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Concentration of Mercury in Hair of Indigenous Mothers and Infants from the Amazon Basin

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Abstract. Hair mercury concentration, as an indicator of mercury body load, was studied in 251 samples of indigenous women and children living in selected areas of the Amazonian region. The mothers or women of child-bearing age, either non-Indians or Indians, and their children were sampled along the Madeira River and in the Kayapó reservation (Fresco River), respectively. Among the sampled individuals there were mothers with infants less than 2 years old. Total mercury in hair was determined by cold vapor atomic absorption spectrometry after alkaline digestion. The distribution of hair mercury concentration greater than 10 µg/g occurred in 67.4% of non-Indian women and 25% of Indian women; overall only 1% of non-Indian women had concentrations of hair mercury above 50 µg/g. In women of child-bearing age, the median and range of hair mercury concentration was 14.08 µg/g, and 0.8–94.7 µg/g for non-Indians, and 8.30 µg/g, and 0.8–13.3 µg/g for Indians. The correlation between maternal hair mercury and mercury in hair of infants (less than 2 years of age) still breast-feeding, was statistically significant only for non-Indians ($r = 0.555$ $p < 0.001$). The correlation between length of breast-feeding and mercury concentration in infant's hair was significant for Indian children ($r = 0.512$; $p = 0.029$) but not for non-Indian children ($r = 0.025$; $p = 0.832$). A subsampling of 30 mothers had segmented hair analysis that showed a mean decrease of 20% in body burden during pregnancy, thus indicating the extent of placental transference of mercury to fetuses.

Gold mining in the Amazon basin is widespread. The method employed in gold mining operations, *garimpos*, is crude and environmentally destructive. Because miners (*garimpeiros*) still use the primitive technique of mercury amalgamation to extract the alluvial gold, enormous quantities of mercury are disposed haphazardly in the environment. It is conservatively estimated that 1.3 kg of metallic mercury is used for the extraction of 1 kg of gold. As a result, it has been calculated that 1,500 to 3,000 tons of mercury was released in the Amazon ecosystem between 1977 and 1992 (Pfeiffer *et al.* 1993).

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All mercury used in gold mining finds its way into the Amazon ecosystem either as direct manipulation losses in the river (20%) or as a result of burning amalgam (80%), and eventually reaches the native population through the food chain. Mercury enters the food chain directly as metallic precipitation in crops or after methylation in animals and plants that serve as food for the indigenous people. The metal is bioconcentrated in the trophic fish chain, mainly as methylmercury, which constitutes the main protein source of the indigenous people either Indian or non-Indian (Barbosa *et al.* 1995).

Once absorbed, methylmercury accesses the hair follicle through the blood that bathes the hair root. The assumption that concentration of mercury in hair is proportional to the amount of mercury in blood, and should therefore reflect the body load of the metal, has been extensively studied and demonstrated (Cernichiari *et al.* 1995). Therefore, it is not only possible to estimate the body burden of mercury, but also, knowing the rate of hair growth, it is feasible to recapitulate the body burden in past months (Cernichiari *et al.* 1995).

The toxic effect of mercury poisoning is well known, especially after the environmental disaster of Minamata and several accidental poisonings due to mercury pesticides used in agriculture (Koos and Longo 1976). Preoccupation with the environmental damage caused by gold mining in the Amazon Basin has prompted us to join the effort in studying the extent of mercury contamination and health effects on the indigenous population. In this study we focus on the extent of the mercury load in vulnerable groups such as women of child-bearing age and infants.

Materials and Methods

As part of a major project to assess environmental mercury pollution due to gold mining, we studied the hair mercury concentration in women and children at two sites close to an intensive gold mining area of the Amazon Basin (Figure 1). The chosen areas were selected on the basis of length of operation of important gold mining sites. The mining operations in the Madeira River date back to the 1980s and is therefore older than the *garimpo* of Maria Bonita in the Fresco River (an important tributary of the Xingu River in the northern part of the state of Para. This last mining site started recently and is located 1400 km from the sampling site of the Madeira River in the state of Rondonia.

