Determinants of Elevated Blood Lead during Pregnancy in a Population Surrounding a Lead Smelter in Kosovo, Yugoslavia

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We are prospectively examining the relation between environmental lead exposure and pregnancy outcome in cohorts of women exposed to a wide range of air lead concentrations. Titova Mitrovica, Yugoslavia, is the site of a large lead smelter, refinery, and battery factory. At midpregnancy, 602 women in T. Mitrovica and 900 women in Pristina, a non-lead-exposed control town, were interviewed. Blood was obtained for blood lead (PbB), hemoglobin, erythrocyte protoporphyrin, and serum ferritin measurements. Women were seen again at delivery, at which time maternal and umbilical cord blood samples were obtained. While many demographic and social characteristics were similar across the two towns, women in Pristina were more likely to report employment outside the home, cigarette smoking, and alcohol use during pregnancy. As expected, PbB levels were substantially higher in the smelter town. At midpregnancy, PbB geometric means were 17.1 μg/dL in T. Mitrovica and 8.1 μg/dL in Pristina; 86% of the pregnant women in T. Mitrovica, compared to 3.4% of those in Pristina, had PbB levels >10 μg/dL. Within T. Mitrovica, distance between the home and the smelter was the most important predictor of PbB at mid-pregnancy and delivery. Husband’s employment in the lead industry was associated with a significant increase in maternal PbB levels independent of place of residence. Higher maternal serum ferritin concentrations were associated with lower PbB levels, suggesting that dietary iron inhibits lead absorption. Overall, the placenta was a poor barrier to lead; the relationship between maternal PbB and umbilical cord PbB was linear across a wide range of PbB levels.

Introduction

The effects of environmental lead have only occasionally been examined in relation to human reproduction, most often in populations with extremely high exposure. Evidence of possible effects of lead on pregnancy first appeared in the literature near the turn of the century (1). Women employed in the lead industry were reported to have more miscarriages, stillbirths, premature infants, and neonatal deaths as compared to women in other occupations. Several more recent reviews have reported an excess of spontaneous abortions among lead-exposed women working in the printing industry in Italy and Germany (2–4). Nordstrom et al. (5), in Sweden, found a significantly increased frequency of spontaneous abortions and low birth weight infants (apparently intrauterine growth retardation [IUGR]) among female employees working in a lead smelter. Similar observations were made on populations living in close proximity to that smelter (6,7). Needleman and co-workers have described an increased rate of minor malformations among newborns with cord blood lead (PbB) levels of 8 μg/dL or more (8), the first evidence of such an effect in association with extremely low exposure. More recently, however, a prospective study of women living in Port Pirie, a smelter town in Australia, reported no association between PbB concentration and spontaneous abortion, low birth weight (for term...
births), IUGR, premature rupture of membranes, and congenital anomalies (9).

With the exception of the latter two reports, these studies have not adequately controlled for important factors known to be associated with reproductive outcomes, such as maternal weight, height, parity, smoking, and socioeconomic status (SES). With this in mind, we embarked upon a prospective study of lead exposure during pregnancy that will ultimately follow growth and development during early childhood.

We have examined the relation between environmental lead exposure and pregnancy outcome in cohorts of women exposed to relatively high and low air lead concentrations. The Autonomous Province of Kosovo, Yugoslavia, is a mountainous farm region currently in the process of industrialization due to its vast mineral and coal reserves. A large lead smelter, refinery, and battery factory are major elements of the economy of the city of Titova Mitrovia, which is approximately 40 km north of Pristina, the capital city of Kosovo (Fig. 1). Surveys conducted in 1978 and 1980 found the population of T. Mitrovia to have markedly elevated PbB levels due to industrial air lead emissions (10). In contrast, lead exposure in Pristina is minimal; mean PbB levels in children (10) are lower than those reported for U.S. children (11).

This paper has two objectives. First, we introduce the study design and methods used in the study of pregnancy outcomes. Second, we explore predictors of elevated PbB during pregnancy. Two subsequent papers will describe the actual pregnancy outcome findings, the preliminary results of which were presented at this conference.

**Methods**

The study recruited subjects from areas in and around the Yugoslavian cities of T. Mitrovia and Pristina. Near the center of each town, there is a single out-patient clinic that provides routine prenatal care. Attendance in the clinics is largely determined by distance from the home. During the period from May 1985 through December 1986, women who were in approximately 12 to 20 weeks gestation were invited to participate in the study. Because trained staff were occasionally unavailable, not all women who came to the clinic were asked to participate. The promise of a photograph of the baby at birth influenced the vast majority of women to participate. The final sample size consisted of 900 women in Pristina and 602 in T. Mitrovia; the unequal sample sizes reflect the differences in the sizes of the clinics, Pristina being the larger of the two.

