TCDD BLOOD LEVELS, POPULATION CHARACTERISTICS, AND INDIVIDUAL ACCIDENT EXPERIENCE

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1. BACKGROUND

After the July 10th, 1976 accident in Seveso, the contaminated area was divided into three zones (A, B, and R with about 750, 5,000, and 30,000 inhabitants, respectively) based on decreasing soil levels of 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD)[1]. Within the frame of a collaborative molecular epidemiology study, individual exposure to dioxin and its relationship to demographic and personal characteristics were evaluated on 126 healthy subjects from zones A and B and a surrounding non-contaminated territory.

2. MATERIAL AND METHODS

Sixty-three subjects from the highly exposed area (zones A and B) and 64 from the outside non-contaminated area were randomly selected from computerized rosters. Sixty-two subjects were males and 64 females; approximately, half of the subjects were smokers and half non-smokers.

TABLE 1. POPULATION CHARACTERISTICS

	ZONES A + B				OUTSIDE			
AGE	M	F	SMK^+	SMK^-	M	F	SMK^+	SMK^-
20-29	6	6	7	5	7	8	8	7
30-39	6	2	5	3	9	4	8	5
40-49	7	10	6	11	2	9	3	8
50-59	3	3	3	3	3	5	4	4
60-69	6	5	4	7	5	5	5	5
70+	4	5	3	6	5	2	5	2
Total	32	31	28	35	31	33	33	31

SMK^+ = Current smokers SMK^- = Non-smokers

After signing an informed consent, subjects responded to a questionnaire on residence history, smoking habits, occupational exposures, health history, medications, and accident experience, and donated a blood sample. Personal characteristics of study subjects are illustrated in Table 1.
TCDD blood levels were measured in serum following the method by Patterson and collaborators (1987)[2]. Results were lipid adjusted. TCDD measurements were performed on 122 samples. After quality control tests, 12 samples were discarded. Of the remaining samples, TCDD levels for 23 subjects were not detectable. Extrapolated TCDD levels for subjects of the polluted zones (A and B) at the time of the accident were calculated using a half-life of 7.1 years[3] without correction for background TCDD levels. Multiple regression analyses were performed after logarithmic transformation of TCDD values; for samples with non-detectable values, serum TCDD was estimated by dividing the lipid-adjusted detectable limits by $\sqrt{2}$[4].

3. RESULTS

As expected, the highest TCDD serum levels were seen in zone-A, but also in zone-B TCDD levels were definitely higher than in the area outside the accident scenario ($p<0.01$, Kruskal-Wallis test) (Table 2). The levels of the other congeners were not different across zones. The pattern of TCDD serum levels was almost identical when individual zone assignment was performed according to any of the following criteria: i) official residence (local vital statistics); ii) place of living in the period of the accident (questionnaire); iii) presence in the area on the day of the accident (questionnaire). These findings support the exposure categorization by zone of residence which has been adopted in the mortality[5] and cancer incidence[6] follow-up, and in the clinical laboratory investigations[7,8].

TABLE 2. SERUM TCDD BY RESIDENCE

ZONE	No.	TCDD (ppt) CURRENT MEAN (Extrapolated)*	S.D. (Extrapolated)*	MEDIAN (Extrapolated)*
A	6	61.5 (333.8)	29.5 (163.2)	71.5 (388.7)
B	52 (43)#	16.8 (111.4)	15.8 (83.6)	12.5 (77.6)
Outside	52	5.3	4.0	5.5

* Assumed half-life = 7.1 yrs (Pirkle, 1989)
# Subjects with detectable TCDD levels only

TCDD median levels were significantly higher in females ($p<0.01$). Interestingly, several other polychlorinated dibenzo-p-dioxins and furans were also found to be statistically significantly increased in women. Table 3 shows TCDD levels by zone and gender. Differences in zones B and Outside were statistically significant. TCDD also varied with age ($p<0.01$) (Table 4).
Gender, age and zone were significantly associated with TCDD levels in a multiple regression model ($p<0.01$).
Analysis are being performed on the relationship of TCDD levels to reproductive factors (menopausal status, parity, oral contraceptives, etc.); consumption of vegetables grown in the

kitchen-garden and /or meat from courtyard animals bred in the area at the time of the accident or shortly after; body mass index, and other factors (tobacco smoke, occupational exposures, etc.) potentially relevant to dioxin inter-individual variability and gender difference.

