Developmental dental defects in children exposed to PCBs in eastern Slovakia

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Abstract

The effects of long-term exposure to polychlorinated biphenyls (PCBs) on developmental dental defects of deciduous and permanent teeth in children in eastern Slovakia, where PCBs from a chemical plant manufacturing Delors contaminated the surrounding district were evaluated. Four hundred and thirtytwo children, lifelong residents, aged 8–9 years were examined. Children’s caries susceptibility and gingival health was assessed by standard dental indices, and developmental enamel defects by the FDI index. Data from the PCBRISK project data set and questionnaires completed by the parents provided information on exposure and various confounding factors. The proportion of teeth with different types and extensions of developmental enamel defects correlated with serum PCB concentration. The proportion of deciduous teeth affected with enamel defects was significantly higher in higher exposed children ($\chi^2 = 8.35; p = 0.03$) according to their serum PCB concentration (group 0: <200; group 1: 200–600; group 2: >600 ng PCBs g$^{-1}$ serum lipids). The proportion of permanent teeth affected with any enamel defect was significantly higher in higher exposed children ($\chi^2 = 7.237; p = 0.027$). Furthermore, the extent of the enamel defects was also greater ($\chi^2 = 10.714; p = 0.005$). In multivariate linear regression analysis PCB exposure was significantly related to developmental enamel defects of permanent teeth only. No associations between PCB exposure and caries susceptibility, gingival health or number of teeth were observed. This study demonstrated a dose–response relationship between PCB exposure and developmental enamel defects of permanent teeth in children, the evidence for deciduous teeth was not conclusive.

Keywords: PCBs; Developmental enamel defects; Children; Environmental exposure; Slovakia

1. Introduction

Polychlorinated biphenyls (PCBs) are highly persistent and ubiquitous organochlorine environmental pollutants. Being lipophilic, they increase in concentration up the food chain and bioconcentrate in animal and human tissues. Humans are exposed to PCBs mainly via diet, they are also transferred to the fetus and infants transplacentally and lactationally (Ahlborg et al., 1992). PCB-induced developmental defects that may be irreversible are raising concern due to high environmental exposure and possible greater sensitivity of children (Brouwer et al., 1995). Evidence is building that human tooth development may be a sensitive endpoint of PCB toxicity.

Developing tooth is sensitive to a wide range of local and systemic disturbances. Because of the absolute metabolic stability of enamel structure, changes in enamel during its development are permanent in nature. Insults occurring during the earliest stages of enamel development, that is matrix formation, will result in the clinical appearance of an enamel hypoplasia. In contrast, insults occurring during the calcification and maturation stages of enamel development may lead to deficiency of mineralization and usually manifest as enamel opacities.
Developmental dental defects were first described in two episodes in Asia of epidemic PCB poisoning, Yusho and YuCheng. Excess of ectodermal defects and developmental delay (Ahlborg et al., 1992), including a variety of dental changes such as mottled, chipped and carious teeth, perinatally erupted teeth, distortion of tooth roots, retarded eruption, and lack of permanent teeth have been reported (Rogan et al., 1988; Wang et al., 2003). These children born to exposed mothers would have had transplacental exposure and exposure through breast milk. Nevertheless, co-contamination with polychlorinated dibenzo-furans (PCDFs) was largely responsible for the overall toxicity (Safe, 1994; Masuda, 1996).

Alaluusua et al. (1999) found that developmental enamel defects on first permanent molar teeth undergoing mineralization during the first 2 years of life were correlated with the total exposure to polychlorinated aromatic hydrocarbons via mother’s milk. The correlation was strong with exposure to prevailing levels of polychlorinated dibenzo-p-dioxins (PCDDs) and furans (PCDFs) but weak with exposure to PCBs alone.

In our previous study we have shown developmental enamel defects of permanent teeth in children exposed to PCBs alone (Jan and Vrbicˇ, 2000), suggesting that the developing human teeth are vulnerable to PCBs. There is no data on the adverse effects on deciduous teeth that develop before and a few months after birth.