Two predominant languages are spoken in the region: Serbo-Croatian and Albanian. Therefore, at intake, women were interviewed by a bilingual nurse on sociodemographic characteristics, medical, reproductive, and occupational histories of both parents, and cigarette smoking and alcohol consumption during pregnancy. Height, weight, and blood pressure were measured and a urine specimen obtained and examined for blood, bilirubin, ketones, sugar, protein, and pH.

Within approximately 12 hr of delivery, each woman was again interviewed. Maternal blood and umbilical cord blood were obtained for four measurements described below. The placenta was trimmed of membranes, weighed, and a piece frozen for subsequent biochemical analyses. A trained neonatologist examined every baby, using a structured examination for gestational age (12) and malformations. For those women who did not deliver in a hospital, a visiting nurse was sent to the home to determine the outcome of the pregnancy.

Blood samples were obtained at intake and at delivery for the measurement of PbB (13), erythrocyte protoporphyrin (EP) (14), serum ferritin (15), and hemoglobin concentrations. Blood specimens were refrigerated on site and transported on wet ice to Columbia University, where all blood analyses were conducted. The Columbia University lab participates in the CDC’s PbB and EP quality control programs and is certified by the Occupational Safety and Health Administration (OSHA). Over the course of the study, agreements with CDC values of PbB and EP, measured by intraclass correlation coefficients, were 0.95 and 0.99, respectively.

Address location in relation to the smelter served as an indicator for exposure to airborne lead. Zones were defined by concentric circles of 2, 4, 6, and 8 km in radius drawn around the lead smelter-refinery complex; these were designated as zones 1 through 4, respectively. The city of T. Mitrovia fell within zone 1. Zone 5 consisted of all territory

![Figure 1](image-url)
from 8 km of T. Mitrovica to Pristina; zone 6 was Pristina itself and all areas to the south. Of the 1502 subjects in the study, we were unable to assign a zone to 24 women.

**Statistical Analysis**

The relationship between PbB (at both midpregnancy and delivery) and social, demographic, and biologic variables was examined using least square multiple regression analysis. Independent variables of interest were address zone of residence, husband’s place of employment, and maternal iron stores (i.e., serum ferritin concentrations); reference categories for these variables were the Pristina zone (zone 6), employment outside the lead industry, and serum ferritin > 10 ng/mL, respectively. Potentially confounding variables were identified *a priori* and assessed by evaluating changes in the regression coefficients of the independent exposure variables. These include cigarette smoking, maternal age, ethnic group, and maternal education.

**Results**

Table 1 compares demographic and social characteristics of women in the two towns. At entry to the study, the gestational age of women in Pristina was 131.4 days, as compared to 118.3 days in T. Mitrovica. Women in T. Mitrovica were, on average, 6 months younger and more likely to be of neither Serbian nor Albanian ethnicity. Their spouses were of similar age and ethnicity to spouses of women in Pristina. Mitrovica women were slightly shorter, a result consistent with the fewer number of Albanians among them. Alcohol use (defined as consumption of any alcohol during pregnancy) was less common in T. Mitrovica (5.1%) than in Pristina (9.3%). In each town, on average, the women had had more than two previous pregnancies. More women in Pristina reported having a job outside the home, although of those with jobs, comparable proportions, about 75%, were still working at the time of the interview. Mean monthly household income was therefore slightly lower in T. Mitrovica. The women were similar, however, with respect to other measures of socioeconomic status, not all of which are shown.

The mean monthly PbB levels at intake in T. Mitrovica fluctuated substantially from month to month, presumably a consequence of the highly variable industrial lead emissions, and were higher than in Pristina, where they varied little (Fig. 2). The frequency distribution of PbB levels revealed that 86.0% of the pregnant women in T. Mitrovica, but only 34.3% in Pristina, had PbB levels > 10 μg/dL (Fig. 3). The geometric means for PbB were 17.1 μg/dL in T. Mitrovica and 5.1 μg/dL in Pristina (Table 2).

Elevated EP concentrations are considered to be indicative of lead toxicity and/or iron deficiency. The mean serum ferritin concentration, a measure of the adequacy of tissue iron stores, was slightly higher in T. Mitrovica than in Pristina. Despite the higher iron stores, the mean EP was elevated in T. Mitrovica (Table 2), clearly reflecting some degree of metabolic inhibition of red cell heme synthesis due to lead.

