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Lifestyle and Mercury Contamination of Amerindian Populations along the Beni River (Lowland Bolivia)

Selma Ximena Luna Monrroy, M.Sc.; Ronald Wily Lopez, M.Sc.; Marc Roulet, Ph.D.; Eric Benefice (Corresponding author), M.D., Ph.D.

Abstract

The objective of this paper was to document mercury contamination of Amerindian populations living along the Beni River in Bolivia and to examine risk factors related to their lifestyle. A cross-sectional survey was performed among 15 communities on the flood plains of the Beni River at the foothills of the Andes. Hair mercury content (H-Hg) served as a bioindicator of mercury contamination. Mercury values were available for 556 people. Four indicators of lifestyle were analyzed: community accessibility, subsistence activity, fish consumption, and ethnicity (i.e., members of the Tacana or Ese Ejja ethnic group). The median of H-Hg was equal to 4.0 μg/g (95% *CI* [confidence interval] = 3.6–4.4). Approximately 86% of the subjects had H-Hg values lower than 10 μg/g. No significant differences existed in H-Hg between adult women and children, nor according to age group. Subjects belonging to the Ese Ejja ethnic group had higher H-Hg than subjects from the Tacanas ethnic group. Communities accessible only by canoe were more frequently contaminated than those accessible by road. Subjects who ate at least one serving of fish per day had higher H-Hg, and families who maintained substantial fishing activity were more strongly contaminated. Contamination levels were found to be low compared with other Amazonian studies. The most strongly affected groups, however, were those which preserved a traditional way of life and were the most economically and socially disadvantaged.

Introduction

Mercury is a widespread element that acts as a potent neurotoxin after conversion into methylmercury (MeHg). Mercury in aquatic environments is methylated through the action of anaerobic bacteria, and enters the food chain in this form. As a consequence, the main path to contamination of the general population is through fish consumption.

Contamination of populations in the Amazon basin has been recognized since the 1990s and was attributed to gold-mining activity (Branches, Erickson, Aks, & Hryhorczuk, 1993). Mercury is used to extract gold found in river beds and is evaporated from the amalgam by heating. Less than 30% of the mercury is recovered, while more than 60% is released into the atmosphere and returns to the soil with the rains or is directly rejected into the river (Maurice-Bourgoin & Quiroga, 2002).

Various studies reported that riverside Amazonian populations, especially fish-eating groups, were exposed to mercury contamination (Santos et al., 2000). Moreover, a neurological impact was discernible in adults as well as in children (Grandjean, White, Nielsen, Cleary, & de Oliveira Santos, 1999; Lebel et al., 1998).

The situation in the Andean tributaries of the Amazon has not been extensively elucidated despite the fact that huge quantities of mercury have been used for centuries (Maurice-Bourgoin & Quiroga, 2002). Gold-mining activity is not the only source of mercury: erosion of contaminated soils and volcanic activity must also be considered (Maurice-Bourgoin, Quiroga, Chincheros, & Courau, 2000; Roulet et al., 1999). Substantial gold mining took place in the upper Beni, however, and involved about 200 cooperatives. It was estimated that each year, 250 to 500 kg of mercury were used and that 50%–70% of the mercury was directly released into the environment (Maurice-Bourgoin & Quiroga, 2002). Contamination of populations living downstream could therefore be suspected. A survey performed in 1995–2000 reported fairly high concentrations of mercury in the hair of members of an Amerindian community living on the bank of the Beni (Maurice-Bourgoin et al., 2000). The expansion of the Beni just at the exit point of the Andes, its contribution to forming the Rio Madeira (one of the main tributaries of the Amazon), and the existence of sites favorable to mercury methylation justified a more in-depth survey of the process of mercury contamination of riverside populations.

Hence, the purpose of our study was twofold: first, to document mercury contamination in a group of communities living in the flood plain of the Beni River and second, to examine risk factors associated with their lifestyle.

Subjects and Methods

Study Context

Within the framework of a multidisciplinary study on mercury contamination in the flood plains of the Beni River, it was decided that a study be performed of human populations living in the area, from the Andean piedmont to 120 km downstream. The survey was performed from March to August 2004.

