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## Breast milk lead and cadmium levels from suburban areas of Ankara

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#### ARTICLE INFO

Article history:
Received 1 November 2010
Received in revised form 8 February 2011
Accepted 22 February 2011
Available online 14 April 2011

Keywords:
Lead
Cadmium
Breast milk
Edinburgh Postpartum Depression Scale
Smoking
Anthropometric measurement

#### ABSTRACT

The objectives of this study were (1) to evaluate levels of lead (Pb) and cadmium (Cd) in the breast milk at 2 months postpartum, (2) to investigate the relationship between Pb and Cd levels in breast milk and some sociodemographic parameters and (3) to detect whether these levels have any influence on the infant's physical status or on postpartum depression in the mothers. Pb and Cd levels in breast milk were determined by Inductively Coupled Plasma Mass Spectroscopy (ICP-MS). The median breast milk concentrations of Pb and Cd were 20.59 and 0.67 µg/l, respectively. In 125 (87%) of 144 samples, Pb levels were higher than the limit in breast milk reported by the World Health Organization (WHO) (>5 µg/l). Breast milk Cd levels were >1 µg/l in 52 (36%) mothers. The mothers with a history of anemia at any time had higher breast milk Pb levels than those without a history of anemia (21.1 versus 17.9  $\mu$ g/l; p=0.0052). The median breast milk Cd levels in active and passive smokers during pregnancy were significantly higher than in non-smokers (0.89, 0.00 µg/l, respectively; p = 0.023). The breast milk Cd levels of the mothers who did not use iron and vitamin supplements for 2 months postpartum were found to be higher than in those who did use the supplements (iron: 0.73,  $0.00 \mu g/l$ , p = 0.023; vitamin: 0.78,  $0.00 \mu g/l$ , p = 0.004, respectively). Breast milk Cd levels at the 2nd month were correlated negatively with the z scores of head circumference and the weight for age at birth (r = -0.257, p = 0.041 and r = -0.251, p = 0.026, respectively) in girls. We found no correlation between the breast milk Pb and Cd levels and the Edinburgh Postpartum Depression Scale scores. Breast milk monitoring programs should be conducted that have tested considerable numbers of women over time in view of the high levels of Pb in breast milk in this study.

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#### 1. Introduction

The growing population, rapid industrialization and urbanization have increased the exposure to environmental pollutants of all living systems (Järup, 2003; Wong and Lye, 2008). Lead (Pb) and cadmium (Cd) are reported among the top 10 toxic metals in the Priority List of Hazardous Substances announced by the Agency for Toxic Substances and Disease Registry (ATSDR, 2007).

Pb is one of the neurotoxicants that seems to be involved in the etiology of psychological pathologies. Experimental studies have shown that exposure to Pb during pregnancy and/or lactation results in depression in female rats (Lisboa et al., 2005), whereas an antidepressant-like effect related to subchronic exposure has been reported in mice (Soeiro et al., 2007). In adults, exposure to Pb has

been found to be associated with anxiety and depression (Wang et al., 2006; Dang et al., 2008). Similarly, Cd has caused neurotoxicologic and behavioral changes in both humans and experimental animal studies (Wang et al., 2006; Sinha et al., 2008; Dési et al., 1998). Cd exposure may be implicated in some neurological disorders including hyperactivity and increased aggressiveness in humans (Matés et al., 2010). Nonetheless, the observational clinical data in the literature on the relationship between anxiety or depression and Cd exposure are inadequate.

Pb and Cd can negatively influence growth in newborns. Several studies have reported an inverse relationship between anthropometric measurements of the newborn and the placental or umbilical cord Pb/Cd levels (Atabek et al., 2007; Zentner et al., 2006; Llanos and Ronco, 2009; Ronco et al., 2009). Previous researches have shown that anthropometric measurements during the infancy period and childhood are influenced by exposure to Pb (Frisancho and Ryan, 1991; Ballew et al., 1999).

