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ORIGINAL PAPER

Mercury hair levels and factors that influence exposure for residents of Huancavelica, Peru

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Abstract Between 1564 and 1810, nearly 17,000 metric tons of mercury (Hg) vapor was released to the environment during cinnabar refining in the small town of Huancavelica, Peru. The present study characterizes individual exposure to mercury using total and speciated Hg from residential samples, total Hg in hair, and self-reported questionnaire data regarding factors influencing exposure (e.g., frequency of fish consumption, occupation). Total Hg concentrations in hair from 118 participants ranged from 0.10 to 3.6 μg/g, similar to concentrations found in the USA and lower than concentrations in other Hg-exposed populations around the world. Pearson's correlation coefficients for data in this study suggest that there is a positive correlation between concentrations of total Hg in hair and concentrations of total Hg in adobe bricks, dirt floors, and surface dust; however, these correlations are not statistically significant. Results of a one-way analysis of variance (ANOVA) identified that total Hg concentrations in hair were significantly related to gender ($p < 0.001$), living in a neighborhood where smelters were previously located ($p = 0.021$), smoking status ($p = 0.003$), frequency of house cleaning ($p = 0.019$), and frequency of fish consumption ($p = 0.046$). These results highlight the need for further studies to better characterize Hg

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exposure in Huancavelica, particularly as related to residential contamination. A comprehensive analysis of residential Hg contamination and exposure in Huancavelica will guide the development and implementation of mitigation and remediation strategies in the community to reduce potential health risks from residential Hg exposure.

Keywords Adobe · Hair · Health · Mercury · Soil

Introduction

Huancavelica is located in the Andean highlands of Peru and was the site of large-scale cinnabar (HgS) refining from 1564 to 1810 (Robins 2011; Robins and Hagan 2011). Cinnabar refining ended in the 1970s but mercury (Hg) contamination persists in the community today. A previous study has reported total Hg concentrations in ambient soils up to 1,200 μg/g, among the highest concentrations measured in urban areas around the world (Robins et al. 2012). Residents in Huancavelica built their adobe brick houses from this same contaminated soil and often leave interior walls and floors uncovered and unsealed. Total Hg concentrations in adobe bricks, dirt floors, and surface dust were found to be strongly correlated with one another and ranged from 8.00 to 1,070 μg/g, 3.06 to 926 μg/g, and 0.02 to 9.69 μg/wipe, respectively (Hagan et al. 2013). Speciation and bioaccessibility analyses of adobe brick and dirt floor samples suggest that most Hg dissolves only in the presence of strong acids (e.g., concentrated nitric and hydrochloric acid). However, a small percentage (<10 %) of Hg may be bioaccessible following ingestion, as indicated by leaching tests with simulated gastric fluids (Hagan et al. 2014). While the percentage of the total Hg in adobe bricks and dirt floors that is bioaccessible is small, concerns remain for potential health impacts following ingestion of Hg-contaminated particles from adobe bricks and dirt floors, particularly for small children (Hagan et al. 2014).

Hair is routinely used as a biomarker for Hg exposure, particularly for methylmercury exposure from dietary consumption of fish and other foods (Díez et al. 2011; Nuttall 2006). Mercury is incorporated into hair follicles during growth and arises from the presence of Hg in blood (Díez et al. 2011). Hair grows at a rate of 0.6–3.36 cm per month, but is often reported as 1 cm per month in the literature for convenience (Nuttall 2006). Although whole blood and urine may be better indicators of more recent Hg exposure than hair, there are advantages to using hair specimens. Mercury has a relatively long half-life in hair of 1.5–2 months, which allows for the evaluation of past exposures (Díez et al. 2011; Nuttall 2006). The stability of Hg in hair samples makes their transport and storage easier than blood and urine (Nuttall 2006). Moreover, higher concentrations of Hg accumulate in hair compared with blood and urine, making measurement less complicated (Nuttall 2006).

The purpose of this study was to evaluate individual exposures to Hg in Huancavelica, Peru. The objectives of this study were to: (1) quantify total Hg in hair samples from 118 participants in Huancavelica and compare the concentrations with other Hg-exposed populations, and (2) identify factors that influence exposure as represented by total Hg concentrations in hair, including residential Hg contamination and environmental factors. The results of this study will be used to prioritize future research efforts in Huancavelica for evaluating and reducing exposure.

