UPTAKE OF MERCURY BY PLANTS AND ITS DISTRIBUTION IN LIVING ORGANISMS IN AN ENVIRONMENT WITH INCREASED CONCENTRATION OF THIS ELEMENT

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Abstract

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The extent of biological transformation of inorganic mercury was measured on fish at a typical inorganic source (mine and distillation plant in Idrija, Yugoslavia) using a newly developed isolation method for methyl mercury. Results are tabulated on the uptake levels and distribution of mercury in the organs of plants, animals and humans. For the latter beard has been shown to be an advantageous indicator of the exposure.

BIOLOGICAL TRANSFORMATION OF INORGANIC MERCURY INTO METHYL MERCURY IN NATURE

Recent evidence [1-3] indicates that the occurrence of methyl mercury in fish might be the result of natural metabolic transformations and not primarily a consequence of the extensive use of pesticides or pollutants on a methyl mercury basis.

It was therefore considered of interest to study the possible occurrence of methyl mercury in an environment in which its presence at the source of contamination can be practically excluded.

Earlier results from the Idrija area, which were based on the distribution of total mercury only, have been extended to include in addition methyl mercury determinations.

A very selective isolation method has been devised based on the reaction of methyl mercury cyanide vapour with either cysteine or glutathione followed by its extraction into benzene and subsequent determination by gas chromatography. Tracer studies using labelled methyl mercury have shown good recoveries in the range of 80 - 90%.

The original technique, as described in the IJS Report No. R-599, has been modified to enable considerable improvements to be made with respect to the sensitivity of the method and allow the detection of methyl mercury at the one part per billion level using samples typically between 0.1 and 1.0 g.

Comparative results on fish caught at the source of contamination and at various points downstream in a relatively fast flowing river, the Idrijca, show a rapid decrease in the total mercury contamination but an increase in the methyl to total mercury ratios, which at a distance of about 35 km already approaches 100%, demonstrating that most of the mercury in muscle is in the form of methyl species. Liver, measured at the same time, shows a different pattern of distribution, though again the ratio increases with distance downstream.

The geographical situation and sampling points are shown in Fig. 1. Detailed results for groups of fish collected at the respective points are presented in Table I(a). The Hg content of some river organisms, potential fish food, are shown in Table I(b).

Among possible sampling material from human subjects, we first considered hair, but in view of the uncertainties associated with the relatively long exposure of hair to possible contamination and likely variation of mercury concentration depending on the longitudinal section taken for analysis, we have decided — when surveying the male population (miners) — to take beard as a more suitable material.

Comparative analysis of hair and beard taken from some male persons did not show significant differences. Beard, however, offers obvious practical advantages: taking beard samples does not disturb the daily routine of the subject, the rate of growth is fairly constant and the quantity collected daily (~ 50 mg) is sufficient to allow precise and accurate determinations if applying a sensitive technique such as activation analysis. Its exposure is short and by washing the face prior to shaving the outer contamination can be practically eliminated.

The histograms in Fig. 2 show the elimination of mercury in the beard of two subjects who were occasionally exposed to increased levels of mercury in their working duties. Beard provides a useful instantaneous indicator for disturbances associated with the equilibration of mercury in the body.

More time will be required to select and analyse appropriate groups large enough to give statistical meaning to these observations, but work is now in progress to substantiate these findings, and some of the recent results are shown in Table II.

Investigations related to vegetation have been extended to include new species and to cover, apart from mercury, six other elements (Zn, Cu, Mn, Fe, K and Ca) in plants and in the adjacent soil, in an attempt to find eventual correlation with respect to mercury. Typical analyses showing concentrations of these elements are shown in Table III (a) and (b). Comparative results for mercury are also being obtained for the Podljubelj Area, on the site of the abandoned mercury workings, shown in Table IV (a) and (b).

Additional experimental animals, i.e. rabbits, have been transferred from a noncontaminated area to Idrija, and mercury levels are being regularly checked in blood, as well as the excretion of mercury through urine and faeces. The animals are fed on forage grown and collected in the contaminated area and analysed for mercury. These measurements should provide correlation between intake, uptake and excretion of mercury. A picture of the increase and distribution of mercury as a function of time is evident from Table V.

