ANALYTICAL QUALITY CONTROL IN STUDIES OF ENVIRONMENTAL EXPOSURE TO MERCURY

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Abstract

The work of the laboratory for quality control in this co-ordinated project for the period from November 1993 to June 1994 is presented. The major effort was devoted to assisting in establishing the homogeneity and total methylmercury levels in two new hair reference materials prepared as control materials for the project, numbered 085 (spiked) and 086 (natural level). Results for some hair materials from participants are also given.

1. INTRODUCTION

The general aims of the programme and the role of the Nuclear Chemistry Department, J. Stefan Institute, as a reference laboratory for purposes of ensuring the achievement of the quality assurance programme of this CRP were discussed in our first and second annual reports, and at the previous research co-ordination meetings (RCMs) [1,2].

Most of our activities have been performed in collaboration with the IAEA in Vienna (Dr. S. Stone) or the IAEA Marine Environmental Laboratory in Monaco (Dr. M. Horvat). The topics discussed in this report include co-operative analysis of the new mercury in hair standards, 086 natural level and 085 high level (spiked), prepared by the IAEA, during and after its preparation, analysis of human hair samples received from participants in the programme, and improvements and developments in analytical procedures for mercury analysis.

2. METHODS

In the report period, the method used for hair analysis was basically the technique described by May *et al.* [3], which uses an anion exchange separation of extracted inorganic from organic mercury species, followed by destruction of organic species by UV irradiation, with the usual CV-AAS finish.

About 100 mg of hair was shaken with 10 ml 6 M HCl for 24 h in the dark and centrifuged. Protected from the light, the sediment was washed twice, re-centrifuged and the washings combined with the centrifugate, which was then passed down a Cl^- form Dowex-1 anion exchange column to absorb inorganic Hg^{++}. The presence of Hg^{++} in the eluate was tested for by reduction with $SnCl_2$ and CV-AAS; none was found. The eluate was then subjected to 24 h irradiation from a UV lamp to decompose MeHg to Hg(II), and Hg(II) determined by CV-AAS.

Total mercury in hair was determined by destruction of up to 100 mg of hair with 2 ml conc. HNO_3 in a sealed tube by heating in a block for several hours (or preferably overnight) at 90°C, followed by CV-AAS determination [4].

3. RESULTS AND DISCUSSION

3.1. Analysis of IAEA Hair reference materials 086 and 085

The analytical control function of our laboratory was heavily engaged in the report period in assisting in the establishment of total mercury and methylmercury levels during the preparation of two new human hair reference materials for the CRP, prepared at IAEA Seibersdorf by spiking a natural, low-level mercury content human hair batch from India (Sample 086) with methylmercury to form a high level standard (Sample 085).

The preliminary results for methylmercury after spiking and initial homogenization are shown in Table I and revealed satisfactory homogeneity, with a MeHg level of about 22 μg/g.

Results for total and MeHg in three bottles each of two subsamples (*i.e.* 6 bottles in all) of the homogenized non-spiked hair 086 are shown in Table II. Agreement for both analytes between the bottles and between the two batches was excellent. The total mercury level is about 0.60 $\mu g \cdot g^{-1}$, of which, about 47% (0.28 $\mu g \cdot g^{-1}$) is present as MeHg. Quality control analyses using BCR-A human hair and the Canadian CRC certified reference material TORT-1 (Lobster haematopancreas) gave excellent results.

The spiked hair sample 085 was further homogenized and three sub-batches H-46, H-47 and H-48 bottled. Three bottles of each sub-batch were then analyzed in our laboratory for total Hg and MeHg as shown in Table III. The results indicate that the homogeneity both within and between the batches is excellent, and the results were also in very good agreement with the preliminary results after spiking (Table I). The quality control analyses were also in excellent agreement with the certified values.

After the final stage of preparation of the two new hair reference materials 085 and 086, which involved radiation sterilization and remixing, followed by bottling, a final control of the as-bottled samples was performed as shown in Table IV. As for the spiked material (085), the results are virtually identical to those obtained in Table III, while for the unspiked base material 086, the results are in very close agreement with those obtained before, as shown in Table II.

Thus, on the basis of these extensive analyses, both materials appear to be very homogeneous and stable samples, which should enable participants to test their methods and carry out routine control analyses, even if unfortunately towards the end of the programme, rather than nearer the beginning.

Further analyses of total mercury by radiochemical neutron activation using the volatilization technique [5] are in progress to check the results reported here by CV-AAS.

3.1.1. Note on moisture content of 085 and 086

Some evidence was obtained from periodic determination of moisture that there is a tendency for moisture to increase and some variation between samples 085 and 086. Therefore, we suggest as a precaution, that moisture content be determined on a separate sub-aliquot on every occasion that the hair materials are used for quality control, or at least if the interval between the last determination of moisture is more than one month.