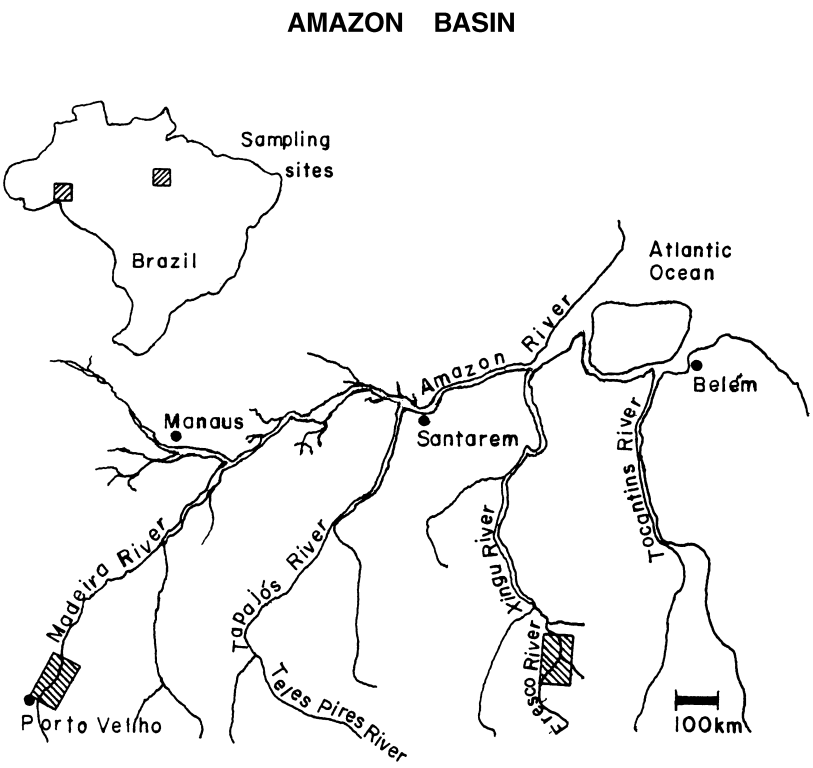

Fig. 1. Map of Amazon Basin showing sampling sites

Sampling and Hair Collection

The subjects sampled were 126 women of child bearing age (19–45 years old) and 125 children. These individuals were either Kayapo Indians (28 women and 54 children) of the Kikretum tribe living on the Fresco River, or non-Indian people (98 women, 71 children), also called *ribeirinhos* or *caboclos*, that lived along the Madeira River.

The hair was sampled from the occipital area, cut close to the scalp with stainless steel scissors, bundled together with cotton thread, placed in an envelope properly identified, and taken to the Laboratory of Environmental Chemistry of the University of Brasilia for analysis. Details of sample collection were given in a previous publication (Barbosa *et al.*1995).

Table 1. Mercury concentration (μg/g) in hair of indigenous women and children

	Indians		Non-Indians	
	Women	Children	Women	Children
N	28	54	98	71
Mean	8.11	7.30	14.08	10.82
Median	8.30	6.55	12.80	7.80
SD	3.16	3.50	10.67	8.46
Range	0.8–13.70	2.0–20.40	2.6–94.70	0.8–44.4

No statistically significant difference was observed between means within racial groups

Sample Preparation

All glassware was washed clean, rinsed consecutively with KOH, double-distilled water, left to rest in 50% HNO_3 for 24 h, again rinsed with double-distilled water, and then dried at 100°C for 12 h.

