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Report

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The Catrimani River basin in northern Brazil is the home of the Yanomama and has been the site of renegade gold mining since 1980. Gold-mining operations release inorganic mercury (Hg) into the environment where it is organified and biomagnified in aquatic ecosystems. Ingestion of mercury-contaminated fish poses a potential hazard to fish-eating populations such as the Yanomama. We surveyed Hg levels in Yanomama villagers living near mined and unmined rivers in 1994 and 1995, and analyzed Hg levels in piranha caught by villagers. In 1994, 90 Yanomama Indians from 5 villages and in 1995, 62 Yanomama Indians from 3 villages participated in the studies. Four villages surveyed in 1994 were located directly on the Catrimani River, approximately 140-160 km downstream from past gold-mining activities. The other village surveyed in 1994 was situated on the unmined Ajaraní River. In 1995, 2 of the Catrimani River villages were revisited, and a third Yanomama village, on the unmined Pacu River, was surveyed. Blood organic mercury levels among all villagers surveyed ranged from 0 to 62.6 μg L^{-1} (mean levels in each village between 21.2 μg L^{-1} and 43.1 μg L^{-1}). Mercury levels in piranha from the mined Catrimani River ranged from 235 to 1084 parts per billion (ppb). Nine of 13 piranhas, measuring 30 cm or longer had total mercury levels which exceeded mercury consumption limits (500 ppb) set by both the World Health Organization and the Brazilian Ministry of Health. Unexpectedly, high mercury levels were also observed in fish and villagers along the unmined Ajaraní and Pacu Rivers suggesting that indirect sources may contribute to environmental mercury contamination in the Amazon basin.

INTRODUCTION

In the early 1980s, the Amazon region experienced a large increase in gold mining activities. In this process riverbanks and sediments are aspirated and then passed over sieves impregnated with metallic mercury. Five to 30% of this mercury is lost in the process, leaching into the waterways (1, 2). The resulting gold-mercury amalgam is heated, resulting in the vaporization of approximately 55% of inorganic mercury into the atmosphere (1).

Because mercury use is neither regulated nor monitored, the total amount of mercury discharged into the Amazonian ecosystems can only be estimated. Pfeiffer and Lacerda estimate that 1.32 kg of Hg is lost to the atmosphere per kg gold produced (1). These authors use the reported annual gold recovery (which may be as low as 10% of the actual amount obtained), and estimate the average annual discharge of mercury into the atmosphere to be approximately 52 tonnes (t) Hg yr^{-1}. This amount equals 0.9-2.1% of the total global anthropogenic atmospheric emissions of mercury (1). Contamination of the ecosystem increases to 130 t yr^{-1} if discharges into inland waters and soils are also considered (1). Due to rough field conditions, miners frequently use mercury:gold ratios as high as 10:1, thus increasing the actual total amount of mercury used (1). The inorganic mercury oxidizes, and once in the waterways (either from direct runoff or from atmospheric deposition) is converted into organic mercury by aquatic microorganisms. Bioaccumulation then occurs in the aquatic ecosystem with piscivorous fish showing the highest levels of organic mercury. Human exposure to organic mercury occurs mainly through ingestion of contaminated fish (3).

Much of the original gold-mining activities began in the Madeira River (central eastern Brazil) and in the Tapajós River areas. Environmental sampling in the Madeira River basin in the state of Rondônia indicates high mercury content in both sediment and fish. Piscivorous fish consistently have the highest levels of mercury, indicating biomagnification in the food chain (4-7). Mercury exposure among miners and the population in the surrounding areas has been assessed by several researchers.

Although total mercury levels are important, fractionation of the sample into inorganic and organic mercury is essential. People involved with gold mining and gold processing have elevated inorganic mercury levels, whereas people who consume fish have elevated organic mercury levels (8-18). Barbosa et al. (17) found mean total hair mercury levels among the Apiaká to be 34.2 ppm with 87.2% in the organic form. Kehrig et al. (16) fractionated hair samples obtained from the Tapajós and Madeira River basin finding an organic mercury level of 59.4 ppm in one individual. Among the riparian population in the Madeira River basin, with a very high fish diet, Boischio et al. (10) found one individual with a total mercury level in hair of 303.1 ppm; fractionation was not carried out in the study.

