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Maternal Dietary Intake of Polyunsaturated Fatty Acids Modifies the Relationship between Lead Levels in Bone and Breast Milk[1,2]

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Abstract

Whereas dietary fats are known to influence bone mineral density, little is known about their effect on the skeletal stores of lead that are a pervasive source of fetal and infant lead exposure from heightened mobilization during pregnancy and lactation. This cross-sectional study examined the potential influence of maternal dietary intake of saturated and unsaturated fats on the relationship of lead levels in bone and breast milk during lactation. Lead was measured in blood, breast milk, and bone (patella and tibia) at 1 mo postpartum in 310 women in Mexico City. Dietary nutrient intake was assessed using a validated FFQ. Multivariate linear regression analyses were used to study the influence of dietary saturated and unsaturated fats on the association between bone and breast milk lead. In multivariate models that included both the dietary intake of SFA and PUFA, an interquartile range increase in patella lead [~20 μg/g (0.097 μmol/g)] was associated with a 24% (95% CI = 5–43) higher increase in breast milk lead in women in the lowest tertile of PUFA intake compared with those in the highest tertile of PUFA intake. Monounsaturated fatty acids did not modify the relationship between lead levels in patella and breast milk. In conclusion, higher maternal dietary intake of PUFA may limit the transfer of lead from bone to breast milk. J. Nutr. 138: 73–79, 2008.

Introduction

Over the past 2 decades many countries have experienced a decline in blood lead levels, reflecting a reduction in exposure from the environment (1). However, for those individuals previously exposed to lead, remobilization of skeletal stores of lead continues to pose a potential health risk (2). This scenario is particularly relevant during lactation when 3–7% of bone mass is mobilized (3–5) and bone lead is transferred via breast milk to the infant (6–9).

It is well recognized that diet plays an important role in the maintenance of bone health in children and adults (for reviews, see 10–13). Among the numerous components of a normal diet, the effects of saturated and unsaturated fat intake on bone health have received considerable attention. Both animal experiments and epidemiological studies have reported an inverse association between SFA intake and bone mineral density (BMD)[11] (14–16). Although data on the relationship between dietary monounsaturated fatty acids (MUFA) and bone health are limited, Trichopoulou et al. (17) have reported a positive association between dietary MUFA and BMD in adult men and women in Greece. In contrast to MUFA, a much larger volume of literature is available on the effects of PUFA on skeletal health. Two randomized placebo-controlled clinical trials have shown that PUFA supplementation is beneficial in maintaining BMD and encourages bone formation in adults (18,19). PUFA levels in maternal and cord blood are also positively associated with bone mineral content in infants (20). Furthermore, data from a number of animal models and in vitro experiments have supported the role of PUFA in bone metabolism (21–23).

The influence of dietary fats on bone remodeling is particularly relevant during lactation, because the nutritional needs of the infant are met in part by mobilization of calcium and other

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[11] Abbreviations used: BMD, bone mineral density; DRI, dietary reference intake; MUFA, monounsaturated fatty acid.

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minerals from the maternal skeleton (4). Maternal breast milk also provides lipids, including PUFA, which serve an important role in early infant development (for reviews, see 24,25). While breast milk is clearly beneficial for infant health, it may also serve as a route for a number of environmental contaminants, including lead (26,27). It is, therefore, important to better understand factors that influence the transfer of lead and other toxins from maternal stores to breast milk.

Although it is known that dietary saturated and unsaturated fats are important determinants of BMD, their effect on the mobilization of lead from maternal skeletal stores remains largely unexplored. Based on current biological evidence, it is our hypothesis that dietary intake of saturated and unsaturated fats (MUFA and PUFA) will influence the transfer of maternal bone lead to breast milk. We have, therefore, examined the additive and interactive effects of maternal dietary intake of fatty acids and biomarkers of bone lead on levels of lead in breast milk in ~300 lactating women in Mexico City, Mexico.

