PCDD/ Fs in mother's milk may cause developmental defects in childrens' teeth

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1. Introduction

Dental hard tissues, like bone, are mineralized connective tissues. Unlike bone, enamel and dentin are not remodeled. Thus disturbances in the function of odontoblasts (dentin forming cells) and/or ameloblasts (enamel forming cells) lead to definite morphological consequences, by which the time and sometimes the nature of the damage can be determined. This makes the dental hard tissues unique compared with other tissues in humans. The etiology behind the dental disturbances can be genetic, acquired (e.g. nutritional deficiencies, high fluoride intake, drugs such as cyclophosphamide and tetracyclines) or, most often, idiopathic.

Polychlorinated dibenzo–p–dioxins (PCDDs) and polychlorinated dibenzofurans (PCDFs) can be detected in human milk and thus infants may be exposed to high cumulative doses via breast feeding[1]. Dental changes after accidental exposure of human infants to polychlorinated biphenyls or other dioxin–like compounds have been reported[2,3]. These include the presence of erupted teeth in newborn babies, mottled, chipping and carious teeth, altered eruption of permanent teeth, and abnormally shaped tooth roots. Animal studies on the continuously growing incisors of rats and mice have also indicated dental changes after exposure to 2,3,7,8–tetrachlorodibenzo–p–dioxin (TCDD). These include accelerated tooth eruption[4] and impaired dentin and enamel formation [5]. The aim of the present study was to investigate whether any correlations between PCDD/F exposure and changes in the dentition of a normal breast–fed child population are found. Especially we were interested in seeing whether PCDD/F compounds could be among the

causative agents in enamel hypomineralization, often found in newly erupted permanent first molars. The permanent first molars were chosen as the target teeth since they are mineralized during the first two years of life, the time when the child is exposed to PCDD/Fs via mother's milk.

2. Methods

Milk Sample Collection

Women were recruited between January and May 1987 from one of the maternity hospitals in Helsinki, the capital, and the maternity clinic of the hospital serving the surroundings of the city of Kuopio, Eastern Finland. The study is part of a WHO/EURO coordinated follow-up studies on levels of PCDDs and PCDFs in human milk. All women giving birth to a child during that time period were invited and about 150 mothers from both areas promised to collect a milk sample after four weeks provided they were still nursing. Altogether 168 milk samples were obtained, 77 samples from Helsinki and 91 samples from Kuopio.

Determination of PCDDs and PCDFs

About 40–80 mL of each breast milk sample, equivalent to 1.4 g fat, was spiked with 100 pg of ^{13}C-labelled PCDD and PCDF standards (ED-998 tetra-octa chlorodioxin standard solution and EF-999 tetra-octa chlorofuran standard solution, Cambridge Isotope Laboratories). Milk fat was extracted with diethyl ether/hexane and the fat content determined. The extract was defatted in a silica gel column and cleaned up with activated carbon column (Carbopack C, 60/80 mesh) containing Celite (Merck 2693) to separate PCDD/Fs from PCBs and further cleaned with an activated alumina column (Merck 1097, standardized, activity level II-III). The quantitation was performed by selective ion recording using a VG 70 SE mass spectrometry (resolution 10,000). Levels of 17 most toxic PCDD/Fs were expressed in TCDD toxic equivalents (TEQ) calculated by using the international equivalency factors (NATO/CCMS, 1988). The laboratory reagent and equipment blank samples were treated and analyzed by the same method as the proper samples, one blank for five samples. Detection limits for the different PCDD/F congeners were 0.3 – 1.0 pg/g in fat samples. Recoveries for internal standards were more than 60% for all congeners. The laboratory has participated successfully in international quality control studies for the analysis of PCDDs and PCDFs in cow milk samples organized by EU/BCR-project in 1993[6,7].

The total PCDD/F exposure of a child via mother's milk was calculated using the formula: I-TEQ/0.2877 x (1-exp(0.2877 x duration of lactation months/12)) which takes into account the duration of breast feeding and the milk I-TEQ value with 25% decrease per year in the concentration.

Dental examination

Six years later 136 children of mothers whose milk was analyzed and who still lived in Helsinki or Kuopio area were invited to a dental examination. Of those invited 91 participated the study at the age of 6 (mean age 6.3 years, range 6.1-6.3 years). Mineralization changes in the primary first and second molars and permanent first molars (provided that they were erupted) were recorded. Those children who had not their permanent first molars erupted at the age of 6, as well as those who had not responded to the first invitation were invited one year later. Altogether data on mineralization changes of the primary teeth were available from 91 children and of the permanent teeth from 102 children.

3. Results

A total of 17 children (17%) showed mineralization changes in the permanent first molars and 32 children (35%) in the primary first and/or second molars (Table 1). One out of the four permanent first molars was affected in eight children, two in three, three in four, and four molars in two children. Of the affected molars 16 were in maxilla and 19 in mandible. In the primary dentition, one or two out of eight primary molars were affected in 21 children, two to five molars in seven children and six to eight molars in four children, respectively. Most lesions in the permanent and primary molars were white chalky enamel lesions of moderate size (Table 1).

In the mother's milk, I-TEQs ranged from 3.8 to 99.4 pg/g milk fat with the mean being 19.8 pg/g (S.D. 10.9). The duration of breast feeding ranged from 1 to 36 months, the mean was 10.5 months (S.D. 5.5). Mineralization changes in the permanent first molars occurred more often (Mann-Whitney U-test, P=0.017) and they were more severe (regression analysis, r=0.3, P=0.003, Mantel Hänszel chi square P=0.010, Table 2) in children who were exposed to a greater amount of PCDD/Fs via mother's milk than in those who were exposed to a lesser extent. The duration of breast feeding alone was not associated with mineralization changes (Mann-Whitney U-test, P=0.17). Neither were I-TEQs or the log(I-TEQ) values significantly associated with the severity of mineralization changes in primary molars which are mineralized before birth.

None of the mothers reported an exposure to putative harmful compounds in their work environment. Twelve mothers had a history of cigarette smoking during the last 12 months before the delivery. All children of these mothers had normally mineralized first permanent molars and two children had mineralization defects in primary molars.

Table 1. Number of children with different hypomineralized teeth: severity and extent of enamel hypomineralization in the permanent first molars and primary molars.

	Number of children (%)	Chalky lesions	Chalky lesions with loss of enamel	Chalky lesions with affected dentin	Moderate-sized lesion(s)	Large lesion(s)
Hypomin-eralized permanent first molar(s)	17/102 (17%)	14	1	2	10	7
Hypomin-eralized primary molar(s)	32/91 (35%)	26	6	0	25	7

Table 2. Number of children with mineralization defects of the permanent first molars in relation to the total PCDD/F exposure of the child via mother's milk.

Mineralization of the permanent first molars	Low exposure (<8.0)*	Moderate exposure (8.0–16)*	High exposure (>16)*
Normal	22	41	22
Mild defect in only one tooth	1	5	2
Moderate defect or mild defect in more than one tooth	0	3	4
Severe defect	0	0	2

* Exposure value is calculated from the formula I–TEA/0.2877 x (1–exp(0.2877 x duration months/12))

4. Conclusions

The results suggest that PCDD/Fs may be a causative agent of enamel hypomineralization and that the prevailing levels of PCDD/F compounds in human milk may cause enamel hypomineralization in the developing teeth of children. As PCBs and other chlorinated compounds found in human milk may also be involved in disturbing tooth development, further studies are needed to evaluate their role.

5. References

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