Few studies have investigated the environmental impact of gold mining in the remote area of Roraima, northern Brazil. We previously surveyed blood-mercury levels among the Makuxi people (Savannah region) and found elevated organic mercury levels in blood (19). This study focuses on the Yanomama, an indigenous preliterate stone-age people, living in the Catrimani River basin (Fig. 1), in a remote area of the tropical rainforest. These people have little contact with the outside world.

The Catrimani River was actively mined between 1980 and May 1992. Accurate estimates of the numbers of gold miners are not available because monitoring was difficult as a result of the remote terrain. Although gold miners were ejected from the area by governmental decree in 1992, renegade mining was noted from January to October 1994 (300-400 miners), and periodically still occurs within the Yanomama reserve. The Yanomama villagers in the reserve do not participate in the gold-mining activities. In 1994 and 1995, we surveyed blood organic mercury levels among Yanomama villagers and mercury levels in fish from the Catrimani River basin, Roraima, Brazil. The aim was to determine whether these mining activities had an impact on the environment and health of these indigenous people.

MATERIALS AND METHODS

Sampling Procedures

In January 1994, during the dry season, we surveyed villagers living in 4 villages (Haximapiiporatheri, Wapokohipiitheri, Ukuxipiitheri, and Maamapiitheri) along the Catrimani River, 140-160 km downstream from past mining sites and 1 village (Rohahipiitheri) in an area distant from the mining along the Ajaraní River (Fig. 1). In February 1995, villagers living in Maamapiitheri and Ukuxipiitheri were resurveyed, as well as villagers living in Pookohipiitheri, along the unmined Pacu River. In both 1994 and 1995, all villagers over 1-year old, who were in residence at the time of the survey, were invited to participate. All residents who were eligible participated in the surveys. Informed consent, demographics, occupational history, medical history and dietary history were obtained. This study was approved by the Institutional Review Boards of both the University of Wisconsin at Madison and the University of Illinois at Chicago.

Sample Treatment and Analyses

Three milliliters (ml) of venous blood (1.5 ml from children less than 5-years old) were collected from each participant in separate Vacutainer tubes containing liquid EDTA. Ten Vacutainer tubes with the same lot number were retained to evaluate the mercury blank. The samples were stored in a solar powered refrigerator at approximately 4°C. They were then transported in a styrofoam cooler with ice to the United States and analyzed for total and inorganic mercury content by cold vapor spectrophotometry (20). Using this method, total mercury is determined by reduction of all species of mercury (inorganic and organic) with a stannous chloride/cadmium chloride mixture. Inorganic mercury is determined in a separate sample with stannous chloride. The organic mercury (primarily methylmercury) is then determined as the difference in these 2 measurement results. Quality control for these determinations was established by the duplicate analysis of 2 bench whole-blood pools. One whole-blood pool contained inorganic mercury and methylmercury in approximately equal concentrations. The whole-blood bench pools were "normal" whole blood spiked to about 15-25 ng ml^{-1}. All means and ranges for bench controls were within previously established 95% confidence limits. Technical assistance was provided by the Center for Disease Control and Prevention.

