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Association of Passive Smoking with Dental Caries and Cotinine Biomarker among 8 to 12-Year-Old Children

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Abstract

Background: The present study was designed to determine the association of passive smoking with dental caries and cotinine biomarkers among 8- to 12-year-old children in Semnan, Iran.

Methods: This cross sectional study was conducted on two groups of 8-12-year-old children exposed (n=100) and non-exposed to passive smoking (n=100) in Semnan (Iran). The sampling method was two-stage cluster sampling design. The data collection tool was a standard checklist consisted of demographic, laboratory and dental characteristics. The examinations were performed by a dentist on a dental unit and the dental indicators were measured for two groups. Also, salivary cotinine was measured and recorded using ELISA method by special cotinine kit (De salimetrics medi etc, USA). Univariate and Multivariable Logistic Regression models; and area under the ROC curve were used for data analysis by STATA 14 software.

Results: Univariate logistic regression model showed a statistically significant difference between the groups, exposed and unexposed to passive smoking, in terms of the salivary cotinine level, age, sex , decayed, missing, and filled teeth (dmft) index, decayed, and filled teeth (dft) index and missing teeth (mt) (P-Value ≤0.20). However, multiple logistic regression model showed that the decayed, missing, and filled teeth (dmft) index were significantly correlated with passive smoking (OR=1.28; 95% CI: 1.05 – 1.38).

Conclusion: This study revealed that the decay risk of the deciduous teeth is higher in children exposed to passive smoking; therefore, passive smoking should be considered in the implementation of programs to prevent tooth decay in children.

Key Words: Children, Cotinine Biomarker, Dental Caries, Iran, Passive Smoking.

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1- INTRODUCTION

Tooth decay is a microbial infectious disease affecting the calcified tissue of the tooth, due to which the minerals in the tooth are reduced; and the organic compounds are destroyed and decomposed. The most common cause of tooth loss is usually tooth decay and periodontal disease. The caries process results from the interaction of primary and secondary factors such as microbial plaque containing cariogenic microorganisms, cariogenic substrate, time, saliva, immune mechanisms, tooth tissue and ecological conditions of the oral and microbial flora (1, 2).

Smoke generated by burning the tip of a cigarette and inhaled by nonsmokers is considered passive smoking (PS). Studies show that approximately 600,000 non-smokers die each year from passive smoking in the world, most of them occurring in children (3). According to the World Health Organization, 42.9% of Iranian children are exposed to secondhand smoke which is almost as harmful as active smoking; and children in the early years of development are more vulnerable than adults (4). Cigarette smoke can cause lung cancer, cardiovascular diseases and respiratory infections (5, 6). Evidence suggests that children exposed to secondhand smoke are more likely to develop dental caries (7, 8). Environmental cigarette smoke contains more than 4,000 chemicals that are released into the air and can be effective on oral health (9). Smoking in family members can be associated with tooth decay in children and poor oral hygiene (10).

A useful indicator of exposure to tobacco smoke is the cotinine biomarker (the main metabolite of nicotine), which can be measured in plasma, urine, and saliva. Normal concentrations of cotinine is usually <5 ng / ml in the saliva of inactive smokers and between 100 and 105 ng / ml for the active smokers (11, 12). Therefore, exposure to passive cigarette smoke can be screened by the cotinine as this biomarker is stable against temperature changes and has a longer half-life than nicotine (13, 14). In general, non-smokers who are exposed to secondhand smoke absorb nicotine and other components like smokers; and the more they are exposed to passive cigarette smoke, the higher the level of these components is in their body. On the other hand, the level of salivary cotinine and the level of streptococcus mutans and lactobacilli in people exposed to salivary smoke, salivary flow rate and pH of peripheral cigarettes increase and salivary buffering property decreases significantly (15-18).

Therefore, due to the importance of oral health of children exposed to secondhand smoke and due to the limited studies conducted in this field, especially in Iran, the aim of this study was to determine the association of passive smoking with dental caries and cotinine biomarker among 8-to-12-Year-old children in Semnan, Iran.