Subjects and Sampling

According to a national census in 2001, 14,000 people lived in the project area, but only 4,000 actually lived along the river bank. Since the present study considered the risk in the general population, we decided to include only the women and their children. The rationale for this choice was that prenatal mercury contamination could severely impair cerebral development of infants due to the capacity of methylmercury to cross the placenta barrier (Gilbert & Grant-Webster, 1995). The population was scattered throughout small communities and hamlets with frequent displacement for their subsistence. It was not possible to draw a random sample, and we preferred to examine each mother and her children present at the site during the visit. In all, 173 mothers, 354 children 0–10 years of age, and 104 children 10–15 years of age were surveyed.

Ecological and Cultural Setting

The area under study is part of the northern Amazon region of lowland Bolivia. It encompasses 10 millions hectares with a population of around 170,000 inhabitants. This region has a tropical climate with an annual rainfall ranging from 1,800 to 2,200 mm. Two ethnic groups live in this area. The largest ethnic group is the Tacanas, and the smaller ethnic group is a closely related tribe, the Ese Ejjas. The Tacana population is made up of around 3,400 people, on the basis of self-designation (*Diagnóstico Socioeconómico*, 2005).

Tacanas are mainly farmers who grow maize, rice, tubers (cassava roots), plantains, and bananas. They practice a system of slash and burn to cultivate pieces of land that can

reach one hectare. They obtain a substantial part of animal foods from hunting and fishing. They are well integrated into the Bolivian society. The majority of Tacanas speak Spanish and are able to read and write. The Ese Ejjas form the second group. They are believed to currently number 600–700 members. The clan living in the study area had approximately 200 people. Few adults have gone to school or speak Spanish, and they continue to use their vernacular language. Their productive activities are oriented towards use of forest and river resources. They cultivate the same products as the Tacanas (rice, maize, tubers, plantains) but in smaller parcels. They dedicate a large part of their time to fishing either in groups or individually. They have preserved a semi-nomadic way of life, following migration of fish and returning to their village during the flooding period.

Health Characteristics

Information collected during the visits indicated a mediocre health status in this population. Results have been partly published elsewhere (Benefice, Monroy, Jimenez, & Lopez, 2006). Fifty percent of the mothers surveyed had lost at least one child. Mothers usually breastfed their children for more than one year. The prevalence of anemia among women was high: 42% (95% *CI* [confidence interval] = 34.6%–49.8%) presented moderate to severe forms of anemia. This figure was 1.5 times higher than the national level. Chronic malnutrition represented by growth retardation was very frequent in children: 41% (95% *CI* = 33.9%–49.0%) of preschoolers were affected. Intestinal parasitism was very frequent in children.

Ethical Considerations

This study was approved by the "Comité Nacional de Bioética de Bolivia" and by the "Comité Consultatif de Déontologie et Bioéthique" (IRD, France). Subjects or their parents were individually informed concerning the aims of the study, and they or their parents signed an informed consent form. At the end of the study, results were individually provided to each participant with explanations.

Variables in the Study

Mercury

In the present study, total mercury content of hair (H-Hg) was used as a bioindicator of contamination. Hair strands were cut at the occipital region. Analyses of mercury were carried out in the Laboratory of Environment Quality (LCA) of the Ecology Institute of La Paz University. Hair samples were rinsed in 0.01% ethylenediaminetetraacetic acid (EDTA). Analyses were performed on total length of the hair. A portion of 20 mg of hair was digested using acids (two volumes of nitric acid [HNO_3] and one volume of sulfuric acid [H_2SO_4]). Measurements were performed using cold vapor coupled with atomic absorption spectrometry (Perkin Elmer 3110®). All analyses were performed in duplicate. In case of a difference greater than 10% between measurements, a third analysis was done. When this was not possible (e.g., not enough material), values were discarded. For this reason 63 values were rejected. Twelve people did not accept hair cutting. Mercury values were available for 556 people (88% of the subjects; 163 mothers and 393 children).

Independent Variables

In this study, living conditions of communities were represented by four variables.

1. Village accessibility. Communities were divided into three groups—those accessible via a non-asphalted track (representing four communities located four to 15 km from the small town of Rurrenabaque); six communities located about two to five hours by canoe downstream from Rurrenabaque; and five communities eight to 12 hours downstream from the towns accessible only by canoe.
2. Main subsistence activity. Families were divided into three groups—predominance of farming and logging; practice of fishing as the main activity or secondary activity; and wage-earning, storekeeping, or handicrafts.
3. Ethnicity. Self-classification in one of the ethnic groups of the area: Tacana (149 families) or Ese Ejjas (25 families).
4. Fish consumption. Each mother was interviewed concerning the frequency of fish consumption: 1) at least one serving per person per day; 2) less than one serving per day but at least one serving per week; 3) less than one serving per week. Mothers were also requested to name, describe, and identify the fish they consumed and give an order of frequency.

