Bull. Environ. Contam. Toxicol. (2003) 71:1–5

DOI: 10.1007/s00128-003-0121-2

Bulletin of Environmental Contamination and Toxicology

Passive Exposure to Tobacco Smoke: Hair Nicotine Levels in Preschool Children

N. Kalinić, Lj. Skender, V. Karačić, I. Brčić, V. Vadjić

Institute for Medical Research and Occupational Health, Ksaverska c. 2, 10000 Zagreb, Croatia

Received: 15 December 2002/Accepted: 14 April 2003

Hair nicotine levels were measured in children to establish the utility of the method in assessing exposure to environmental tobacco smoke (ETS).

There is increasing evidence of an association between children's exposure to ETS and diseases and disorders such as airway infections, asthma and reduced lung function (Martinez et al.,1992; Ehrlich et al., 1992; Forastiere et al., 1994). Methods for determining the degree of ETS exposure has received much attention in recent years, and various biological markers have been studied such as urine cotinine, whose measurements were accepted world wide (Haufroid and Lison, 1998). However, these measurements can reflect a day or two old exposure to nicotine (Benowitz and Jacob, 1993) and samples may be difficult to obtain, especially in young children. Moreover, metabolism and elimination of cotinine are influenced by factors such as age, sex, diurnal variation, liver enzyme activity, and kidney function. The measurement of nicotine in hair is an effective noninvasive method. It has many advantages over the urine cotinine measurement (Kintz et al., 1992; Mizuno et al., 1993). In particular, hair may provide information on long-term exposure. Each centimeter of hair reflects approximately one month of exposure, as hair has a growth rate of 1±0.3 cm/month (Uematsu et al., 1995). Nicotine is preserved in the hair shaft throughout the life of the hair, and after the hair is cut, samples can be stored for years at room temperature in a closed envelope without loss or degradation of nicotine (Zahlsen and Nilsen, 1994).

This study compared nicotine concentrations in children's hair with the reported smoking status within the households where children live.

MATERIAL AND METHODS

The participants in this study were 22 girls and 11 boys, aged 5 to 7 years, from one kindergarten in Zagreb, Croatia. Parents who gave their informed consent on behalf of their children were asked to answer questions about the smoking habits in their household as well as about their socio-economic status and education. The hair samples were collected in March 2001.

A tress of hair of about 5 mm in diameter was cut near the root from the rear of the scalp. Hair growth in this area is reported to be relatively uniform, so that each centimeter represents approximately one month of exposure to ETS. The hair samples were stored in paper envelopes at room temperature until analysis. 50 mg hair samples were used for analysis and dissolved by incubation in 2 mL of 5 M sodium

Correspondence to: N. Kalinić

hydroxide for 20 min at 70 °C. After cooling 3 mL of diethyl ether containing 5 mg/L of the internal standard 2,6-di-t-butyl-4-methylphenol, and 100 μL of n-butyl acetate were added. The extraction was performed under a vigorous vortex mixing for 15 minutes. The organic layer was transferred to a conical glass tube and reduced to approximately 100 μL under a stream of nitrogen; 1 μL was injected into GC/MS (Zahlsen and Nilsen, 1994).

The analyses were performed using a Varian 3400 CX GC with Saturn ion trap mass spectrometer. The chromatographic column was Rtx-5 (5% diphenyl-95% dimethyl polysiloxane) 30 m, 0.25 mm i.d. capillary column with a 0.25 μm thick film. The initial column temperature of 50 °C was held for 1 minute, then programmed to 260 °C at 50 °C/min and held for 1 minute. Ultra-pure grade helium was used as the transport gas at a flow rate of about 1 mL/mm. Septum-equipped programmable injector (SPI) was used; the initial temperature of 40 °C was held for 0.1 min, then programmed to 280 °C at 200 °C/min and held for 8 minutes. The transfer line temperature was 260 °C.

Nicotine and the internal standard 2,6-di-t-butyl-4 methylphenol were detected by selected ion monitoring. For nicotine, a mass to charge ratio (m/z) of 162 was used for quantitative calculation.

RESULTS AND DISCUSSION

Table 1 shows household smoking habits and the most common situations in which children were exposed to ETS. According to parents' reports, 20 of the 33 children (60.6%) were exposed to ETS.

Table 1. Summary of reported exposure to enviromental tobacco smoke for 33 children in Zagreb, Croatia in 2001

	Children No	*(%)*	*Range*[x]
Mother smokes	7	21.2	1-20
Father smokes	10	30.3	1->20
Mother and father smoke	2	6.1	1->20
Other person smoke	1	3.0	1-10
Unexposed children	13	39.4	

[x]Number of cigarettes a day

Figure 1 shows ion chromatogram of nicotine in the control hair. Control hair was fortified with 2 ng/mg nicotine. Hair samples of children whose parents did not smoke had 0.56 ng/mg nicotine, what is presented at Figure 2, whereas the hair of children with the parents who smoked had 7.69 ng/mg nicotine (Figure 3).

Nicotine hair levels ranged from below the detection limit to 7.89 ng/mg of hair (Figure 4).

