 2.5 studentized residuals above the mean. The mean of the 2,3,4,7,8-penta-CDF studentized residuals among the 16 subjects was 1.17 above the mean of the referent population (see Table 1).

Table 1 also includes an estimate of the percentage contribution to each subject's TEQ from 'extra' 2,3,4,7,8-penta-CDF, i.e., the amount of 2,3,4,7,8-penta-CDF in serum above the mean of the referent population after adjustment for age, age^2 and BMI. The percentage contribution of extra 2,3,4,7,8-penta-CDF to the TEQ ranges from -1.7% to +20.4%, with a mean and median contribution of 4.9% and 2.6%, respectively, among the 16 cases.

Subjects were asked about the average number of beef meals consumed per week or per month during the time period in which they ate the beef from the index case, and then this number was multiplied by the total number of weeks or months of beef consumption to estimate the total number of beef meals consumed. The estimated number of beef meals ranged from zero to 2248, with a mean and median of 1109 and 1056 beef meals, respectively (see Table 2). Subjects were also asked about the number of meals with vegetables from the index case's garden consumed per week or per month during the time period in which they ate vegetables from the garden of the index case, and a total number of vegetable meals was computed in a similar manner. The estimated number of meals with vegetables from the garden of the index case ranged from 44 to 1350, with a mean and median of 570 and 506 meals, respectively. Since consumption of potentially contaminated beef stopped in 1996 (and most consumption of vegetables stopped in about 1997), but serum measurements were not performed until 2008 (2005 for the index case, 2004 for case #5) it is possible that the present results underestimate past serum levels of 2,3,4,7,8-penta-CDF.

For comparison, the overall mean, median, 95th percentile and maximum for serum 2,3,4,7,8-penta-CDF in the University of Michigan Dioxin Exposure Study control population were 6.0 ppt, 5.4 ppt, 13.0 ppt, and 26.2 respectively (not adjusted for age or BMI).

Most cases included in this follow-up investigation had higher serum concentrations of 2,3,4,7,8-penta-CDF than the mean of the control population (adjusted for age, age^2 and BMI), but, with two exceptions, all were less than 2.5 studentized residuals above the mean. The mean extra or excess contribution of 2,3,4,7,8-penta-CDF to the TEQ was 4.9%, and was less than 21% in all cases. Overall, prolonged regular consumption of beef and/or vegetables raised in the flood plain of the Tittabawassee River can be an important route of exposure to dioxin contamination in the soil. However, the contribution of such exposure to TEQ above background appears to be modest in most cases, which is consistent with previous studies.[7,8]

Acknowledgements and References

Financial support for this study comes from the Dow Chemical Company through an unrestricted grant to the University of Michigan. The authors acknowledge Ms. Sharyn Vantine for her continued assistance and Drs. Linda Birnbaum, Ronald A. Hites, Paolo Boffetta and Marie Haring Sweeney for their guidance as members of our Scientific Advisory Board.

1. Hilscherova K, Kannan K, Nakata H, Hanari N, Yamashita N, Bradley PW, McCabe JM, Taylor AB, Giesy JP. 2003. *Environ Sci Technol* 37:468–474.
2. Demond A, Adriaens P, Towey T, Chang S-C, Hong B, Chen Q, Chang C-W, Franzblau A, Garabrant D, Gillespie B, Hedgeman E, Knutson K, Lee CY, Lepkowski J, Olson K, Ward B, Zwica L, Luksemburg W, Maier M. 2008. *Environ Sci Technol* (in press).
3. Ogura I. 2004. *Organohalogen Compounds*. 66:3376-3384.
4. U.S. EPA (United States Environmental Protection Agency). 1994. Available: http://www.epa.gov/sw-846/pdfs/8290.pdf [accessed 16 August 2007]
5. U.S. EPA (United States Environmental Protection Agency). 1999. EPA Publication No. EPA-821-R-00-002. United States Environmental Protection Agency.
6. Van den Berg MLS, Birnbaum LS, Denison M, De Vito M, Farland W, Feeley M, Fiedler H, Hakansson H, Hanberg A, Haws L, Rose M, Safe S, Schrenk D, Tohyama C, Tritscher A, Toumisto J, Tysklind M, Walker N, Peterson RE. 2006. *Toxicological Sciences* 93(2):223-241.
7. Ewers U, Wittsiepe J, Hens-Bischoff G, Balzer W, Alger B, Urban U. 1997. *Gesundheitswesen*. 59(1):41-50.
8. Ewers U, Wittsiepe J, Schrey P, Gatzert U, Hinz S, Csicsaky M. 1996. *Gesundheitswesen*. 58(8-9):465-469.

