

BEHAVIOR OF PCBS/PCDFS IN PATIENTS WITH YUSHO PCB POISONING AND CLINICAL SYMPTOMS

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Introduction

Two mass-food poisoning called Yusho and Yucheng occurred in Japan¹ and Taiwan² in 1968 and 1979, respectively, and officially approved patients were counted for more than 1850 and 2000, respectively. Yusho and Yucheng patients steadily consumed the particular rice oil for up to about 210 and 260 days, respectively, and average latent periods before the typical illness became apparent were estimated as 71 and 80 days, respectively. According to the survey on 141 Yusho patients³ and 98 Yucheng patients⁴, average total intake per capita of PCBs, PCDFs and PCQs were estimated to be 633, 3.4 and 596 mg, respectively, to Yusho patients and 973, 3.84 and 490 mg, respectively, to Yucheng patients. Average total intakes of TEQ (toxic equivalent quantity for 2,3,7,8-tetrachlorodibenzo-p-dioxin), calculated from the concentrations of PCDF/PCB congeners and individual TEQ factors, are estimated to be 0.62 mg in a Yusho patient and 0.27 mg in a Yucheng patient. Two groups of patients with Yusho or Yucheng ingested similar levels of dioxin toxicants and showed severe typical symptoms like acneiform eruption, dermal pigmentation, increased eye discharge and others. It is therefore important to examine the longtime behavior of PCBs and PCDFs in Yusho and Yucheng patients and persistent state of these symptoms for understanding the behavior of PCBs and PCDFs in human body and the toxic effects of these chemicals on humans.

Materials and methods

Blood sample During the period 1982 to 1998 for Yusho and 1980 to 1995 for Yucheng, serial samples of whole blood were collected from affected individuals usually at the health center in Fukuoka or Taichung for their periodic health examination. Ten ml of blood was collected from Yusho and Yucheng patients and kept frozen at -20°C until analysis. The concentrations of PCBs and PCDFs in the blood of the following individuals were used for evaluating the elimination profile in human body. Five Yusho patients: all female, age from 31 to 51 at the time of exposure and three Yucheng patients: 2 male and 1 female, age from 17 to 33 at the exposure. All the 8 Yusho and Yucheng patients had typical Yusho-type severe chloracne at the time of exposure. Blood samples analyzed were collected from the five Yusho patients 8 to 11 times in the 16 years and from the three Yucheng patients 7 to 8 times in the 15 years.

PCB analysis Blood sample was added with 5 ng of 2,2',3,4,5,5',6-hepta-CB or four ^{13}C labeled PCBs as internal standards and saponified in 1N NaOH ethanol solution. The mixture was extracted with n-hexane 5 ml twice. The n-hexane extract was washed with 2 ml of water, dried over sodium sulfate and chromatographed on 1 g of silica gel, eluting with 20 ml of n-hexane. The n-hexane eluate was concentrated to a small volume and analyzed for PCB congeners by gas chromatograph/electron capture detector or gas chromatograph/quadrupole mass spectrometer.

PCDF analysis To the blood sample added the internal standards consisting of six ^{13}C -PCDDs, seven ^{37}Cl -PCDFs and three ^{13}C -planarPCBs, and then added ethanol, n-hexane and aqueous saturated ammonium sulfate. The mixture was homogenized and, after separation, the upper n-hexane portions were combined, washed with water, and evaporated to dryness for obtaining the extractable lipid. The lipid material was redissolved in n-hexane and treated with concentrated sulfuric acid. The organic extract passed through multi layer column of cesium hydroxide/silica gel and sulfuric acid/silica gel, separated chromatographically the PCDDs/PCDFs from the major portion of PCBs on Florisil column, and separated the toxic planar components from non planar analytes on a column of Carbopack C carbon dispersed silica. The final extract was added with the recovery standards and injected to high separation gas chromatography / high resolution mass spectrometry (VG Analytical MS or Finnigan MAT 90)⁵.

Results

The concentrations (fat base) of three PCDF congeners were determined in the blood of three Yucheng patients from 1980 to 1995 and five Yusho patients from 1982 to 1998. The whole base concentrations of six PCB congeners were also determined in the same blood samples of Yucheng and Yusho patients. Table 1 lists the biological half-lives of PCDF and PCB congeners in Yucheng and Yusho patients calculated on assuming first order exponential elimination in one compartment model⁶. The three PCDF congeners eliminated in Yucheng patients at about 3.0 years of biological half-life for 5700 days (15.6 years) after the contamination. As observed in Yusho patients, from 5000 days after the contamination, half-lives of these PCDF congeners were prolonged to 3.5-7.7 years. Half-lives of the five PCB congeners (4.2-6.0 years), except for 2,3',4,4',5-penta-CB, were longer than those of PCDF congeners in the same Yucheng patients. After the concentrations of PCB decreased to about 10 ppb (whole base), the PCB congeners eliminated more slowly from the human body as observed in Yusho patients from the time of 5000 days after the contamination. Among the PCBs determined, 2,3',4,4',5-penta-CB was a unique PCB. Its half-life was shorter than those of PCBs and PCDFs in the Yucheng patients, and its concentration decreased to lower than the control level at 4000 days after the contamination.