The difference in hair nicotine levels between children who lived with smokers (range=1.29 to 7.89 ng/mg of hair) and those who did not live with smokers (range=0.27 to 4.42 ng/mg of hair) was highly significant ($t=5.456001$; $P<0.01$).

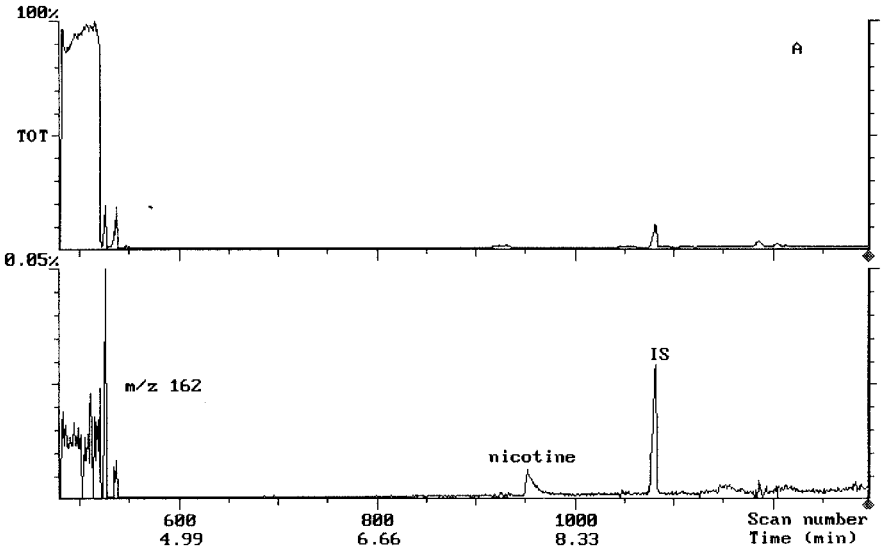

Figure 1. Ion chromatogram of nicotine in control hair
IS=internal standard

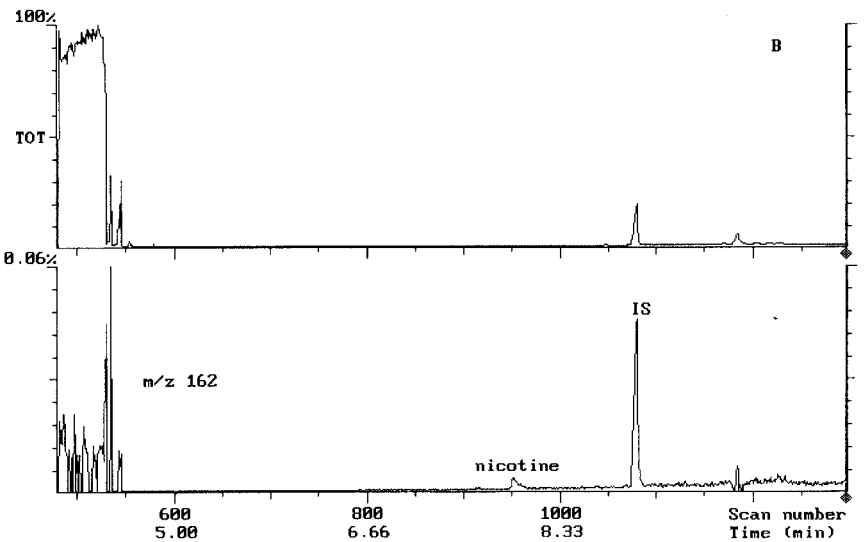

Figure 2. Ion chromatogram of nicotine in nonexposed children

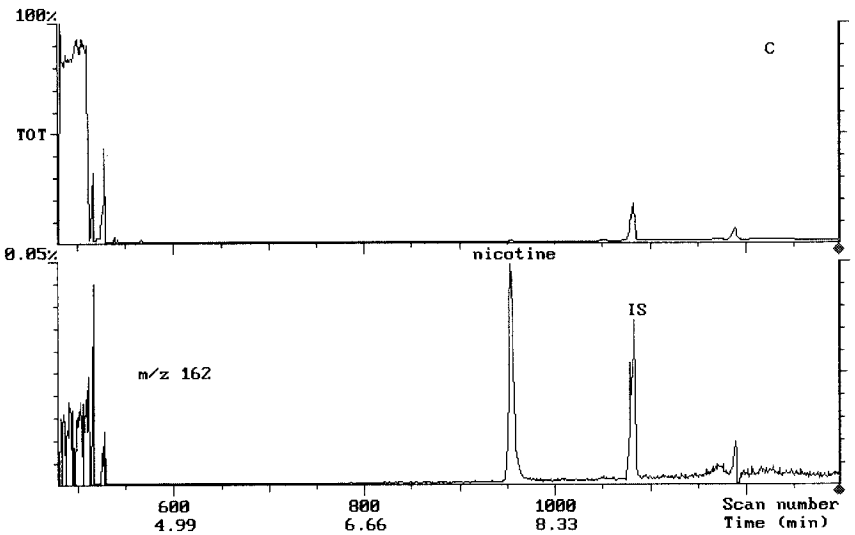

Figure 3. Ion chromatogram of nicotine in exposed children

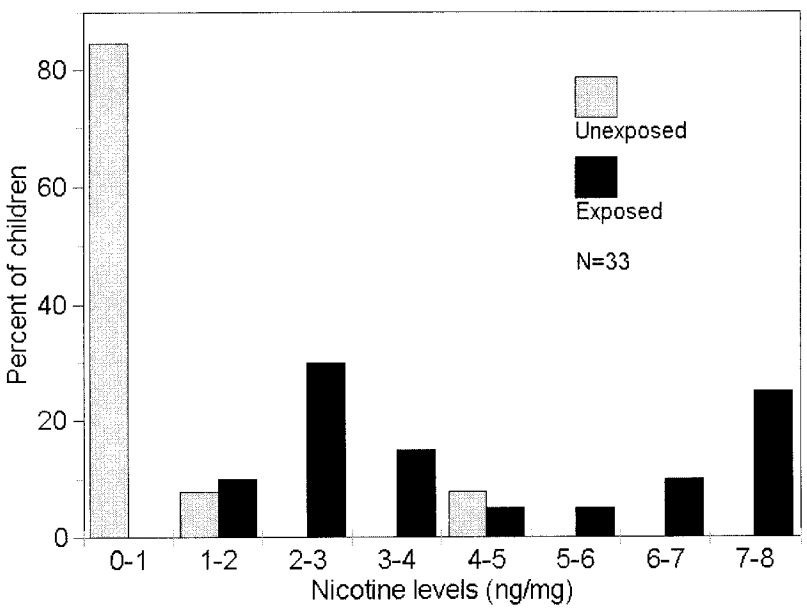

Figure 4. Nicotine levels in the hair of children reported to be exposed or unexposed to environmental tobacco smoke in the questionnaire

In this study, children who shared their household with smokers showed much higher nicotine concentrations in hair than children who lived with nonsmokers. For example, there were two children (only) whose both parents smoked. One showed the highest (7.89 ng/mg) and the other very high (7.20 ng/mg) hair nicotine level. The concentrations were similar to the concentrations found among 94 children, aged 12-36 months by Nafstad, Botten, Hagen, et al., 1995, and to 297 children, aged 3-27 months (Mahoney and Al-Delaimy, 2001). They were, however, higher than those reported by Al-Delaimy, Crane, and Woodward in 2001 for 117 children aged 3 months to 10 years. Only one nicotine concentration of a child who lived with parents nonsmokers (4.42 ng/mg of hair) deviated from other non-exposed children. Additional questions did not give an answer that would explain this result. Perhaps, the parents provided inaccurate information and/or the child was exposed to ETS in some other way. At the same time, similar results found in the hair of two sisters living in the same household suggest that the measurement of ETS exposure was accurate (0.59 and 0.36 ng/mg of hair). Hair nicotine analysis has the advantage of being objective, individual and quantitatively specific and sensitive.

REFERENCES