Materials and Methods

Study subjects. A detailed description of data collection methods has been reported earlier (28,29). Briefly, pregnant women were recruited between January 1994 and June 1995 from 3 maternity hospitals in Mexico City (National Institute of Perinatology, Manuel Gea Gonzalez Hospital, and Mexican Social Security Institute). Potential study participants ($n = 2910$) were interviewed and, of the 1398 found to be eligible at the interview, 629 agreed to participate. The present study is limited to the 310 participants who provided breast milk samples at 1 mo postpartum and had adequate volume for analysis remaining after the pilot phase of the study that investigated methods for improved digestion procedures (7). Comparisons between the subgroup of women included in this study and the wider cohort have been reported earlier (7).

Data on demographic characteristics and known risk factors for environmental lead exposure were collected at the start of the study. All study participants received a detailed explanation of the study and counseling on reduction of lead exposure. Informed consent was obtained from the subjects and the research protocol was approved by the human subjects committees of the National Institute of Public Health of Mexico, Harvard School of Public Health, and the participating hospitals.

FFQ. Maternal dietary intake was assessed at 1 mo postpartum using a 116-item semiquantitative FFQ designed to estimate the usual dietary intake over the previous 12-mo period, including pregnancy (30). Specifically, this questionnaire was designed to classify individuals by the relative intake of 20 nutrients from 82 foods of the Mexican diet. The FFQ also included 9 beverages and collected specific information on the consumption of fats, sugar, and salt. Questions relevant during pregnancy such as use of vitamins and pregnancy-related health problems that may affect food consumption were also included. The validity and reproducibility of this questionnaire to assess dietary intake of a number of nutrients, including MUFA, PUFA, and SFA, has been assessed previously in this population (30). The average nutrient content of each food item was derived from the USDA food consumption tables and the database of the National Institute of Nutrition (31). To derive the relative intake of a specific nutrient, the amount of the nutrient in each food was multiplied by the subject's reported frequency of intake of that food item and the products were summed over the 82 foods. The nutrient values obtained from the FFQ and food tables were adjusted for energy intake using the nutrient residual model approach. The rationale behind energy adjustment of nutrient intake in epidemiological studies has been detailed by Willett et al. (32).

Bone lead. A spot-source ^{109}Cd K-X-ray fluorescence instrument was used to measure maternal bone lead. In vivo measurements of each subject's mid-tibial shaft (representing cortical bone) and patella (trabecular bone) were taken for 30 min each. A detailed description of this technique has been reported by Hu et al. (33). In brief, the instrument used a Cd γ-ray source to provoke the emission of fluorescent photons from target tissue that were then detected, counted, and arrayed on a spectrum. A net lead signal is determined in μg lead/g bone mineral. The instrument also provides an estimate of uncertainty associated with each measurement. For quality control, we excluded bone lead measurements with uncertainty measurements >10 μg/g[12] for tibia ($n = 4$) and 15 μg/g for patella ($n = 15$). High uncertainty measurements generally reflect excessive patient movement outside of the measurement field or excessive thickness of overlaying tissue and do not produce acceptable results. Bone lead measurements have previously been shown to be good markers of cumulative lead exposure as indicated by increases in patella and tibia lead with age and duration of residence in Mexico City (34).

Breast milk lead. Breast milk samples were collected at 1 mo postpartum from lactating women using techniques that minimize environmental contamination and determine lead concentrations in breast milk with a high percentage of recovery. Before manually expressing milk, the breast was washed with deionized water that was also collected and analyzed for lead contamination. Ten milliliters of milk was collected in preleached polypropylene tubes. Samples were frozen, shipped to the Channing Laboratory (Boston, MA), and stored at −30°C (Fisher IsoTempPlus) until analysis. The breast milk samples were digested with HNO_3 in a high-pressure asher and lead content was measured by isotope dilution-inductively coupled plasma MS (Sciex Elan 5000; Perkin-Elmer). The limit of detection for lead analysis in breast milk by high-pressure asher digestion and isotope dilution-inductively coupled plasma MS was 0.001 μg/L[13] of milk. Sample preparation was performed at the University Research Institute for Analytical Chemistry (Amherst, MA) and instrumental analysis was performed at the Trace Metals Laboratory of the Harvard School of Public Health (Boston, MA). A detailed description of the analytical procedures has been reported by Ettinger et al. (7).

