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A Contribution to the Establishment of Reference Values for Total Mercury Levels in Hair and Fish in Amazonia

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Studies on mercury levels in the Amazonian Region have typically lacked background or reference parameters. A sectional study on Hg concentration in hair and fish was conducted, together with an assessment of the prevalence of signs and symptoms related to Hg poisoning, in four communities in the Amazon Basin not impacted by gold mining, located either by a river course (Santana do Ituqui and Caxiuanã) or by a lake (Aldeia do Lago Grande and Vila do Tabatinga). Mercury determinations in hair and fish were made by flameless atomic absorption spectrophotometry. Mean total Hg in hair was 4.33 μg/g (0.40–11.60 μg/g) in 321 individuals from Santana do Ituqui, 3.98 μg/g (0.40–11.76 μg/g) for 316 persons in Aldeia do Lago Grande, 5.46 μg/g (0.37–49.85 μg/g) for 504 individuals from Vila do Tabatinga and 8.58 μg/g (0.61–45.59 μg/g) for 203 inhabitants from Caxiuanã. Fish consumption was very high in all those communities but no signs or symptoms associated with Hg poisoning were found. Mean Hg concentration in fish varied from 0.006 to 2.529 μg/g for carnivores and from 0.008 to 0.871 μg/g for noncarnivores. These values suggest that further studies including a larger number of communities would eventually lead to values of "normal" Hg concentration in the Amazonian Region quite above the limits suggested by the World Health Organization.

Key Words: **mercury; background; boundary values; gold mining; amazonia.**

INTRODUCTION

The politics of Amazonian occupation initiated in the 1970s facilitated the establishment of several economic activities, especially gold mining, considered a potential source of mercury (Hg) release into the environment. This particular activity resulted in the significant increase of point emissions of that pollutant both into the atmosphere and into aquatic systems, thus contributing to the dispersion of Hg in different environmental compartments and to the possibility of its incorporation in the aquatic food chain through the processes of bioaccumulation (Lacerda *et al*., 1988; Lebel *et al.*, 1997; Malm, 1998; Hacon *et al.*, 1997).

Over the years, through many projects focused locally or involving broader geographical areas, academic works have contributed decisively to an increase in awareness of the risk of mercury pollution derived from gold mining in the Amazonian region (Pfeiffer *et al.*, 1993; Malm *et al.*, 1995). However, the establishment of levels of exposure in these areas has been hindered by the lack of reference or preexisting background parameters for comparison, with respect to both environmental matrices and biological indicators of exposure.

In this sense, the present study seeks to contribute to the establishment of such background parameters for the Amazonian region, by investigating exposure to Hg and the general health conditions of four communities located outside the area of influence of gold mining, but sharing similar food habits with other areas affected by prospecting activities—namely, relying on fish as their main source of protein.

MATERIALS AND METHODS

The methodology adopted had a standardized structure for the four communities under study, consisting of a sectional study for evaluating mercury determination in hair and fish species and the health status in the communities described below.

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The first site was the riverside community of Santana of Ituqui, in the municipal district of Santarém, located on the right margin of the Amazon river, near its confluence with the Tapajós river, in the western portion of the State of Pará. It lies some 625 km west of Belém, the State capital, and it is accessible from Santarém via the Santarém–Curuauna highway. Site number two was Aldeia do Lago Grande, at the edge of the Lago Grande lake, Amazon river basin, in the municipal district of Monte Alegre, some 600 km far from Belém. It can be reached by waterway or bus from the district seat. The lacustrine/riverine community of Vila do Tabatinga, located in the municipal district of Juriti by the Lago do Salé lake, was the third site chosen; its boundaries include the Amazon river (north), the municipal district of Aveiro (south), that of Santarém (east), and the State of Amazonas (west), and it can be reached from the district seat by road. The fourth community chosen was Caxiuanã, where the Ferreira Penna Scientific Station is located. This research station occupies an area of 33,000 hectares in the Caxiuanã National Forest, in the western portion of the State of Pará, and it includes the bay of Caxiuanã and neighboring lands belonging to the municipal districts of Portel and Melgaço (Costa *et al.* 1997). It is a preserved ecosystem and, as such, it is an ideal area for medium- and long-term scientific studies. With a low population density, local families are settled mainly around the edge of the bay.

Drinking water in all those communities originates from small rivers or lakes next to the respective village and is consumed without any kind of treatment. This water is also used for bathing and household chores. Sanitary conditions are very poor and wastes are either buried or discharged into the water.