Statistical Analysis

Data were entered and double-checked using Epi Info software. All continuous variables were checked for normality of distribution. Bivariate comparisons were done using Student's *t*-test or the one-way analysis of variance test. Scheffe and Newman-Keuls post hoc multiple comparison tests were applied. In case of absence of normality of distributions, non-parametric tests were employed. Multiple associations between mercury exposure and indicators of lifestyle were studied through a multinomial logistic regression procedure. Main effects and two-way interaction terms were included.

Results

Mercury Content of Hair

Table 1 shows H-Hg values in different groups of subjects. As a whole, the median was equal to 4.03 µg/g (95% *CI* = 3.6–4.4). The distribution was asymmetric, with a heavy tail towards the right. The median was 4.4 µg/g for mothers and 3.9 µg/g for children, but the difference between medians was not statistically significant ($Z = 1.03$, $p = .30$). Among children and adolescents, no gender difference existed. The correlation coefficient between H-Hg of mothers and children was moderate to high (Spearman rank correlation [r_s] = 0.68 for $n = 390$ pairs). Interestingly, correlation coefficients were apparently higher for adolescents ($r_s = 0.74$) than for children ($r_s = 0.67$).

The distribution of H-Hg in mothers varied significantly according to their status. Pregnant mothers had the lowest H-Hg concentrations and lactating mothers the highest (Kruskall-Wallis one-way analysis of variance test, $p < .001$).

As a whole, H-Hg was not significantly correlated with age. When considering breastfed infants and weaned children separately, however, a negative correlation existed between H-Hg and age from birth to18 months of age ($r_s = -0.32$, $p < .04$), and a positive correlation, though not significant, from 18 to 36 months of age ($r_s = 0.28$, $p = .06$). Hence, the relationship appeared curvilinear, with a slow decrease during the breastfeeding period, followed by a rise thereafter.

Mercury hair content was not normally distributed. After appropriate log transformation, the hypothesis of normality could be accepted (Martinez-Iglewicz test). Hence, we used this log-transform value (Ln-Hg) in subsequent analysis.

Fish Consumption

Among a total of 173 mothers, seven were not able to provide reliable answers concerning fish consumption in their family. Among the 166 responding mothers, 20% said they ate at least one serving of fish per day; 62% said more than once a week; and 18% said less than once a week. Among these mothers, only 154 gave accurate responses to the questionnaires. They identified 18 different varieties of fish. The four most frequently consumed fish were "sábalo" or *Prochilodus nigricans* (43.5% of answers); pacú or *Colossoma macropomum* (9.1% of answers); "pintado" *Pseudoplastystoma fasciatum* (9% of answers); and palometa or *Pygocentrus nattereri* (6.4% of answers). Sábalo and pacú are herbivorous fishes, pintado is a piscivorous fish, and palometa is a carnivorous fish. Consumption of herbivorous and omnivorous fish accounted for 63.6% of answers, and piscivorous and carnivorous fish for the remaining 36.4%. We did not find significant differences in the nature of fish consumed between the two ethnic groups. A significant difference existed, however, according to village accessibility: communities with road access ate the highest proportion of herbivorous fish (75%), while remote communities, accessible only by canoe, consumed the highest proportion of carnivorous fishes (54.5%) ($\chi^2 = 12.5$, $p < .001$).

Relationships between H-Hg and Community Characteristics

Figures 1 to 4 represent the cumulative distribution of H-Hg according to various groupings of subjects. The arrows dissect the 50% line.

- Grouping according to ethnicity (Figure 1): the Ese Ejjas curve was swerved to the right and its median was superior to that of the Tacanas. Mean values were significantly different ($t = 7.9$, $p < .001$).
- Grouping according to village accessibility (Figure 2): communities close to the city but accessible only by canoe presented the highest H-Hg distribution. Differences between means were statistically significant ($F = 129.5$, $p < .0001$). Villages close to the city with road access had lower H-Hg values than those accessible only by canoe.
- Grouping according to fish consumption (Figure 3): subjects with at least one serving of fish per day had higher H-Hg values than subjects with one or less servings per week. Mean differences were significant ($F = 20.1$, $p < .0001$).

