

FREQUENCY OF SISTER CHROMATID EXCHANGES IN JAPANESE INFANTS PERINATALLY EXPOSED TO CHLORINATED DIOXINS AND RELATED COMPOUNDS

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Introduction

Breast milk has been contaminated with extremely toxic dioxins which are polychlorinated dibenzo-*p*-dioxins, polychlorinated dibenzofurans and coplanar polychlorinated biphenyls, due to their pollution of mother's body^{1,2,3}. Consequently, these compounds contaminate the foetus and infants through the placenta and from the mother's milk, respectively.

Formation and induction of sister chromatid exchanges (SCEs) have been considered a good index to the synthetic and sharp genotoxicity for several mutagenic and carcinogenic chemicals^{4,5}.

In this study, the relationship between the SCE frequency of the infant lymphocytes around postnatal of 1 year and the levels of the dioxins in mother's milk was statistically investigated in order to evaluate their genotoxicity to the most sensitive stages of human life, which are the foetus and suckling.

Materials and Methods

Fifty to 100 ml of mother's milk at the postpartum period of 2 ~ 4 month were collected from 124 healthy mothers, mean age : 29 years old and the range : 22 ~ 41 years old, in July ~ October, 1994, June ~ October, 1995 and June ~ October, 1996. These samples of the mother's milk were analyzed for the dioxins by HRGC-HRMS technique using a Finnigan MAT-95 mass spectrometer (Germany) directly interfaced with Varian Model 3400 gas chromatograph^{3,6}.

Toxic equivalent (TEQ) concentrations of the dioxins were calculated by using 1998 WHO TEF values⁷. TEQ-sum of all congeners of the dioxins determined in the milk samples was summarized as the total 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (2,3,7,8-TCDD) TEQ concentration or level.

Five to 10 ml of the peripheral blood of 105 infants born of these mothers were individually obtained by venipuncture in January ~ March of 1995, 1996 and 1997. Among them, SCE frequency of the lymphocytes was measured in 64 infants.

Lymphocytes in the whole blood were stimulated with phytohemagglutinin and cultured for two replicative cycles in the presence of bromodeoxyuridine (100 μ M) as detailed elsewhere^{8,9}. Differential staining of sister chromatids was obtained by a fluorochrome plus Giemsa technique and the frequency of SCEs was evaluated.

Relationship between control, solvent (DMSO) treated, SCE frequency (SCE_{control}), 7,8-benzoflavone (ANF) treated SCE frequency (SCE_{ANF}) or Δ SCEs (SCE_{ANF} - SCE_{control}) and

the dioxins levels in breast milk was statistically examined by Spearman rank correlation method.

Results and Discussion

The frequency distributions of $3CE_{control}$ and $3CE_{ANF}$ of lymphocytes in the blood of infants are indicated in Fig. 1. Mean frequencies \pm standard errors of mean per cell of $3CE_{control}$, $3CE_{ANF}$ and $\Delta 3CEs$ were 8.3 ± 0.13 , 11.9 ± 0.19 and 3.6 ± 0.18 , respectively.

The distributions of the contamination levels of the dioxins in the breast milk are shown in Fig. 2. Average concentrations \pm standard errors of mean were 0.96 ± 0.04 pg-TEQ/g and 24.0 ± 0.8 pg-TEQ/g on whole and lipid weight bases, respectively.

As indicated in Fig. 3, we could not find any significant correlation of dioxin concentrations in the breast milk with the frequency of $3CE_{control}$ or $3CE_{ANF}$ in the lymphocytes of infants around postnatal of 1 year. Frequency of $\Delta 3CEs$ also seemed not to correlate with the dioxin levels.

Based upon these findings, perinatal exposure to dioxins through the placenta or from the mother's milk was considered not to show genotoxicity such as the induction of SCEs at the current contamination levels of Japanese mothers.

This study, however, was carried out with rather small number of infants, so in order to get more conclusive results concerning their SCE induction, further large-scale studies are required.

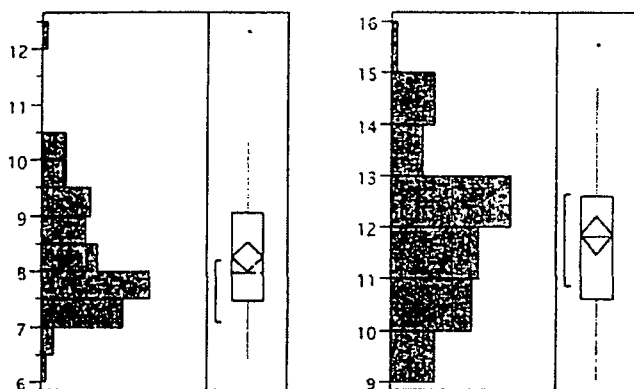


Fig. 1. Frequency distributions of $3CE_{control}$ and $3CE_{ANF}$ in lymphocytes of infants left : $3CE_{control}/Cell$, right : $3CE_{ANF}/Cell$

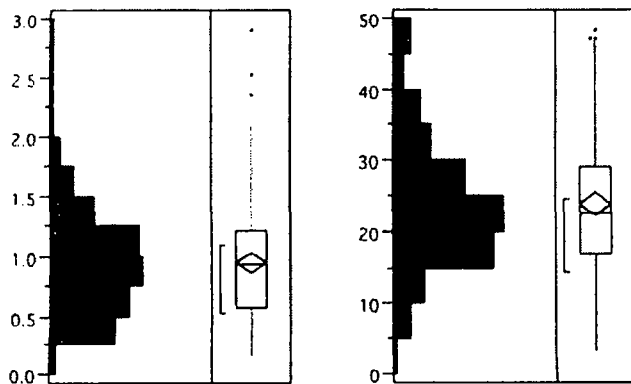


Fig. 2. Distributions of contamination levels of dioxins in the breast milk left : whole weight basis (pg-TEQ/g), right : lipid weight basis (pg-TEQ/g)

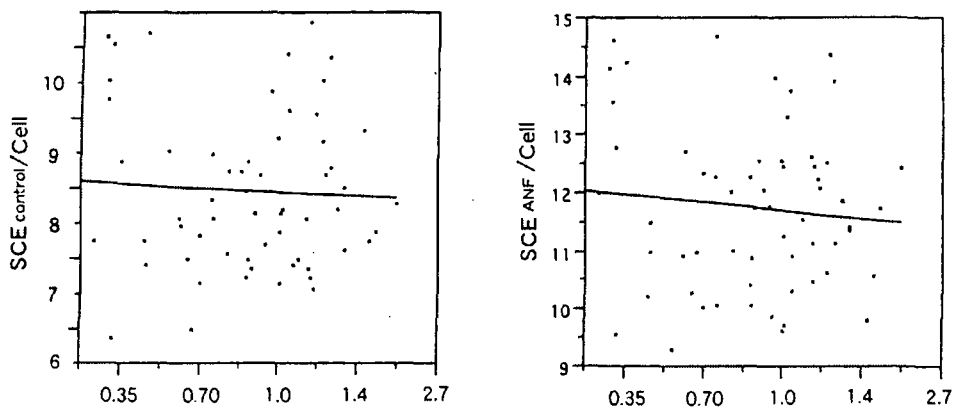


Fig. 3. Relationship between the dioxins in breast milk and frequency of SCE_{control} or SCE_{ANF}
 SCE_{control} : p=0.69, SCE_{ANF} : p=0.45

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