Methods

Residential samples

Samples of adobe brick, dirt floor, surface dust, and vapor were collected from 60 residences in four distinct neighborhoods (Ascención, San Cristóbal, Santa Ana, and Yananaco) in August 2010, as described by Hagan et al. (2013). Verbal consent for participation was obtained from an adult resident at each household and was recorded in the field log. Institutional review board (IRB) approval for the study was obtained through Duke University.

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Residential samples were analyzed for total Hg concentrations according to EPA Method 1631 (US EPA 2002) as described in Hagan et al. (2013). Briefly, adobe brick, dirt floor, and surface dust wipe sample were hot-block extracted in 4:1 HCl:HNO_3 at 85 °C for 5 h. Following digestion, samples were diluted in 1 % bromine monochloride and analyzed for total Hg concentrations using a Brooks Rand MERX-T (Brooks Rand Labs, LLC, Seattle, WA). Adobe brick and dirt floor samples underwent additional analyses for Hg speciation (Bloom et al. 2003) and bioaccessibility (Schaider et al. 2007) as described in Hagan et al. (2014). Hg speciation was determined by performing sequential selective extractions on adobe brick and dirt floor samples, which uses five extractant solutions that separate Hg compounds based on chemical behavior (Bloom et al. 2003; Hagan et al. 2014). Bioaccessibility extractions were performed on adobe brick and dirt floor samples using a simulated gastric fluid comprised of 0.4 M glycine and adjusted to a pH of 1.5 with HCl (Hagan et al. 2014; Schaider et al. 2007).

Hair samples and questionnaire data

Hair samples and self-reported questionnaire data were collected from 118 adult participants in five distinct neighborhoods (Ascención, San Cristóbal, Santa Ana, Santa Barbara, Yananaco) in July 2012. Ascención, San Cristóbal, and Yananaco are neighborhoods with historical cinnabar smelting, while Santa Ana and Santa Barbara did not have historical cinnabar smelting activity. Written consent for participation was obtained from an adult resident; IRB approval for the study was obtained through Duke University and the Ethics Committee at the National University of Huancavelica.

Hair samples were collected from the occipital region of the scalp by isolating a 0.75–1 cm diameter bundle of hair, cutting the hair as close to the scalp as possible, then securing the scalp end of the hair in a folded piece of paper with a paper clip prior to labeling with the sample identification number (UNEP 2008). Hair within 1–2 cm of the scalp region of the sample (mass of 0.01–0.02 g) was analyzed for total Hg content by thermal decomposition using a RA-915 + Lumex Mercury Vapor Analyzer equipped with a RP-M324 attachment (Ohio Lumex Co., Twinsburg, OH) according to EPA Method 30B (US EPA 2008).

Health assessment questionnaires were adapted from the United Nations Environment Programme (UNEP 2008). Information was collected through a face-to-face interview with an adult resident at the same time as collection of the hair sample. Information was collected related to occupational exposure, diet, confounders, and health effects (e.g., sleep disturbances, physical and mental fatigue, and well-being). Questions were also included related to hair washing, product use, and hair treatments such as use of hair dye.

Statistical analyses

Data for concentrations of Hg in residential samples and in hair samples were tested for normality prior to statistical analysis, and both were found to be log-normally distributed. Relationships between total Hg concentrations in hair samples and for total, speciated, and bioaccessible Hg concentrations in residential samples were evaluated with Pearson's correlation coefficients. Correlations were calculated for households with paired hair and residential data ($n = 34$). One-way analysis of variance (ANOVA) was performed between total Hg concentrations in hair and responses from questionnaires to identify associations between factors other than residential concentrations that might influence exposure and body burden. One-way ANOVAs were performed on the 118 participants with hair and questionnaire data, although sample size varied if a participant did not respond to a particular question on the questionnaire. The values represented in the boxplots are the first quartile, median, and third quartile. The whiskers are the minimum and the maximum, and the diamonds represent the arithmetic means. Statistical analyses were completed using SAS 9.2 software (SAS Institute Inc., Cary, NC).