- When fishing was the primary or secondary subsistence activity of the family, the distribution curve shifted to the right in comparison with farmers, loggers, or wage earners (Figure 4). Because distributions were not normal, a non-parametric test was used (Kruskall-Wallis one-way analysis of variance test). Differences between medians were statistically significant ($\chi^2 = 113.2$, $p < .0001$).

Redundancies between lifestyle variables are clear cut. Thus, to disentangle the relationships, a logistic regression model was run between H-Hg as dependent variable, divided in two discrete categories: low H-Hg ≤ 1.18 μg/g (1st quartile of the distribution) and high H-Hg ≥11.4 μg/g (4th quartile of the distribution) and the same categorical variables analyzed over village accessibility, ethnic group, fish consumption, and subsistence activity. A model without subset selection and without interaction was first run. The model had a normal completion and classified correctly 86.9% of the subjects. Village accessibility and fish consumption produced significant effects but not ethnicity or subsistence activity. Then a hierarchical model with forward selection of terms was used (the term with the largest log-likelihood was entered first) (Table 2). Village accessibility and fish consumption still significantly predicted the classification in high or low H-Hg groups. Indeed, interactions existed between accessibility and fish consumption and subsistence activity and fish consumption.

Discussion

The present study raises two different conclusions. First, the mercury contamination level of these populations seemed to be relatively low: 86% of subjects were below 10 μg/g, a proposed limit for doubling of the 5% standard risk for neurological abnormalities in infants (World Health Organization, 1990). Hopefully, among the 14% of subjects with H-Hg higher than 10 μg/g, no pregnant women were found. Second, results pointed to greater exposure among populations that continue to preserve an "Amazonian way of life." These groups have managed to survive in a somewhat aggressive environment with poor resources (Dufour, 1992). They are disadvantaged due to difficulties in integration into the national population, encroachment into their territory by colonialists, and poor access to medical and educational facilities. And they now must survive in a contaminated environment.

A pioneer study on mercury contamination of the Ese Ejjas community was published some years ago (Maurice-Bourgoin et al., 2000). The authors found a mean H-Hg of 9.8 μg/g, which was very close to that in the present study (median = 9.2; min = 0.5 to max = 34.2 μg/g

for the Ese Ejjas). Our sample did not include adult males, however. We directed our efforts toward mothers and their children, because they represented the most vulnerable group. Nevertheless it is possible that non-inclusion of males lowered the observed mean H-Hg values.

In the Amazon region, levels of exposure to mercury in populations living in mining activity areas are extremely variable. A recent study from the Tapajos basin (Eastern Amazonia) indicates that H-Hg varied widely according to fish consumption habits (Dorea, de Souza, Rodrigues, Ferrari, & Barbosa, 2005). In that article, a review of 22 studies performed in the Amazon basin on adult populations was presented: 12 studies displayed average H-Hg greater than 10 μg/g. Those observations denote incommensurably higher mercury exposure than what is reported here.

Frequency of fish servings was associated with higher mercury burden. While it was not possible to quantify the amounts of contaminated fish eaten, this risk factor holds independent predictive power in regression analysis. This is in line with other Amazonian studies (Dorea, Barbosa, Ferrari, & de Souza, 2003; Harada et al., 2001; Santos et al., 2002; Santos et al., 2000).

This risk factor points to the existence of a substantial methylation process in the flood plain of the Beni River (Maurice-Bourgoin & Quiroga, 2002). An interesting issue concerns the elimination of an ethnicity effect when other variables were taken into account. Ethnicity may work as a social, economic, and ecological indicator but did not carry here any explanator power per se. The nature of fish species eaten is also of importance. In the present study, more carnivorous and piscivorous fish were eaten in remote communities accessible only by canoe than in easy-to-reach villages. Indeed, elevated mercury concentrations were found in piscivorous and omnivorous fish of this area (Maurice-Bourgoin, Quiroga, Chincheros, & Courau, 2000).

In our study, we found a moderate to high correlation between H-HG of mothers and that of their children. This suggests that fish consumption was fairly homogeneous within the household. While no clear relationship between H-Hg and age, nor between adult women and children, was found, a tendency toward an increase with age or length of residence could be expected, as observed in other studies (Santos et al., 2002). More important is the curvilinear relationship between age and H-Hg observed in children less than 36 months old. It attests to a decrease in contamination after birth and a progressive rise during the weaning period when the child begins to eat fish. It also suggests that breastfeeding is not a risk factor for Hg

contamination and should be promoted in light of its positive effects on child development (Dorea & Donangelo, 2006).