Experimental studies have confirmed that the specific cells forming the teeth are sensitive to polychlorinated aromatics which lead to permanent changes in dental hard tissues. Rhesus macaques exposed to PCDDs and dioxin-like compounds, including PCBs via food showed squamous metaplasia of ameloblasts and the development of jaw cysts (McNulty, 1985). Studies on continuously growing rat incisors reported the selective toxic effects on ameloblasts in adult rats treated with PCB mixture KC-400 (Hashiguchi et al., 1985).

Recent studies have revealed high PCB contamination in eastern Slovakia, in the Michalovce region, where PCBs from a chemical plant manufacturing Delors contaminated the surrounding district (Kocˇan et al., 2001). The total serum PCB levels in samples from the general population there, exceeded by several times the background levels. Mean levels of serum TEQPCB were 40 times higher if compared with the background levels (Kocˇan et al., 2004). PCB levels in breast milk samples there were the highest in Slovakia (Petrik et al., 2001).

The goal of the present study was to investigate the effect of long-term PCB exposure on the developmental dental defects of children in eastern Slovakia. Exposure was measured at the individual level, with a close control on other possible confounding factors.

2. Materials and methods

Subjects described in the present report have been enrolled in the European PCBRISK project. Their description, study methodology, and exposure characteristics have been published previously (Kocˇan et al., 2004; Trnovec et al., 2004). In short, 208 children from the Michalovce, and 224 from the less exposed Svidnik area, aged 8–9 years, were examined in September 2002 by two calibrated dentists. They had to be born and living in the study area, and children’s mothers should have permanently lived in the respective area for at least 5 years before the child’s birth.

Presence of deciduous and permanent teeth was recorded, dental caries was assessed using the decayed, missing and filled tooth (dmft) index of the World Health Organization (1987), and gingival health by the Loe–Sillness index (Silness and Loe, 1964). Developmental defects of enamel were assessed using the FDI index (FDI, 1992) on buccal surfaces of deciduous and permanent teeth. Three main types of developmental defects of enamel were recorded: demarcated opacities, diffuse opacities, and hypoplasia. The extent of the defects was recorded in thirds of the surface area. Questionnaires that were completed by the parents and other data from the PCBRISK project data set provided information on various confounding factors and modifiers (e.g. place of residence during tooth development, parity, duration of breast-feeding, eating fish from rivers and/or lakes, children’s diseases, medications, fluoride, metal, and metalloid exposure). Analyses of blood samples for organochlorines were made by high resolution gas chromatography using electron capture detection (Kocˇan et al., 2004).

Children were categorized into three groups according to their serum total PCB concentration (group 0: <200; group 1: 200–600; group 2: >600 ng PCBs g⁻¹ serum lipids).

All the data were analysed using the SPSS 9.0 statistical software package.

3. Results

The mean number of decayed, missed and/or filled deciduous teeth (dmft) per child was 1.5 (±1.8), and for permanent teeth (DMFT) it was 3.2 (±2.4).

Of the 3523 deciduous teeth evaluated, 4.5% were affected with developmental enamel defects. Of the 6340 permanent teeth evaluated, 65.0% were affected with developmental enamel defects, 49.4% with demarcated opacities and/or hypoplasia, and 33.4% with diffuse opacities. Of the 487 children examined, 57.5% of the children had at least one permanent tooth affected with an enamel defect, 43.5% had at least one permanent tooth affected with demarcated opacity and/or hypoplasia, and 29.6% with diffuse opacity. The percentage of children with at least one deciduous tooth affected with any enamel defect was 9.4%, 7.2% of children had at least one deciduous tooth affected with demarcated opacity and/or hypoplasia and 4.1% with diffuse opacity.