Within T. Mitrovica, zone of residence was the most important predictor of elevated maternal PbB, both at midpregnancy (Table 3) and delivery (Table 4); PbB levels declined as the distance from the smelter to the home increased. Husband’s employment in the lead smelter, refinery, or battery factory was associated with an increase in adjusted mean maternal PbB, both at midpregnancy (Table 3) and at term (Table 4), independent of address zone. This finding is illustrated graphically in Figure 4. Cigarette smoking

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

| Characteristic                   | T. Mitrovica | Pristina |
|----------------------------------|--------------|----------|
| Number of women in study         | 602          | 900      |
| Gestation at interview, days     | 118.3 (26.0) | 131.4 (29.6) |
| Maternal age, years              | 26.4 (5.3)   | 26.9 (5.0) |
| Paternal age, years              | 30.5 (6.1)   | 30.5 (5.7) |
| Maternal ethnicity               |              |          |
| % Albanian                       | 56.2         | 61.2     |
| % Serbian                        | 25.3         | 24.8     |
| % Other                          | 18.5         | 14.0     |
| Paternal ethnicity               |              |          |
| % Albanian                       | 59.4         | 61.8     |
| % Serbian                        | 25.0         | 22.9     |
| % Other                          | 15.5         | 15.3     |
| Maternal weight, kg              | 63.5 (10.9)  | 63.9 (10.2) |
| Maternal height, cm              | 159.3 (7.4)  | 161.6 (5.9) |
| % Cigarette smokers              | 24.8         | 28.3     |
| % Alcohol users                  | 5.1          | 9.3      |
| Number of previous pregnancies   | 2.1 (2.2)    | 2.3 (2.4) |
| % With a job                     | 32.5         | 45.6     |
| % Working during pregnancy *     | 76.4         | 75.8     |
| Maternal education, years        | 8.9 (4.2)    | 9.0 (4.1) |
| Paternal education, years        | 11.7 (2.8)   | 11.4 (3.1) |
| Monthly income in home (U.S. dollars) | 237.7 (161.8) | 259.9 (176.1) |

* Data are means with standard deviation in parentheses.  
* Of those with a job.  
* In order to adjust for inflation, the mean monthly dinar income was converted to U.S. dollars using the rate effective during the month of the interview.
Table 2. Hematologic findings at midpregnancy.*

| Measurement                      | T. Mitrovica | Pristina |
|---------------------------------|--------------|----------|
| PbB, µg/dL                      | 17.1 (6.9, 42.6) | 5.1 (2.5, 10.6) |
| EP, µg/dL                       | 36.0 (9.9, 130.3) | 25.4 (10.4, 61.7) |
| Serum ferritin, ng/mL           | 14.1 (2.0, 98.0) | 11.5 (1.5, 88.3) |
| Hemoglobin, g/dL                | 12.4 (10.3, 14.5) | 12.3 (10.0, 14.7) |

*Data for PbB, EP, and serum ferritin are geometric means with 95% confidence intervals in parentheses. Geometric means were calculated as 1/n [log_{10} (x_1) + log_{10} (x_2) + ... + log_{10} (x_n)]. The means for hemoglobin concentrations are arithmetic.

Table 3. Regression model for the prediction of midpregnancy blood lead concentration.*

| Variable                      | Estimate | SE  | 95% CI | p-Value |
|-------------------------------|----------|-----|--------|---------|
| Address zone                  |          |     |        |         |
| <2 km vs. Pristina            | 0.5555   | 0.0115 | 0.5780, 0.5330 | <0.0001 |
| 2-4 km vs. Pristina           | 0.4506   | 0.0311 | 0.5116, 0.3896 | <0.0001 |
| 4-6 km vs. Pristina           | 0.3913   | 0.0240 | 0.4383, 0.3443 | <0.0001 |
| 6-8 km vs. Pristina           | 0.4085   | 0.0319 | 0.4710, 0.3460 | <0.0001 |
| ≥8 km vs. Pristina            | 0.1072   | 0.0166 | 0.1397, 0.0747 | <0.0001 |
| Husband employed in the lead industry | 0.0491   | 0.0160 | 0.0805, 0.0177 | 0.0022 |
| Ethnic group                  | 0.0335   | 0.0111 | 0.0553, 0.0117 | 0.0025 |
| Serbian vs. Albanian          | 0.0433   | 0.0132 | 0.0992, 0.0174 | 0.0010 |
| Serum ferritin                | 0.0255   | 0.0123 | 0.0496, 0.0004 | 0.0393 |
| <5.0 vs. 5.0-9.9              | 0.0078   | 0.0115 | 0.0303, -0.0147 | 0.4961 |
| Inteceptor                    | 0.6707   |       |        |         |
| Model n = 1452                |          |     |        |         |
| Model R² = 69.1%              |          |     |        |         |

*Log (base 10) of blood lead (µg/dL) adjusted to hemoglobin of 12.0 (g/dL).

*Confidence interval.

was not a particularly important predictor of PbB, either in the entire study population or in T. Mitrovica alone.

