Serum estrogen and its metabolites in pregnant women exposed to dioxins and polychlorinated biphenyls (PCBs)

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Introduction

Dioxins and PCBs are environmental endocrine disruptors that have half-life of 7-10 years in human bodies and have toxicities including carcinogenesis. Studies showed a high estrogen 4-/2-hydroxylation ratio appears to be a marker for neoplasm. The aim is to examine dioxin and PCBs body burden[1] in relation to estrogen metabolites and catabolites[2].

Methods and Materials

Data and specimen collections: The participants were pregnant women of no clinical complication, aged 25-34 years, and delivered their babies in a medical center located in central Taiwan, during December 1, 2000 and November 30, 2001. All pregnant women who came in for a routine check-up in the obstetrics clinic of this hospital and whose pregnancies were over 18 weeks were invited and interviewed for occupation and life-style including smoking and cooking habits. Maternal blood was collected during the third trimester and placenta upon the delivery.

Specimen analyses: Seventeen dioxin congeners, 12 dioxin-like and 6 indicators PCBs in placenta were measured by high-resolution (HR) gas chromatography and HR mass spectrometry (MS) in the ERGO laboratory, Germany. Estrogen metabolites were hydrolyzed with glucuronidase and extracted by C18 solid phase extraction. After evaporation, the residues were dissolved in 50 μL loading buffer and 20 μL was injected and analyzed by liquid chromatography coupled with tandem mass spectrometry. Concentrations of the metabolites were calculated using the specific peak area, and corrected with the peak area of the internal standard.

Results and Discussion

Figure 1 and 2 shows that the ratio of 4-OH-E_2/2-OH-E_2 decreased with increasing tertiles of 2.3.7.8-tetrachlorodibenzo-p-dioxins (TCDD) from 0.74 (C.I.= 0.58-0.89), 0.48 (0.39-0.56), to 0.46 (0.37-0.55) ($p<0.001$, $\beta= -0.16$, $R=0.45$ by general liner regression). This is consistent with results from in vitro test[3] and Seveso TCDD exposed women with decreased breast cancer risk[4]. Table 1 demonstrates that concentration of 4-OH-E_2 increased with increasing concentrations of high-chlorinated PCDFs (i.e. 1.2.3.4.6.7.8-Hepta-CDF: $p=0.03$, $\beta=0.454$, $R=0.30$). It is implicated that CYP1A2 enzyme activity may decrease with increasing TCDD levels particularly when relative to

the CYP1A1 activity. The current study not only validated the association between TCDD and E_2 catabolites also disclosed the different relationships among TCDD, PCDF and PCB to estradiol catabolites in humans. 4-OH-E_2 DNA adducts was suggested to be more suitable biomarker for breast cancer risk in the pre-menopausal women.

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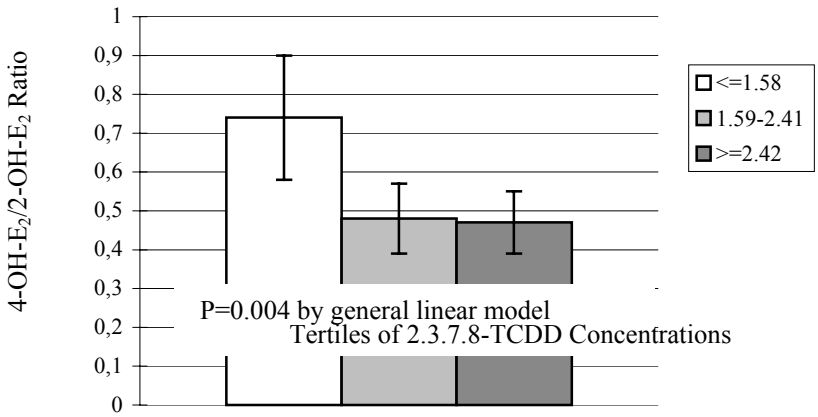
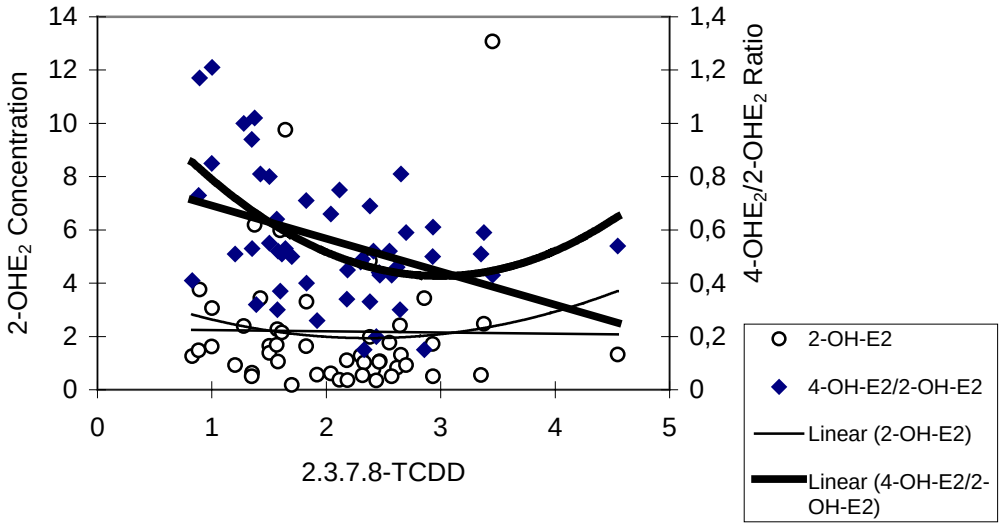
Figure 1: Ratio of 4-OH-E2 to 2-OH-E2 concentrations (pg/g lipid) according to tertiles of Tetra-chlorinated dibenzo-p-dioxins (TCDD)