The first exposure to toxic metals in humans takes place in the intrauterine period and exposure lingers thereafter, primarily through breast milk, breathing air and drinking water. In the lactation period, toxic metals such as Pb and Cd mobilize from the maternal stores and

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transfer from the maternal blood into the breast milk, which is the ideal nutritional source for the infant (Gundacker et al., 2002; Hallén et al., 1995; Leotsinidis et al., 2005; Sternowsky and Wessolowski, 1985; Ursinyova and Masanova, 2005). The mobilization that begins during pregnancy has been shown to continue during the lactation period (Barbosa et al., 2005). Learning about the toxic metal levels in the breast milk is important for two reasons: first, as a pathway of exposure, and second, as an indicator of likely prenatal exposure (Järup, 2003).

A limited number of studies are available in Turkey regarding the determination of the toxic metal concentrations in breast milk (Dursun, 2008; Ermiş et al., 1994; Hakan, 2002; Kirel et al., 2005; Turan et al., 2001). In our study, we aimed to detect the Pb and Cd levels in breast milk during lactation at the 2nd postpartum month and to investigate the effect of some sociodemographic parameters on these concentrations. Furthermore, we planned to evaluate the influence of these toxic metals on the infant's physical growth and on postpartum depression in the mothers.

#### 2. Materials and methods

The mothers residing in a suburban area who delivered their babies at the Zekai Tahir Burak Maternity Hospital in Ankara, Turkey were invited at 2 months postpartum as a part of a previous study (Yalcın et al., 2010). A total of 144 voluntarily enrolled healthy mothers were evaluated at İhsan Doğramacı Children's Hospital, and all subjects provided breast milk samples at 2 months postpartum. These mothers were not occupationally exposed to toxic metals. They were living in a suburban area but not a non-industrial area of Ankara. All the mothers completed a questionnaire to provide details regarding some of the maternal and infant characteristics, including occupation, smoking habits, reproductive history, history of anemia at any time, and intake of iron and vitamin supplementation during pregnancy and within 2 months after delivery. The level of maternal hemoglobin on 1st day postpartum was taken from the hospital records. In addition, all mothers were evaluated using the Edinburgh Postpartum Depression Scale (EPDS) to identify postpartum depression risk (Engindeniz, 1996). The infants were examined, and their weight, length and head circumference were recorded. The study was approved by the Ethical Committee of the Faculty of Medicine, Hacettepe University (HEK 07/97-10). The mothers were informed about the purpose of the study and a written consent was obtained from all participants.

#### 2.1. Analysis of the breast milk

The breast milk samples were collected by manual suckling 2 h following the latest feeding session in the morning. The mothers provided 10 ml of milk directly into clean polyethylene tubes. All the samples were frozen immediately after collection and kept frozen  $(-20\,^{\circ}\text{C})$  until they were analyzed.

In order to minimize the polyatomic interferences, 65% nitric acid, (concentrated grade (MERCK 452)), reagent water equivalent to ASTM Type 1 (ASTM D 1193; >18 m $\Omega$  cm resistivity), 0.5% (v/v) nitric acid, a tuning solution (for sensitivity tuning: 10 µg/l for each of the metals Li, Y, and Tl in 1% HNO<sub>3</sub>), and a single-element standard stock solution for Pb and Cd (1000 ppm) were used. 1 g of breast milk sample was accurately weighed, put into a dry XP1500 vessel, and 5 ml of HNO<sub>3</sub> along with 5 ml of distilled water was added. For digestion, this mixture was placed in a microwave oven. Following digestion, the samples were diluted with 25 ml distilled water. The standard working solutions and the milk samples in the polyethylene tubes were placed in the automatic sequencer of the Inductively Coupled Plasma Mass Spectrometry (ICP-MS). The lowest limits of quantitation [LOQ] for Pb and Cd were 0.2 µg/l (Yokogawa Analytical Systems Inc., 2005).