Results

Correlations between Hg in hair and Hg in residential samples

Total Hg concentrations in hair ranged from 0.10 to 3.6 μg/g. Pearson's correlation coefficients were calculated for total Hg concentrations in participants' hair and total Hg concentrations in residential samples for these participants, as shown in Table 1. The associations

Table 1 Pearson's correlation coefficients, p values, and sample sizes for log-transformed mass of total Hg in hair and residential samples

	Adobe bricks	Dirt floors	Surface dust	Indoor air
Hair				
r	0.226	0.332	0.057	−0.089
p	0.198	0.055	0.747	0.617
n	34	34	34	34

between total Hg concentrations in hair and total Hg concentrations in adobe bricks, dirt floors, and surface dust were positive but not statistically significant. Similarly, but not shown in Table 1, Pearson's correlation coefficients were calculated for total Hg concentrations in hair and speciated or bioaccessible fractions of Hg in residential samples. Again, no significant correlations were found, with the exception of the "elemental" fraction of Hg in adobe bricks ($r = 0.493$, $p = 0.023$).

Relationships between Hg in hair and other factors

The relationships between total Hg concentrations in hair and questionnaire responses related to occupational exposure, diet issues, confounders, and health effects were evaluated using one-way ANOVA. Responses that were statistically significantly related

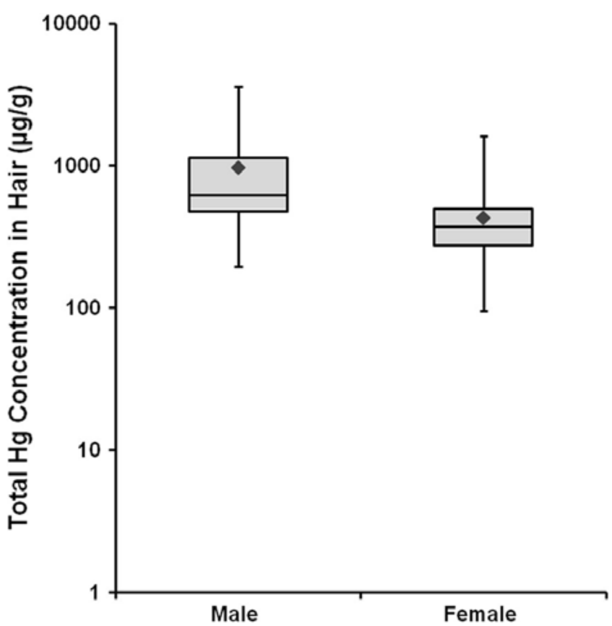

Fig. 1 Total Hg concentrations in hair based on gender (males: $n = 21$, females: $n = 95$)

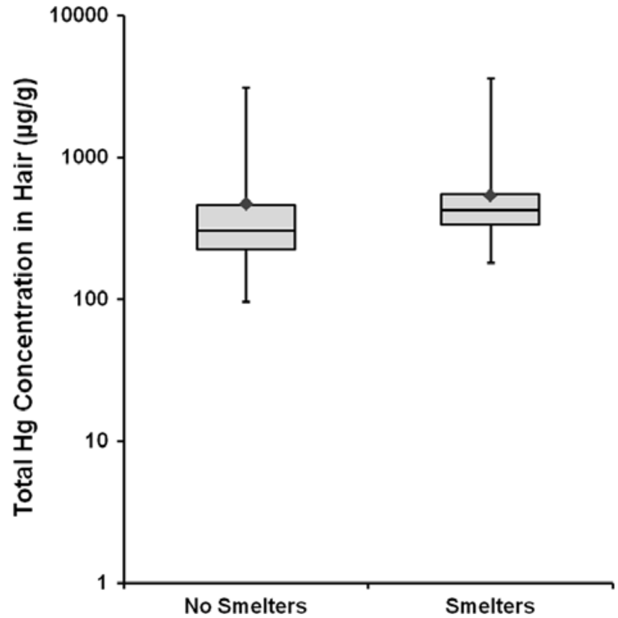

Fig. 2 Total Hg concentrations in hair based on living in a neighborhood with historical cinnabar smelter activity (smelters: $n = 87$, no smelters: $n = 30$)