Study procedures on each site included the following steps: (1) Preliminary contacts were made with the community, including a local population census and visits to all residences 30 days before the actual survey. (2) The survey began with an invitation to all residents to participate. Each individual signed a registration form and a release document ensuring informed consent for ethical purposes and was given a questionnaire that included data on identification, home and life conditions, occupational history, and dietary habits with emphasis on fish intake and morbidity. (3) A small local laboratory was set up for the collection, identification, and conservation of biological specimens. (4) Medical exams were performed by qualified MDs so as to include the observation of signs and general symptoms, and a special physical exam focused on features related to the central nervous system (CNS) (speech and sight, static and dynamic equilibrium, motor coordination, tactile sensitivity, muscle tone, and posture).

Biological samples collected for different purposes included hair, for quantification of total mercury, urine, when requested by clinical exams, blood, for diagnosis of malaria, determination of a hematocrit, and assessment of select blood chemicals and mercury levels, and feces, for direct assessment of the presence of intestinal parasites.

Hair samples were taken from different areas of the scalp, including at least 100 strands for each area, cropped about 1 cm off the scalp with stainless steel scissors and stored in white envelopes at room temperature for later analysis.

Fish samples were collected in lakes or rivers, and small portions of their edible tissue were removed from each specimen in the field, frozen and individually stored in plastic bags for later analysis. Fish species were identified and divided into carnivorous and noncarnivorous forms, the former category including fish-, crustacean-, mollusk- and worm-eaters and the latter mainly herbivores, mud-eaters, and plankton-feeders.

Analytical procedures for the determination of total Hg concentration in hair and fish samples were performed at the Evandro Chagas Institute (SAMAM/IEC) following the method proposed by Akagi *et al.* (1995, 1996). Mercury level was determined by flameless atomic absorption spectrophotometry using a Hg-3500 Mercury Analyzer.

Data obtained were stored using dBase software. Nonparametric methods were employed for statistical analysis. Pearson's test was applied to evaluate correlations between Hg level distributions in different groups (Armitage, 1971).

RESULTS AND DISCUSSION

Diet in all four communities studied consisted mainly of fish, with occasional additions of seasonal fruit, green vegetables, milk, and eggs. Maternal breast-feeding was a frequent habit. Clinical investigation did not reveal any signs or symptoms in the examined individuals indicating particular dysfunctions of the CNS that could be associated with mercury intoxication, such as changes in speech pattern, static and dynamic balance, motor coordination, tactile and painful sensitivity, etc. Individuals found to require attention during clinical examination were medicated accordingly. Health conditions observed were similar to those found in a community in a

TABLE 1

Distribution of Total Hg in Hair by Age Group and Weekly Fish Intake in the Riverside Community of Santana do Ituqui, State of Pará, Brazil, 1996

Age group (years)	*N*	Average weekly fish intake	Mean total Hg (μg/g)	Range (μg/g)	Standard deviation
0–4	25	11.0	3.78	0.50–8.50	2.07
5–9	76	12.8	4.14	0.40–10.50	2.37
10–14	75	12.7	4.60	1.50–11.60	2.08
15–24	44	13.9	4.29	1.30–9.60	2.00
25–34	32	13.1	4.82	1.70–9.60	2.84
35–44	30	12.4	4.12	1.20–9.20	1.95
45–54	20	12.7	4.59	1.90–9.00	2.01
> 54	19	13.5	4.12	0.70–9.00	2.14
Total	321	12.7	4.33	0.40–11.60	2.18

TABLE 2

Distribution of Total Hg in Hair by Age Group and Weekly Fish Intake in the Riverside Community of Aldeia do Lago Grande, State of Pará, Brazil, 1996

Age group (years)	*N*	Average weekly fish intake	Mean total Hg (μg/g)	Range (μg/g)	Standard deviation
0–4	34	10.0	3.36	0.40–8.68	1.90
5–9	44	12.0	3.98	0.92–11.54	2.36
10–14	62	12.0	3.93	0.46–7.56	1.66
15–24	51	12.0	3.79	0.70–7.77	1.65
25–34	37	12.0	4.34	0.79–11.76	2.80
35–44	28	10.0	4.26	0.64–11.72	2.56
45–54	17	12.0	4.89	1.22–11.42	2.71
> 54	43	12.0	3.96	1.91–7.89	2.22
Total	316	12.0	3.98	0.40–11.76	2.14

risk area, often including intestinal parasitism and diarrhea.