In Table VI results on the mercury distribution in organs of a deceased retired mercury smelter are presented. Values for the methyl mercury concentration in muscle, lungs, liver and kidney are also included. In blood the determination of methyl mercury was attempted but could not be detected. The man died of heart failure. During his service with the mercury mine and distillation plant in Idrija he suffered in 1956 from acute mercury poisoning. Since his retirement five years ago, he was living in Idrija on a site where the exposure could not be severe.

The value for Hg concentration in the thyroid gland would indicate a special ability of this organ for accumulating mercury, but it must be yet

subject to confirmation by experiments on animals, which in general show a similar pattern of mercury distribution to humans.

Work is still in progress on all points summarized in this report and additional data are being collected in particular for the aquatic organisms in order to find possible sources of methyl mercury in the food chain of fish.

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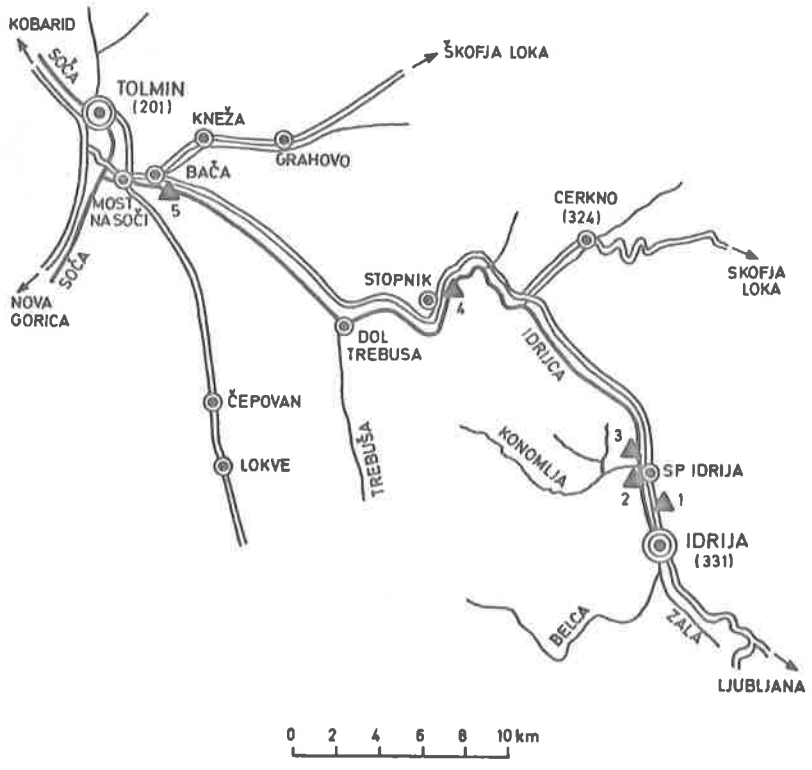

FIG.1. Geographical situation and sampling points (▲).

TABLE I(a). MERCURY CONCENTRATIONS OF FISH MUSCLE AND LIVER FROM THE RIVER IDRIJCA AT SAMPLING POINTS 1, 4 AND 5, FROM APRIL-JUNE 1971 ($\mu g/g$)

Sampling point	No.	Sample (fish) weight (g)	Age (years)	Total Hg in muscle	CH_3Hg^+ in muscle	CH_3Hg^+/Hg_{tot}	Total Hg in liver	CH_3Hg^+ in liver	CH_3Hg^+/Hg_{tot}
1	1	-	-	4.9	0.16	0.03	77.4	0.6	0.01
	2	-	-	2.6	0.52	0.20	28.7	1.9	0.07
	3	-	-	2.7	0.70	0.25	49.2	0.8	0.02
	4	-	-	2.9	0.73	0.25	46.2	1.7	0.04
	5	-	-	6.5	0.29	0.045	118.8	0.6	0.005
4	6	240	4	0.99	0.90	0.91	10.0	1.63	0.16
	7	196	4	0.59	0.43	0.72	16.4	0.93	0.06
	8	270	5	0.66	0.51	0.77	11.9	1.12	0.09
	9	124	-	1.24	1.08	0.87	31.3	2.35	0.08
	10	120	3	1.12	0.92	0.82	15.8	1.84	0.12
	11	120	3	0.87	0.76	0.87	19.2	1.91	0.09
	12	520	4	0.52	0.60		5.13	1.16	0.23
	13	500	4	0.66	0.64	0.97	5.55	0.59	0.11
5	14	210	3	0.39	0.43		2.73	0.44	0.16
	15	275	4	0.30	0.26	0.86	3.41	0.28	0.08
	16	165	3	0.51	0.45	0.88	6.18	1.07	0.17
	17	95	2	0.35	0.33	0.93	2.36	-	-