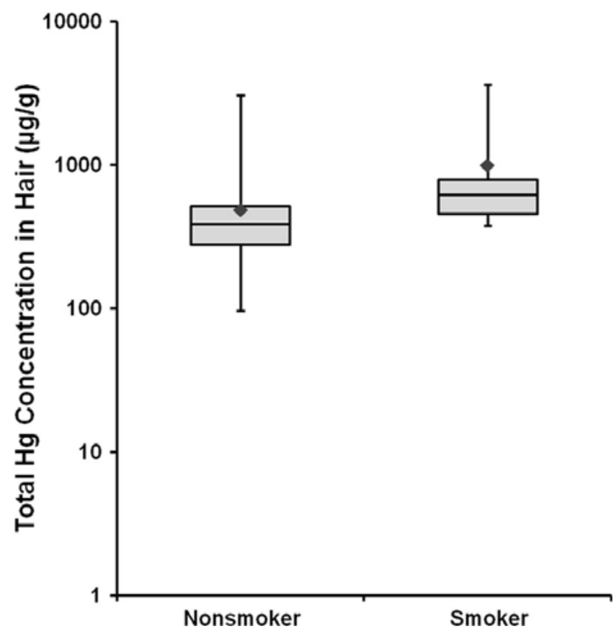

Fig. 3 Total Hg concentrations in hair based on current smoking status (smoker: $n = 9$, nonsmoker: $n = 108$)

to total Hg concentrations in hair included: gender ($p < 0.001$) as shown in Fig. 1; living in a neighborhood where smelters were previously located ($p = 0.021$), Fig. 2; smoking status ($p = 0.003$), Fig. 3; frequency of house cleaning ($p = 0.019$), Fig. 4; and frequency of fish consumption ($p = 0.046$), in Fig. 5.

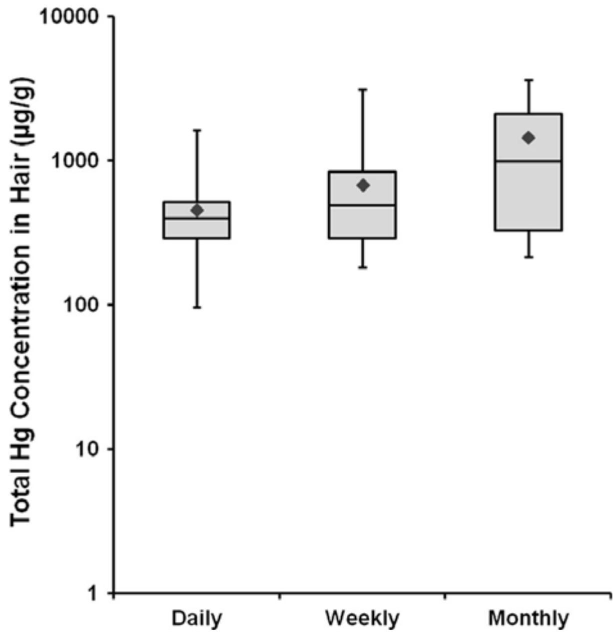

Fig. 4 Total Hg concentrations in hair based on frequency of house cleaning (daily: $n = 86$, weekly: $n = 19$, monthly: $n = 4$)

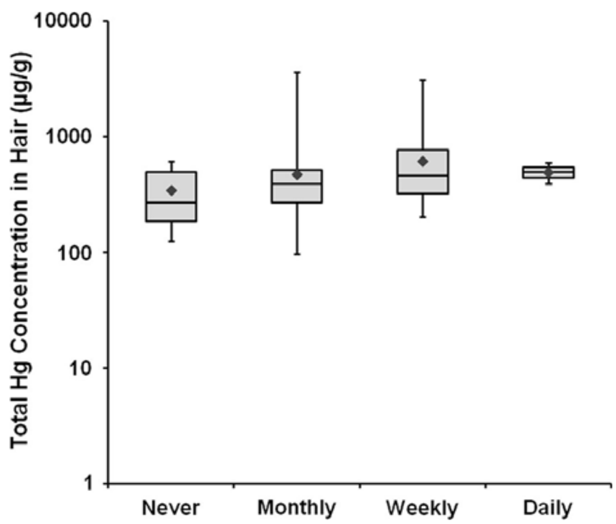

Fig. 5 Total Hg concentrations in hair based on frequency of fish consumption (never: $n = 9$, monthly: $n = 56$, weekly: $n = 50$, daily: $n = 2$)

The relationships between total Hg concentrations in hair and questionnaire responses related to hair product use and chemical processing (i.e., hair dye) were evaluated using one-way ANOVA and were found to not be statistically significant.