Prevention of mercury contamination is perplexing, since population contamination is mainly from fish consumption, and fish is of foremost nutritional importance in these Amazonian areas. This question was addressed by Dorea in the context of the nutritional ecology of native Amazonians (Dorea, 2003; Dorea, 2004). That author analyzed in detail the two major neurotoxins present in the diet of Amazonian populations: cyanogenic glucosides, from cassava (Manihot esculenta), and methylmercury from fish. He weighed the neurotoxins against the nutritional advantages conferred by intakes of high quality proteins and lipids, abundant energy and carbohydrates, and natural antitoxins such as selenium and sulfur amino acids. He concluded that despite elevated toxin levels, no evidence of harmful effects on health was found. Indeed, no reports existed of clinically detectable neurological abnormalities in that region. Nevertheless, in a study which included fishermen and their families from the Tapajos (Brazil), neurological observations were compatible with mild Minamata disease (Harada et al., 2001).

A hair concentration of 50 μg/g would mean a 5% increase in the risk of paresthesia (Dufour, 1992). Fortunately, values reported here were below that limit (the maximum observed was 34.1 μg/g in a 10-year-old boy). As a rule, studies performed in the Tapajos included subjects with higher H-Hg than that observed in the present study. In some cases, however, alterations were observed in motor and visual function at H-Hg content below 50 μg/g (Grandjean, White, Nielsen, Cleary, & de Oliveira Santos, 1999; Lebel et al., 1998). Those studies suggest that functional abnormalities could occur in certain subjects at levels below the proposed 50 μg/g threshold.

These findings should be considered in the light of health problems prevalent in the area. We found that 42% of mothers presented mild anemia and 41% of children had growth retardation attributable to chronic malnutrition (Benefice, Monroy, Jimenez, & Lopez, 2006). These conditions could also be responsible for impairment in cognitive and motor development of children (Chang, Walker, Grantham-McGregor, & Powell, 2002; Lozoff & Georgieff, 2006). The possibility that responses to neuropsychological tests are altered by tropical or nutritional diseases was evoked in other Amazonian studies and should not be discarded (Dorea, 2003; Grandjean, White, Nielsen, Cleary, & de Oliveira Santos, 1999). More studies are needed in these remote and vulnerable communities of the Upper Amazon to determine whether tropical and nutritional diseases carry out a cumulative effect with environmental contaminations on child development.

Our study presents some limitations. It was a cross-sectional study. It was limited to a particular area and therefore does not necessarily reflect the overall situation of the Beni River. Also, the scattering of populations along the river banks did not enable a selection of large samples. One of the strengths of this study, however, is the fact that it compared two different, although closely related, ethnic groups with contrasting exposures. It also should be mentioned that this is the first study which focuses on a public health perspective in Bolivia.

Preventive measures might be based on changes in fish consumption, or, even more importantly, on the nature of the fish consumed at certain periods by vulnerable populations. In view of the high nutritional value and antitoxin characteristics of fish, total suppression of its consumption does not appear reasonable (Clarkson & Strain, 2003; Dorea, 2003; Dorea, 2004). Recommendations should be made with caution and should be primarily focused on pregnant women. Otherwise, their applicability in lowland Bolivia is fraught with complications.

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Table 1 Hair mercury concentrations (µg/g) in Amerindian groups of the Beni River (lowland Bolivia)

Group	Median (IC $_{95\%}$)	Geometric mean (IC $_{95\%}$)	Mean (SD)	Minimum	Maximum
Overall sample (n=556)	4.0 (3.6 ~4.4)	3.7 (3.4 ~4.0)	5.3 (4.3)	0.08	34.1
Children (n=393)	3.9 (3.4 ~4.4)	3.6 (3.3 ~3.9)	5.2 (4.4)	0.08	34.1
Mothers (n=163)	4.4 (3.5 ~5.4)	3.9 (3.3 ~4.5)	5.5 (4.1)	0.15	20.0
Pregnant (n=18) [1]	3.3 (1.3 ~3.9)	2.4 (1.5 ~3.8)	3.2 (2.1)	0.2	7.8
Lactating (n=57)	5.5 (4.4 ~6.4)	4.8 (3.9 ~5.9)	6.2 (4.1)	0.5	18.3
Non-pregnant- non-lactating women (n=93)	4.1 (3.0 ~5.4)	3.7 (3.0 ~4.5)	5.4 (4.1)	0.15	20.0

[1]: The total number of mothers (n=163) was lower than the sum of different maternal conditions (n=168), since we did not include in the groups of mothers the 5 pregnant adolescents who were still living with their mothers.

