TOTAL AND METHYL MERCURY LEVELS IN HUMAN SCALP HAIRS*

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ABSTRACT

The contents of total and methyl mercury in scalp hair samples of 1179 fishermen living at a typical Hg-polluted region in Northeast China and 27 lying-in women and their new born babies in Beijing have been determined by instrumental neutron activation analysis, gas chromatography (electron coupling) and other techniques. Only 18 of all fishermen have the Hg contents above 5 μg/g, which indicates that the Hg pollution there has been substantially alleviated. The longitudinal Hg patterns of the lying-in women show a gradually lowering tendency during pregnant period. Further, the Hg contents of the new-born babies are generally above or close to those of their mothers, confirming the mechanism that the methyl Hg, an organic species of Hg with high toxicity, is readily able to penetrate the placental barrier and accumulated in fetus. Thus, the mercury poison has occurred at the early stage of pregnancy.

Keywords Chemical species of mercury, Total Hg, Methyl Hg, Neutron activation analysis, Gas chromatography, Human hair, Hg pollution

1 INTRODUCTION

Mercury is a toxic element to human being. Its specific toxicity lies in the methylated species, i.e. methylmercury (Me-Hg), which is readily able to enter human body via food chain[1,2] and long-term accumulated in it through various biological membranes. Recent clinical observation has indicated that the Me-Hg has effects on early stage of newborn baby development and mental ability of the children, whose mothers were exposed during pregnancy to 3–4 times the tolerable weekly intake set by WHO and FAO.

Hg pollution is still an environmental problem to be solved in some regions of China[3]. In order to study the level of Hg pollution in the Second Songhuajiang River

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System, a typical Hg-polluted area, we systematically determined the Hg contents in 1179 hair samples taken from fishermen living at the immediate vicinity of the river by NAA (neutron activation analysis) and Gas chromatography (GC) (electron coupling (EC)) in the framework of the IAEA Coordinated Research Project on assessment of environmental exposure to mercury in selected human populations as studied by nuclear and other techniques.

It is known that during prenatal period the methylated species of Hg is easily transferred from mothers to fetus through placental tissue. In order to study the hereditary toxicity of Hg, the correlation between Hg contents in scalp hairs of lying-in women and their new-born babies has been investigated in this work. Also, the variation of Hg contents in pregnant women's hairs during their pregnancy was studied by NAA and synchronous radiation-based XRF (X ray fluorescence) as well.

2 EXPERIMENTAL

2.1 Sampling

The sampling procedure outlined in the United Nations Environment Programme (UNEP) on the determination of methylmercury, total mercury and total selenium in human hair, and the Reference Methods for Marine Pollution Studies No.46 (draft), October 1987, prepared in co-operation with WHO and IAEA[4] are followed.

Head hair samples of 1179 fishermen living at the Second Songhuajiang River System in northeast China and 27 pairs of mothers' and their new-born babies' hairs immediately or at a couple of days after delivery to the Beijing Zhongguanchun Hospital were collected, together with the necessary information, *e.g.* name, age, sex, occupation and nutritional habits. All hair samples were taken from the occipital area and as close as possible to the scalp. At least 10 cm long and about 10 g hairs were cut from the lying-in women without waving or dying in the latest 10 months. Due to a little hair available, all hairs of the babies were sampled and their weights ranged from 0.5 to 1.0 g. The hair washing procedure recommended by IAEA was followed to remove dirty materials, *i.e.* washing by acetone and 3 times by bidistilled water-acetone. Then, the hair was left to dry in a clean desiccator for use.

2.2 Analysis of total mercury

NAA, mercurymetry (MM) and atomic fluorescence spectrometry (AFS) were used to determine the total mercury in the hairs in this work. Here only the methodology of NAA is briefly described.

Hg has two stable nuclides, ^{196}Hg and ^{203}Hg. Both of them are able to be readily determined by NAA. We count the two peaks of 68.8 and 77.3 keV from ^{197}Hg produced by ^{196}Hg (n,γ) in a heavy water reactor with neutron flux of 4×10^{13}–5×10^{13} n/cm^2·s for 30 min irradiation. The counting was done after 3 d cooling of hair samples by using a planar high-pure Ge detector with a better energy resolution in low energy region than normal HPGe detectors.

