



Mercury, lead and cadmium in human milk in relation to diet, lifestyle habits and sociodemographic variables in Madrid (Spain)

Esther García-Esquinas^{a,b,*}, Beatriz Pérez-Gómez^{a,b}, Mario Antonio Fernández^c, Ana María Pérez-Meixeira^d, Elisa Gil^d, Concha de Paz^d, Andrés Iriso^d, Juan Carlos Sanz^d, Jenaro Astray^d, Margot Cisneros^d, Amparo de Santos^d, Angel Asensio^d, José Miguel García-Sagredo^e, José Frutos García^d, Jesus Vioque^{b,f}, Marina Pollán^{a,b}, Gonzalo López-Abente^{a,b}, Maria José González^c, Mercedes Martínez^g, Pedro Arias Bohigas^h, Roberto Pastor^a, Nuria Aragonés^{a,b}

^a Environmental and Cancer Epidemiology Unit, National Center for Epidemiology, Carlos III Institute of Health (ISCIII), Madrid, Spain

^b Consortium for Biomedical Research in Epidemiology and Public Health (CIBERESP), Spain

^c Instrumental Analysis and Environmental Chemistry Department, Organic Chemistry Institute, CSIC, Spain

^d Madrid Regional Health and Consumer Affairs Authority, Madrid, Spain

^e Medical Genetics Department, Ramon y Cajal University Teaching Hospital, Madrid, Spain

^f Public Health Department, Miguel Hernandez University, Alicante, Spain

^g Health Prevention and Environmental Health Department, Madrid, Spain

^h Institute for Health Information, Ministry of Health and Social Policy, Madrid, Spain

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ABSTRACT

Background: Although breastfeeding is the ideal way of nurturing infants, it can be a source of exposure to toxicants. This study reports the concentration of Hg, Pb and Cd in breast milk from a sample of women drawn from the general population of the Madrid Region, and explores the association between metal levels and socio-demographic factors, lifestyle habits, diet and environmental exposures, including tobacco smoke, exposure at home and occupational exposures.

Methods: Breast milk was obtained from 100 women (20 mL) at around the third week postpartum. Pb, Cd and Hg levels were determined using Atomic Absorption Spectrometry. Metal levels were log-transformed due to non-normal distribution. Their association with the variables collected by questionnaire was assessed using linear regression models. Separate models were fitted for Hg, Pb and Cd, using univariate linear regression in a first step. Secondly, multivariate linear regression models were adjusted introducing potential confounders specific for each metal. Finally, a test for trend was performed in order to evaluate possible dose–response relationships between metal levels and changes in variables categories.

Results: Geometric mean Hg, Pb and Cd content in milk were 0.53 $\mu\text{g L}^{-1}$, 15.56 $\mu\text{g L}^{-1}$, and 1.31 $\mu\text{g L}^{-1}$, respectively. Decreases in Hg levels in older women and in those with a previous history of pregnancies and lactations suggested clearance of this metal over lifetime, though differences were not statistically significant, probably due to limited sample size. Lead concentrations increased with greater exposure to motor vehicle traffic and higher potato consumption. Increased Cd levels were associated with type of lactation and tended to increase with tobacco smoking.

Conclusions: Surveillance for the presence of heavy metals in human milk is needed. Smoking and dietary habits are the main factors linked to heavy metal levels in breast milk. Our results reinforce the need to strengthen national food safety programs and to further promote avoidance of unhealthy behaviors such as smoking during pregnancy.

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Abbreviations: Hg, mercury; Cd, cadmium; Pb, lead; $\mu\text{g L}^{-1}$, micrograms per liter.

* Corresponding author. Address: Área de Epidemiología Ambiental y Cáncer, Centro Nacional de Epidemiología, Instituto de Salud Carlos III, C/Sinesio Delgado 6, 28029 Madrid, Spain; Tel.: +34 91 822 26 88; fax: +34 91 387 78 15.

E-mail address: egarciag@isciii.es (E. García-Esquinas).

1. Introduction

Breastfeeding is the recommended way of feeding and nurturing infants worldwide (WHO, 2003). Nevertheless, breast milk can also be a pathway of maternal excretion of toxic elements.

The mere presence of these chemicals in breast milk would not necessarily indicate a health risk for lactating infants, but since health effects are biologically plausible, biomonitoring surveillance and research in this area is considered a public health priority (Landrigan et al., 2002).

Exposure of newborns to mercury (Hg), lead (Pb) and cadmium (Cd) is of special interest due to its widespread use and toxicity (Jarup, 2003). Hg and Pb can adversely affect infants' nervous systems (Dorea, 2004; Bellinger, 2008). Though pregnancy is reported to be the most hazardous period for mercury exposure via trans-placental transference (Dorea, 2004), lactating females have also an increased Hg clearance rate, explained at least in part, by the excretion of this metal (Ramirez et al., 2000). Even if methylmercury, the most neurotoxic form, is barely transferred to milk (Solomon and Weiss, 2002), this organic form of Hg is absorbed almost completely from the breast-fed infants gastrointestinal tract and can readily cross through the brain barrier (Bottcher et al., 1987; Jarup, 2003). Inorganic forms, in contrast, are easily excreted in milk, but only a small percentage is absorbed by the neonate and they rarely penetrate brain barriers since they are not lipophilic.

