Association of passive smoking with dental development in young children

**ABSTRACT**

**Aim** To assess dental development in young children who have been exposed to passive smoking by comparing dental ages with the corresponding features in a healthy control group.

**Materials and methods** A total of 90 passive smokers (PS) with a mean age of 5.49 years and 90 healthy age-matched controls were included in this study. The children were investigated for stimulated salivary cotinine level. Three categories were formed with respect to the number of cigarettes smoked. Dental development was analysed using panoramic radiographs.

**Results** The dose-response relationship between the cotinine levels of the PS subjects and the number of cigarettes smoked per day was found to be significantly different for the three exposure categories. A significant difference was found between the chronological and dental ages of PS subjects when compared with those of the control group, but there were no significant differences in dental ages related to gender in both groups.

**Conclusion** We could clearly conclude that young children who were PS subjects had delayed dental development.

**Keywords** Chronological age; Dental age; Passive smoking; Salivary cotinine levels.

**Introduction**

Passive smoking exposure may start in utero and may continue after birth throughout the childhood. The relationship between both pre- and post-natal passive smoking exposure and the health status of children has been reported in many investigations [Ferris et al., 2010]. In addition, there are a number of studies on oral health in relation to passive smoking in young children [Avsar et al., 2008; Avsar et al., 2009; Hanioka et al., 2008; Leroy, 2008].

Use of the biomarker cotinine can potentially reduce misclassification, allowing one to compare a high-exposure group with a low passive smoking exposure group. Cotinine, a primary metabolite of nicotine, has a much longer half-life (about 18-20 h) than nicotine (half-life ~2 h), resulting in higher and more stable plasma concentrations [Benowitz, 1996; Binnie et al., 2004].

Dental development is an important indicator of disturbances during odontogenesis, and factors such as diseases, chemotherapy and radiation therapy can affect teeth at any phase prior to their complete formation and calcification [Condo et al., 2011; Holderbaum et al., 2005; Vasconcelos et al., 2009]. Several methods for estimating the dental age according to the degree of calcification of teeth from radiographs have been described [Moorrees et al., 1963; Demirjian et al., 1973; Gustafson and Koch, 1974]. No previous study has examined the effect of PS on dental development by performing a dental age assessment. Therefore, the aim of this investigation is to determine the dental age of children exposed to passive smoking in relation to the number of exposures per day.

**Materials and methods**

This study was approved by the Ethical Committee of Ondokuz Mayis University, and written informed consent was obtained from each parent.

**Study population**

Three categories were formed with respect to the children’s daily exposure to cigarette smoke at home according to the total sum of the number of cigarettes smoked per day by each smoking household member. A sample of 30 PS subjects from each exposure group (15 boys and 15 girls) (4–6 years of age; n = 90) were selected randomly and were enrolled in the study as the study population.

Children who attended kindergarten and did not have stay-at-home mothers were excluded from the study because of all of the possible sources of exposure to PS outside of the home. The exclusion criteria that were used for the sample also included the presence of systemic conditions that could affect the salivary gland physiology, such as Sjogren’s syndrome, obesity, cachexia, and diabetes mellitus, as well as additional fluoride prophylaxis (other than with the use of fluoride toothpaste), antibiotics, or antimicrobial agents in the previous 3 months.

For the control subjects, children of the same gender and age as the PS subjects were selected randomly from
patients who received their dental treatment at the same facility and lived in a nonsmoking households (n = 90).

The questionnaire contained items, administered by the same interviewer, about personal, familial and environmental characteristics such as tooth-brushing habit (less than once, once or more than once per day), daily dietary sugar exposure (less than three times per day or more than three times per day), parental education levels (no education, primary school, middle school, high school or university), and family income ($600/month or >$600/month).

**Assessment of salivary cotinine level**

Saliva samples were collected from all subjects between 09:00 am and 12:00 pm to minimise the effects of circadian rhythms and were stored at -80°C until cotinine analysis. To determine the cotinine level, samples were brought to room temperature on the day of testing and were centrifuged at 3000 rpm for 15 min. The clear top phase of the sample was pipetted into the appropriate test tubes. The cotinine level was measured using a microplate enzyme immunoassay according to the manufacturer’s protocol (Salimetrics, State College, PA, USA) and was expressed as ng/mL. The test used 20 mL of sample, had a lower sensitivity limit of 0.05 ng/mL, a range of sensitivity from 0.05 to 200 ng/mL, and average intra- and interassay coefficients of variation of <5.8 and 7.9%, respectively.

**Assessment of dental development**

All of the radiographs were taken using a Planmeca Proline PM unit. Samples with digital panoramic radiograph of adequate quality, and without a history of medical or surgical disease that could affect the presence and development of mandibular permanent teeth, were investigated. Exclusion criteria included image deformity affecting the estimation of mandibular permanent teeth, hypodontia, or gross pathology, as well as missing mandibular permanent teeth except the third molar. The chronological age, converted to a decimal age, was based on the date of the panoramic radiograph and the date of birth.

Each panoramic radiogram was assessed twice by observers to minimise the evaluation error. The first evaluation was carried out using a light-box, and the development stages of particular teeth were entered into tables in a computer database. Then, the images were scanned and stored in a computer database, and the calcification evaluation was carried out on the computer screen. In accordance with the rules of the Demirjian’s method, [Demirjian et al., 1973] the development of seven left permanent mandibular teeth was evaluated.

**Reproducibility**

To assess reproducibility, 50 randomly selected radiographs were re-examined 2 months after the initial examination by the second observer. The percentage agreement of the two readings was calculated by examining 50 radiographs of 350 teeth. The agreement between duplicate scores of the mineralisation stages of 350 teeth was 87%. The difference between the two scores did not exceed one stage for any tooth.