Discussion

Relationship between Hg in hair and Hg in residential samples

Total Hg concentrations are relatively high in adobe brick, dirt floor, and surface dust samples in residences in Huancavelica. High residential Hg contamination could be related to high Hg concentrations in hair for the 34 participants who had paired hair and residential data. Of the 34 households with paired data, average adobe brick, dirt floor, surface dust, and elemental Hg vapor concentrations were 222 µg/g, 178 µg/g, 0.878 µg/wipe, and 0.81 µg/m^3, respectively. On the other hand, a previous study found that only a small percentage of the total Hg in residential samples was bioaccessible (Hagan et al. 2014). Pearson's correlation coefficients for data in the present study suggest that, while there is a positive association between concentrations of total Hg in hair and concentrations of total Hg in adobe bricks, dirt floors, and surface dust, these correlations are not statistically significant. A larger sample size might provide greater statistical power to better elucidate any potentially significant correlations.

Relationships between Hg in hair and other factors

The relationship between gender and total Hg concentrations in hair was highly significant ($p < 0.001$) as shown in Fig. 1. Males ($n = 21$) were found to have significantly higher total Hg concentrations in hair than females ($n = 95$). The average age between males and females was nearly identical, at 45 years old and 44 years old, respectively, suggesting gender differences are not being driven by age differences between males and females. This finding may reflect different exposures between males and females. In Huancavelica, males are more likely to be employed directly in mining or in other occupations that would likely increase exposure (e.g., construction projects that would increase exposure to contaminated dirt and dust in the ambient environment). Díez et al. (2011) also found greater Hg concentrations in hair for males and attributed the difference to males consuming larger portions of fish. The present study, however, did not collect specific information related to portion size when consuming fish.

Table 2 Total Hg hair concentrations compared for populations with different exposure situations

Location	Mean (μg/g)	Range (μg/g)	Remarks	Reference
Huancavelica, Peru	0.53	0.10–3.6	Cinnabar mining region	Current study
Almadén, Spain	2.4	0.67–4.8	Cinnabar mining region	Díez et al. (2011)
Castilla-La Mancha, Spain	1.7	0.20–5.6	Cinnabar mining region	Díez et al. (2011)
Diwalwal, Philippines	2.7	0.03–38	Gold mining region	Drasch et al. (2001)
Beni river, Bolivia	3.8	0.42–16	Gold mining region	Barbieri et al. (2009)
Gulf of Gabes, Tunisia	6.5	1.3–14	Frequent fish consumers	Mezghani-Chaari et al. (2011)
Taiji, Japan	2.5	0.4–6.1	Non-fish consumers	Endo and Haraguchi (2010)
United States	0.47	0.35–0.58	NHANES study, 16-49 year old women	McDowell et al. (2004)

The presence of previous cinnabar refining in the neighborhood was significantly related to total Hg concentrations in hair ($p = 0.021$), as shown in Fig. 2. Participants living in a neighborhood that historically had smelters operating (Ascención, San Cristóbal, Yananaco; $n = 87$) had significantly higher total Hg concentrations in hair than those living in a neighborhood that did not historically have cinnabar refining operations (Santa Ana, Santa Barbara; $n = 30$). A previous study found statistically significant differences in total Hg concentrations in adobe bricks, dirt floors, and surface dust by neighborhood (Hagan et al. 2013), which would suggest that higher total Hg concentrations in hair are related to legacy contamination from historic cinnabar refining.

Smoking status was also significantly related to total Hg concentrations in hair ($p = 0.003$), as shown in Fig. 3. Smokers ($n = 9$: six males, three females) had significantly higher total Hg concentrations in hair than nonsmokers ($n = 108$). The reason for the positive relationship between smoking and higher total Hg concentrations in hair is not clear. Perhaps an unknown, physiological reaction occurs as a result of smoking that increases the body burden of Hg. Mucociliary clearance in a smoker's respiratory tract tends to be impaired, thereby allowing more Hg-contaminated particles to reach a smoker's lungs. This result may also be a consequence of the hand-to-mouth activity that occurs when smoking.

The frequency of house cleaning was found to be significantly related to total Hg concentrations in hair ($p = 0.019$), as shown in Fig. 4. Participants who cleaned their houses daily ($n = 86$) had lower total Hg concentrations in hair than participants who cleaned their houses monthly ($n = 4$); there was no significant difference in Hg hair concentrations between those who cleaned their houses weekly ($n = 19$) and those who cleaned more or less frequently. The difference between more and less frequent house cleaning is likely a result of having less Hg-contaminated dust on surfaces within the home, thus reducing potential ingestion from hand-to-mouth activity or from food surfaces.