#### 2.2. Statistics

The data were analyzed using the SPSS Windows 14.0 (SPSS Inc, Chicago, IL, USA). The results are presented with median values and 25th–75th percentiles. Because, the distribution of Pb and Cd levels was normally skewed, the Mann–Whitney U test was applied to compare the Pb and Cd levels between the subgroups, which were defined by various maternal and infant characteristics. The Pearson correlation coefficient was used for the relationship between breast milk Pb and Cd concentrations and numerical variables (anthropometric measurements, EPDS scores, etc.). The anthropometric measurements among children, the z scores of weight-for-age (WAZ), length-for-age (LAZ), head circumference for age (HCZ), and body mass index for age (BAZ) were calculated on the basis of recent World Health Organization Growth References (WHO, 2006). The statistical significance was accepted at p<0.05.

#### 3. Results and discussion

The mean maternal age  $(\pm\,\mathrm{SD})$  was 25  $(\pm\,5)$  years (minmax = 17-41). Seven (4.9%) mothers were older than 35 years. Ninety-nine (68.8%) of the mothers and 72 (50%) of the fathers had been educated for less than 8 years. Only 11 (7.6%) mothers were working. The monthly income was <US\$250 in 25 (18%) families, between US\$250-\$500 in 77 (53.5%) families, and >US\$500 in 42 (28.5%) families (Table 1). Mean gravity  $(\pm\,\mathrm{SD})$  was 2.0  $(\pm\,0.9)$  and mean parity  $(\pm\,\mathrm{SD})$  was 1.6  $(\pm\,0.8)$ . The birth interval was less than 2 years in 10 (15.9%) mothers who had parity >1. The mean maternal Hb level  $(\pm\,\mathrm{SD})$ , analyzed at the postpartum 1st day, was 11.6  $(\pm\,1.8)$  (min-max = 6.5-15.7) g/dl.

Seventy-eight (54.2%) infants were female and 66 (45.8%) were male. Nine (6.3%) of the infants had birth weight <2500 g and 17 infants were born at <37 weeks (Table 1). The mean birth weight ( $\pm$ SD) was 3226 $\pm$ 445 g (min-max=2030-4080) and the mean head circumference was 35.0 $\pm$ 1.4 cm (min-max=32.0-38.0). At two months of age, the mean infant weight ( $\pm$ SD) was 4755 $\pm$ 684 g, the mean infant length ( $\pm$ SD) was 55.2 $\pm$ 2.6 cm and the mean infant head circumference ( $\pm$ SD) was 38.0 $\pm$ 1.5 cm (Table 1).

The levels of Pb and Cd in 144 breast milk samples, taken at 2 months postpartum, are presented in Table 2. The Pb level in 137 (95%) breast milk samples was above the detection limit ( $>0.2 \,\mu g/l$ ) and in 125 (87%) samples, levels were higher than the Pb limit ( $>5 \,\mu g/l$ ) as reported by the World Health Organization (WHO) in breast milk for the first 3 months of lactation under normal conditions. The median (25–75 percentile) Pb level was 20.6 (12.1–29.2)  $\mu g/l$ . In one case, the Pb concentration in breast milk was 1515  $\mu g/l$ . This value was confirmed by a duplicate analysis. The Cd level in the same breast milk sample was 43.02  $\mu g/l$ . This was also the

**Table 1**Characteristics of the study population.

Characteristics	Mean ± SD (range), N (%)
Mothers	<u> </u>
Age (y)	$25 \pm 5 (17-41)$
Housewife	133 (92.4)
Education level ≤8 years	99 (68.8)
Gravity	$2.0 \pm 0.9 \ (1-10)$
Parity	$1.6 \pm 0.8 \; (1-7)$
Infants	
Gender, girls	78 (54.2)
Gestational age (wk)	$38.6 \pm 1.7 \ (32.0 - 41.0)$
Birth weight (g)	$3226 \pm 445 \ (2030 - 4080)$
Head circumference at birth (cm)	$34.7 \pm 1.4 \ (32.0 - 38.0)$
Weight at 2 months of age (g)	$4755 \pm 684 \ (2650 - 6800)$
Length at 2 months of age (cm)	$55.17 \pm 2.58 \ (47.5 - 61.0)$
Head circumference at 2 months of age (cm)	$38.0 \pm 1.5 \ (33.0 - 41.0)$

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