Sampling point 1: (Marof) at source of inorganic mercury contamination.
Sampling point 4: (Stopnik) 20 km downstream from the source.
Sampling point 5: (Most na Soči) 35 km downstream from the source.

TABLE I(b). MERCURY CONTENT OF ORGANISMS FROM THE RIVER IDRIJCA AT SAMPLING POINTS 4 AND 5 (μg/g fresh wt)

Sampling point	Classification									
	Ephemeroptera		Crustacea	Trichoptera					Diptera	Gastropoda
	larvae	larvae	imago	pupae	pupae in shields	larvae in quivers	larvae	larvae	imago	
4	2.18	1.10	-	1.18	6.65	-	4.56	1.88	0.66	-
	-	1.95	7.34	-	8.86	-	7.24	-	-	-
5	-	-	-	0.40	0.35	3.62	1.15	0.45	-	-
	-	3.07	-	0.75	0.24	-	3.45	0.57	-	11.55

Sampling point 4: (Stopnik) 20 km downstream from the source of inorganic mercury contamination.
Sampling point 5: (Most na Soči) 35 km downstream from the source of inorganic mercury contamination.

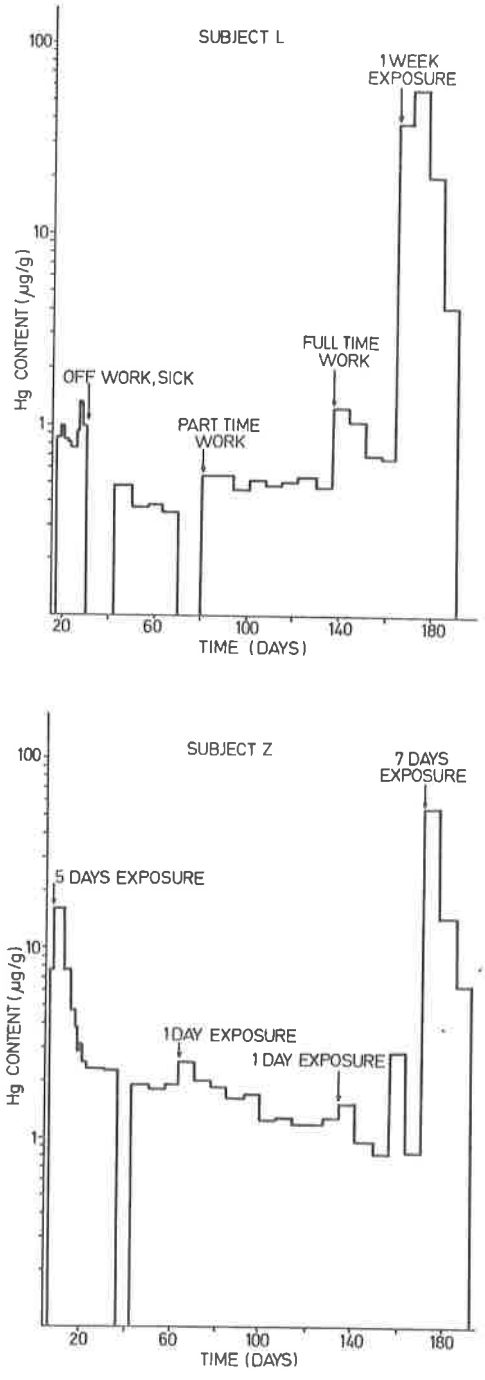

FIG. 2. Elimination of mercury in the beard of two subjects.