Table 2: Result of a logistic regression between high and low H-Hg and families characteristics (Forward selection with 2 interaction terms)

	Regression coefficient	Standard error	Wald z-value	p	Odds ratio	Lower 95% IC	Upper 95% IC
Intercept	-2.69	0.39	-6.88	0.00			
Accessibility: road	4.11	0.55	7.40	0.00	60.70	20.46	180.08
Fish consumption (< 1 serving per day)	1.84	0.79	2.32	0.02	6.31	1.33	29.83
Economic activity (no fishing)	-0.03	0.50	-0.06	0.95	0.96	0.36	2.59
Accessibility * fish consumption	-2.34	1.06	-2.20	0.03	0.10	0.01	0.77
Fish consumption* Subsistence activity	2.64	1.28	2.07	0.04	14.03	1.15	171.37

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Figure 1: Cumulative distribution of H-Hg according to ethnicity

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Figure 1

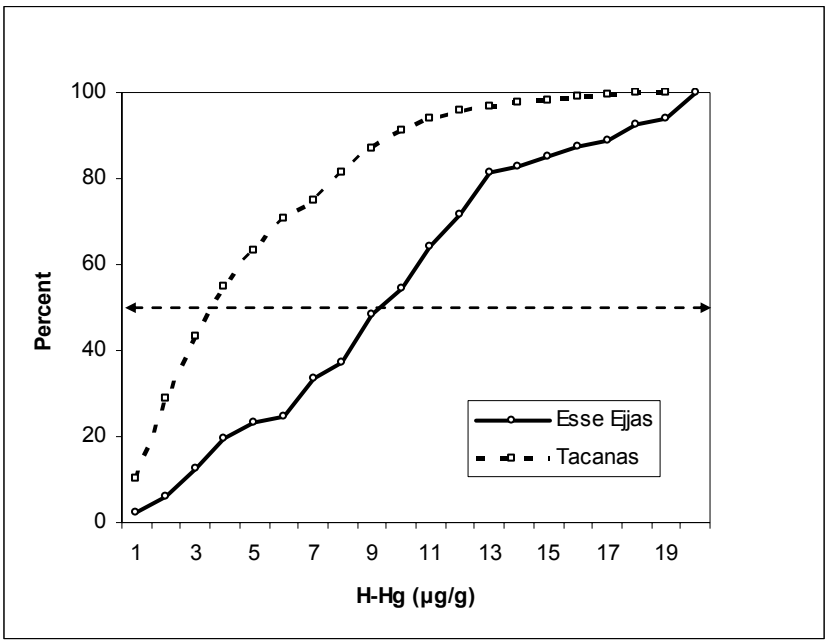


Figure 2

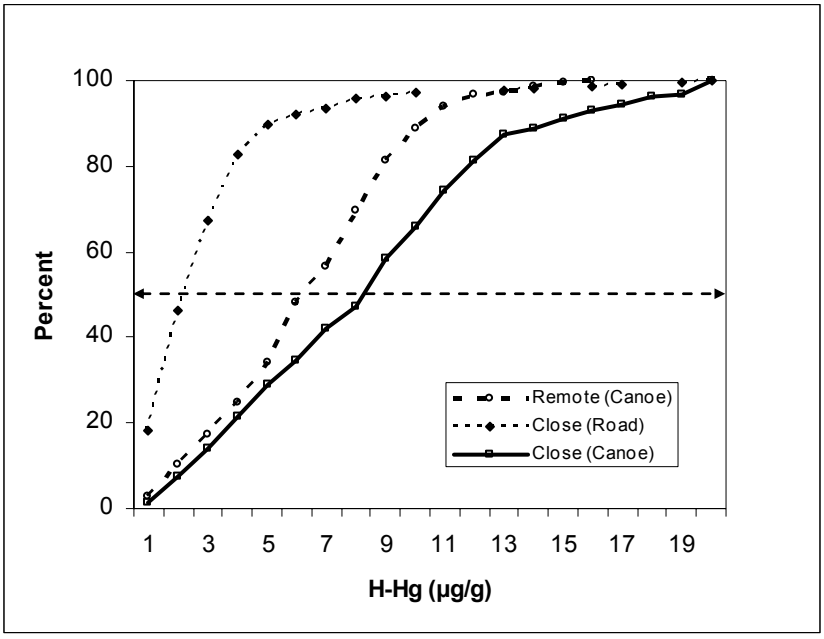

100
80
60
40
20
0
Percent
1
3
5
7
9
11
13
15
17
19
H-Hg (µg/g)
Remote (Canoe)
Close (Road)
Close (Canoe)