Statistical analysis. Summary statistics and scatter plots of bone lead (patella and tibia), breast milk lead, maternal dietary intake of SFA, PUFA, MUFA, and other dietary variables, including carbohydrate, protein, total fat, and calcium, were examined to study bivariate relationships and identify outliers. We calculated Spearman correlation coefficients with 2-sided *P*-values and compared mean breast milk lead levels within key subject characteristics using ANOVA with Scheffé multiple comparison procedure.

Questionnaire items were used to assess breast-feeding practice at 1 mo postpartum. A response to the question "Have you fed your infant only breast milk (over the 1-mo period)"? was used to distinguish exclusive and partial breast-feeding practices. Other questions that were used to determine duration of breastfeeding included, "Are you currently breast-feeding your infant"? and "What was the age (days) of infant when breast-feeding was initiated?"

A series of multivariate linear regression models was estimated with breast milk lead (log *e* transformed) as the outcome variable and bone lead (patella or tibia) as the predictor variable. Separate models were estimated for patella and tibia lead. Each model incorporated covariates previously associated with breast milk lead levels in this population (7), including breast-feeding practice (partial vs. exclusive), previous pregnancies (0, 1, and ≥2), infant weight change (g), and maternal dietary intake of calcium (≥1000 mg/d vs. <1000 mg/d). In addition to these variables, total maternal fat intake (g/d) was also included, because dietary fat has been associated with increased lead absorption (35–38). To this core model, either maternal dietary SFA, PUFA, or MUFA intake was added as an independent covariate (Model 1). In Model 2, an interaction term for these dietary fat variables and bone lead (patella or tibia) was included. Tertiles of SFA, MUFA, and PUFA intake were used in the regression analyses. To compare the influence of each dietary fat variable on the relationship between lead levels in bone (patella or tibia) and breast milk, we estimated the change in breast milk lead (log *e* transformed) corresponding to an interquartile range increase in bone lead. Finally, we also stratified our multivariate regression analyses by breast-feeding practice to compare differences among mothers who

[12] SI unit conversion factor: bone lead (patella or tibia) (μmol/g) = 0.004826.
[13] SI unit conversion factor: breast milk lead (μmol/L) = 0.004826.

practiced exclusive and partial breast-feeding. We used Studentized residual plots and Cook's D statistic to identify outliers and influential points. All statistical analyses were performed using SAS release 9.1.3 (SAS Institute). Values in text are means ± SD. Statistical tests were considered significant at $P < 0.05$.

Results

Bone lead levels. The mean patella lead levels of the mothers in our study were generally higher than lead levels in tibia (**Table 1**). In agreement with previously reported results on this study population (34), both patella and tibia lead levels increased with duration of residence in Mexico City. Patella lead concentrations of mothers residing in Mexico City for >20 y (16.5 ± 15.5 μg/g) were higher than levels for mothers living there for 5–20 y (11.2 ± 13.5 μg/g) or for <5 y (10.5 ± 11.7 μg/g) ($P < 0.05$). Similarly, tibia lead levels were higher in women whose period of residence in Mexico City exceeded 20 y (11.0 ± 10.0 μg/g) compared with women who had lived there for 5–20 y (7.7 ± 10.6 μg/g) or <5 y (5.1 ± 8.0 μg/g) ($P < 0.05$).

Maternal dietary intake. Based on the FFQ data, it was estimated that 67% of participants had a daily energy intake below the recommended dietary reference intake (DRI) of 11,288 kJ/d for lactating women aged 24 y (39). Furthermore, all participants had carbohydrate intake above the DRI (210 g/d) and 18.1% of the mothers were deficient in dietary protein intake (DRI = 71 g/d). Approximately 50% of the participating mothers had calcium intake below the DRI of 1000 mg/d. With the exception of a positive relationship between total fat intake and patella lead, none of the dietary variables included in our analyses correlated with maternal lead biomarkers (**Table 2**).