Tables 1 to 4 show distribution of total Hg in hair and weekly fish intake by age groups in those four communities. In all situations, the number of individuals tabulated is lower than the total number investigated in each community, as it refers to the individuals who actually allowed hair sample collecting.

In Santana of Ituqui, 425 residents and 64 houses were registered, 326 individuals (77.0%) being interviewed. Sex ratio of the population was 153 (46.9%) male individuals for 173 (53.1%) females. Mean values of Hg found for fish species consumed locally were 0.124 μg/g (0.032 to 0.367 μg/g) for carnivores and only 0.052 μg/g (0.010 to 0.186 μg/g) for noncarnivores. Table 1 indicates that mean fish ingestion for the population was 12.7 fish specimens per week, being lowest for infants from 0 to 4 years of age (11.0) and highest among 15- to 24-year-old people (13.9). Mean Hg value in hair overall was 4.33 μg/g. It is important to notice that the lowest values for hair (3.78 μg/g) were found among individuals who consumed less fish (infants), while the highest (4.60 μg/g) were observed among teenagers and adults with a high rate of fish consumption.

In the community of Aldeia do Lago Grande, a preliminary census registered 350 residents in the village, distributed in 88 households, of which 325 (93%) were interviewed. Sex ratio was 181 (55.4%) males for 146 (44.6%) female individuals. Mercury concentration in carnivorous fish varied between 0.049 and 0.489 μg/g, with a mean value of 0.300 μg/g; for noncarnivores the variation was from 0.008 to 0.183 μg/g, with a mean value of 0.044 μg/g. Table 2 shows that mean weekly fish ingestion there was 12.0 specimens. Mean Hg values in hair was 3.98 μg/g, with the lowest (3.36 μg/g) again among people up to 4 years of age and the highest (4.89 μg/g) in those 45 to 54 years old.

In Vila do Tabatinga, for a population of 700 people registered in the preliminary census, 563 people—41.9% male and 58.1% female—were included in the survey. Mercury values found in fish revealed a mean of 0.036 μg/g (0.010–0.489) for carnivores and 0.096 μg/g (0.011–0.087) for noncarnivores. The mean weekly fish ingestion was 10.47 fish specimens, being lowest (8.5) among the up to 4-year-old group and highest (12.37) in a person aged 54 years. Table 3 shows that mean values of Hg in hair was 5.37 μg/g, being lowest in those up to

TABLE 3

Distribution of Total Hg in Hair by Age Group and Weekly Fish Intake in the Riverside Community of Tabatinga, State of Pará, Brazil, 2000

Age group (years)	*N*	Average weekly fish intake	Mean total Hg (μg/g)	Range (μg/g)	Standard deviation
0–4	83	8.50	4.78	0.37–15.21	3.23
5–9	71	10.60	4.77	0.91–13.43	2.80
10–14	81	10.95	4.77	1.03–15.36	2.86
15–24	81	10.11	5.30	1.58–11.30	2.32
25–34	59	10.39	5.65	1.05–15.93	3.52
35–44	53	11.25	7.23	2.34–16.96	3.44
45–54	39	11.05	6.53	2.83–14.68	2.38
> 54	32	12.37	6.40	2.13–16.14	3.03
Total	499	10.47	5.37	0.37–16.96	3.09

TABLE 4

Distribution of Total Hg in Hair by Age Group and Weekly Fish Intake in the Riverside Community of Caxiuanã, State of Pará, Brazil, 2000

Age group (years)	*N*	Average weekly fish intake	Mean total Hg (μg/g)	Range (μg/g)	Standard deviation
0–4	38	10.74	8.28	0.61–36.70	7.55
5–9	37	13.06	8.47	2.11–45.59	7.31
10–14	34	12.94	8.21	3.65–19.46	3.85
15–24	45	12.31	9.45	3.65–44.49	7.32
25–34	17	13.41	9.49	2.56–23.28	6.30
35–44	15	12.27	7.61	1.97–15.90	4.15
45–54	15	12.07	6.16	1.53–12.13	2.93
> 54	13	12.15	10.46	3.92–22.52	5.39
Total	214	12.31	8.58	0.61–45.59	6.30

14 years of age (about 4.77 μg/g) and highest among local inhabitants aged 35 to 44 years (7.23 μg/g).