Al-Delaimi WK, Crane J, Woodward A (2001) Passive smoking in children: effect of avoidance strategies at home as measured by hair nicotine levels. Arch Environ Health 56:117-122.

Benowitz NL, and Jacob P (1993) Nicotine and cotinine elimination pharmacokinetics in smokers and non-smokers. Clin Pharmacol Therapeut 53:316-323.

Ehrlich R, Katten M, Godbold J et al. (1992) Childhood asthma and passive smoking. American Rev Resp Dis 145:594-599.

Foastiere F, Agabiti N, Giuseppe M et al. (1994) Passive smoking as a determinant of bronchial responsiveness in children. American J Resp Crit Care Med 149:365-370.

Haufroid V, Lison D (1998) Urinary cotinine as a tobacco-smoke exposure index: a minireview. International Arch Occup Environ Health 71:162-168.

Kintz P, Ludes B, and Mangin P (1992) Evaluation of nicotine and cotinine in human hair. J Foren Sci 37:72-76.

Mahoney GN, and Al-Delaimy W (2001) Measurement of nicotine in hair by reversed-phase high-performance liquid chromatography with electrochemical detection. J Chromatog B 753:179-187.

Martinez FD, Cline M, and Burrows B (1992) Increased incidences of asthma in children of smoking mothers. Pediatrics 89:21-26.

Mizuno A, Uematsu T, Oshima A et al. (1993) Analysis of the nicotine content of hair and its utilization for assessing individual cigarette-smoking behavior. Therapy Drug Monit 15:99-104.

Nafstad P, Botten G, Hagen JA et al. (1995) Comparison of three methods for estimating environmental tobacco smoke exposure among children aged between 12 and 36 months. Int J Epidemiol 24:88-94.

Uematsu T, Mizuno A, Nagashima S et al. (1995) The axial distribution of nicotine content along hair shafts as an indicator of changes in smoking behaviour: evaluation in a smoking cessation programme with or without the aid of nicotine chewing gum. British J Clin Pharmacol 39:665-669.

Zahlsen K, and Nilsen OG (1994) Nicotine in hair of smokers and non-smokers: sampling procedure and gas chromatographic/mass spectrometric analysis. Pharmacol Toxicol 75:143-149.