Figure 3

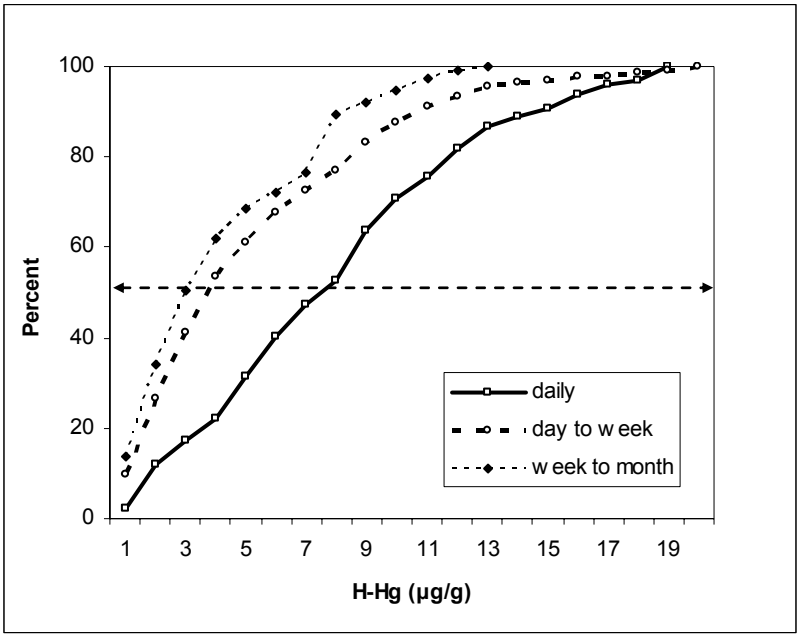
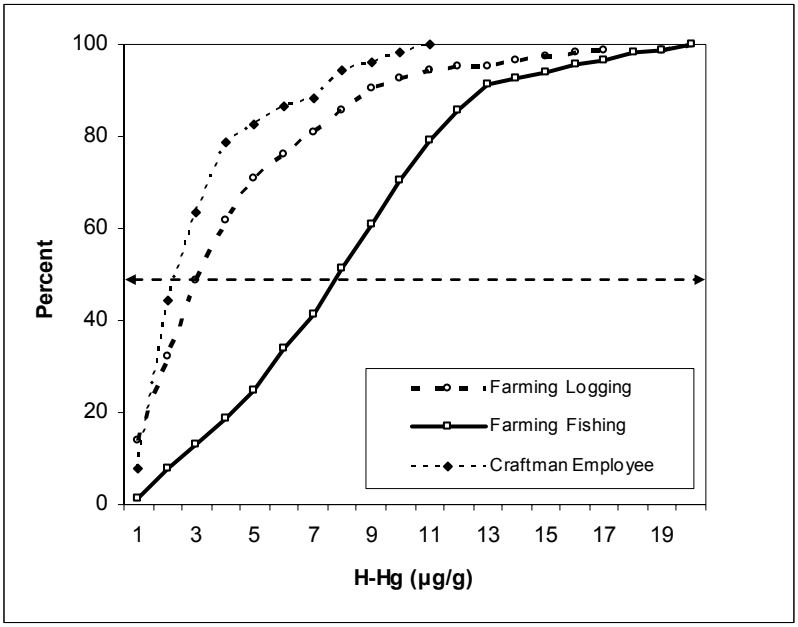

100
80
60
40
20
0
Percent
1
3
5
7
9
11
13
15
17
19
H-Hg (µg/g)
daily
day to w eek
w eek to month

Figure 4


100
80
60
40
20
0
Percent
1
3
5
7
9
11
13
15
17
19
H-Hg (µg/g)
Farming Logging
Farming Fishing
Craftman Employee