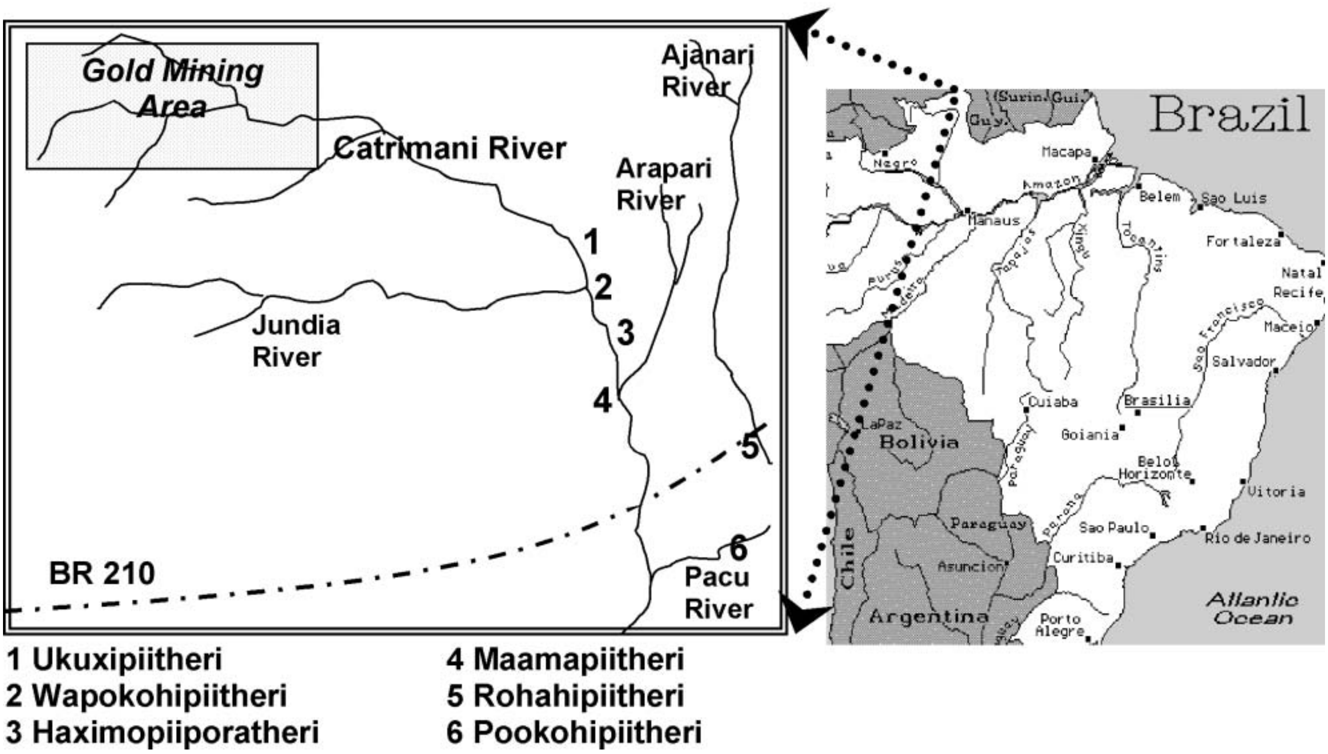
Fish Sampling and Analyses

In January 1994, 18 dietary fish from waters near each village on both the Catrimani and the Ajaraní Rivers were sampled from normal catches. In February 1995, 22 dietary fish were sampled from normal catches along both the Catrimani and Pacu Rivers. Each fish was measured for length and photographed for identification. Approximately 2 grams (g) of edible tissue were excised, weighed, and then stored in 95% isopropyl alcohol. This method of preservation was tested for reproducibility of mercury analysis prior to sampling and found to be acceptable (< 10% error). Isopropyl alcohol preservation was chosen because freezing of the samples was not possible. Analysis for total mercury was by cold vapor atomic absorption spectroscopy using standard EPA methods (21). Results are expressed on an original wet weight basis.

Data Analyses

Mean ages among villages were compared using the 1-way analysis of variance (ANOVA) method. The same method with age as a covariate was applied to compare mean mercury levels among villages. Scheffé's multiple contrasts were used in *a posteriori* comparisons. To compare mean Hg levels in Ukuxipiitheri villagers between 1994 and 1995 the 2-sample t-test was applied. Mercury levels in Maamapiitheri villagers between 1994 and 1995 were compared using the paired t-test. Mercury levels of families across the villages were compared using the nested ANOVA with age adjustment. The relationship between mercury level and size of fish was analyzed using the linear regression technique with log-transformed data of the 2 measurements. For Catrimani River, the multiple regression method was separately applied to test equal slopes or coincidence of 2 regression lines, to compare the relationship between mercury level and size of fish in 2 rivers and 2 different years.

Figure 1. Yanomama villages.



Piranha (*Serrasalmus rhombeus*). Photo: K. Sing.