Table 1. Summary of TEQ and 2,3,4,7,8-penta-CDF Concentrations (in ppt on a lipid adjusted basis) in Serum Among People Who Consumed Beef and Vegetables

Case Number	Age at Blood Draw	WHO-TEQ	WHO-TEQ Studentized Residuals*	2,3,4,7,8-Penta-CDF	Penta-CDF Studentized Residuals*	Predicted Penta-CDF**	Percent Contribution to TEQ
1 (Index)	≥60	52.4	2.22	42.5	4.29	6.8	20.4
2	30-44	17.6	1.21	14.7	3.48	3.5	19.3
3	30-44	18.1	1.02	9.5	2.25	3.8	9.4
4	≥60	44.0	0.32	21.4	1.79	9.9	7.9
5	≥60	69.4	1.45	20.1	1.59	10.2	4.3
6	≥60	33.1	0.81	12.9	1.47	7.2	5.1
7	45-59	24.1	0.53	8.84	1.02	6.0	3.5
8	45-59	24.3	0.21	8.42	0.63	6.7	2.1
9	45-59	13.7	-0.41	6.34	0.69	4.9	3.1
10	45-59	31.0	1.02	7.11	0.41	6.4	0.7
11	45-59	19.6	0.02	6.72	0.46	5.9	1.3
12	≥60	22.5	-0.29	8.45	0.45	7.3	1.5
13	45-59	14.8	-0.46	6.03	0.38	5.4	1.4
14	45-59	20.4	0.10	6.29	0.21	6.0	0.5
15	≥60	19.3	-0.72	6.94	-0.05	7.3	-0.6
16	45-59	19.2	-0.28	5.34	-0.37	6.4	-1.7

*Distance from the lognormal mean of the referent population after adjustment for age, age^2 and BMI;
**Predicted mean 2,3,4,7,8-penta-CDF (in ppt) for persons with the same age and BMI of each subject based on the referent population

Table 2 Reported Number of Beef and Vegetable Meals

	Floodplain Beef Consumption			Floodplain Vegetable Consumption		
Case Number	Years Consumed	Avg Meals per Year	Approx. Total Beef Meals	Years Consumed	Avg Meals per Year	Approx Total Veg. Meals
1 (Index)	1985-1996	48-96	792	1982-2008	40	1040
2	1985-1996	144-192	1848	1982-2008	46	1196
3	1985-1996	144-180	1848	1982-2008	6	156
4	1985-1996	104-280	1559	1982-2008	26	676
5	1985-1996	2	22	1997-2008	4	44
6	1985-1996	156-20	2002	1982-1993	46	506
7	1985-1996	144	1584	1982-1997	59	885
8	1985-1996	208	2288	1982-1997	26	390
9	None	0	0	1998-2008	52	520
10	1985-1996	52	572	1982-1997	13	195
11	1985-1996	52	572	1982-1997	33	495
12	1986-1990	96	384	1982-1997	90	1350
13	1990-1996	240-300	1656	1998-2008	91	910
14	1985-1996	52	572	1982-1997	3	45
15	2 yrs	360	720	2000-2006	unsure	-
16	1985-1996	120	1320	1982-2006	6	144