Mercury Determination

The hair samples were comminuted with stainless steel scissors, weighed, and digested before analysis. The method of Magos and Clarkson (1972) for mercury analysis of hair was employed as described previously (Barbosa *et al.* 1995). To a known weight of approximately 10 mg of hair, 1.0 ml 8.3 mM cysteine and 2.0 ml of 11.3 M NaOH were added and heated at 90°C for 15 min. Care was taken to avoid boiling. The digests were cooled in an ice bath and let stand to reach room temperature and then diluted with 7.0 ml of 171 mM NaCl. The weight of the digests were registered.

The determination of total mercury in hair was performed by cold vapor atomic absorption spectrometry (CV-AAS). The instrument used was a Mercury Monitor LDC Analytical, Model 1255 (LDC, Riviera Beach, FL) connected to a multimeter HP Model 3435 (Hewlet Packard, Palo Alto, CA) and a Neptune Dyna Pump (Magnetek Universal Electric, Owosso, MI). A 1-ml aliquot of digest was transferred to the reaction vessel and 1.0 ml of 8.3 mM cysteine, 20 ml of 171 mM NaCl, 10 ml of 8.0 M H_2SO_4, 1.0 ml of $SnCl_2 \cdot 2H_2O$/$CdCl_2$, and 20 ml of 13.3 M NaOH were added. The reaction produced a vapor that passed through a water solution of tri-n-butyl-phosphate in ice next to the absorption chamber where the absorption reading took place.

For recapitulation of hair mercury concentration during pregnancy, we analyzed segmented hair cut at intervals of 3.3 cm. Hair strands were aligned on a double-faced tape fixed on a graduated ruler where measurements of 3.3 cm were made corresponding to the previous 3 months (1.1 cm/month) of pregnancy as recommended by Cernichiari *et al.* (1995).

Precision and accuracy of mercury determinations was assured by the use of internal standards of hair, provided by the Hair Mercury-Interlaboratory Comparison Program, Ottawa, Ontario, Canada. As part of this intercalibration program, which started in 1992, our results so far have been considered acceptable. Out of 27 samples analyzed 25