Maternal education was associated with the PbB at delivery (Table 4), but not PbB at midpregnancy (Table 3). Ethnic group was an important predictor of both the midpregnancy and delivery PbB levels; Albanian women had the lowest PbB levels. We suspect that the Albanian custom of removing shoes at the entrance to the home reduces the tracking of soil and dust lead into home. The non-Albanian and non-Serbian women, a group consisting primarily of Gypsies, exhibited higher PbB levels than Albanians or Serbians after holding constant other variables.

The role of nutritional factors on gastrointestinal lead absorption has been addressed in earlier presentations at this symposium by Mahaffey (16) and Ziegler (17). We have had the opportunity to examine two nutritional factors in these analyses: iron, as assessed by serum ferritin concentrations, and milk consumption during pregnancy. At midpregnancy, women who had serum ferritin concentrations of < 5 ng/mL had significantly higher PbB levels than those with ferritins of 10 ng/mL or more (Table 3). [WHO considers 10 ng/mL...
to be the lower limit of normal in nonpregnant women (18). By delivery a strong dose-response effect was observed (Table 4); the higher the serum ferritin, the lower the PbB.

When we examined the influence of milk consumption during pregnancy on maternal PbB, the data initially suggested that PbB was positively associated with milk consumption (data not shown). This association persisted in multiple regression analyses. We hypothesized that the effect of milk would be strongest in those address zones most heavily exposed to lead. However, contrary to expectations, the relationship between PbB and milk consumption was not consistent in all address zones. Further analysis showed that this apparent relationship was due to extremely elevated PbB levels in several women who were milk drinkers; their elevated PbB levels proved to be due to other factors.

Finally, we examined the relationship between maternal PbB at delivery and the umbilical cord PbB in 888 mother-infant pairs who were between 28 and 44 weeks of gestation. The strong relationship between maternal PbB and umbilical cord PbB (Fig. 5) (slope = 0.928, r = 0.920) was linear across a wide range of PbB values.

Discussion

Because lead readily distributes from blood into bone, PbB is considered to be marker of recent exposure to lead, rather than total body lead burden. It is therefore not surprising that the PbB levels of pregnant women in T. Mitrovica were higher among those residing nearest the smelter, where air lead concentrations are highest. A 1980 survey of PbB in residents of zone 1 reported a mean (arithmetic) PbB of 32.4 μg/dL in nonpregnant women, reflecting the extremely high mean monthly air lead concentrations, which ranged from 21.3 to 29.2 μg/m³ (10); the allowable concentration of air lead is 0.7 μg/m³ in Yugoslavia. Immediately prior to the current study, as a result of the construction of a new smokestack and air filtration system at the smelter, air lead concentrations fell into the range of 0.9 to 12.8 μg/m³ (data not shown). Although this represents some improvement in air quality, and PbB levels have declined somewhat, residents of T. Mitrovica are obviously exposed to unacceptably high air lead levels.

The transfer of lead dust on the clothing, skin, and hair of lead workers from workplace to the home has been documented as a major source of hazard to children of lead workers (19–21). We have observed a risk of undue lead absorption in pregnant wives of lead workers. Thus, since lead freely crosses the placenta, paternal employment in the lead industry poses a risk of lead exposure to the unborn child. We therefore urge the lead industry to institute highly effective occupational hygiene measures capable of preventing the transport of lead from the workplace into the home. Among our subjects in T. Mitrovica, wives of battery workers had higher PbB levels than those of smelter or refinery workers.

In his presentation at this symposium, Ziegler correctly pointed out that nutritional supplementation is not a solution to the problem of undue lead absorption (17). Our finding concerning the apparent protective effect of adequate iron stores against lead absorption, however, should not be overlooked. Clearly, certain subsets of the population, such as pregnant women and children of low SES, require iron supplementation simply to prevent the syndrome of iron deficiency. Those same subsets, however, are most at risk for the consequences of undue lead absorption. Since iron deficiency increases the risk for elevated lead absorption, the health care system could reduce the prevalence of elevated PbB by redoubling its efforts to prevent iron deficiency in populations at risk. Bagnhurst and co-workers, in a study of pregnant women living near the Port Pirie lead smelter, have previously reported that women who received iron supplementation during pregnancy had lower PbB levels than those not supplemented (22).

Observations made more than a century ago suggested that milk consumption prevented the occurrence of plumblism in workers in the white-lead industry (23). Since that time, animal studies have revealed that some components of milk, such as lactose (24–27) and fat (24,28), increase gastrointestinal lead absorption, while other components such as calcium (29) decrease absorption; the Port Pirie study has reported lower means PbB among women who consumed less than 500 mg dietary calcium/day in comparison to those who consumed larger amounts (22). To our knowledge, the present study represents the first attempt to examine the influence of milk consumption on PbB in humans. The failure of this study to document an association between milk consumption and PbB suggests that the effects of the biochemical constituents of milk on lead absorption may counteract each other.

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