2- MATERIAL AND METHODS

2-1. Study Design and Subjects

This cross sectional study was designed to determine the association of passive smoking with dental caries and cotinine biomarker among 8-12-year-old children of Semnan, Iran. The study population was children aged 8-12 studying in schools of Semnan city (Iran).The sample size required for the present study was estimated to be 200 students. They were divided into two groups: exposed to passive smoking (n=100) and non-exposed to passive smoking (n=100). The children with the following characteristics were considered as exposed: 1- having a person addicted to smoking in the family 2- exposure to passive cigarette smoke for at least one year. Children who did not have a smoking addict in the family were considered non-exposed. In the present study, two-stage

cluster sampling design was used. In the first step, the city of Semnan was divided into 4 regions: north, south, east and west. Then, two schools from each region (one boys' school - one girls' school) were randomly selected. Next, 25 students from each school were randomly selected. Inclusion criteria consisted of the age range of 8 to 12 and informed consent of parents to participate in the research. Exclusion criteria were being under 8 and over 12 years of age, having any systemic disease (diabetes, hypertension, kidney, lung, cardiovascular, hepatic, rheumatic and gastrointestinal diseases), cancers, autoimmune diseases (HIV/AIDS), having a history of taking any medication, being under periodontal treatments in the past year, and having a plaque index more than 40%.

2-2. Data Collection

The data collection tool was a standard checklist consisted of demographic, laboratory and dental characteristics. The demographic variables were obtained through interviews with the child's parents. For dental variables, examination was performed by a dentist on a dental unit and decayed, missing, and filled teeth (dmft) index, decayed, and filled teeth (dft) index and missing teeth (mt) for deciduous teeth and DMFT, DFT and MT were measured for the permanent teeth. To measure cotinine, at the beginning of the day, the children were asked to rinse their mouths thoroughly and then to swallow and finally collect their saliva in a special sterile container within half an hour. These saliva samples were then inserted into test tubes containing the intermediate substance. They were quickly transferred to the laboratory by maintaining a cold chain (-2.5 ° C) and in a container containing ice packs. They were stored at -70 ° C and then evaluated by ELIZA Reader (HumaReader HS, Germany) with a special cotinine kit (De salimetrics medi tec, USA).

2-3. Statistical analysis

Data were analyzed using Stata software version 14.0 (Stata Corp, College Station, TX, USA). For descriptive analyses, the mean, standard deviation (SD), and number (%) were reported. Then, univariate and multivariate logistic regression models were used to determine the relationship between decay of deciduous and permanent teeth and passive cigarette smoke in children aged 8-12 years; and finally crude and adjusted odds ratios (OR) with 95% confidence interval (CI) were estimated. Also, the area under the ROC curve was used for assessing the discriminative ability of the Multivariable Logistic Regression model. P-values <0.05 were considered to indicate statistical significance.

2-4. Ethics considerations

Before data collection, the aims of the research were explained to the child's parents; then informed consent was obtained from them. In addition, this study was performed according to the principles expressed in the Declaration of Helsinki and was approved by the Deputy of Research and Ethics Committee of Semnan University of Medical Sciences (Iran) (IR.SEMUMS.REC.1398.177).

3- RESULTS

This cross sectional study was performed to determine the association of passive smoking with dental caries and cotinine biomarkers among 8-12-Year-old children of Semnan, Iran. A total of 100 children exposed to passive smoking and 100 children unexposed to passive smoking were enrolled in the study. **Table 1** shows the demographic and dental characteristics of the children under study in the two groups of exposed and unexposed to passive smoking. As can be seen, the mean age of children in exposed and unexposed groups with passive smoking were 9.64 ± 1.31 and 10 ± 1.64 years, respectively. The number of boys