3.2. Results for hair samples from participating laboratories

Two batches of hair were sent from Brazil (Dr. M. Vasconcellos) for analysis, being collected from Indians in the Amazon basin (for a fuller description of the samples and an interpretation of the results, see M. Vasconcellos *et al.* these Proceedings). As shown in Table V and VI, both groups are high and the first group in particular has a very high and constant percentage of MeHg (89 ± 6 %).

We regret that these samples were the only hair material we have received in the report period, and that participating laboratories still seem reluctant to make use of the opportunity to have their results checked by independent analysis. Perhaps, this is mainly due to the absence so far in the programme, of a readily available hair reference material for laboratories to use to establish their own quality assurance. Nevertheless, just because of the absence of such materials, quality control by comparative analysis with the reference laboratory is more desirable.

3.3. Analytical developments

In co-operation with the group of N. Bloom, two pulications on the speciation of merucry using the ethylation-GC-CVAFS techniques have appeared [6,7], allowing sensitive and simultaneous determination of inorganic and organic species.

3.4. Supplementary programme

Due to the continuing unrest and war situation in Croatia, our programme on pregnant women has been unable to progress further. Recently, in co-operation with Swedish scientists, we showed that there was no detectable in-vivo methylation of mercury in chloralkali workers exposed to merucry vapour [8]. Previous work on this topic had produced conflicting or insufficient evidence.

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TABLE I. PRELIMINARY RESULTS FOR MeHg IN FOUR BOTTLES OF IAEA SPIKED HAIR AS $\mu gHg \cdot g^{-1}$ (dry weight)[a], DETERMINED NOV. 1993

Sample	Results	Mean ± σ	n
A	(13.5)[b], 21.8, 22.6, 22.0	22.1 ± 0.4	(3)
B	18.6, 21.6, 22.1, 21.5	21.0 ± 1.6	(4)
C	22.5, 21.6, 22.0, 21.7	22.0 ± 0.4	(4)
D	24.1, 24.6, 21.6, 22.1	23.1 ± 1.5	(4)
	overall mean	22.0 ± 1.3	(15)

[a]corrected for moisture factor f_{H2O} (freshweight/dry weight) of 1.1086
[b]rejected on statistical grounds since outside overall mean of all 16 results minus 3σ (21.5 ± 2.48 (16): 3σ = 7.44)

TABLE II RESULTS FOR TOTAL Hg AND MeHg (as ng $Hg \cdot g^{-1}$, dry weight) IN THREE BOTTLES EACH OF TWO SUBSAMPLES OF UNSPIKED HAIR, CODE 086, DETERMINED IN MARCH 1994

Sample	Total Hg results	MeHg results	f H_2O
IAEA 086/1			
1/1	619.5 , 585.2	254.9 , 292.2	1.1215
1/2	591.3 , 583.1	273.6 , 251.8	
2/1	563.8 , 620.2	317.9 , 260.5	1.1201
2/2	605.3 , 622.9	295.7 , 283.6	
3/1	571.6 , 632.9	257.1 , 292.2	1.1203
3/2	608.0 , 588.4	290.7 , 303.4	
mean	599.4 ± 22.0	281.1 ± 21.3	
IAEA 086/2			
1/1	619.2 , 597.1	281.0 , 287.3	1.1271
1/2	623.7 , 639.2	271.3 , 277.5	
2/1	634.7 , 622.3	274.9 , 249.4	1.1324
2/2	579.0 , 589.9	291.5 , 262.4	
3/1	610.8 , 619.8	288.0 , 276.0	1.1324
3/2	590.1 , 608.8	266.7 , 272.5	
mean	605.0 ± 21.4	278.0 ± 17.2	
BCR-397 Human Hair	12.1, 11.6, 12.1, 13.3* mean: 12.3 ± 0.7 (4)* certified: 12.3 ± 0.5*		
CRC TORT-1		121.9, 122.2, 127.1 mean: 124 ± 2.9 (3) certified: 128 ± 14	

*units here are $\mu g\ Hg \cdot kg^{-1}$

TABLE III. RESULTS FOR TOTAL Hg AND MeHg IN IAEA HAIR 085, THREE BOTTLES EACH OF THREE BATCHES AFTER HOMOGENIZATION, IN $\mu g \cdot g^{-1}$ (dry weight), DETERMINED IN FEBRUARY 1994