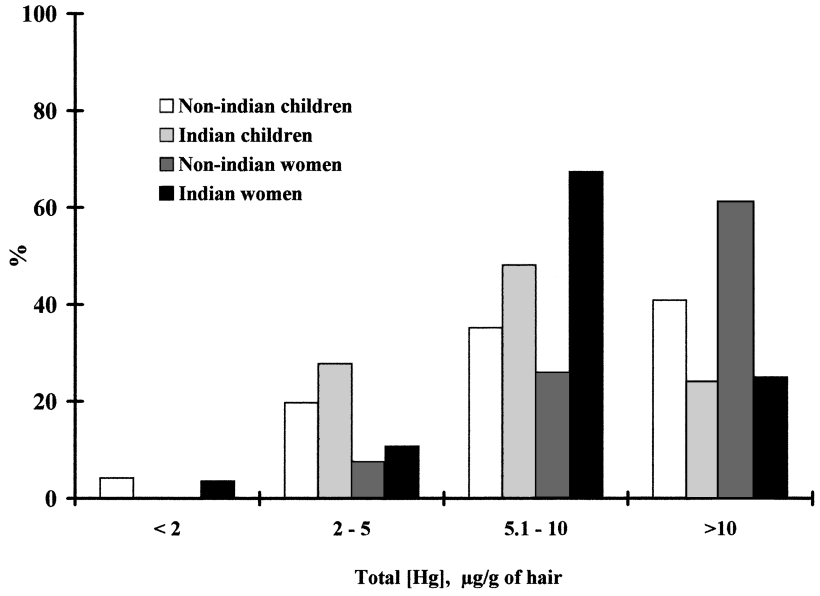

Fig. 2. Frequency distribution of total hair mercury concentrations

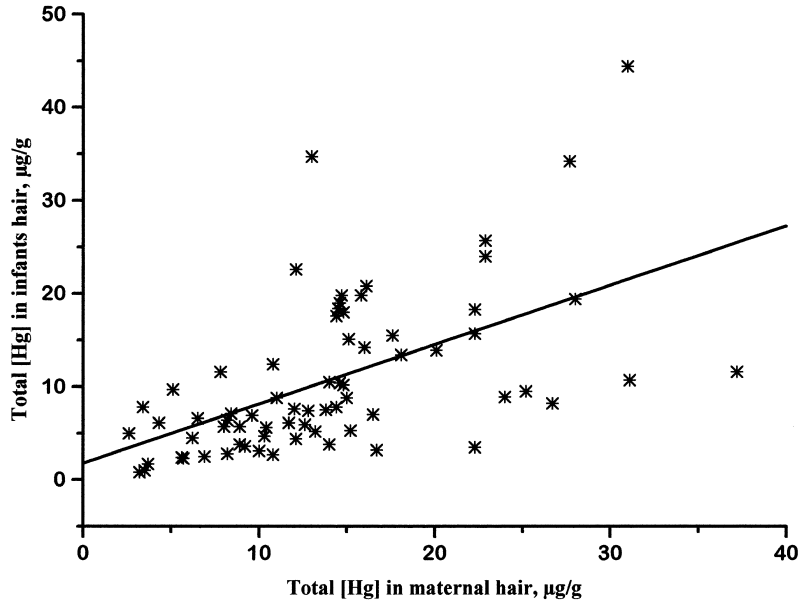

Fig. 3. Correlation of total mercury concentration in hair of infants and respective mothers among the Indian population ($r = 0.228$; $p = 0.412$)

(93%) gave results within 2 SD, and 18 (67%) within 1 SD of the corrected mean.

Summarized data (mean, SD, and ranges), Pearson correlation between variables, and analysis of variance, were performed using a SAS (SAS Institute, Cary, NC) computer program for PC. A *p* value of less than 0.05 was considered statistically significant.

Results

Concentrations of mercury in hair are summarized in Table 1 for the women and children, both Indians and non-Indians. No statistically significant difference was found within the groups of Indians and non-Indians. The distribution of hair mercury concentration (Figure 2) shows that most of the samples for the Indian women and children were below 10 µg mercury/g hair, while 60% of the non-Indian women and children were above it.

The relationship of the body burden of mercury between mothers and their breast-fed infants are shown in Figures 3 and 4. A statistically significant correlation was seen only for non-Indian ($r = 0.556$; $p < 0.001$) mother–infant pairs. The hair mercury of infants still breast-feeding (<2 years of age) was further studied as a function of age (length of breast-feeding) and the results are shown in Figures 5 and 6 for Indian and non-Indian children, respectively. In the Indian infants a significant correlation ($r = 0.512$; $p = 0.029$) between age and total mercury in their hair was observed but not in the non-Indian children ($r = -0.029$; $p = 0.813$).

With breast-feeding women it was possible to assess and differentiate the role of pregnancy and lactation in the body load