Pb levels in breast milk are usually higher than those of Hg. Pb from past exposures accumulates in the mother's bones, is released along with calcium into her blood, and subsequently makes its way into her breast milk (Ettinger et al., 2006). Some authors have estimated that, for the first three months post-delivery, 36–80% of all blood Pb in breast-fed infants comes from mother's milk (Gulson et al., 2003).

Regarding Cd, this heavy metal is a well-known human carcinogen. Cd levels can also affect brain development in infants, and it has also been suggested that it might increase the risk of premature delivery (Nishijo et al., 2002; Jarup, 2003; Schoeters et al., 2006). Once absorbed, this metal is transported into the liver, where it induces the synthesis of Metallothionein (MT). Cd bound to MT is released into blood and excreted by the kidney, accumulating in the renal cortex over the woman's lifetime (Jarup et al., 1998). Unlike Hg and Pb, very little Cd is transferred across the placenta and only a small percentage reaches breast milk (Vather et al., 2001).

In Spain, the Madrid Regional Health Authority has conducted a biomonitoring surveillance project, known as BioMadrid (Aragones et al., 2008), in order to estimate the levels of selected pollutants in different biological substrates in a sample of father–pregnant woman–newborn trios drawn from the general population. Some of this study's results have already been published (Diez et al., 2009; Lope et al., 2010). This paper reports the level of Hg, Pb and Cd in breast milk samples taken from female BioMadrid participants, and the relationship between these levels with diet, lifestyle habits and socioeconomic variables.

2. Materials and methods

2.1. Study population

The study population was a sub-sample of 100 women who participated in the BioMadrid Project and among whom Hg, Pb and Cd levels were assessed in breast milk samples. The BioMadrid project was a descriptive cross-sectional study, designed to conduct biomonitoring of exposure to environmental pollutants in the general population of Madrid. Both the design and field work have been outlined in detail elsewhere (Aragones et al., 2008). The BioMadrid study sought to recruit 150 couples residing in two geographical areas of the Madrid Region, with half the sample being recruited from the Madrid City district of Vallecas, and the other half being drawn from the Greater Madrid Metropolitan belt (Parla and

Getafe). All women attending pre-natal courses held by the public health system in the participant areas were invited, along with their respective partners, to participate in the BioMadrid Project until the predefined sample size was attained. Enrollment lasted from October 2003 through May 2004. Two women gave birth before the first scheduled date, and three couples failed to attend the scheduled appointments. As a consequence, only 145 couples formally entered the study. Subsequently, one father and 10 newborns were lost to follow-up due to delivery circumstances and complications in logistic procedures. Finally, we recruited 145 pregnant women, 144 fathers and 135 newborns. The study was approved by the Ethics Committee of the Carlos III Institute of Health.

Of the 145 female participants, 129 women (89%) reported that they had started breast feeding their babies. Of these, 22 could not donate a sample of breast milk, due to not having enough milk or experiencing problems in collecting the sample. Finally, 107 women (74%) were able to provide milk samples, though in seven cases there was not enough for laboratory analysis purposes.

2.2. Epidemiologic data and breast-milk collection

Women who decided to participate were given an appointment around the 38th week of pregnancy. Trained interviewers administered individual questionnaires to collect sociodemographic data, information on lifestyles, current and previous pregnancies (including the duration of breast feeding of earlier children), medical history, occupation and environmental exposures. Dietary information was obtained using a food-frequency questionnaire validated in Spain (Vioque, 2006).

At the end of the third week postpartum, women were asked to complete a short lactation questionnaire and to collect a sample of 20 mL of breast milk according to a standardized procedure, using the breast pump provided by the research team. Before sample collection, mothers were asked to avoid using soaps and creams for 3 d. Breasts had to be washed before lactation using tap water, and then dried with gauze. After having breast-fed the baby, expression of milk into the standard sterilized polypropylene pump-collection container was performed. Milk samples were kept refrigerated (2–8 °C) in 50 mL 210 261 EU polypropylene tubes (Soria Genlab, Madrid, Spain) until collected by study personnel, and then frozen and stored at –20 °C at the Regional Public Health Laboratory for subsequent laboratory analysis.

2.3. Laboratory methods

After lyophilization (Telstar Cryodos equipment), an aliquot of 0.5 g of the sample underwent digestion for 5 h in hermetic Teflon containers, using an acid medium (HNO₃ and H₂O₂).

Total Pb and Cd levels were determined using a Perkin Elmer AAnalyst 600 Atomic Absorption Spectrometer (Perkin Elmer Hispania, Madrid, Spain), fitted with a transversely heated graphite atomizer furnace assembly and longitudinal Zeeman-effect background correction. Total Hg was measured by a Perkin Elmer FIMS-400 Atomic Absorption Spectrometer (Perkin Elmer Hispania, Madrid, Spain) using the cold vapor technique (Cold Vapor Atomic Absorption Spectrometer).

Detection limits were as follows: 0.06 µg L⁻¹ for Hg; 0.50 µg L⁻¹ for Cd; and 3.40 µg L⁻¹ for Pb. Lyophilized control material Sero-norm™ (Trace Elements Whole Blood 2, SERO, Billingstand, Norway) was used to verify the precision and accuracy of the analytical measurements. In all cases, analyses were performed after the correct values had been confirmed by the instrumentation. Also, the laboratory tested the milk containers for Hg, Cd and Pb traces by analyzing blanks, and all of them were under the instrumental limit of detection.

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