Table 1 Biological Half-life of PCDF and PCB Congeners in Yusho and Yucheng Patients

	Half-life (Years)									
	Yucheng patient				Yusho patient					
	0.6 ~ 15.6 years after onset				14.0 ~ 29.1 years after onset					
	BS	SS	RK	Median	KK	TS	YUM	TH	HH	Median
2,3,4,7,8-Penta-CDF	2.7	3.6	2.9	2.9	14.3	7.7	6.1	5.2	11.4	7.7
1,2,3,4,7,8-Hexa-CDF	2.7	3.6	3.5	3.5	6.5	4.5	3.9	5.1	6.9	5.1
1,2,3,4,6,7,8-Hepta-CDF	2.6	2.5	2.2	2.5	6.6	2.6	3.5	3.5	3.4	3.5
Average	2.7	3.2	2.9	3.0	9.1	4.9	4.5	4.6	7.2	5.4
2,3',4,4',5-Penta-CB	1.6	1.9	1.5	1.6	19.5	6.9	33.7	17.6	10.4	17.6
2,2',4,4',5,5'-Hexa-CB	3.4	4.2	4.2	4.2	9.1	7.4	16.0	12.9	7.4	9.1
2,2',3,4,4',5'-Hexa-CB	4.4	4.5	5.5	4.5	12.8	8.9	13.7	31.0	9.5	12.8
2,3,3',4,4',5'-Hexa-CB	3.8	5.6	5.3	5.3	9.4	8.5	21.5	13.2	14.4	13.2
2,2',3,3',4,4',5'-Hepta-CB	4.7	6.0	5.9	5.9	18.4	12.3	237.5	13.3	443.7	18.4
2,2',3,4,4',5,5'-Hepta-CB	4.3	6.0	6.0	6.0	16.7	12.2	20.4	10.3	224.6	16.7
Average	3.7	4.7	4.7	4.6	14.3	9.4	21.1	16.4	118.3	14.6

Discussion

PCBs in Yusho patients have peculiar gas chromatographic pattern and different from those of

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control persons, that is, concentrations of 2,3',4,4',5-penta-CB are relatively low and concentrations of 2,3,3',4,4',5-hexa-CB are relatively high in Yusho patients. The difference of pattern has been used as one of important criteria for Yusho. As shown in Table 1, biological half-life of 2,3',4,4',5-penta-CB is relatively short as 1.6 years in Yucheng patients and that of 2,3,3',4,4',5-hexa-CB is relatively long in Yusho and Yucheng patients. Typical Yusho PCB pattern was therefore produced in 1-2 years after the contamination probably due to the strong enzyme induction by the PCDFs ingested together with PCBs in the patients.

Comparing the levels of PCBs and PCDFs in both the patients at 15 years after the intake, average concentrations of 2,2',4,4',5,5'-hexa-CB and 2,3,4,7,8-penta-CDF in the three Yucheng patients were 2.2 times higher and 3.8 times less than those of the five Yusho patients, respectively. The concentrations of 2,2',4,4',5,5'-hexa-CB and 2,3,4,7,8-penta-CDF in Yusho blood just after the onset were consequently estimated to be 37 ppb in whole blood and 60 ppb in fat, respectively. As the human blood usually contains 0.3% of extractable lipid, fat base concentrations of 2,2',4,4',5,5'-hexa-CB in the blood of Yusho patients were calculated to be 12 ppm just after the onset. As total PCBs in Yusho patients were reported to include 16 % of major PCB congener, 2,2',4,4',5,5'-hexa-CB, total PCB concentrations are estimated to be high as 75 ppm in blood fat of the Yusho patients 0.6 year after the ingestion. These high concentrations were slowly decreased to 2.3 ppm during 30 years after the intake of PCBs, biological half-life being 4.2 years in the first 15 years and 9.1 years in the next stage of 15 years. TEQ factor for 2,3,4,7,8-penta-CDF is 0.5 and 2,3,4,7,8-penta-CDF contributed about 70 % of the dioxin toxicity in Yusho rice oil and adipose tissue of Yusho patients⁶, TEQ concentration (fat base) in the Yusho patients just after the onset was calculated to be 40 ppb from the concentration (60 ppb) of 2,3,4,7,8-penta-CDF. This high TEQ concentration was gradually decreased to 0.6 ppb during 30 years after the ingestion with biological half-life being estimated to be 4.5 years. Fig 1 illustrates the changes of these concentrations during 30 years of intoxication in Yusho patients. In 1995, TEQ levels in the blood of 83 Yusho patients were determined to be 156 ppt/lipid on average, ranging from 86 to 1016 ppt/lipid and the level was 12.8 fold higher than in the control⁷.