TABLE 1 Characteristics, lead biomarkers, and dietary nutrient intakes of women at 1 mo postpartum[1]

	n	
Characteristics		
Age, *y*	310	24.4 ± 5.0 (14–42)
Duration of living in Mexico, *y*	308	20.1 ± 8.7 (0.5–40)
Schooling, *y*	304	9.3 ± 3.1 (1–18)
Married, *%*	310	70.0
Prior pregnancies, *n*	308	2.0 ± 1.2 (1–7)
Previous lactation >12 mo, *%*	307	21.2
Exclusive breast-feeding, *%*	308	32.1
Current use of lead-glazed ceramics, *%*	310	41.9
Past use of lead-glazed ceramics, *%*	310	76.1
Lead biomarkers		
Blood lead[2], *μg/dL*	310	9.3 ± 4.4 (1.8–29.9)
Breast milk lead[3], *μg/L*	310	1.4 ± 1.1 (0.2–8.0)
Patella lead[3,4], *μg/g bone mineral*	293	14.5 ± 14.9 (< 1–67.2)
Tibia lead[3,5], *μg/g bone mineral*	303	9.6 ± 10.1 (< 1–76.5)
Dietary nutrient intake		
Energy, *kJ/d*	304	10356 ± 2560 (5431–18,529)
Calcium, *mg/d*	304	1030.1 ± 245.4 (492.3–1723.8)
Protein, *g/d*	304	80.0 ± 9.6 (51.9–110.8)
Carbohydrate, *g/d*	304	346.5 ± 34.9 (228.9–439.4)
Total fat, *g/d*	304	85.4 ± 13.7 (54.3–137.9)
Saturated fat, *g/d*	304	28.0 ± 5.2 (14.2–47.8)
Monounsaturated fat, *g/d*	304	31.6 ± 5.6 (18.4–52.4)
Polyunsaturated fat, *g/d*	304	20.7 ± 6.1 (10.1–53.9)

[1] Values are means ± SD (range) or percent.
[2] SI unit conversion factor: blood lead (μmol/L) = 0.04826.
[3] SI unit conversion factor: breast milk lead (μmol/L) and bone lead (patella or tibia) (μmol/g) = 0.004826.
[4] Fifteen women were excluded due to uncertainty measurements >15 μg/g bone mineral (0.72 μmol/g).
[5] Four women were excluded due to uncertainty measurements >10 μg/g bone mineral (0.048 μmol/g).

Breast milk lead. Mean levels of breast milk lead were higher for women aged 20–30 y (1.44 ± 1.19 μg/L) than women aged <20 y (1.20 ± 0.83 μg/L) and also those older than 30 y (1.11 ± 0.65 μg/L). Breast milk lead levels also increased with duration of residence in Mexico City, with those residing there for >20 y having higher levels (1.43 ± 1.17 μg/L) than those living there for 5–20 y (1.29 ± 1.03 μg/L) and <5 y (0.99 ± 0.43 μg/L). However, these differences were not significant, and, similarly, no significant differences in breast milk lead were observed with the number of prior pregnancies, duration of previous lactation, or breast-feeding practice ($P \geq 0.19$).

In relation to dietary variables, mothers in the lowest tertiles of dietary PUFA intake had lower breast milk lead levels (1.14 ± 0.64 μg/L) than mothers in the 2nd tertile (1.45 ± 1.24 μg/L) and 3rd tertile (1.44 ± 1.22 μg/L). Lead levels in breast milk in the 2nd tertile of MUFA intake (1.51 ± 1.34 μg/L) were higher than those in the first tertile (1.32 ± 1.08 μg/L) and those in the 3rd tertile (1.21 ± 0.75 μg/L). Mothers in the 2nd and 3rd tertiles of SFA intake had breast milk lead levels (1.45 ± 1.25 μg/L and 1.36 ± 0.92 μg/L, respectively) that were higher than those in the lowest tertile (1.23 ± 1.06 μg/L). However, these differences in breast milk lead levels within categories of maternal dietary fatty acid intake were not significant. Breast milk lead was significantly correlated with mother's blood lead and patella lead at 1 mo postpartum but not with any of the dietary fat variables (Table 2).

