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DETERMINATION OF TOTAL MERCURY AND METHYLMERCURY IN HUMAN HEAD HAIR BY RADIOCHEMICAL METHODS OF ANALYSIS

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1. INTRODUCTION

In the first period of the project, efforts were carried out in order to detect population groups in Brazil that could be at risk with respect to mercury contamination, mainly by ingestion of contaminated fish.

These efforts resulted in the localization of two possible regions of interest:

1. Region of Billings Dam, which is localized at one of the most heavily industrialized parts of the country, where there is suspicion of pollution by chlor-alkali and other industries. People living in several parts of this Dam consume frequently fish caught by themselves, without much control from public health authorities. The population group of interest was determined by CETESB, which is the State Company responsible for environmental control in Sao Paulo.

2. Xingu Park, located in the Amazonic region, where several Indian tribes live and where the gold exploration activities have risen much concern due to the use of mercury in the process. Tons of mercury are thrown in the rivers of the region annually.

The study of the Indian populations in this region is being conducted in collaboration with physicians of the School of Medicine of Sao Paulo, who for many years have been working with the local inhabitants, taking care of their health. It is important to note that the Indians consume fish daily, caught in the rivers of the Park.

Besides these two groups, also a control group was analyzed, of people with no suspicion of contamination by mercury, such as friends, colleagues and students from the University of Sao Paulo.

2. EXPERIMENTAL

2.1. COLLECTION AND WASHING OF HAIR SAMPLES

Hair samples were collected from the following groups up to now:

1. Control group, consisting of 25 people, in total. Of these, 16 were females, with ages between 21 to 74 and 9 were males, with ages between 20 and 39.

2. Group of people living near the Billings Dam, in a small village called "Santa Cruz", of which 28 samples were collected up to now. In this group, 17 were females with ages between 17 to 57, and 11 were males, with ages from 18 to 65.

3. Group of Indians living in the Xingu Park, of which 14 were females, with ages between 21 to 69 and 13 were males, with ages between 24 to 59.

All the samples were collected and washed according to the procedure recommended by the IAEA [1].

2.2. ANALYSIS OF TOTAL MERCURY IN HAIR SAMPLES AND REFERENCE MATERIALS

2.2.1. Irradiation and measurements

About 100 to 200 mg of the prepared hair samples and reference materials Fish Flesh Homogenate, MA-A-2/TM (IAEA) and Chinese Human Hair, SHINR-HH, were weighed in polyethylene envelopes.

Irradiations were carried out for a period of one hour, in a pneumatic station, under a thermal neutron flux of about 10^{12} $n \cdot cm^{-2} \cdot s^{-1}$.

The standards were prepared by pippeting about 1 mg of mercury, in the nitrate form, onto sheets of Whatman No. 40 filter paper, previously impregnated with a solution

of thioacetamide, to prevent mercury losses by volatilization before and during irradiation, as recommended by Noguchi et al. [2].

After a decay period of about 70 hours, samples, reference materials and mercury standards were measured in a GMX 20195 ORTEC Ge detector, with a resolution of 1.9 keV in the 1332 keV peak of ^{60}Co. The detector is coupled to an ADCAM 918A Multichannel Buffer and associated electronics.

Spectrum analysis was performed by means of the VISPECT2 software, developed by D. Piccot, from Saclay, France.

For calculation of mercury concentrations, the 77 keV peak of ^{197}Hg ($t_{1/2}$ = 64.1 h) was used. Experiments were carried out with Hg tracer to check for self absorption, which was not encountered for hair samples up to about 200 mg.

2.3. ANALYSIS OF METHYL MERCURY IN HAIR OF INDIANS FROM THE XINGU PARK

Half of the amounts prepared of the 27 hair samples that were collected from the group of Indians from the Xingu Park were sent to the Department of Nuclear Chemistry of the Institute "Josef Stefan", from Ljubljana, Slovenija, for analysis of methylmercury.

Methylmercury was determined through the kind offering of Dr. Milena Horvat.

3. RESULTS

3.1. ANALYSIS OF REFERENCE MATERIALS

Two different reference materials were analyzed to check the analytical procedure employed: Fish Flesh Homogenate, MA-A-2/TM (IAEA) (Hg = 0.47 ± 0.02 ppm) and Chinese Human Hair RM, SHINR-HH (Hg = 2.16 ± 0.21 ppm). A minimum of six determinations were carried out for each RM and the results obtained were: 5.7% relative error and 9.4% relative standard deviation for MA-A-2/TM and 1.8% relative error and 6.9% relative standard deviation for SHINR-HH.

Table I presents a summary of the results obtained for the mercury contents in hair of the three population groups studied: control group, group of Indians living in the Xingu Park and group of people living near the Billings Dam in the State of Sao Paulo.

In the case of the group of Indians studied, also the contents of methylmercury in hair are presented.