Finally, in Caxiuanã, 222 people—118 males and 104 females—took part in the survey. Mercury values obtained revealed a mean of 0.287 μg/g (0.006–2.529) for carnivorous fish and 0.093 μg/g (0.009–0.871) for noncarnivores. Mean weekly ingestion of fish was 12.31, lowest (10.74) once again among the up to 4-year-old group and highest (13.41) in individuals aged 25 to 34 years. Mean value of Hg in hair (Table 4) was 8.58 μg/g, higher than in other communities, this time being lowest in adults between 45 and 54 years of age (6.16 μg/g) and highest (10.46 μg/g) in inhabitants aged over 54 years.

Overall, considering all four communities as a whole, the mean number of fish examples taken up weekly was 11.55. Values of Hg in hair varied between 0.372 and 49.590 μg/g, with a mean value of 5.360 μg/g and a standard deviation of 3.95 μg/g.

Table 5 consolidates the main data obtained in the survey with regard to Hg values in hair and fish, in addition to the average ingestion of fish in each one of the four communities investigated. The community of Caxiuanã differs from all others with reference to Hg values in hair and in the range of Hg values in carnivorous fishes. There, for the determination of Hg in hair, in addition to the mean, the median also registers a high value, suggesting that one should not use border values, as such a practice could imply an undue rise of the mean value.

In the analysis of Hg values in carnivorous fish in Caxiuanã, although the mean value there is similar to that in other communities, the value range is much wider, reaching up to 2.529 μg/g. Detailed analysis of the ranking of the results obtained for carnivorous fish in this community shows that, while in the other three sites the highest value obtained was 0.489 μg/g, in Caxiuanã there were 34 samples with values between 0.500 and 0.987 μg/g,

TABLE 5

Distribution of Total Hg in Hair and Fish and Weekly Fish Intake in the Four Communities Studied, State of Pará, Brazil

	Santana do Ituqui	Aldeia do Lago Grande	Tabatinga	Caxiuanã
Hg in hair (μg/g)				
Number	321	316	499	214
Mean	4.33 μg/g	3.98 μg/g	5.37 μg/g	8.58 μg/g
Range	(0.40–11.60)	(0.40–11.76)	(0.37–16.96)	(0.61–45.59)
St. deviation	2.18	2.14	3.09	6.30
Median	4.00 μg/g	3.61 μg/g	4.75 μg/g	6.97 μg/g
Average weekly fish intake	12.7	12.0	10.47	12.3
Hg in carnivorous fishes (μg/g)				
Number	22	213	23	222
Mean	0.124 μg/g	0.300	0.036	0.287
Range	(0.032–0.367)	(0.049–0.489)	(0.010–0.489)	(0.006–2.529)
Hg in non-carnivorous fishes (μg/g)				
Number	43	296	55	97
Mean	0.052 μg/g	0.044 μg/g	0.096 μg/g	0.093 μg/g
Range	(0.010–0.186)	(0.008–0.183)	(0.011–0.087)	(0.009–0.871)

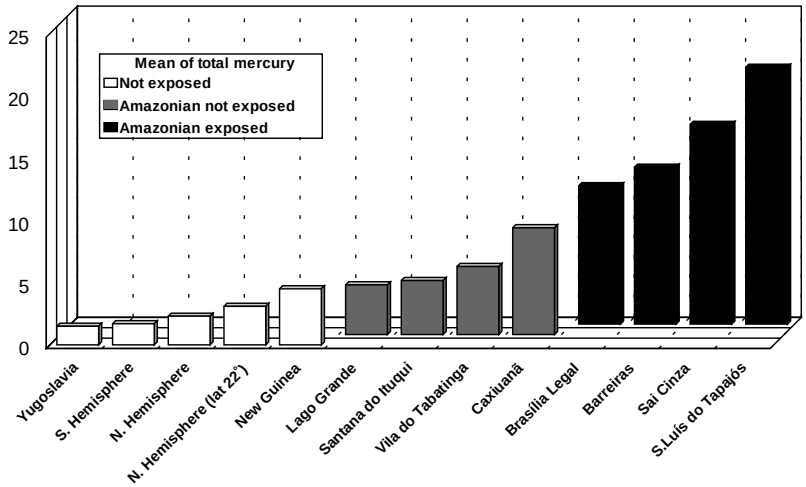

FIG. 1. Comparative levels of total Hg in hair in populations exposed and not exposed to mercure related to gold mining. Source: Horvat *et al.*, 2000; Ayrey, 1993; Suzuki *et al.*, 1998; Santes *et al.*, 2001.