Association of lead levels in bone and breast milk. Multivariate linear regression analyses with breast milk lead as the outcome variable showed that tibia lead was not a significant predictor of breast milk lead (data not shown). However, patella lead was a significant predictor of breast milk lead in all models (**Table 3**). In relation to the dietary variables, MUFA intake was not significantly associated with breast milk lead (Model 1, MUFA, Table 3) and the relationship between lead levels in patella and breast milk (Model 2, MUFA, Table 3) did not differ in different tertiles of MUFA intake, indicating no effect modification by maternal dietary MUFA intake.

TABLE 2 Correlation of lead biomarkers and dietary fat variables of women at 1 mo postpartum[1]

	Patella lead	Tibia lead	Blood lead	Total fat	MUFA	SFA	PUFA
Breast milk lead	0.13*	0.05	0.46***	0.05	−0.02	0.04	0.09
Patella lead		0.33***	0.29***	0.12*	0.08	0.04	0.10
Tibia lead			0.22***	0.08	0.08	0.06	0.09
Blood lead				0.04	0.04	0.02	0.07
Total fat					0.68***	0.53***	0.55***
MUFA						0.64***	0.18**
SFA							−0.03

[1] Values are Spearman correlation coefficients, $n = 282–284$. *$P < 0.05$; **$P < 0.01$; ***$P < 0.001$.

TABLE 3 Association of patella lead with lead concentrations in breast milk of women at 1 mo postpartum[1,2]

	MUFA		PUFA		SFA	
Variables	Model 1[3] ($n = 282$)	Model 2[4] ($n = 282$)	Model 1 ($n = 281$)	Model 2 ($n = 275$)	Model 1 ($n = 282$)	Model 2 ($n = 276$)
Intercept	−0.21 ± 0.29	−0.21 ± 0.30	−0.07 ± 0.26	−0.27 ± 0.26	−0.06 ± 0.27	0.01 ± 0.26
Patella lead[5], *μg/g*	0.01 ± 0.003**	0.01 ± 0.005	0.01 ± 0.003***	0.02 ± 0.005***	0.008 ± 0.003***	0.003 ± 0.005
Fat variable[6], *g/d*						
1st tertile	Reference	Reference	Reference	Reference	Reference	Reference
2nd tertile	0.11 ± 0.10	0.12 ± 0.14	0.15 ± 0.09	0.27 ± 0.13*	0.08 ± 0.09	−0.03 ± 0.12
3rd tertile	−0.02 ± 0.12	−0.02 ± 0.15	0.16 ± 0.10	0.56 ± 0.13***	0.14 ± 0.11	−0.10 ± 0.14
Patella lead × fat variable						
1st tertile	— —	Reference	— —	Reference	— —	Reference
2nd tertile		−0.0008 ± 0.007		−0.003 ± 0.007		0.008 ± 0.006
3rd tertile		0.0002 ± 0.006		−0.02 ± 0.006**		0.02 ± 0.007*
Total adjusted R^2	0.066	0.066	0.072	0.150	0.064	0.113

[1] Values are parameter estimates ± SE, $n = 283$; numbers in models differ due to exclusion of outliers identified using Cook's D statistic. Value is significantly different from parameter estimate for reference group: *$P < 0.05$; **$P < 0.01$; ***$P < 0.001$.
[2] Multivariate linear regression models used to estimate the effect modification by maternal dietary intake of MUFA, PUFA, and SFA of the relationship between lead levels in patella and breast milk. Models were adjusted for breastfeeding practice (partial vs. exclusive), previous pregnancies (none, 1 and ≥2), infant weight change (g), maternal dietary calcium intake (≥1000 mg/d vs. <1000 mg/d), and maternal total fat intake (g/d).
[3] Model 1 = multivariate linear regression model in which covariates previously associated with breast milk lead and the maternal dietary intake of MUFA, PUFA or SFA are included.
[4] Model 2 = Model 1 with added term for the interaction between patella lead and the respective fatty acid variable.
[5] SI unit conversion factor: patella lead (μmol/g) = 0.004826.
[6] Fat variable is either MUFA, PUFA, or SFA.