Hair is typically used as a measurement of exposure to methyl mercury from fish consumption. In Huancavelica, total Hg concentrations in hair were also significantly related to the frequency of fish consumption, as shown in Fig. 5. Residents of Huancavelica consume fish less frequently than other populations, with a majority of participants reporting monthly fish consumption.

Comparison of total Hg concentrations in hair in Huancavelica and elsewhere

Hair has been used as a biological indicator in other studies evaluating varying routes of exposure to Hg, as shown in Table 2. Díez et al. (2011) measured total Hg concentrations in hair in Almadén and Castilla-La Mancha, Spain, for those exposed as a result of historic cinnabar mining and refining. The range of total Hg concentrations for hair in Almadén and Castilla-La Mancha is similar to the range in Huancavelica. However, the mean concentrations in the two Spanish sites are 3 and 4.5 times greater than the mean concentration in Huancavelica. Populations exposed to Hg from gold mining and refining in the Philippines (Drasch et al. 2001) and Bolivia (Barbieri et al. 2009), had hair concentrations orders of magnitude higher than those in Huancavelica. Mezghani-Chaari et al.

(2011) measured total Hg concentrations in hair for frequent fish consumers in Tunisia and found a mean concentration of 6.5 μg/g, over 12 times greater than the mean total Hg concentration in hair for the study participants in Huancavelica. As a comparison, non-fish consumers in Japan (Endo and Haraguchi 2010) had total Hg concentrations in hair nearly five times greater than participants in Huancavelica. In the USA, total Hg concentrations in hair measured as a part of the National Health and Nutrition Examination Survey (NHANES) found a lower range of hair concentrations than in the present study, but the mean concentration of 0.47 μg/g was comparable to the mean concentration in Huancavelica (McDowell et al. 2004).

Limitations of the present study

Although positive associations were found for the relationship between total Hg concentration in participants' hair and Hg concentrations in samples of adobe bricks, dirt floors, and surface dust taken in participants' residences, these relationships were not statistically significant. One reason for the lack of significance for these positive relationships may be that the number of participants with paired hair and residential data was low ($n = 34$).

This study measured total Hg concentrations in hair from adults only. Adults in general have lower hand-to-mouth activity than children and as such would likely be exposed less frequently to Hg-contaminated particles within the home. Children have special health risks due to exposure to Hg that were not be evaluated in the present study.

A control population from an uncontaminated area of Peru with similar fish consumption rates would have provided insight regarding "normal" ranges of Hg concentrations in hair. The lack of a control population is an additional limitation of this study and should be addressed in future biomarker studies in Huancavelica. Moreover, there may be limitations of the sample population in terms of selection method (e.g., biases introduced by volunteer availability and interest).

While hair is routinely used as a biomarker for exposure to methylmercury, other chemical forms of Hg may not be represented adequately in hair concentrations. Inhalation of elemental Hg vapor and ingestion of Hg-contaminated particles might be more accurately characterized using other biomarkers, such as whole blood or urine. Elemental and inorganic forms of Hg are incorporated into hair much less efficiently than methylmercury (Nuttall 2006) so that exposure to these forms is potentially underestimated by using hair samples alone.

Conclusions

Total Hg concentrations were measured in hair samples from 118 participants in Huancavelica, Peru, a site of historic cinnabar refining. Total Hg concentrations in hair averaged 0.53 μg/g (range 0.10 to 3.6 μg/g) similar to those found in the USA but lower than some other Hg-exposed populations around the world. Although a positive relationship was found between total Hg concentrations in hair and Hg-contaminated residential samples, the relationship was not statistically significant. This suggests that Hg exposure to residents, as indicated by total Hg measurements in hair, is relatively low despite elevated Hg in soil and dust in both residential and ambient environments. Total Hg concentrations in hair were significantly related to gender, living in a neighborhood with smelters, smoking status, frequency of house cleaning, and frequency of fish consumption.

Further studies are needed to better characterize Hg exposure in Huancavelica, particularly as related to residential contamination. Whole blood and urine can be used in parallel with hair samples to measure recent exposures to other forms of Hg. Future research efforts may require larger sample sizes and evaluation of children's exposures; these studies would require additional IRB approvals. A more complete understanding of exposure will assist future efforts to reduce individual exposure to Hg contamination in Huancavelica, Peru.

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