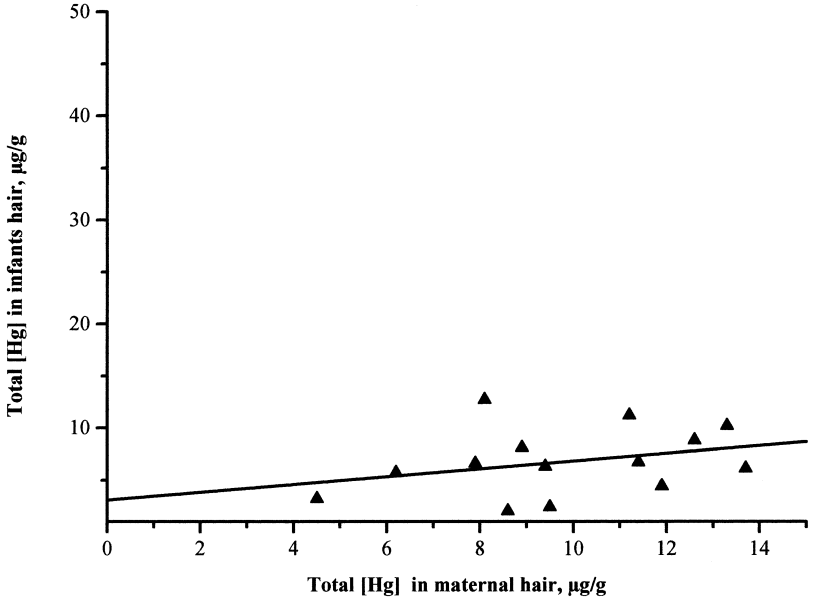

Fig. 4. Correlation of total mercury concentration in hair of infants and respective mothers among the non-Indian population ($r = 0.553$; $p = 0.0001$)

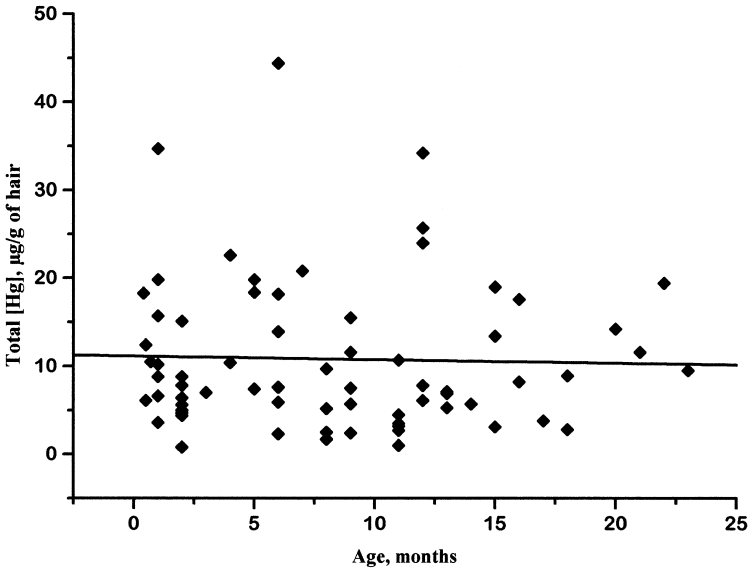

Fig. 5. Correlation of mercury concentration in hair of infants as a function of length of breast feeding (age of infants) among the non-Indian population ($r = -0.025$; $p = 0.832$)

of mercury and its transfer to the fetus (Figure 7). Using 1.1 cm/month as a guide, segments of hair that represent the gestation period showed a mean decrease in mercury concentration (20%). The level of mercury in hair attains pregestation levels in the first trimester immediately postpartum.

Discussion

Mercury gains access to hair mainly as methylmercury after the ingestion of contaminated food. For the native human population of the Amazon basin the contamination with methylmercury is through the ingestion of food as plants, animals, and fish grown in the polluted ecosystem. Fish is by far the most important dietary item consumed by these people and also the most important food bioconcentrator of mercury. However, it has been estimated that non-Indians consume relatively more fish than the Indians who prefer hunting (Barbosa *et al.* 1995). The per capita intake of fish for the non-Indian population has been estimated in 200 g/day (Boischio *et al.* 1995). The mercury concentration in the fish caught in the Amazon Basin varies depending on the trophic position of the fish species. Concentrations of mercury > 0.5 mg mercury/kg wet weight were reported in 45% of piscivorous fish samples from Madeira River (Barbosa *et al.* 1995).

Evidence from our own studies indicates that food consumption of contaminated staple items is more important than occupational exposure to gold mining activities. In a previous publication, Barbosa *et al.* (1995) reported that direct involvement with gold mining showed lower levels of hair mercury concentration compared to the native population of Indians and non-Indians. Leino and Lodenius (1995) also found an elevation of hair mercury associated with consumption of contaminated fish.