Similar analyses of maternal dietary PUFA showed that, although PUFA intake was not a significant predictor of breast milk lead (Model 1, PUFA, Table 3), the interaction between patella lead and dietary PUFA was significant for the highest tertile of PUFA intake (Model 2, PUFA, Table 3). This result indicated that the association between lead levels in patella and breast milk was weaker in subjects whose PUFA intake was in the highest tertile than those in the lowest category of dietary PUFA intake. Inclusion of the interaction term also substantially increased the predictive power of the model.

Analyses of influence of maternal dietary SFA intake on the relationship between lead levels in patella and breast milk showed that the interaction between SFA intake and patella lead was significant for the highest tertile of SFA intake (Model 2, SFA, Table 3). This indicated that, in contrast to the PUFA results, the association between lead levels in patella and breast milk was stronger in subjects whose SFA intake was in the highest tertile compared with those in the first tertile of dietary SFA intake. Addition of the interaction term to the regression analysis increased the predictive power of the model.

Because the parameter estimates for SFA and PUFA indicated effect modification in opposite directions, a model incorporating both the PUFA and SFA term and their respective interactions with patella lead was also estimated (**Table 4**). In this model, the interaction between PUFA and patella lead remained significant for the highest tertile of PUFA intake. An IQR increase in patella lead (~20 μg/g) was associated with 0.34 μg/L higher breast milk lead level in the lowest tertile of PUFA intake compared with the highest tertile of PUFA intake. This accounts for a 24% higher increase in breast milk lead in the mothers whose dietary intake of PUFA was in the lowest tertile (95% CI = 5–43). However, the interaction between SFA and patella lead was not significant (Table 4).When this analysis was stratified by breast-feeding practice, the parameter estimates for the interaction between PUFA and patella lead did not show any significant change from the nonstratified analysis. For both the partial breast-feeding ($n = 185$) and the exclusive breast-feeding ($n = 89$) groups, parameter estimates for the interaction between patella lead and the 3rd and 2nd tertile of PUFA intake were −0.02 ± 0.01 and −0.01 ± 0.01, respectively. In additional analyses, dietary intakes of SFA, MUFA, and PUFA did not

TABLE 4 Combined influence of maternal dietary intake of PUFA and SFA on the relationship between lead levels in patella and breast milk[1,2]

Variable	Parameter estimate ± SE
Intercept	−0.34 ± 0.17*
Patella lead[3], *μg/g*	0.02 ± 0.007 *
PUFA, *g/d*	
1st tertile	Reference
2nd tertile	0.26 ± 0.13*
3rd tertile	0.50 ± 0.13***
SFA, *g/d*	
1st tertile	Reference
2nd tertile	0.08 ± 0.13
3rd tertile	0.07 ± 0.14
Patella lead × PUFA	
1st tertile	Reference
2nd tertile	−0.005 ± 0.007
3rd tertile	−0.02 ± 0.007**
Patella lead × SFA	
1st tertile	Reference
2nd tertile	−0.0004 ± 0.007
3rd tertile	0.005 ± 0.007

[1] $n = 277$. Six outliers, identified using Cook's D statistic, were excluded from analysis. Value is significantly different from parameter estimate for reference group: *$P < 0.05$; **$P < 0.01$; ***$P < 0.001$.
[2] Model adjusted for breast-feeding practice (partial vs. exclusive), previous pregnancies (none, 1, and ≥2), infant weight change (g), maternal dietary calcium intake (≥1000 mg/d vs. <1000 mg/d), and maternal total fat intake (g/d). Adjusted $R^2 = 0.149$.
[3] SI unit conversion factor: patella lead (μmol/g) = 0.004826.

modify the relationship between lead levels in blood and breast milk (data not shown).

Discussion

In our cross-sectional study of ~300 women, maternal dietary intake of PUFA modified the relationship between lead levels in patella and breast milk at 1 mo postpartum. Higher dietary intake of PUFA corresponded to a weaker association between lead levels in patella and breast milk and this result remained consistent in models that examined PUFA and SFA concomitantly. SFA intake modified the relationship between lead levels in patella and breast milk only in the model that excluded maternal PUFA intake, suggesting that PUFA exert a stronger influence on the relationship between patella and breast milk lead.