was 58 (58) and 50 (50) for exposed and unexposed groups, respectively. Also, the means (±S.D) of the salivary cotinine level, decayed, missing, and filled teeth (dmft) index, decayed, and filled teeth (dft) index and missing teeth (mt) for exposed and unexposed groups with passive smoking were 11.37 ± 0.27 vs. 11.16 ± 0.31, 2.06 ± 2.07 vs. 1.92 ± 1.92, 1.87 ± 1.69 vs. 1.44 ± 1.54 and 0.74 ± 0.96 vs. 0.59 ± 0.84; respectively. In addition, for permanent teeth, the means (±S.D) of decayed, missing, and filled teeth (DMFT) index, decayed, and filled teeth (DFT) in exposed and unexposed groups were 1.26 ± 0.97 vs. 1.22 ± 0.98. Other details of these variables can be seen in **Table 1**.

Table-1: Demographic and dental characteristics of the children under study in the case and control groups

Quantitative Variables	Groups	Number	Mean	S.D***	Min	Max
Age (year)	Exposed *	100	9.64	1.31	8	12
	Un-exposed **	100	10	1.46	8	12
Salivary cotinine level	Exposed	100	11.37	0.27	10.85	11.96
	Un-exposed	100	11.16	0.31	11.50	10.48
decayed, missing, and filled teeth (dmft) index	Exposed	100	2.06	2.07	0	8
	Un-exposed	100	1.92	1.92	0	7
decayed, and filled teeth (dft) index	Exposed	100	1.87	1.69	0	8
	Un-exposed	100	1.44	1.54	0	7
missing teeth (mt)	Exposed	100	0.74	0.96	0	4
	Un-exposed	100	0.59	0.84	0	4
Decayed, Missing, and Filled Teeth (DMFT) index	Exposed	100	1.26	0.97	0	4
	Un-exposed	100	1.22	0.98	0	4
Decayed, and Filled Teeth (DFT) index	Exposed	100	1.26	0.97	0	4
	Un-exposed	100	1.22	0.98	0	4
Missing Teeth (MT)	Exposed	100	0	0	0	0
	Un-exposed	100	0	0	0	0

Qualitative Variables		Groups	
		Exposed (%)	Un-exposed (%)
Sex	Boy	58 (58)	50 (50)
	Girl	42 (42)	49 (49)

**Case: Exposed to passive smoking
** Control: Un-exposed to passive smoking
***S.D: Standard Deviation

Table 2 shows the relationship between decay of deciduous and permanent teeth and passive cigarette smoke in children aged 8- 12 years by univariate logistic regression model. As can be seen, a statistically significant difference was found between the exposed and unexposed groups with passive smoking in terms of the salivary cotinine level, age , sex , decayed, missing, and filled teeth (dmft) index, decayed, and filled teeth (dft) index and missing teeth (mt) (P-Value ≤0.20).

Table-2: The relationship between decay of deciduous and permanent teeth with passive cigarette smoke in children aged 8- 12 years by univariate logistic regression model

Variable		Crude OR	95% Confidence Interval	P-Value
Age (year)	8	Reference	-	-
	9	0.78	0.34 – 1.75	0.545
	10	0.99	0.41 – 2.40	0.982
	11	0.86	0.35 – 2.15	0.754
	12	0.34	0.13 – 0.88	0.026
Sex		1.42	0.81 – 2.28	0.199
Salivary cotinine level		14.32	0.85 - 242	0.065
decayed, missing, and filled teeth (dmft) index		1.19	1.03 – 1.37	0.018
decayed, and filled teeth (dft) index		1.18	0.99 – 1.41	0.064
missing teeth (mt)		1.21	0.88 – 1.65	0.195
Decayed, Missing, and Filled Teeth (DMFT) index		1.05	0.79 – 1.40	0.740
Decayed, and Filled Teeth (DFT) index		1.05	0.79 – 1.40	0.740
Missing Teeth (MT)		-	-	-

*P-Values ≤ 0.20 were considered significant to enter the Multivariable Logistic Regression Model

Then, in order to eliminate potential confounding variables, the variables with P-Value ≤0.20 in the univariate logistic regression model were introduced in the multivariate logistic regression model, simultaneously. **Table 3** shows OR and 95% CI derived from multivariate logistic regression model for the relationship between decay of deciduous and permanent teeth with passive smoking in children aged 8 – 12 years. As can be seen, after adjusting for the confounding variables, a statistically significant relationship was observed between the decayed, missing, and filled teeth (dmft) index and passive smoking (OR=1.28; 95% CI: 1.05 – 1.38) in children aged 8 – 12 years (P-Value<0.05). This means that exposure to passive smoking increases the odds of dmft by about 1.28 times after adjusting for the confounding variables.