Sample	total Hg, $\mu g \cdot g^{-1}$ (dry weight)		MeHg, $\mu g \cdot g^{-1}$ (dry weight)	
H-46				
1/1	24.6 , 24.6	25.6	21.1 , 21.1	21.4
1/2	26.8 , 26.3		21.0 , 22.2	
2/1	26.3 , 25.9	26.4	23.3 , 23.3	21.7
2/2	26.9 , 26.6		19.8 , 20.4	
3/1	25.7 , 26.4	26.2	21.7 , 22.9	21.1
3/2	27.0 , 25.7		19.7 , 20.1	
H-47				
1/1	26.2 , 25.9	25.5	21.8 , 22.0	22.0
1/2	24.3 , 25.5		21.6 , 22.4	
2/1	25.6 , 26.8	25.9	23.8 , 24.2	22.9
2/2	25.5 , 25.6		22.1 , 21.6	
3/1	26.1 , 26.3	26.1	22.6 , 22.0	22.5
3/2	26.1 , 25.8		21.7 , 23.5	
H-48				
1/1	25.7 , 23.5	25.3	21.2 , 21.4	21.7
1/2	27.2 , 24.6		21.6 , 22.6	
2/1	25.3 , 25.1	25.5	22.0 , 22.2	22.0
2/2	27.3 , 24.4		21.8 , 21.8	
3/1	25.3 , 25.1	26.0	21.8 , 21.0	22.0
3/2	25.7 , 28.0		23.7 , 21.6	
	Grand mean:	25.8 ± 0.4 (9)	Grand mean:	21.9 ± 0.5 (9)
Quality control				
BCR-397 Human Hair	12.5, 11.3, 12.3, 12.7 12.2 ± 0.6 (4) certified: 12.3 ± 0.5		0.631, 0.756, 0.742 0.71 ± 0.07 (3) not certified	
CRC TORT-1	0.338 ± 0.004 (3) certified: 0.330		0.126, 0.130, 0.128, 0.120 0.126 ± 0.004 (4) certified: 0.128 ± 0.014	

TABLE IV. FINAL RESULTS FOR TOTAL AND MeHg IN IAEA 085 SPIKED and 086 UNSPIKED HAIR SAMPLES, IN $\mu g \cdot g^{-1}$ (dry weight) AND $ng \cdot g^{-1}$ (dry weight), RESPECTIVELY, DETERMINED IN MAY 1994, AFTER RADIATION STERILIZATION AND FINAL BOTTLING

Sample	total Hg	MeHg
085 (No. 10)[a]		
1	24.9 , 25.6	22.3 , 21.7
2	25.0 , 25.9	23.3 , 23.1
3	26.1 , 26.4	21.8 , 22.2
4	24.5 , 26.7	22.1 , 23.6
5	26.0 , 25.6	21.4 , 21.7
	mean: 25.7 ± 0.7	22.3 ± 0.8
086 (No. 105)[b]		
1	564 , 636	290 , 282
2	628 , 608	272 , 282
3	596 , 589	277 , 290
4	637 , 593	276 , 289
5	597 , 597	262 , 293
	mean: 607 ± 20	mean: 282 ± 10
Quality control:		
BCR-397	12.4, 11.2, 12.0,	TORT-1 131.3, 124.6,
Human Hair	12.5, 12.4	126.3, 130.3
	mean: 12.1 ± 0.5 (5)	mean: 128 ± 14
	Certified: 12.3 ± 0.5	Certified: 128 ± 14

[a]moisture factor : f_{H_2O} = 1.1143
[b]moisture factor : f_{H_2O} = 1.1349

TABLE V. TOTAL Hg AND MeHg IN HAIR SAMPLES FROM BRAZIL IN $\mu g \cdot g^{-1}$ (dry weight). SAMPLE IDENTIFICATION CODES REFER TO THOSE OF BRAZILIAN PROGRAMME.

Sample	Total Hg	MeHg	% Me / Hg
482	21.5	10.0	47
940	12.1	7.97	66
944	10.9	6.67	61
972	11.8	8.41	71
2092	18.9	8.28	44
5028	13.9	6.98	50
5098	22.4	16.4	73
5121	10.4	8.29	80
5219	11.1	6.89	62
5225	10.1	7.83	78
5226	16.6	11.0	66
5279	11.3	7.56	67
5317	17.9	15.5	87
5354	19.1	12.4	65
5355	11.7	8.89	76
5361	29.5	14.2	48
5428	13.2	10.0	76
6031	15.4	12.7	82
6032	10.1	8.48	84
6033	15.8	10.1	64
6097	11.7	8.99	77
6099	7.70	5.94	77
6106	16.9	11.1	66
cocoyea	23.5	16.8	71

TABLE VI TOTAL Hg AND MeHg IN BRAZILIAN HAIR SAMPLES IN $\mu g \cdot g^{-1}$ (dry weight). SAMPLE IDENTIFICATION CODES REFER TO THOSE OF BRAZILIAN PROGRAMME.

Sample No.	Total Hg	MeHg	% Methyl
1225	18.3	16.3	89
1226	15.3	13.1	86
1228	19.2	16.7	87
1230	13.1	12.4	95
1234	21.3	18.4	86
1241	17.7	14.9	84
1242	16.7	13.7	82
1244	18.1	15.0	83
1245	15.9	14.4	91
1247	14.2	14.2	100
1250	14.7	13.4	91
1251	11.5	9.47	82
1253	20.1	18.5	92
1255	26.5	23.3	88
1269	11.3	10.1	89
1274	20.6	18.0	87
1277	5.36	4.79	90
1278	19.1	16.3	85
1280	15.5	14.2	92
1281	17.7	15.4	87
1285	19.9	18.4	92
1286	20.6	20.6	100
1293	14.7	10.2	70
1324	26.4	25.7	97
1341	16.4	15.4	94
1652	24.1	22.7	95