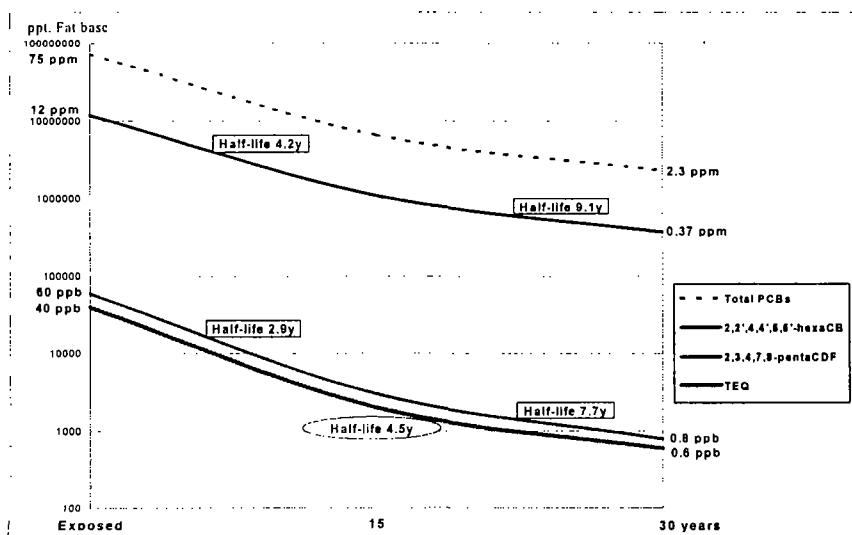


Fig. 1 Estimated elimination of PCB, PCDF and TEQ in the Yusho patients during 30 years (Fat base, ppt)

With these levels of PCB and TEQ exposure, typical Yusho symptoms such as chloracne, pigmentation, cheesy secretions from the meibomian glands of eyes and others were observed in Yusho patients¹. These Yusho symptoms diminished with time as the level of TEQ gradually decreased down to 0.6 ppb with the passage of 30 years. Various internal diseases that might be caused by enzymatic and/or hormonal disorders⁶ are summarized in Table 2. Disorder of serum

Table 2 Effects of enzymatic and/or hormonal disorders in Yusho patients

Organ	Sign	Time of examination
		Yusho outbreak 1968
Liver	Smooth surfaced endoplasmic reticulum : ↑	1969
Female	Irregular menstrual cycle : ↑	1970
Urine	Androsterone : tend ↑ in male, ↓ in female	1970
Serum	IgG : ↓, IgA, IgM : ↓	1970
Serum	Triglyceride : ↑	1973
Serum	Bilirubin : ↓	1974
Serum	Ribonuclease : ↑	1974
Teeth	Anomalies in number and shape : ↑	1978
Serum	helper T/suppressor T : ↑	1983
Serum	Thyroxine, Triiodothyronine : ↑	1984
Lymphocyte	Aryl Hydrocarbon Hydroxylase : ↑	1985
Serum	Triglyceride : ↑	1988
Body	General fatigue, Numbness : ↑	1988
Serum	Triglyceride, Total cholesterol : ↑	1993
Serum	Thyroid Stimulating Hormone : tend ↑	1996
Oral cavity	Pigmentation : tend ↑	1996/1998
Serum	IgA, IgG, IgM : tend ↑	1997

levels of triglyceride, thyroxin and immunoglobulin and higher induction of aromatic hydrocarbon hydroxylase in lymphocyte chronically continued in Yusho patients for 20 years and these enzymatic and hormonal disorders are still observed at present time 30 years after the onset. Taiwan Yucheng children prenatally exposed to PCBs and PCDFs had poorer cognitive development at age of 4 to 7 years⁸. The body height and penis length of Yucheng children were lower than those of controls at age of 11 to 14 years⁹. These changes might be caused by the estrogenic or antiandrogenic effects of the PCBs/PCDFs in Yucheng children, which were 10 to 30 fold higher than the controls.

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