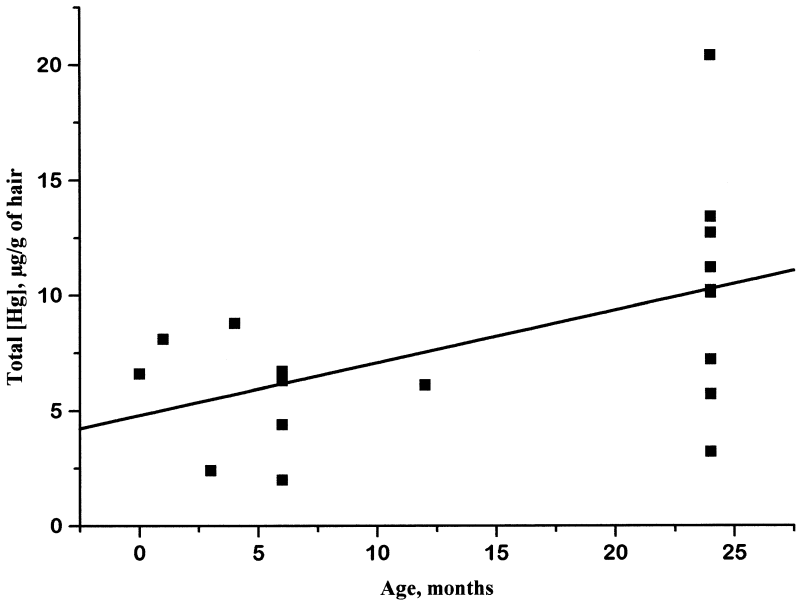

Fig. 6. Correlation of mercury concentration in hair of infants as a function of length of breast feeding (age of infants) among the Indian population ($r = 0.512$; $p = 0.029$)

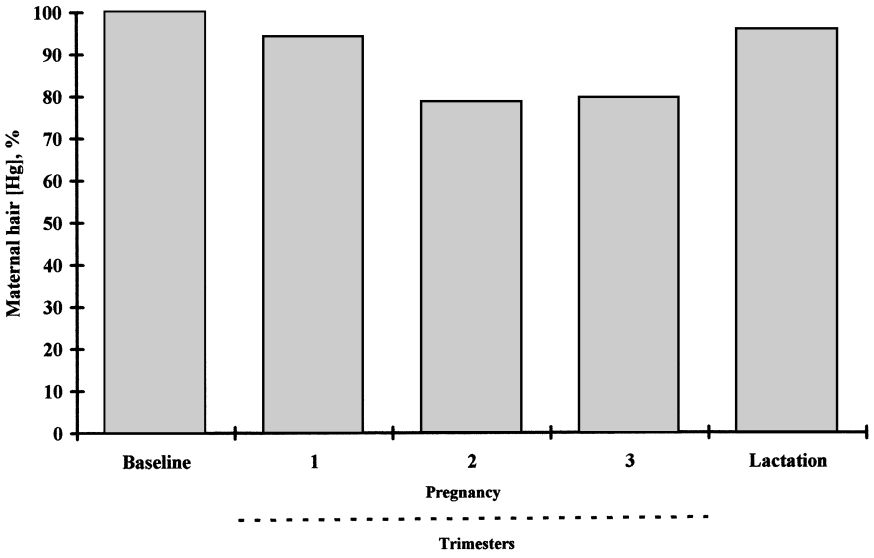

Fig. 7. Relative changes in maternal hair mercury concentration by trimester. Baseline and lactation refers respectively to the trimesters immediately before and after pregnancy

It is important to mention that these indigenous people are not directly involved with gold mining activities. They make a living through fishing and agriculture. Gold rushes usually attract foreign individuals, whose food habits are not necessarily the same as the riverine populations, and who may consume less fish. In addition, gold mining is an almost exclusively adult male activity not suitable for women or children.