The proportion of teeth with different types and extensions of enamel defects correlated with serum PCB concentration (Table 1).
The proportion of deciduous teeth affected with developmental enamel defects was significantly higher in higher exposed children (Kruskal–Wallis $\chi^2 = 8.35; p = 0.03$) according to their serum total PCB concentration categorized in 3 groups. As it’s difficult to differentiate demarcated opacities from fluorosis-like diffuse opacities in deciduous teeth, and the number of developmental enamel defects was small, only data for any type of enamel defects in deciduous teeth are presented in Table 1. The proportion of permanent teeth affected with any developmental enamel defect was significantly higher in higher exposed children (Kruskal–Wallis $\chi^2 = 7.237; p = 0.027$). Furthermore, the extent of the defects was also greater (Kruskal–Wallis $\chi^2 = 10.714; p = 0.005$). When excluding diffuse opacities from the final statistical analysis, there was a significantly higher proportion of permanent teeth affected with demarcated opacities and/or hypoplasia in the children in higher exposed groups (Kruskal–Wallis $\chi^2 = 9.985; p = 0.007$). Distribution of enamel defects among differently exposed children is shown in Fig. 1.

In order to evaluate the relative importance of the various possible etiological reasons for developmental enamel defects of permanent teeth, we have utilized standard multiple regression analysis (Table 2). Variables with $p$-values <0.20 in bivariate analysis were selected for inclusion in a multivariate model. These variables were living of mother in the same place before birth in years, hereditary illness, serum PCB concentration categorized in three groups, serum TSH concentration, and year of birth for child. The best model based on statistical significance for the proportion of permanent teeth with any developmental enamel defect as dependent variable was the model with predictors: hereditary illnesses, living of mother in the same place before birth in years, and serum PCB concentration. With respect to deciduous teeth serum PCB concentration did not qualify to be included in the final multivariate model.

No association was found between caries susceptibility, gingival health, or number of teeth and PCB exposure.

### 4. Discussion

This study demonstrated a dose–response relationship between long-term PCB exposure and developmental enamel defects of permanent teeth in children. PCB exposure was significantly associated with developmental enamel defects of deciduous and permanent teeth in bivariate analysis. In multivariate analysis with linear regression, PCB exposure was statistically significantly related to enamel defects of permanent teeth only.

In deciduous dentition the enamel formation starts around the 14th week of gestation and is completed at the end of the 1st year of life. It may therefore be assumed that enamel defects of deciduous teeth indicate prenatal or early postnatal damage affecting ameloblasts or enamel maturation. Children with developmental enamel defects had both deciduous and permanent teeth affected, suggesting that these defects resulted from a systemic factor acting pre- and post-natally. Since children were life-long exposed to PCBs through the food chain, the dietary intake of PCBs had a larger effect on their total body burden than the exposure in utero or via milk (Patandin et al., 1999). This is in accordance with our findings that the enamel of deciduous teeth showed to be less disturbed during its development compared to permanent teeth.

The difference in the prevalence of developmental enamel defects among differently exposed children was mostly due to demarcated opacities and/or hypoplasia.

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**Table 1**

<table>
<thead>
<tr>
<th></th>
<th>Deciduous</th>
<th>Permanent teeth</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Any defect</td>
<td>Any defect</td>
</tr>
<tr>
<td>Correlation coefficient</td>
<td>0.10</td>
<td>0.13</td>
</tr>
<tr>
<td>Sig. (2-tailed)</td>
<td>0.04</td>
<td>0.01</td>
</tr>
</tbody>
</table>

$^a$ Affected teeth had defects greater in size than one third the buccal area.