Fig. 1 demonstrates the area under the ROC curve for significant variables included in the multivariable logistic regression model; the area under the ROC curve was 0.7620.8133, which demonstrates the high discriminative power of this model.

4- DISCUSSION

Smoking is an important risk factor that contributes to the incidence and mortality of many diseases (19). The dangers of secondhand smoke in passive smokers are almost the same as in active smokers, and most importantly, children in the early years of development are more vulnerable than adults (20). The results of univariate logistic regression model showed a statistically significant difference between the exposed and unexposed groups with passive smoking in term of the salivary cotinine level, age , sex , decayed, missing, and filled teeth (dmft) index, decayed, and filled teeth (dft) index and missing teeth (mt) (P-Value ≤0.20). However, the results of multiple logistic regression model demonstrated a statistically significant relationship only

between the decayed, missing, and filled teeth (dmft) index and passive smoking (OR=1.28; 95% CI: 1.05 – 1.38).

Table-3: The relationship between decay of deciduous and permanent teeth with passive cigarette smoke in children aged 8- 12 years by multivariate logistic regression model

Variable		Adjusted OR	95% Confidence Interval	P-Value
Age (year)	8	Reference	-	-
	9	0.41	0.04 – 4.31	0.456
	10	1.41	0.06 – 35.83	0.832
	11	1.58	0.05 – 38.40	0.994
	12	0.09	0.001 – 5.58	0.260
Sex		0.36	0.05 – 2.42	0.313
Salivary cotinine level		14.20	0.42 – 475.02	0.139
decayed, missing, and filled teeth (dmft) index		1.28	1.05 – 1.38	0.015
decayed, and filled teeth (dft) index		1.43	0.98 – 1.58	0.197
missing teeth (mt)		1.33	0.82– 1.68	0.255

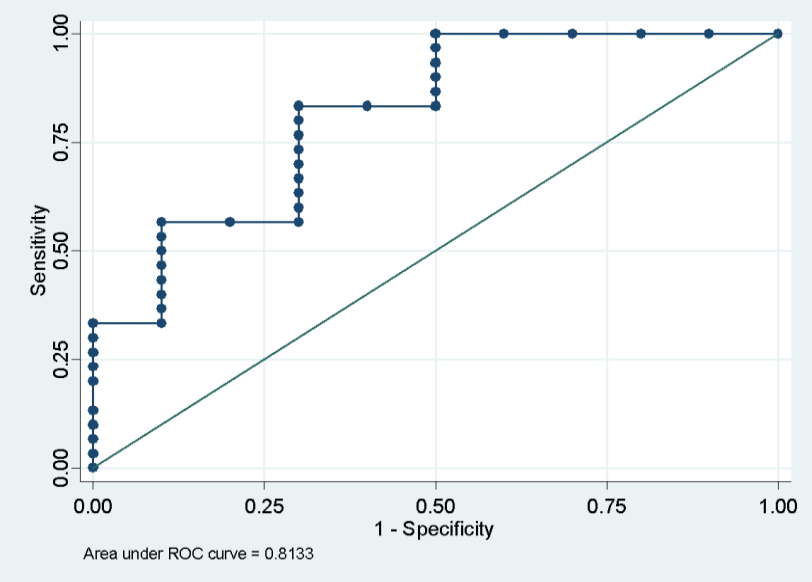

Fig. 1: The area under the curve for the variables entered in multivariable logistic regression model