RESULTS

Blood Samples

Table 1 shows the results of samples collected in 1994 for blood organic and total mercury levels in villagers. A total of 60 people were sampled in the mined Catrimani area, and 30 in the unmined Ajaraní village. In the village of Ukuxipiitheri, only 4 individuals were available since the rest of the inhabitants (n = 27) were visiting a village outside the area. There was no significant difference in mean ages among the villages (1-way ANOVA, $F_{4,85} = 1.23$, $p = 0.30$). The means for both blood organic and total mercury for the 5 villages were not all equal (1-way ANOVA with age adjusted; $F_{4,84} = 14.83$, $p < 0.001$; $F_{4,84} = 16.25$, $p < 0.001$. Age adjustment was not important; $F_{1,84} = 0.84$, $p = 0.36$; $F_{1,84} = 0.43$, $p = 0.51$). Mean blood organic and total mercury levels were significantly higher among villagers living along the Catrimani River (Haximapiiporatheri, Wapokohipiitheri, Ukuxipiitheri and Maamapiitheri) compared to villagers living in Rohahipiitheri along the unmined Ajarani River (Scheffé's multiple contrasts using the average of 4 villages along the Catrimani River; both significant at $\alpha = 0.05$). Maamapiitheri had significantly higher mean mercury levels than each of the 4 other villages (Scheffé's pairwise multiple contrasts; both significant at $\alpha = 0.05$).

Table 2 presents the results of the surveys of 3 villages conducted in February 1995. The village of Ukuxipiitheri was resurveyed with all of the villagers present (n = 31). Because Maamapiitheri had elevated mercury levels in 1994, a second set of blood samples was taken (n = 11). Pookohipiitheri, on the Pacu River, was the last village tested in 1995. Again, there was no significant difference in mean age among the 3 villages (1-way ANOVA, $F_{2,59} = 0.78$, $p = 0.47$). The mean organic and total blood mercury levels in these villages were not all equal (1-way ANOVA with age adjusted; $F_{2,58} = 17.43$, $p < 0.001$; $F_{2,58} = 16.65$, $p < 0.001$. Age adjustment was not important; $F_{1,58} = 0.13$, $p = 0.72$; $F_{1,58} = 0.30$, $p = 0.58$) (Table 2). Again, Maamapiitheri had the highest mean total blood mercury level (Scheffé's pairwise multiple contrasts; both significant at $\alpha = 0.05$), as well as the individual with the highest level (60.3 $\mu g\ L^{-1}$).

In the 2 villages that were resurveyed, Ukuxipiitheri and Maamapiitheri, there were no significant differences in mean blood organic and total mercury levels between 1994 and 1995 (Ukuxipiitheri: 2-sampled t-tests; $t_{33} = 0.44$. $p = 0.66$; $t_{33} = 0.78$, $p = 0.44$. Maamapiitheri: paired t-test, 10 pairs; $t_9 = 0.35$, $p = 0.73$; $t_9 = 0.66$, $p = 0.52$). Mercury levels among all villagers surveyed in 1994 were significantly clustered within families (nested ANOVA: $F_{26,58} = 2.53$, $p < 0.002$ for organic mercury; $F_{26,58} = 2.59$, $p < 0.002$ for total mercury).

Fish Samples

The results of mercury levels in piranha (*Serrasalmus rhombeus*) caught in each river as a function of the size of fish are shown in Figure 2. Mercury levels in these piranha increase as the size of the fish (and therefore age) increases. Following previous methods (22), both mercury level and size of fish were log transformed and regression lines were fitted with the following results.

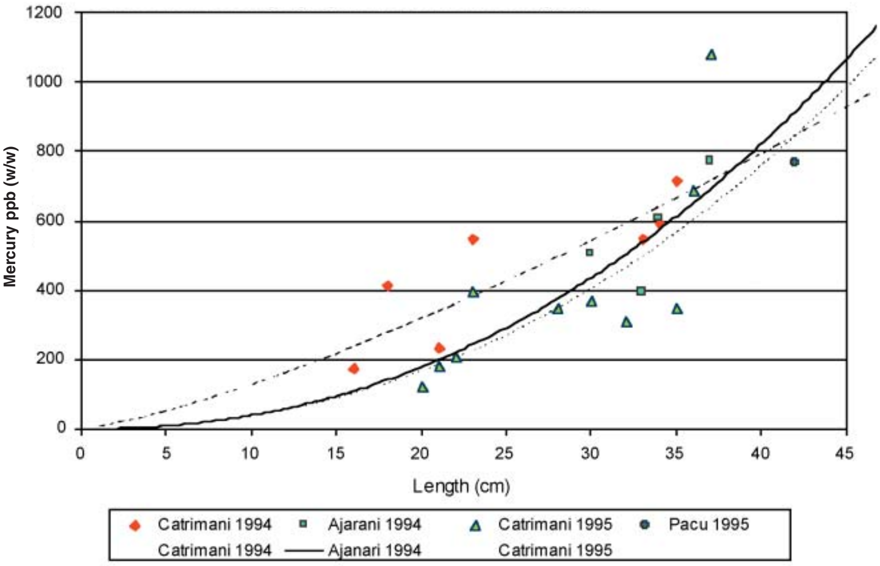
Catrimani River 1994:
$\text{Log (Hg)} = 0.779 + 1.323\ \text{Log (length)}$ [n = 7, Adj $R^2 = 0.604$, $p = 0.024$]