7 samples in the 1.000–1.571 μg/g range, and 2 specimens—1 tucunaretinga or peacock cichlid (*Cichla temensis*, family Cichlidae) and 1 mandubé or barbelless catfish (*Ageneiosus brevifilis*, family Ageneiosidae)—with Hg contents above 2.000 μg/g.

Figure 1 presents a comparison between the mean values of Hg in hair found in each one of those four communities with those reported for other areas—three gold mining sites in Amazonia and four places in other continents free of gold mining-related Hg. It can be observed that Hg concentrations in hair in the Amazonian areas studied with no exposure to mining-related Hg represent an intermediate pattern. They are higher than those in nonexposed regions overseas, with the single exception of the mean value for a population in New Guinea—a region also located below the Equator and with a dense rainforest vegetation and tropical climate. On the other hand, as expected, those values were lower than the results for Amazonian areas actually under the influence of gold mining.

A correlation analysis between the weekly consumption of fish and the Hg value in hair was performed through Pearson's test of linear correlation, where coefficient r measures the linear relationship between two variables (Fig. 2). If $r = 1$ and $r = 0$ mean perfect correlation and total absence of correlation, respectively, a weak ($r = 0.1491$) yet statistically significant ($P = 0.0000$) correlation was observed in the communities under study, showing a slight tendency of increase in the amount of Hg in hair following an increase in the frequency of fish intake. Additionally, in accordance to the coefficient of determination $r^2 = 0.0330$, it is considered indeed a weak correlation: in other words, only 3.3% of the variation in the values of Hg could be considered related to fish consumption.

CONCLUSIONS

The populations under study presented a similar overall pattern that serves as a criterion for

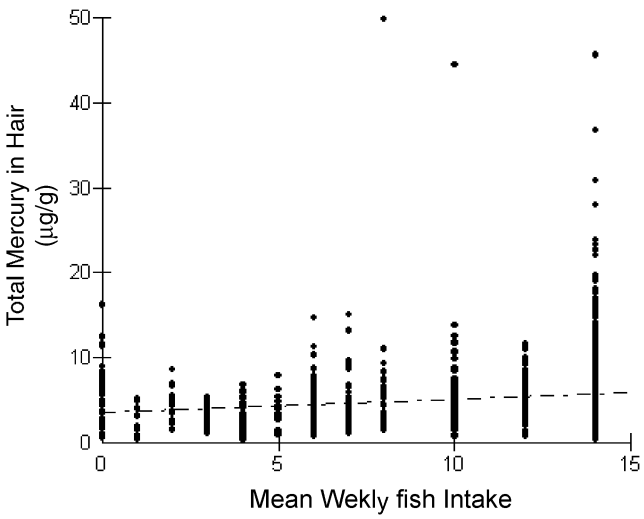

FIG. 2. Linear correlation between total hg in hair and mean weekly fish intake in the four communities studied, State of Pará, Brazil.

comparison with those described in surveys carried out in areas under the influence of gold mining-related mercury—that is, they all consist of riverine populations with reasonably similar social and economical profiles and feeding habits characterized by a high weekly consumption of fish.

The mean values of total Hg in fish in the sites under study were lower than those reported for areas actually influenced by mercury, except those registered for the community of Caxiuanã, which, although presenting a profile similar to the other three areas, had fish samples with values of up to 2.529 μg/g, an observed fact still lacking a plausible explanation.

As for the Hg contents in hair, except again for Caxiuanã, the communities under study presented mean values about 3 times below those reported for Brasília Legal, 4 times lower than those from Sai Cinza, and 4.5 times lower than those from São Luís of the Tapajós, all communities affected by gold mining activities. However, in some individuals, observed values seem to indicate that they were subjected to some degree of exposure: at the moment it is not possible to confirm the possible existence of an exposure nucleus or whether this value distribution is characteristic for the area. Further studies should be carried out for better understanding of the reason behind the high Hg values in the environmental and biological samples found in Caxiuanã.

Discussions on reference values for indicators of a "normal" condition should include several other items in addition to the definition of reference limits. It is quite possible that, following the survey of a larger number of communities, one ends up concluding that the boundaries of "normal" values in Amazonia should be higher than the standards accepted by the World Health Organization (WHO). Amazonian populations, particularly those living along rivers and lakes, could have a feeding profile different from that referred to by the WHO, and the physical features of their people (weight, height, etc.) could be of paramount importance for the accurate definition of reference values.

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