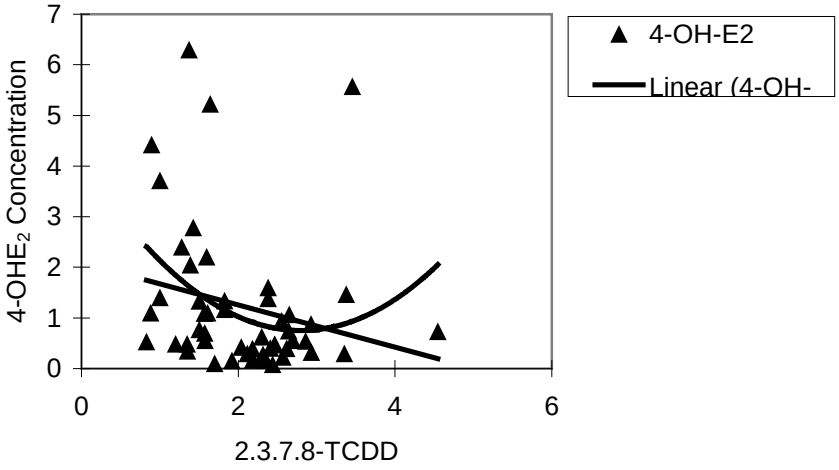
Acknowledgements
The National Health Research Institutes, Taiwan, funded the study. The authors would like to thank to Dr. L.-Y. Lin for his assistance on subject recruitment, to Ms H.-Y. Yu on specimen collection, and to professor O. Papke on specimen analyses of dioxins and PCBs.

Figure 2: Relations of 2.3.7.8-tetra-chlorinated dibenzo-p-dioxins (TCDD) concentration to 2-OH-E_2 concentration (left axis of upper panel), 4-OH-E_2/2-OH-E_2 ratio (right axis of upper panel), and 4-OH-E_2 concentrations (lower panel)



2-OH-E_2=-0.04*(TCDD)+2.28 (p=0.93, R^2=0.0002)
Quadratic model:
2-OH-E_2=0.37*$(TCDD)^2$-1.77*(TCDD)+4.03
(p=0.66, R^2=0.02)

Ratio=-0.124*(TCDD)+0.82 (p=0.004, R^2=0.16)
Quadratic model:
Ratio=0.09*$(TCDD)^2$-0.55*(TCDD)+1.25
(p=0.001, R^2=0.27)



4-OH-E_2=-0.42*(TCDD)+2.09 (p=0.12, R^2=0.05)
Quadratic model:
4-OH-E_2=0.43*$(TCDD)^2$-2.39*(TCDD)+4.10 (p=0.06, R^2=0.12)

Table 1. Correlations between dioxins/PCBs body burden and steroid hormones in maternal venous serum

Hormone \ Exposure	2.3.7.8-TCDD	1.2.3.7.8-PeCDD	1.2.3.4.6.7.8-HpCDF	1.2.3.4.7.8.9-HpCDF	Total non-ortho-PCBs	Total mono-ortho-PCBs	Total Indicator PCBs
Androstenedione (ng/ml)	-0.058	0.038	0.175	0.115	-0.002	0.067	0.095
E_1 (ng/ml)	-0.122	0.038	0.257#	0.316*	-0.018	0.155	0.262#
E_3 (ng/ml)	0.053	0.263#	0.114	0.136	-0.105	-0.063	0.013
Progesterone (ng/ml)	0.073	0.303*	0.017	0.140	0.000	0.184	0.123
2-OH-E_2 (ng/ml)	-0.013	0.080	0.281*	0.023	0.004	0.036	0.006
4-OH-E_2 (ng/ml)	-0.222	-0.121	0.204	-0.026	-0.039	-0.038	-0.043
E_2	-0.309*	-0.313*	0.116	0.259#	0.028	0.024	0.112
Testosterone	-0.099	-0.099	-0.064	-0.055	0.122	-0.030	0.036
4-OH-E_2/2-OH-E_2	-0.400**	-0.316*	0.078	0.013	-0.098	-0.044	0.027
4-OH-E_2/E_2 Ratio	0.110	0.230	0.088	-0.033	0.023	0.088	0.028
2-OH-E_2/E_2 Ratio	0.189	0.197	0.103	-0.126	-0.033	-0.071	-0.066

#p<0.1, *p<0.05, **p<0.01, ***p<0.001 by Pearson correlation analyses

TCDD: Tetra-chlorinated dibenzo-p-dioxin
PeCDD: Penta-chlorinated dibenzo-p-dioxin
HpCDF: Hepta-chlorinated dibenzo-furan
PCDDs: Polychlorinated dibenzo-p-dioxins
PCDFs: Polychlorinated dibenzo-furans
PCB: Polychlorinated biphenyles

References

1. Clemons M. and Goss P. (2001) N. Engl. J. Med. 344, 276.
2. Wang S.-L., Lin C.-Y., Guo Y.-L., Lin L.-Y., Chou W.-L., Chang L.W. (2004) Chemosphere 54, 1459.
3. van Duursen M.B., Sanderson J.T., van der Bruggen M., van der Linden J., van den Berg M. (2003) Toxicol. Appl. Pharmacol. 190, 241.
4. Warner M., Eskenazi B., Mocarelli P., Gerthoux P.M., Samuels S., Needham L., Patterson D., Brambilla P. (2003) Environ. Health. Perspect. 110, 625.