Prenatal mercury exposure of infants causes adverse neurological effects. The human fetus was found to be five to 10 times more sensitive than adults to mercury poisoning (Clarkson 1989, 1992). Clinical signs in uterine-exposed infants include delayed development and mild neurological disturbances. In situations of acute poisoning due to mercurial pesticides such as in Iraq (Shahristani *et al.* 1976), analysis of hair segments in adults with mild symptoms of mercury intoxication showed mercury hair concentration between 120–600 µg mercury/g corresponding to an average body burden of 0.8–4.4 mg mercury/kg, of body weight (ratio of 137:1). As indicated by Clarkson (1992), the threshold of maternal hair mercury concentration indicative of adverse effects to the fetus is in the order of 10–20 µg mercury/g hair. In the present study only a few samples (1%) reached levels above 50 µg mercury/g; in the great majority of samples (67% of the women in the Madeira River and 25% of Indian samples) hair mercury concentrations were between 10–20 µg mercury/g. In the heavily polluted Minamata Bay, where the villagers consume mercury contaminated fish (11.4–39.0 µg mercury/g), the levels of mercury in hair was 191–705 µg mercury/g hair (Koos and Longo 1976; Harada 1982). However, in the less contaminated environments of Japan (Fujita and Takabatake 1977), Poland (Sikorski *et al.* 1986), North Sea (Grandjean *et al.* 1992), and coastal Chile (Bruhn *et al.* 1995), the reported median levels of mercury for women were below 4 µg mercury/g hair. In Canada, 30% of Cree women tested (15–45 years old) showed hair mercury concentrations over 10 µg mercury/g (Wheatley and Paradis 1996) and women of James Bay (Girard and Dumont 1995)

showed a range of hair mercury comparable with our results (5–20 µg mercury/g hair). Results from other Amazonian sites have shown even higher mean values for women of childbearing age. A recent study in the Tucuruí River (Leino and Lodenius 1995) showed median hair mercury concentration of 65 µg/g. In the same study, Indians showed hair mercury concentrations comparable to our results. A small group of women from the Teles Pires and Juruena Rivers showed mean concentrations of 41.2 µg mercury/g hair (Barbosa *et al.* 1997).

Considering that body half-life of mercury is estimated to be between 60 and 70 days (Shahristani *et al.* 1976) and that fish are widely consumed, these women are carrying body loads of mercury approaching the threshold for fetal intoxication. No overt signs of mercury intoxication, such as paresthesia, was reported in this population, neither were they expected to occur at the present level of exposure. A genotoxicity survey of the same population did not show gene frequency alteration greater than observed for the general population (Ferrari *et al.* 1992). However neurological examination conducted by Lebel *et al.* (1996) in men and women living in other Amazonian regions showed evidence of neurological disturbances with hair mercury concentrations ranging from 5.6–38.4 µg/g.

Prenatal exposure to methylmercury has been evaluated by recapitulation analysis of maternal hair segments. All the women sampled showed significant levels of mercury contamination during pregnancy. Like Girard and Dumont (1995), this study found a decrease in mercury concentration in maternal hair during pregnancy, with decreases of 20% from the first to the third trimester of pregnancy. Mercury levels return to preconception levels rapidly, in the first trimester postpartum. These results indicate that placental transfer of mercury to the fetus is more important than the transfer during lactation. In fact, in our study, as in others (Sikorkski *et al.* 1986), there was a significant correlation between mercury concentration in maternal and neonatal infant hair. Such statistical significance was not seen in the correlation between infant's hair and length of breast-feeding. Moreover, considering hair mercury as a proxy for blood mercury concentration, the mean transfer of maternal mercury was substantial. It reached means of 10 to 20% in the second and third trimester, respectively. Because there was a return of hair mercury concentration to prepregnancy levels during lactation, it is safe to assume that the transfer of mercury to the infant through breast milk is relatively negligible.

Chronic mercury ingestion through the food chain has brought wide contamination to the Amazonian indigenous population. Although there is no predictive linear relationship between indices of body mercury load and adverse health effects for the exposed population, it is feasible to speculate that the present levels of contamination are sufficiently high to require strict policies to protect the environment and its inhabitants.

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