Ajarani River 1994:
$\text{Log (Hg)} = -0.618 + 2.205\ \text{Log (length)}$ [n = 4, Adj $R^2 = 0.175$, $p = 0.329$]

Catrimani River 1995:
$\text{Log (Hg)} = -0.524 + 2.111\ \text{Log (length)}$ [n = 9, Adj $R^2 = 0.586$, $p = 0.010$]

The regression lines of mercury levels on fish sizes (both log transformed) between Catrimani River (mined) and Ajanari River (unmined) in 1994 were similar (multiple regression analysis $F_{1,8} = 2.34$, $p = 0.16$ for the rivers; $F_{1,8} = 13.79$, $p < 0.01$ for log transformed length). No significant change in the slopes was observed between 1994 and 1995 for Catrimani River

Figure 2. Relationships between length of piranha *(Serrasalmus rhombeus)* and a total mercury in piranha by river and year (Data shown in original scale; curves are fitted using log-log transformed for each group).



Yanomama of this remote Catrimani River basin are lower than the levels found in the Madeira and Tapajós. The village of Maamapiitheri had the highest mean blood organic mercury levels (43.1 μg L^{-1} in 1994 and 42.2 μg L^{-1} in 1995), as well as the individual with the highest levels (62.6 μg L^{-1} blood or approximately 25 ppm in hair) in both 1994 and 1995. It is the only village statistically different from all the other villages studied. This village is small with only 11 people, 2 of the men hunt and fish, one of whom has a below the knee amputation from a previous accident. The village itself is located on the convergence of a small stream abundant with fish, and the Catrimani River, thus increasing the availability of fish. Although we could not quantify the amounts consumed, from the villagers' descriptions, it appears that their diet is primarily fish throughout the entire year with game being very scarce.

The results of this study differ from 2 other previous studies. Kehrig, et al. (16) in 1997 analyzed 14 hair samples from Yanomama individuals from 1990 and found a mean of 3.31 mg kg^{-1} (blood mercury approximately 13.2 μg L^{-1}). Malm et al. (14) analyzed 162 hair samples collected from February to March of 1990 and found values from 1.4-8.14 μg gm^{-1} (blood mercury approximately 5.6-32.6 μg L^{-1}) with an average of 3.61 μg gm^{-1} (blood mercury approximately 14.4 μg L^{-1}). Both papers point out that these particular Yanomama eat few fish. However, neither dietary history nor fish surveys for mercury levels were carried out in either of these studies. This present study notes much higher levels of blood organic mercury levels in all the Yanomama villages (along the mined Catrimani River, the unmined Pacu and unmined Ajaraní Rivers). This may be due to the high fish diets in this area, to a more marked environmental contamination in the Catrimani River basin, or a combination of both.

(Fig. 2), although the slope of 1995 had increased from 1.323 to 2.111 (multiple regression analysis, $F_{1,12} = 1.16$, $p = 0.30$ for the interaction term between the year and log transformed length). Only one specimen was obtained from the Pacu River in 1995. The highest mercury level (1084 ppb) was observed in a 39 cm long piranha, which was caught in 1995 in the Catrimani River.

Dietary History

Retrospective dietary history was found to be unfeasible because the Yanomama only express numbers of one, two, one couple, two couples and many. Therefore, one of the authors, Giovanni Saffirio, conducted a 17-day prospective dietary survey during the wet season in October 1994, and a 12-day period during the dry season, in December 1994, at Ukuxipiitheri (not during the collection of blood samples). Women fish in small creeks using poison from leaves, whereas the men use lines and hooks when fishing in the Catrimani River. Thus, men caught primarily piranha and the women caught herbivorous fish. Hunting was performed with bows and arrows. Both fish and game were weighed prior to cooking.