Due to the high volatility of Hg, the volatile loss of Hg during sample preparation and irradiation process must be first determined. Our experimental results showed no

significant loss of Hg at 150℃ drying for 3 h, but considerable loss of Hg above 150℃, particularly for hair samples. The less loss of Hg for the chemical standard of Hg is attributed to the addition of thioacetamide as a stabilizer of Hg. Within 1 h irradiation time the volatile loss of Hg can be neglected. However, the extension to over 2 h will cause severe loss of Hg. Thus, the irradiation time of hairs is kept within 30 min. The detailed results refer to Ref.[5].

2.3 Analysis of methylmercury

Our procedure for Me-Hg analysis of hair is shown in Fig.1. The first step is to prepare the sulphyhydryl cotton as follows: 15 g defatted cotton were rinsed in a mixing solution of 50 ml thioglycollic acid, 35 ml acetic oxide, 16 ml 36% acetic acid, 0.15 ml concd. H_2SO_4 and 5 ml bidistilled water for 4 d at 37–38 ℃. Then, the cotton was washed to pH=7 by bidistilled water, squeezed and dried at 38℃ in an oven. The final cotton was stored in a desiccator for use.

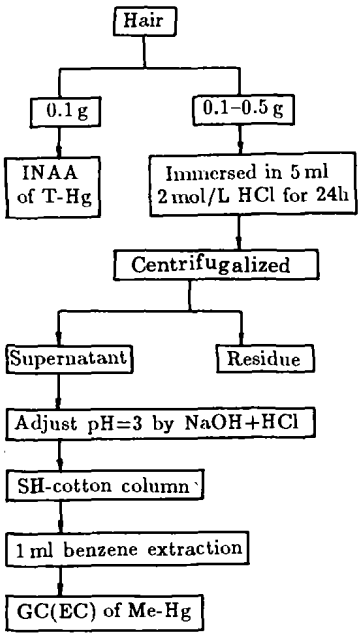

Fig.1 Experimental procedure for Me–Hg analysis of hair sample

Two sets of gas chromatography devices are available. One is Model Varian Vista 6000. The other is Model Shimadzu GC-9A with a C-92A data processor. The chromatographic column is 10% PEG-20M (ϕ3.2 mm×1.1 m). The 60–80 mesh Chromosorb W is used as carrier. The detector is ^{63}Ni of 370 MBq (10 mCi).[6]

2.4 Synchronous radiation-based XRF (SRXRF)

In this work it is also attempted to use SRXRF to study longitudinal variation of Hg contents along hairs in comparison with sectional NAA. A Beijing Electron-Positron Collider (BEPC) is available with 2.8 GeV energy and 150 mA maximum currency intensity. A two-dimensional scanning unit with a size-adjustable slit from 10μm×10μm to 1 mm×1 mm was used. The preliminary experiments indicate that the 9.987 keV L_αX line of Hg can be applied.

3 RESULTS AND DISCUSSION

3.1 In-house analytical quality control

Five working hair samples were prepared as in-house quality control materials. Further, 2 Chinese human hair reference materials (GBW 09101 and GBW 07601) were used to examine the reliability of NAA, MM and AFS for total Hg analysis. Table 1 lists experimental results and the comparison of 3 analytical methods is satisfactory.

Table 2 lists the results of INAA for 2 Chinese hair CRMs and other two NIST biological CRMs, citrus leaves SRM-1572 and pine needle SRM-1575. It can be seen that the INAA technique used in this work is reliable for sub-mg/kg to mg/kg Hg analysis.

Table 1
In-house analytical quality control of total Hg μg/g

No.	INAA	MM	AFS
1	0.53±0.05	0.58±0.02	0.68±0.03
2	1.14±0.03	0.99±0.03	1.08±0.03
3	0.43±0.01	0.44±0.01	0.43±0.02
4	0.48±0.02	0.52±0.03	0.51±0.03
5	0.48±0.03	0.51±0.01	0.52±0.01

Table 2
Examination of analytical accuracy of total Hg by INAA μg/g

Sample	This work	Certified
GBW09101	2.18±0.03	2.16
GBW07601	0.38±0.05	0.36±0.05
SRM1572	0.070	0.080
SRM1575	0.20	0.15±0.05

3.2 External analytical quality control

Because the biological CRM for Me-Hg is not available in China, we participate in the external analytical quality control project organized by IAEA and sent 7 hair samples to the "Josef Stefan Institute" at Ljubljana, Slovenia, pointed as the reference laboratory of the CRP. Table 3 lists the results from the inter-laboratory comparison. The data in Table 3 state that the agreement for total Hg is excellent, but for Me-Hg is good for two samples (A,B), fair for D and E, and poor for C and F.