The results of our study are consistent with most studies in this field. A study by Jakhete et al. aimed to investigate the relationship between environmental cigarette smoke and the prevalence of dental caries in low-income children showed that the risk of dental problems in children with low socioeconomic status was twice as high as children with high socioeconomic status. Moreover, the prevalence of dental problems in them was over 50%. Passive cigarette smoke, lead exposure and food deprivation have been cited as important and effective factors for this difference (21). Another study in the United States aimed to determine the association between environmental tobacco smoke and dental caries in

children 4-7 years of age. It showed that the prevalence of tooth decay was higher in children with active family smokers. In addition, the results of multiple logistic regression model after adjusting the effect of age, socioeconomic status, total fluoride uptake and soft drink consumption showed a statistically significant relationship between smoking and tooth decay (OR=3.38; P-Value =0.001) (22). Mattheus et al. in a cross-sectional study have shown that caries prevalence in children exposed to secondhand smoke at home is 1.59 times more than children un-exposed (23). Consistent with the studies mentioned and our study, a conducted study by Tanaka et al. also found that the dmft score in children exposed to passive smoking was significantly higher than that in the control group (24).

Various indicators have been proposed to determine the importance and extent of exposure to passive smoking. Cotinine is one of the valid and suitable biomarkers for determining the amount of nicotine absorption and exposure to environmental cigarette smoke (25-27). In the study, the mean concentration of cotinine in children exposed to passive smoking was higher than that in children who were not exposed to it; however, the difference was not statistically significant, which was consistent with a number of studies conducted in this field. For example, a study by Ahmadi, et al. in Iran demonstrated that there is no significant difference between the two groups of children exposed and non-exposed to environmental smoking in terms of the level of cotinine (7). In contrast, the results of some other studies were not consistent with those of our study. Nishida et al. (28) and Erdemir et al. (15) reported that the mean salivary cotinine concentration in children exposed to passive smoking was higher than in children not exposed. In addition, the study by Menon et al. aimed to determine the relationship between passive smoking, dental caries and salivary biomarkers among 5–10-year-old children. It indicated that the salivary cotinine levels in passive smoking subjects (case group) were significantly higher than the control group (1.08 ± 0.265 vs. 0.0 ± 0.00) and streptococcus colonies, lactobacillus colonies, dmft and gingival index (GI) scores, and smoking exposure were the most important factors that were directly related to salivary cotinine levels (14). Perhaps one of the reasons for the inconsistency of the results of our study with these studies is the air pollution caused by factories and vehicles. In addition, wind direction should also be considered as an effective factor in the accumulation of air pollutants (28, 29).

These studies have a number of strengths and weaknesses. One of the strengths of this study is the comparison of two groups with equal sample sizes, which increases the statistical efficiency. The second one is the measurement of salivary cotinine levels in 2 groups. However, the study has some limitations, too. First, smoking behavior and subsequent division of children into exposed and non-exposed groups was obtained through questions from the children's parents, which may be accompanied by degrees of differential misclassification bias. Second, this cross-sectional study was performed with a relatively low sample size and the measurements for salivary cotinine levels and caries indices were performed only once. Thus, longitudinal studies with large sample sizes are recommended in order to carefully investigate the causal relationships of exposure and outcome. Third, factors such as the exact amount of exposure to secondhand smoke in children according to pack-year, vitamin C concentration and body mass index (BMI) that may be effective on salivary cotinine levels have not been considered. Finally, the amount of saliva flow, which is an

effective factor in tooth decay, has not been measured.

5- CONCLUSION

The present study suggests that the risk of decay of deciduous teeth is higher in children exposed to passive smoking. Therefore, passive smoking should be considered in the implementation of programs to prevent tooth decay in children.

6- AUTHOR CONTRIBUTIONS

OM and HD designed the study. OM and RGH supervised the study. RGH and HD processed the data. RGH did the statistical analysis. RGH and OM interpreted the results. OM and HD wrote the original draft. OM and HD reviewed and editted the final draft. All authors have read and approved the final manuscript.

7- COMPETING INTERESTS

The authors declare that they have no competing interests.

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