It is important to consider the findings of this study in the context of published data on the effects of PUFA and SFA on bone health. Corwin et al. (15) studied the relationship of dietary SFA and BMD in ~14,000 adults enrolled in the NHANES III conducted in the United States from 1988 to 1994. Although total fat intake was not related to BMD, a consistent negative association between SFA intake and BMD in the femoral neck was observed in both men and women. In a double-blind randomized placebo-controlled trial, Kruger et al. (18) showed that PUFA supplementation in older women increased BMD at the femoral neck and maintained BMD at the spine. The placebo group experienced decreased BMD at both sites. Similarly, van Papendorp et al. (19) reported that supplementation with PUFA-rich fish oil and evening primrose oil for 16 wk increased calcium absorption and stimulated osteoblastic activity in 40 women diagnosed with osteoporosis. In contrast, Bassey et al. (40) observed no significant effect of PUFA supplementation on markers of bone turnover in 85 pre- and postmenopausal women. Although these and other studies have primarily focused on older adults, Weiler et al. (20) observed a positive association between maternal and cord blood PUFA levels and bone mineral content in infants. However, limited data are available on the effects of PUFA intake and BMD during lactation, when bone turnover is high.

The results of our study suggest that higher dietary intake of PUFA may limit the transfer of lead from bone to breast milk. Lead displaces calcium in the hydroxyapatite of calcified tissues and shares some of the metabolic pathways of calcium (41). It is, therefore, plausible that dietary constituents that affect bone mineral content may also modify the release of lead from maternal skeletal stores. We measured maternal dietary intake as well as lead levels in bone (patella and tibia), blood, and breast milk at 1 mo postpartum. Although these measurements were recorded at the same time point, they reflect different periods of exposure. Data on variations in breast milk lead concentrations over the course of lactation are limited and do not indicate any clear change in lead levels during the first month of lactation (42,43). In this study, we used breast milk measurements at 1 mo postpartum as an indicator of lead concentrations from the start of parturition to the time of sample collection. Bone is a long-term storage compartment for lead and, due to increased remodeling during lactation, is thought to release lead stores that have accumulated for years to decades. Skeletal lead stores are a major contributor of lead in breast milk (7,44) and, therefore, any factors, including maternal diet, that affect bone remodeling may influence lead concentrations in breast milk. By measuring maternal diet over the duration of pregnancy using a validated FFQ, we have been able to estimate the influence of maternal intake of saturated and unsaturated fats on the relationship between lead levels in the skeletal compartment and breast milk.

In our study, maternal fatty acid intake did not modify the association between lead concentrations in blood and breast milk. Using structural equation models, it has been proposed that the skeletal compartment contributes lead to plasma independently of erythrocyte lead (45). In the absence of direct plasma lead measurements for all our study participants, we are unable to assess the influence of maternal diet on the pathway by which lead released from bone may be transferred to breast milk. Maternal plasma lead during pregnancy has been shown to be a better predictor of infant mental development than maternal blood lead (46); its relationship with breast milk lead has, however, received little attention. Analysis of data from 86 women indicated that lead levels in breast milk are generally higher than in plasma (47).

Maternal dietary intake of MUFA did not influence the relationship between patella lead and breast milk lead in our analyses. Trichopoulou et al. (17) observed a positive association between BMD and dietary MUFA intake in adult men and women in Greece where MUFA were primarily derived from consumption of olive oil. In contrast, Macdonald et al. (48) observed an inverse association between BMD and dietary intake of PUFA and MUFA in women residing in northeastern Scotland. In the absence of BMD measurements in this study, specific reasons for the lack of interaction between dietary MUFA and patella lead cannot be given. It is, however, possible that marked differences between the diet consumed by subjects in this study and those of participants in studies by Trichopoulou et al. (17) and Macdonald et al. (48) may account for the observed results.