Table 3
External analytical quality control of total and methyl Hg μg/g

Sample	Total Hg		Me-Hg	
No.	Beijing	Ljubljana	Beijing	Ljubljana
A	0.53±0.05	0.57±0.02	0.43±0.04	0.44±0.03
B	1.14±0.03	1.09±0.03	0.89±0.06	0.85±0.07
C	0.43±0.01	0.40±0.01	0.42±0.04	0.27±0.02
D	0.48±0.02	0.49±0.04	0.42±0.03	0.34±0.03
E	0.48±0.03	0.46±0.03	0.42±0.03	0.33±0.02
F	2.18±0.03	1.92±0.25	1.54±0.04	0.90±0.04
G	0.38±0.05	0.38±0.01	–	0.16±0.01

3.3 Hg distribution of hair samples of 1179 fishermen

The T-Hg and Me-Hg contents in 1179 hair samples taken from the human population living at the Second Songhuajiang River System were determined by INAA, MM, AFS and GC(EC). Our results indicate that most of the hair samples contain low Hg, less than 5μg/g. Only 18 samples have high Hg contents (see Table 4). The No.5 sample contains the highest Hg content, up to 113 μg/g. Thus, the Hg pollution is still present, even though it has been substantially alleviated in comparison with the 1970s.

3.4 Correlation of hair Hg contents of lying-in women with those of their new-born babies

The hair Hg contents of 27 lying-in women and their new borns determined by INAA are listed in Table 5. Fig.2 shows the correlation between them. It can be seen that the hair Hg contents of 22 babies are close to or above those of their respective mothers. Only 5 babies have a slightly lower Hg contents. The statistical treatment states that the average Hg value for all babies is 0.66±0.31 μg/g, while 0.59±0.25μg/g

for mothers. In addition, there is no significant difference between them ($P > 0.05$). Lauwerys *et al*[7] reported higher Hg contents of new borns blood (umbilical cord) than those of mother blood (intravenous) by 10%–15% in a non-Hg-polluted area in Belgium. The similar results were also given by Pitkin *et al.*[8]. Thus, our results agree with those of blood samples. At Hg-polluted districts, the hair Hg contents of babies are likely even high.

Table 4

Total and methyl Hg level of 18 fishermen with higher Hg contents μg/g

Sample No.	T-Hg NAA	T-Hg MM	T-Hg AFS	Me-Hg(as Hg) GC(EC)	Sample No.	T-Hg NAA	T-Hg MM	T-Hg AFS	Me-Hg(as Hg) GC(EC)
1	9.90±0.30	11.9	–	7.92	10	16.7±0.2	15.6	14.9	13.7
2	9.82±0.30	10.7	–	8.68	11	14.5±0.2	16.8	–	1.90
3	11.3±0.3	12.0	–	6.96	12	8.45±0.34	13.0	–	4.64
4	9.70±0.36	12.0	–	7.22	13	16.2±0.1	15.0	–	7.64
5	113±10	121	101	60.9	14	8.64±0.34	10.1	9.9	3.96
6	10.2±0.2	11.3	10.9	5.35	15	13.2±0.1	13.3	–	13.2
7	7.86±0.34	10.0	–	6.84	16	9.87±0.20	12.7	–	4.57
8	9.11±0.27	10.4	–	6.10	17	34.6±0.1	54.1	35.5	–
9	10.5±0.3	11.6	–	1.50	18	13.0±0.1	20.4	15.5	–

Table 5

Hg contents of 27 lying-in women and their new borns by INAA μg/g

Sample	1	2	3	4	5	6	7	8	9	10	11	12	13	14
Mother	0.83	0.52	0.55	0.42	0.36	0.30	0.92	0.67	0.46	1.01	0.79	0.61	0.76	1.12
Baby	1.16	0.76	0.66	0.59	0.40	0.39	1.35	0.65	0.45	1.10	0.66	0.82	1.23	0.98
Sample	15	16	17	18	19	20	21	22	23	24	25	26	27	$\bar{X}$±SD
Mother	0.60	0.40	0.47	0.29	0.95	0.22	0.44	0.41	0.71	0.23	0.80	0.72	0.28	0.59±0.25
Baby	0.46	0.48	0.47	0.42	1.07	0.33	0.43	0.30	0.82	0.29	0.64	0.73	0.30	0.66±0.31