TABLE I. SUMMARY OF THE RESULTS OBTAINED FOR MERCURY CONTENTS IN THE HAIR OF THE POPULATION GROUPS STUDIED (IN PPM)

Population Group	x	s	Median	x_G	s_G	Range
Controls	1.06	0.55	0.96	0.93	1.71	0.20 - 2.5
Indians from Xingu Park	18.5[1]	5.9	18.0	17.6	1.38	6.9 - 34
	17.5[2]	4.6	17.7	16.7	1.38	5.4 - 26.5
	15.6[3]	4.5	15.0	14.9	1.40	4.79 - 25.7
Population from the Billings Dam	0.88	0.68	0.74	0.71	1.85	0.30 - 3.0

x = arithmetic mean, s = standard deviation of the arithmetic mean, x_G = geometric means, s_G = standard deviation of the geometric mean. [1]results obtained at IPEN, [2]results obtained at Ljubljana, [3]results of methylmercury

In Table II, a comparison is made between results obtained for total mercury in hair of control populations in the present work and the ones obtained by other authors, in Brazil and in other countries.

4. DISCUSSION

The results obtained for the reference materials Fish Flesh Homogenate and Chinese Human Hair were in good agreement with literature, showing relative errors below 6%.

The analytical procedure employed, using irradiations of 1 hour at a moderate neutron flux allowed the use of polyethylene for encapsulating samples and standards instead of quartz ampoules, which made the procedure cheaper and quicker. This is very important when population studies are involved, since the number of samples to be analyzed can be quite high.

TABLE II. COMPARISON OF THE RESULTS OBTAINED FOR THE ANALYSIS OF MERCURY IN HAIR, OF A CONTROL POPULATION IN THE PRESENT WORK AND BY OTHER AUTHORS (IN PPM).

Author	Arithmetic Mean	Standard Deviation	Range
Present work	1.06	0.55	0.26 - 2.5
Camara et al. [3]	1.46	3.70	0.12 - 35.2
Carvalho et al. [4]	1.11	0.67	0.30 - 2.36
Mazzilli & Munita [5]	2.02	1.36	0.3 - 5.8

The geometric mean of 0.93 ppm found for mercury concentrations of the control population in the present work can be considered as low if compared with some other countries, (3.8 for Japan, 1.8 for USA and 3.5 for England) and is close to the one found for Pakistan (1.2 ppm), according to Chatt and Katz [6].

The arithmetic mean found for the controls in the present work, of 1.06 ppm, is very close to the one found by Carvalho et al [4] of 1.11 ppm. It is interesting to note that the region of Brazil studied by Carvalho et al. is completely different from Sao Paulo in most aspects. The values found by Camara et al. [5] and Mazzilli and Munita [5] also in the State of Sao Paulo, 1.46 and 2.02 ppm, were higher than the ones obtained by us.

In what regards the two other population groups that are being studied, the concentration of mercury in the hair of the Indians from the Xingu Park was very much higher than in the controls, with arithmetic and geometric means of 18.5 and 17.6 ppm, respectively. This was quite surprising, because the region of the Park is still supposed to be free from mercury contamination arising from the gold extraction activities that are occurring in the Amazonic region. On the other hand, the Indians use several natural products, extracted generally from seeds, to treat their hair, that could constitute sources of mercury.

The methylmercury concentrations in the hair of the Indians were also very high, ranging from 5.36 to 26.5 ppm. The percents of methylmercury as compared to total mercury were from 70 to 100% and in most cases higher than 80%. This means that probably the mercury contamination in the Indians is derived from fish consumption.

Also it should be noted that the consumption of fish by the Indians is generally high

and this could mean that the waters where they are caught have been already contaminated by mercury.

The other population group, that consumes fish caught in the Billings Dam, did not show any abnormality in the content of mercury in their hair, indicating that the group is probably not at risk in this sense.

The case of the Indians deserves further study, since the tribes from Xingu Park have not been previously investigated as regards the content of mercury in hair. Samples from another group of Indians have been already collected and are being analyzed for mercury. This group has fish as its main source of proteins, caught always at the same site.

5. FUTURE EXPERIMENTS

Another group of samples of hair of Indians from the Xingu Park is being collected, from another tribe, and is being analyzed for total mercury.

As regards analysis of methylmercury, it is intended to employ a procedure for separation of inorganic mercury from methylmercury similar to the one developed by May et al. [7]. The separation is performed by ion exchange chromatography in chloride medium, using an anionic resin in which inorganic mercury is adsorbed as a chlorocomplex ($HgCl_4^{-2}$) and MeHg is eluted as MeHgCl. After decomposition of organic mercury by UV irradiation or digestion with HNO_3 /$HClO_4$, Hg is determined by cold vapour AAS. In the case of the present work, it could be determined by NAA.

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REFERENCES

[1] Reference Methods for Marine Pollution Studies, n. 46 (draft), October 1987
[2] NOGUCHI, K., SHIMIZU, M., MORIWAKI, K., Radioisotopes, 29 (1980) 221
[3] CAMARA, V.M., CAMPOS, R.C., PEREZ, M.A., TAMBELINI, A.T., Cadernos de

Saude Publica, Rio de Janeiro, **2**(3) (1986) 349

[4] CARVALHO, F.M., TAVARES, T.M., MENEZES, G.M., Ciencia e Cultura, **37**(12) (1985) 2063

[5] MAZZILLI, B., MUNITA, C.J.S., Ciencia e Cultura, **38**(3) (1985) 522

[6] CHATT, A., KATZ, S.A., VCH Publishers Inc., Hair Analysis Applications in the Biomedical and Environmental Sciences, N.Y., 1988, p.25

[7] MAY, K., STOEPPLER, M., REISINGER, K., Toxicol. Environ. Chem., **13** (1987) 153