**Statistical analysis**

All data were analysed using Statistical Package for the Social Sciences Version 13.0 (SPSS Inc., Chicago, IL, USA). One-way ANOVA and Mann-Whitney nonparametric test were used for the statistical analysis.

**Results**

**Sample characteristics**

The differences in socioeconomic status and education levels of the parents between PS subjects and control subjects were not statistically significant (p > 0.05).

**Salivary cotinine level**

The mean cotinine level of the PS subjects was 1.58 ± 4.3 ng/mL. The salivary cotinine level of the control subjects was under the limit of detection of 0.05 ng/ml. The dose-response relationship between the cotinine level of the PS subjects and the number of cigarettes smoked per day was found to be significantly different between each of the three categories in the PS group (Fig. 1). The difference between boys (1.59 ± 4.3 ng/mL) and girls (1.56 ± 4.3 ng/mL) was not significant (p > 0.05).

[Diagram showing mean level of cotinine (ng/mL) against number of cigarettes smoked by gender.]
Dental Development

As shown in Table 1, there were significant differences between chronological and dental ages in the PS group whereas this difference was not significant in the control group. The dental age of the PS subjects was lower compared with the dental age in the control group, and this difference was found to be statistically significant (Table 1).

When the Mann-Whitney was applied to verify the difference between chronological and dental ages in exposure groups, the difference was significant between the three exposure categories as shown in Table 2. Regarding the exposure intensity, maximum differences between chronological and dental ages were found in the high exposure group.

Overall, the results showed that there were no differences in dental ages related to gender in both groups.

Discussion

Although PS is a serious global health problem affecting at least 700 million children, data on the relationship between PS and dental development are limited. The major finding of the present study is that the dental age of PS subjects was significantly lower compared with the dental age in the control group.

This study is unique in that no other investigation has used biological markers for measuring children’s exposure to passive smoking [Kieser et al., 1996; Heikkinen et al., 1994]. The salivary cotinine levels in PS subjects in the present study were higher than in some previously reported studies. In addition, there are controversial results regarding the relationship between gender and salivary cotinine level [Delpisheh et al., 2006]. No significant difference was found between boys and girls in the present study. In our study, when comparing the dental and chronological ages of PS subjects to the control group, we found that the dental age in PS subjects was lower than the chronological age, and this difference was statistically significant. Similarly, Kieser et al. [1996] observed approximately a 4-month delay in the maturation of permanent teeth in Caucasoid children exposed to tobacco smoke from both parents as compared with the children of nonsmokers. In agreement with Heikkinen et al. [1994], our results showed that when we compared the dental age and chronological age of patients submitted to different exposure groups, a significant relationship was found.

There is inconsistency among the studies about the relationship between dental age and gender. Corroborating our results, some researchers found that there was no statistical gender difference [Flores et al., 2010]. In contrast, Krailassiri et al. [2002] reported that higher overestimation of the dental age observed in the girls. The difference in results amongst the studies may be due to differences in age and/or ethnic groups.

According to previous investigations, the possible effect of passive smoking on dental development results from a specific effect on tooth odontogenesis [Saad et al., 1991]. The calcification of permanent teeth starts approximately at the time of birth with the first molars and incisors. It is important to point out that the family members investigated in our study had smoked at home since the birth of the child. During this active growth, teeth are susceptible to environmental disturbances, and nicotine may interfere with the reciprocal induction between ectomesenchymally derived tissues and oral ectoderm, disrupting subsequent normal odontogenesis [Saad, 1991]. According to Saad [1991] and Chowdhury and Bromage [2000], by providing nicotine during and after the fetal cycle, changes in dental structure occur because of developmental perturbations induced by this toxin.

The disturbance of mineral metabolism could be one possible mechanism leading to deviations in tooth maturation. Ayçiçek et al. [2005] suggested that active
or passive maternal smoking causes potent oxidative stress. According to Heikkinen et al. [1992], it is possible that the extra carbon dioxide caused by the defect in oxygenation may lead to acidosis, which in turn could disturb post-mitotic hard tissue crystallisation. Other pathogenetic mechanisms could be related to nicotine and hydrocarbons and to their mutative capacity, or these mechanisms may be simply related to poorer nutrition because of the loss of maternal appetite from smoking.

Many investigations have reported that maternal smoking is a principal risk factor for prematurity [Li et al., 2010]; low-birth weight illnesses in infancy have also been linked to delayed development [Gyulavári, 1966]. None of the children in both groups were preterm infants in our study. Previous investigations reported that weight and head circumference in children whose mothers smoked were significantly lower than those in children of nonsmoking mothers [Vardavas et al., 2010]. One limitation of this study is that we did not collect data on the body mass index of the children. In addition, body size and socioeconomic status are known to have a positive correlation [Kwok et al., 2010]. In contrast, in terms of the socioeconomic factors studied in this population, socioeconomic status and parental education levels between the PS subjects and the controls were not statistically significant.

Developmental disturbances have been reported in numerous studies of prenatal exposure to teratogens such as nicotine, opiates, alcohol and cocaine [Kieser, 1992]. However, because smokers who smoke often drink and are sometimes also substance abusers, it is often difficult to determine which exposure factor is responsible for the observed destabilization [Kieser, 1996]. Here, we compared the effects of postnatal exposure to tobacco smoke on dental development in children not knowingly exposed to any of the above-mentioned teratogens.

**Conclusion**

In conclusion, PS children seem to have delayed dental development compared with non-PS children. The limited results of this study need to be validated in the future with larger PS populations.

**References**