Dietary intake of fatty acids did not modify the relationship between lead levels in tibia and breast milk. These results are not surprising, because tibia lead, a biomarker of lead in cortical bone, has previously been shown to have a weaker association with breast milk lead in this population (7). Furthermore, trabecular bone undergoes considerably more remodeling during lactation than cortical bone (49). It is therefore plausible that dietary fatty acids may be more strongly associated with mobilization of lead stored in trabecular bone, as observed in this study. The interaction between PUFA and patella lead was significant in the highest tertile of PUFA intake (>21 g/d), suggesting that any potential benefits of PUFA in restricting lead transfer from the skeletal compartment to breast milk may only be apparent at high levels of intake.

The estimated nutrient intakes of participants in this study (Table 1) were generally higher than those reported in the 1999 Mexican National Nutrition Survey (50). In that study, the median daily intake of total calories was 5699 kJ, 49.8 g total fat, 15.5 g SFA, 12.9 g MUFA, and 8.8 g PUFA for women aged 12–49 y residing in Mexico City. These differences in dietary intakes between this study and the Mexican National Survey are possibly due to the fact that our study deals with diet during pregnancy and the postpartum period, whereas the majority of participants in the Mexican National Survey were not pregnant or breast-feeding. Differences in instruments between the 2 studies may also account for the disparate results; the 1999 survey used a single 24-h recall, whereas our study used a FFQ, which is more representative of usual intakes. However, the dietary fat intake in our study agreed well with the results of Parra et al. (51), who used a FFQ and erythrocyte phospholipid measurements to estimate dietary fat intake in pregnant Mex-

ican women. In that study, the main sources of (n-3) and (n-6) PUFA in pregnant Mexican women were canned tuna fish and fresh catfish, and eggs and cow's milk, respectively (51). We observed similar dietary sources for PUFA, and meat, cheese, lard, and oils were the primary sources for SFA in our study.

Our study is limited by its cross-sectional design and cannot evaluate the relationship between lead levels in bone and breast milk over the course of lactation, nor can it delve into the biological mechanisms behind the observed findings. Moreover, the absence of erythrocyte fatty acid levels prevents us from analyzing the effects of individual (n-3) and (n-6) fatty acids. The reported dietary intake of women enrolled in this study was below the DRI for a number of nutrients, limiting the generalizability of our results to populations with markedly different dietary patterns. This study is also limited by the use of a 12-mo recall period to estimate dietary intakes in pregnant women, because pregnancy is associated with changes in diet. In a study of ~1500 women living in Boston, consumption of foods between the first and 2nd trimester changed for ~50% (52). Increases of almost 4–6% in the intake of saturated fat and (n-3) and (n-6) fatty acids were also reported in that study. Similarly, changes in dietary intake were reported in a study following adult women from prepregnancy to 2 y postpartum (53). Our study is further restricted by the assumption that the FFQ-recorded maternal fatty acid intake at 1 mo postpartum reflects dietary intake of these nutrients during pregnancy and up to 1 mo postpartum.

In relation to recall of pregnancy diet, Bunin et al. (54) showed that recall as much as 3–7 y postpregnancy is of similar or slightly lower accuracy as recall of the general adult diet. In that study, when data were ranked, quintile agreement between original diet and recalled diet from self-administered FFQ was of the order of 70–80%. Pearson correlation coefficients of original and recalled diets for energy-adjusted intake of SFA, MUFA, and PUFA were ~0.50. These findings suggest that, although diet changes during pregnancy, the FFQ is a useful instrument to categorize subjects according to intake of specific food items or nutrients.

This study also has several strengths, including the use of in vivo measures of bone lead in a large number of participants. The semiquantitative FFQ used in this study has been validated and interclass correlations between diet recorded 1 y apart ranged from 0.38 for PUFA to 0.47 for SFA (30). Furthermore, in a population of pregnant Mexican women, FFQ measurements of the dietary intake of PUFA correlate reasonably well with erythrocyte cell membrane phospholipid levels (50). This study is, to our knowledge, the first epidemiological study to provide evidence that transfer of lead from maternal skeletal stores to breast milk may be modified by dietary intake of PUFA. Increasing dietary intakes of PUFA, while providing a number of other health benefits, may also reduce the exposure of the breast-fed infant to lead accumulated in maternal bones. Further work is required to determine the effects of (n-6) and (n-3) PUFA on skeletal lead stores and if these dietary components may alter the transfer of lead from mother to infant.

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