TABLE II. MERCURY CONTENT OF HUMAN BLOOD, URINE AND BEARD, FROM APRIL-OCTOBER 1971 (ng/g)

	Date	Worker 1 [a]	Worker 2 [a]	Worker 3 [a]	Student 1 [b]	Student 2 [b]
BLOOD	13 April	99.4	55.7	56.8		
	29 April	77.1	31.0	66.8		
	1 June			37.1	2.2	6.1
	17 June					
	24 September				2.9	2.1
URINE	13 April	1030	489.3	232.8		
	29 April	315.4	111.0	88.3		
	1 June			118.6	3.7	3.2
	17 June		369.8			10.0
	24 September					
BEARD	13 April					
	29 April	4200	4200	2130		
	1 June			2360		
	17 June		505		426.4	1950
	24 September					

[a] Exposed directly to mercury contamination. Worker 2 off work, sick from May to June.
[b] Not exposed, but living at Idrija.

TABLE III(a). ELEMENTAL ANALYSIS OF SOILS AND CARROTS FROM IDRIJA, 24 FEB. 1971 (μg/g)

Sample	Depth (cm)	Mn	Zn	Cu	Fe	Ca	Mg	Na	K	Hg	Weight loss on ashing
5/I	10	675	103	55	39 400	750	2 675	4870	13 500	225	15%
5/II	10	675	100	42	40 000	1375	2 800	5500	16 000	-	-
10/I	15	900	105	142	35 800	7750	11 200	6500	14 000	8.2	12%
10/II	15	825	93	40	35 800	6250	11 000	6100	13 250	-	
Carrots[a]		22	11	3.0	21	2630	2 970	1750	2 900		

Elements were determined by absorption and emission flame spectrophotometry, except mercury.
Sample 5/I and 5/II: Area II 1.5 km distant from Area I, consists of ground where mercury-containing deposits occur on or near the surface.
Sample 10/I and 10/II; 2-3 km away from Area I.
[a] Carrots from Area I.

TABLE III(b). ANALYSIS OF SOIL EXTRACTS FROM IDRIJA, 24 FEB. 1971 ($\mu g/g$)

Sample	Depth (cm)	Mn	Zn	Cu	Fe	Ca	Mg	Na	K	Li	Sr	Hg
1	5	71.2	4.4	2.8	6.0	3550	950	225	200	< 0.01	8.5	52.3
2	10	-	4.8	3.9	44.5	-	-	-	-	-	-	48.9
3	15	131	2.8	2.2	23.0	2550	1000	165	140	< 0.01	4.0	39.5
4	5	84.5	6.8	6.1	9.0	1550	410	140	200	< 0.01	3.6	131
5	10	79.0	3.4	6.3	11.3	1450	385	135	135	< 0.01	3.3	225
6	15	96.0	2.6	5.6	12.7	1450	380	145	150	< 0.01	3.5	153
7	10	106.0	198	36.0	4.0	5400	685	345	425	0.01 - 0.02	9.1	378
8	20	106.0	207	40.0	2.0	5025	760	325	300	0.01 - 0.02	8.6	590
9	15	96.0	32.6	22.0	3.0	3975	615	290	330	< 0.01	7.7	56.2
10	15	108.0	3.4	2.8	30.7	2325	485	175	150	< 0.01	8.0	8.2

Elements were determined by absorption and emission flame spectrophotometry, except mercury.
Samples from 1 to 3 and 9: Area I, near the chimney of the distillation plant, is contaminated by finely divided elemental mercury.
Samples from 4 to 8: Area II, 1.5 km distant from Area I, consists of ground where mercury-containing deposits occur on or near the surface.
Sample 10: 2-3 km away from Area I.