In the wet season in 1994, 35 people were living at Ukuxipiitheri. Each person consumed approximately 819 g per week or 3.5 fish meals per week. Fish comprised 26% of the food gathered, with 57% of the fish being piscivorous (primarily piranha). In the dry season, the protein diet of the villagers consisted almost entirely of fish (95% of protein) with 56% of them being piscivorous fish (primarily piranha). Twenty-four people were in residence and consumed approximately 2 kg of fish per week per person, or 8.7 fish meals per week.

DISCUSSION

In the Madeira and Tapajós River areas where gold mining has occurred for many years, mercury contamination in hair in the environment (4-6, 10, 14) and elevated organic mercury levels have been previously noted among the populations (8-18). Other authors have shown that the blood-to-hair ratio of mercury levels is approximately 1 to 250, although individual variations exist (3). Hair organic mercury levels as high as 59.4 ppm have been reported in certain populations in this area (16) or approximate blood organic mercury levels of 238 μg L^{-1}.

The blood organic mercury levels found among the

Because methylmercury is present in many fish species, investigators have looked at the relationship between fish consumption and blood mercury levels in different populations. Brune et al. (23) reviewed 98 international publications that measured blood mercury levels in fish consumers. Reports were classified in categories according to fish consumption and corresponding mean blood mercury level: I (0 fish meals per day) = 2.0 μg L^{-1} (SD 1.8); II (< 2 fish meals per week) = 4.8 μg L^{-1} (SD 1.9); III (2-4 fish meals per week) = 8.4 μg L^{-1} (SD 4.5); IV (> 4 fish meals per week) = 44.4 μg L^{-1} (SD 29.9). One fish meal was considered to equal 230 g. In this study, the Yanomama ate 3.5 fish meals per week (category III) in the wet season, and 8.7 fish meals per week (category IV) in the dry season. The mean organic blood mercury levels in all villages are similar to the mean blood mercury levels found in other populations of high fish consumers. Thus, the blood organic mercury levels among the Yanomama in this study appear to correlate with the amount of fish they consume.

The consequences of consuming large amounts of methylmercury have been shown in both Minamata, Japan, and in Iraq. Based on these 2 outbreaks, WHO refers to a blood mercury level of 200 ppb as the threshold for the clinical manifestation of Minamata disease (3). Recent studies have explored whether lower levels of methylmercury contamination can lead to subtle adverse neurologic effects. Meyers et al. (24) were unable to show a relationship between deleterious effects on a population of children in the Seychelles, who were exposed to methylmercury prenatally and postnatally, and maternal hair mercury concentrations (mean = 6.8 ppm). Neuro-psychological and neurobehavioral test batteries were administered at 6.5, 19, and 20-months of age. A recent follow-up study at 66 months continues to indicate no adverse outcomes (25). However, in the Faroe Islands, Grandjean et al. (26) studied 112 7-year-old children whose mothers had a hair mercury concentration of 10-20 μg g^{-1}. On 6 neuropsychological test measures, these children showed mild decrements relative to controls (i.e.children with exposures less than 3 μg gm^{-1}), especially in the areas of motor function controls, language and memory. In addition, Lebel et al. (27) studied adult villagers on the Tapajós River with total hair mercury levels < 50 μg gm^{-1}, and found that near contrast sensitivity and manual dexterity decreased significantly with hair mercury levels. Comparisons between these papers are difficult because of the different neuro-behavioral and motor testing procedures used. The mean blood mercury levels found among the Yanomama of this study are similar to the levels among the Faroe Islanders.

One interesting finding in this study is that the blood mercury levels appear to be clustered among Yanomama families. The Yanomama men tend to share their catch (primarily piranha) within the immediate family, and then the extended family. When the women catch fish (using a poisoning technique), they work as a group and often all of the women of the village participate. The total fish catch then tends to be distributed among the entire village. However, the fish obtained from these fishing expeditions are primarily herbivorous fish with predominantly lower levels of methylmercury when compared to levels in piscivorous species.