**Table 2**

<table>
<thead>
<tr>
<th>Source</th>
<th>$B$</th>
<th>95% CI</th>
<th>$\beta$</th>
<th>$p$-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Constant</td>
<td>0.10</td>
<td>0.07–0.12</td>
<td>0.00</td>
<td>0.00</td>
</tr>
<tr>
<td>Living of mother in the same place before birth in years</td>
<td>0.00</td>
<td>0.00–0.00</td>
<td>0.00</td>
<td>0.00</td>
</tr>
<tr>
<td>Hereditary illness</td>
<td>0.17</td>
<td>0.11–0.23</td>
<td>0.26</td>
<td>0.00</td>
</tr>
<tr>
<td>Serum PCB concentration</td>
<td>0.02</td>
<td>0.00–0.04</td>
<td>0.12</td>
<td>0.02</td>
</tr>
</tbody>
</table>

$R^2 = 0.11$. 

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Fig. 1. Distribution of developmental enamel defects of permanent teeth among differently exposed children (group 0: <200; group 1: 200–600; group 2: >600 ng PCBs g$^{-1}$ serum lipids).
and so it confirmed the results with our previous study (Jan and Vrbić, 2000). The observation is in accordance with studies on PCBs treated rats and non-human primates (Hashiguchi et al., 1985; McNulty, 1985), where selective toxic effects on ameloblasts and cells of stratum intermediate in the secretory and maturation stage of enamel development were reported.

The proportion of permanent teeth with diffuse opacities that are likely to be of fluoride etiology (Clarkson and O’Mullane, 1989) and caries experience were relatively low and could not have masked demarcated lesions and hypoplasia during clinical examination.

Analysis also assessed many of the covariates suspected to affect estimates of the PCB – developmental enamel defects relation, including those from the complex data set from the PCB RISK project (Trnovec et al., 2004), but found little evidence of confounding or effect modification. For permanent teeth only hereditary illnesses and living of mother in the same place before birth proved to be statistically significant (Table 2).

In our study no associations between the duration of breast-feeding and enamel defects were found. A possible explanation is that in early-life, accumulated PCB levels did not reach the “critical” levels necessary to cause distinctive adverse effects on enamel development. Also, the duration of breast feeding was much shorter than in the dioxin studies of Alaluusua et al. (1999).

We did not observe any associations between PCB exposure and carries susceptibility, gingival health or number of teeth present, as reported in previous studies (Rogan et al., 1988; Hashiguchi et al., 2003; Wang et al., 2003). This may be due in part to differences in methodology and age of the subjects. In our study no radiographs were taken, so congenital missing of tooth germ could not be evaluated.

The exact pathogenetic mechanism explaining how PCBs cause developmental enamel defects is currently not clearly understood. The alteration of retinoid metabolism, inhibition of thyroid hormone activity, and reduced blood flow have been suggested as the mechanisms leading to dental defects (Hashiguchi et al., 1985). Changes in enzyme levels, hormones, growth factors, and their receptors are the principal known biochemical consequences of exposure to dioxin-like coplanar PCBs (Birnbaum, 1994), that are thought to be mediated by the aryl hydrocarbon receptor (AhR) (Safe, 1994). The role of AhR in dioxin-induced tooth defects has been shown (Sahlberg et al., 2002; Gao et al., 2004). The epidermal growth factor receptor (EGFR) seems to be involved, because the dioxin-induced failure of enamel matrix deposition and mineralization of dentin observed in cultured molars of mouse embryos were not seen in molars of mice lacking EGFR (Partanen et al., 1998). Dioxin also interferes with tooth development by enhancing and accelerating apoptosis in the dental epithelium (Partanen et al., 2004). However, nonplanar PCBs are likely to act through a different mechanism of action (Giesy and Kannan, 1998). There are lacks of data on their role in dental development disturbance.

5. Conclusions

Our results demonstrated a dose–response relationship between the long-term PCB exposure and developmental enamel defects of permanent teeth in children in eastern Slovakia, indicating that enamel development is one of the sensitive endpoints of PCB toxicity. With respect to deciduous teeth multivariate analysis did not confirm the contribution of PCB exposure to the prediction of developmental enamel defects. However, bivariate results suggest that PCBs could play a contributing role. Further evaluation of the mechanism of this toxicity is needed.

Acknowledgements

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References