TABLE IV(a). MERCURY CONTENT OF PLANTS AND SOME PLANT ORGANS FROM IDRIJA AND PODLJUBELJ, FROM APRIL - OCTOBER 1971 (μg/g)

Sample No.		Hg conc.
1	Erica carnea – stalk and leaves	2.25
2	Peucedanum oreoselinum – stalk	7.15
	Peucedanum oreoselinum – leaves	1.01
3	Leontodon hispidus – leaves	1.66
4	Silene cucubalus – leaves	6.80
5	Gallium molugo – leaves	4.45
6	Quercus sp. – leaves	12.14
7	Salvia pratensis – leaves	3.91
8	Polygonatum officinale – leaves	1.52
9	Daucus carota – leaves	0.77
10	Lactuca seriola – leaves	0.06
11	Salvia pratensis – leaves	0.25
12	Tusillago farfara – leaves	0.02
13	Taraxacum officinale – leaves	0.07

Samples from 1 to 8: Area I, near the chimney of the distillation plant, contaminated by finely divided elemental mercury.

Samples 9 and 10: Area II, 1.5 km distant, consists of ground where mercury-containing deposits occur on or near the surface.

Samples from 11 to 13: Area III, at Podljubelj, in the Tržič area, similar in character to Area II, being on the site of abandoned mercury workings, but without aerial contamination.

TABLE IV(b). COMPARISON OF MERCURY CONTENT IN DIFFERENT PARTS OF PLANTS FROM IDRIJA AND PODLJUBELJ, FROM APRIL - OCTOBER 1971 ($\mu g/g$)

Sample No.		Hg conc.
1a	Salvia pratensis – a part of leaf	3.91
1b	Salvia pratensis – radial section of stalk with epidermis	0.61
1c	Salvia pratensis – radial section of stalk without epidermis	0.23
2a	Silene cucubalus – leaves	6.80
2b	Silene cucubalus – stalk	0.91
3a	Peucedanum oreoselinum – leaves	7.15
3b	Peucedanum oreoselinum – stalk	0.96
4a	Gallium molugo – leaves	4.45
4b	Gallium molugo – stalk	1.74
5a	Quercus sp. – leaves	12.14
5b	Quercus sp. – stalk	2.33
6a	Daucus carota – leaves	0.77
6b	Daucus carota – longitudinal section of root centre	0.16
6c	Daucus carota – radial sections of root, washed with water	0.28
7a	Lactuca seriola – leaves	0.06
7b	Lactuca seriola – middle of root	0.008
8a	Ostrya carpinifolia – bark of tree	0.45; 0.45
8b	Ostrya carpinifolia – wood	0.023; 0.029

Samples from 1 to 5: Area I, near chimney of the distillation plant contaminated by finely divided elemental mercury.

Samples 6 and 7: Area II, 1.5 km distant, consists of ground where mercury-containing deposits occur on or near the surface.

Samples 8a and 8b: Area III, at Podljubelj, in the Tržič area, similar in character to Area II, being on the site of abandoned mercury workings, but without aerial contamination.

TABLE V. MERCURY CONTENT OF RABBIT ORGANS (ng/g fresh wt)

Sample	Still-born[a] rabbit	Age 1 day[b]	Age 7 months[c] exposure time 9 weeks
Hair	-	-	293.3
Kidney	32.6	48.0 43.7	26 120
Liver	67.0	140.8 91.3	2 120
Meat	44.9	59.3 25.2	-
Brain	9.0	10.7 15.0	-
Spleen	-	-	191.5
Heart	28.9	21.7 29.6	29.0
Blood	-	-	33.6
Faeces	-	-	4 230
Urine	-	-	164.9
Lungs	24.0	28.4 40.9	98.1

[a] Born to 5 months old mother, transferred to contaminated area 2 months before birth.
[b] Two rabbits, born two months later to the same mother as in [a].
[c] Rabbit from an uncontaminated area, transferred to contaminated area.

TABLE VI. TOTAL AND METHYL MERCURY CONCENTRATIONS OF HUMAN ORGANS FROM IDRIJA, SEPTEMBER 1971 (ng/g)

Sample	Total Hg [a]	Hg as CH_3Hg^+
Blood	1.6	
Lungs	127	5.0
Muscle	8.5	4.0
Kidney	5 500	3.6
Liver	108	1.8
Brain	181	-
Cardiac fat	8.1	
Testicle	295	
Adrenal gland	223	
Thyroid gland	64 000	
Spleen	25	0.8
Pancreas	36	

[a] Average values of duplicate results on one cadaver.