TABLE 3. SERUM TCDD BY RESIDENCE AND GENDER

ZONE	GENDER	No.	TCDD (ppt) MEAN	S.D.	MEDIAN
A	F	2	63.0	25.0	63.0
	M	4	60.7	35.2	71.6
B	F	27	23.5	16.3	18.0
	M	25	9.5	11.6	6.3
Outside	F	27	6.2	3.1	6.3
	M	25	4.4	4.6	4.6

TABLE 4. SERUM TCDD BY AGE

AGE	No.	TCDD (ppt) MEAN	S.D.	MEDIAN
20-29	22	12.1	16.8	6.9
30-39	15	5.6	6.7	4.9
40-49	25	14.9	19.4	8.3
50-59	13	5.5	4.0	5.5
60-69	19	13.2	11.2	9.2
70+	16	29.5	23.0	23.7

4. DISCUSSION

Measurements based on a random sampling design showed that people living in the accident scenario have TCDD blood levels substantially higher than those in the surrounding territory. Consequently, investigations going on in this population (mortality, cancer incidence, molecular epidemiology) might, with high probability, contribute to elucidate effects on human health of TCDD and its mode of action.

The reason why women have higher TCDD and other congeners serum levels is under investigation. TCDD is hypothesized to act as pro- and anti-estrogen[9-11] although the exact mechanism of action is unclear. A possible decrease of estrogen-related tumors have been suggested by incidence and mortality studies on exposed subjects[5-6]. Toxic effects on reproductive system have also been hypothesized[12]. Thus, the finding of increased TCDD levels, particularly in females, can have important implications for public health and calls for further research.

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5. REFERENCES

1. Bertazzi P.A., Di Domenico. Chemical, environmental and health aspects of the Seveso, Italy, accident. In: A. Schechter (ed.) *Dioxin and Health.* Plenum Press, New York, 1994, pp. 587-632.
2. Patterson D.G.Jr., Hampton L., Lapezza C.R.Jr., Belser W.T., Green V., Alexander L., Needham L.L. High-resolution gas chromatographic/high resolution mass spectrometric analysis of human serum on a whole-weight and lipid basis for 2,3,7,8-tetrachlorodibenzo-p-dioxin. *Anal. Chem.* 59, 2000-2005 (1987).
3. Pirkle J.L., Wolfe W.H., Patterson D.G., Needham L.L., Michalek J.E., Miner J.C., Peterson M.R., Phillips D.L. Estimates of the half-life of 2,3,7,8-tetrachlorodibenzo-p-dioxin in Vietnam veterans of operation ranch hand. *J.Toxicol. Environ. Health* 27, 165-171 (1989).
4. Hornung R.W, Reed L.D. Estimation of average concentration in the presence of non detectable values. *Appl Occup. Environ. Hyg.* 5(1), 48-51 (1990).
5. Bertazzi P.A., Zocchetti C., Pesatori A.C., Guercilena S., Sanarico M., Radice L. Ten-year mortality study of the population involved in the Seveso incident in 1976. *Am. J. Epidemiol.* 129, 1187-1200 (1989).
6. Bertazzi P.A., Pesatori A.C., Consonni D., Tironi A., Landi M.T., Zocchetti C. Cancer incidence in a population accidentally exposed to 2,3,7,8-tetrachlorodibenzo-para-dioxin. *Epidemiology* 4, 398-406 (1993).
7. Mocarelli P., Marocchi A., Brambilla P., Gerthoux P.M., Colombo L., Mondonico A., Meazza L. Effects of dioxin exposure in humans at Seveso, Italy. *Banbury Report 35, Biological basis for risk assessment of dioxins and related compounds.* Could Spring Harbour Laboratory Press, 1991.
8. Mocarelli P., Marocchi A., Brambilla P., Gerthoux P.M., Beretta C., Colombo L., Bertona M., Sarto C., Tramacere P., Mondonico A., Crespi C., Signorini S., Brivio R., Carreri V., Meazza L. Human data derived from the Seveso accident - Relevant for human risk assessment. *Toxic Substances Journal* 12, 151-179 (1992).
9. Safe S., Astroff B., Harris M. 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD) and related compounds as antiestrogen: characterization and mechanism of action. Pharmacol. Toxicol. 69, 400-409 (1991).
10. Umbreit T. H., Gallo M.A. Phisiological implication of estrogen receptor modulation by 2,3,7,8-tetrachlorodibenzo-p-dioxin. *Toxicology Let.* 42, 5-14 (1988).
11. Adami H.O., Lipworth L., Titus-Ernstoff L., Hsieh C., Hamberg A., Ahlborg U., Baron J., Trichopoulos D. Organochlorine compounds and estrogen-related cancers in women. *Cancer Causes and Control 6*, 551-566 (1995).
12. Lilienfeld D.E., Gallo M.A. 2,4-D, 2,4,5-T, and 2,3,7,8-TCDD: an overview. *Epidemiologic reviews* 11, 28-58 (1989).