3.5 Longitudinal variations of Hg contents in hairs of pregnant women

It is known that the growing rate of human head hair is about 300–400 μm/d, *i.e.* 1 cm per month. Thus, a strand of 10 cm long hairs can cover the whole pregnant time. INAA was used to analyze the Hg contents of lying-in women's hairs of each 1 cm, while SRXRF for scanning analysis of Hg along the hairs with a step length of 5 mm was made.

Table 6

Variation of Hg contents of lying-in women during pregnancy μg/g

Sample No.	Month (pregnant) 1	2	3	4	5	6	7	8	9	10
1	1.17	1.16	1.03	0.90	0.84	0.86	0.61	0.59	0.63	0.55
2	0.75	0.64	0.62	0.54	0.49	0.40	0.47	0.47	0.42	0.38
3	1.48	1.45	1.03	0.92	0.88	0.75	0.61	0.63	0.69	0.74

Table 6 lists the longitudinal variations of hair Hg contents of 3 representative lying-in women obtained by INAA. Our results show that the hair Hg contents of the lying-in women decrease with increase in pregnant time. The regression analysis of 3 specimens indicates a significant correlation between them ($r_1 = -0.966$, $r_2 = -0.900$ and $r_3 =$

−0.871, $P < 0.001$). (see Fig.3). The SRXRF gave the similar tendency, even though its quantitative calculation was difficult.

Our results confirm the mechanism that Hg is gradually transferred from mothers to their fetus during pregnant period. The animal test claims that the Hg species plays

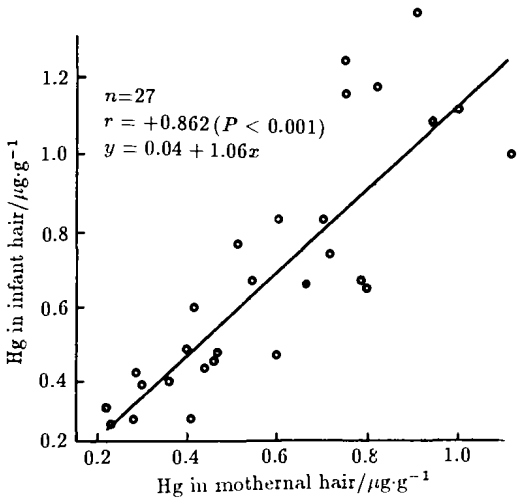

Fig.2 Correlation between hair Hg contents of lying-in women and their new borns

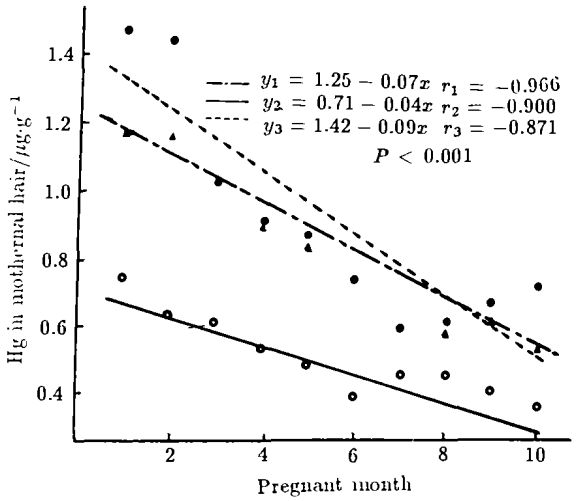

Fig.3 Variations of hair Hg content of pregnant women with pregnant time

great role in its transfer. The inorganic Hg is basically unlikely to go through placental tissue, while the methyl Hg, an ester-loving material, readily penetrates it to enter fetus. It is likely that the fetus is easily able to absorb ester-like materials from placenta[10]. As the early stage of pregnancy is an important period in which infants develop their cerebrum and nervous system, Hg pollution is more dangerous to fetus than to mothers.

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