The dietary fish samples analyzed in this study support the findings of other authors who have noted increased levels of mercury in the environment, with the highest levels being found in piscivorous fish (4-6, 10, 14, 23, 28). As with the blood organic mercury levels, the mercury levels in the fish in the Catrimani, Ajaraní and Pacu Rivers, were not as high as those found in the Tapajós or Madeira Rivers. Surprisingly, the 2 unmined Ajaraní and Pacu Rivers, which are not connected to the mined Catrimani River and do not share watersheds, had piscivorous fish with elevated mercury levels, to the same degree as in the Catrimani River (Fig. 2). This may indicate indirect contamination into the waterways, e.g. from atmospheric deposition. Porvari (29) commented that studies have shown that divalent and gaseous Hg are deposited in areas as far away as 100 km from the emission source. The presence of organic carbon, and the pH of the sampled rivers, the most important parameters in methylmercury formation, were not investigated in this study (30).

In conclusion, the Yanomama, inhabitants of the Catrimani river basin, have elevated blood mercury levels which appear to correlate with their levels of fish consumption. Fortunately, the Yanomama in this study are not as severely affected by Hg pollution as are populations in other parts of Brazil.

There are certain limitations to this study, primarily related to the fact that a prospective dietary history is available for only one of the villages. It is possible that the differences noted between each village is secondary to dietary intake only. However, the Yanomama are hunters and gatherers who obtain protein from their environment. These villages are close enough in proximity that they share the same environmental ecosystems, with game being equally as plentiful or scarce in all the villages. The Yanomama prefer game over fish and eat fish only when game is scarce or other factors are involved as noted for Maamapiitheri. In addition, with known mercury levels in the dietary fish, it is clear that the Yanomama in this area are at risk for increased levels of blood organic mercury. It is unclear if these levels are directly related to the gold-mining activities or from a high background level. In addition, with the devastating fires that occurred in 1998, game will be scarce and the Yanomama will rely more heavily on fish for their dietary needs. Moreover, as some investigators have indicated, burning trees and vegetation may increase the amount of mercury deposition. The effects of this additional environmental strain on the Yanomama people remains to be seen and investigated.

Table 1. Average age, total Hg and organic Hg for 5 Yanomama villages, 1994.

Village	N	Age Mean ± SD (range)	Total Hg μg L^{-1} Mean ± SD (range)	Organic Hg μg L^{-1} Mean ± SD (range)
Haximapiiporatheri[a]	24	22.0 ± 17.0 (1.5 – 64)	29.0 ± 8.2 (16.3 – 48.1)	25.8 ± 8.2 (13.8 – 44.8)
Wapokohipiitheri[a]	21	29.9 ± 22.0 (1.5 – 81)	24.5 ± 9.7 (0 – 41.6)	21.2 ± 8.9 (0 – 37.8)
Ukuxipiitheri[a]	4	38.5 ± 30.2 (10 – 68)	32.3 ± 13.8 (17.5 – 41.6)	27.4 ± 14.8 (11.3 – 40.1)
Maamapiitheri[a]	11	29.2 ± 22.2 (3 – 73)	48.7 ± 10.1 (33.7 – 68.2)	43.1 ± 9.9 (28.9 – 62.6)
Rohahipiitheri[b]	30	23.2 ± 13.2 (3 – 56)	26.3 ± 7.0 (15.8 – 45.2)	22.4 ± 6.4 (12.3 – 37.3)

[a] Along the Catrimani River 140 - 160 km downstream from past mining sites.
[b] Along the unmined Pacu River.

Table 2. Average age, total Hg and organic Hg for 3 Yanomama villages, 1995.

Village	N	Age Mean ± SD (range)	Total Hg μg L^{-1} Mean ± SD (range)	Organic Hg μg L^{-1} Mean ± SD (range)
Ukuxipiitheri[a]	31	24.7 ± 17.4 (4 – 70)	28.7 ± 7.8 (17.7 – 48.1)	25.5 ± 7.0 (15.4 – 43.4)
Maamapiitheri[a]	11	30.8 ± 22.6 (5 – 75)	46.2 ± 10.1 (25.8 – 60.3)	42.2 ± 10.1 (22.8 – 56.6)
Pookohipiitheri[b]	20	30.4 ± 17.4 (4 – 67)	30.9 ± 8.8 (15.8 – 51.2)	27.7 ± 8.4 (13.4 – 47.3)

[a] Along the Catrimani River 140 - 160 km downstream from past mining sites.
[b] Along the unmined Pacu River.

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