MERCURY AND METHYLMERCURY IN SEDIMENTS AND SUSPENDED PARTICLES FROM THE RIVER ELBE, NORTH GERMANY

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ABSTRACT. Total Hg and MeHg concentrations have been determined in sediments and suspended particulate matter from one of the most Hg-polluted rivers of the world, the Elbe river in Northern Germany. Results of total Hg and MeHg are presented. Total Hg concentrations in suspended particulate matter (SPM) are 30 mg kg^{-1} in the average and up to 150 mg kg^{-1} as the maximum value. Total Hg-fluxes are calculated from measurements of an automatically operating station upstream Hamburg as about 17 t a^{-1} in suspended particles. A new MeHg determination by HPLC chromatography and photometric detection was developed and verified. Methylmercury concentrations were up to 10% of the total Hg, i. e. 2.7 mg kg^{-1} (dw).

1. Introduction

The Elbe river, situated in Northern Germany, is one of the most contaminated rivers with regard to Hg for many years (Banat et al. 1972). The river drainage area (Figure 1) is 148,268 km^2 (in Czechoslovakia (CSFR) 51,336 km^2 and in the Federal Republic of Germany (FRG) 96,932 km^2). The average discharge is about 722 $m^3 s^{-1}$.

The main inputs of Hg are mainly from the chlor-alkaline-plants in the former GDR, situated on the tributaries of the Elbe, the Saale and the Mulde. Compared to the natural background of about 0.4 mg Hg kg^{-1} (dw) (Merian, 1984) in sediments and suspended particulate matter (SPM), average concentrations in the range of 30 mg Hg kg^{-1} (dw) and high concentrations of up to 157 mg Hg kg^{-1} (Wilken and Weiler, 1986a) have been found. Under these circumstances some results of the Hg pollution of the Elbe are required.

2. Methods

2.1 COLLECTION AND ANALYSIS FOR TOTAL MERCURY

Samples for a time series (Figure 3) were collected with a precleaned bucket just upstream of the weir in Geesthacht, FRG, (Elbe-km 585.9, about 50 km upstream from Hamburg) and transferred into pre-cleaned polyethylene flasks. The samples were brought directly to the nearby laboratory and the SPM and the water phase was separated by vacuum filtration (< 0.45 µm, Sartorius membrane filter). The SPM was digested for 4 hrs with conc. HNO_3 at 80°C. Hg was measured in all samples by cold vapor atomic absorption spectrometry (CVAAS) according to Hatch and Ott (1968). A Perkin Elmer Model 2380 atomic absorption spectrometer equipped with a deuteri-

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um background corrector and a MHS 20 cold vapor generator was used. The water phase usually contains < 1% of the Hg content of the sample and was therefore not analyzed.

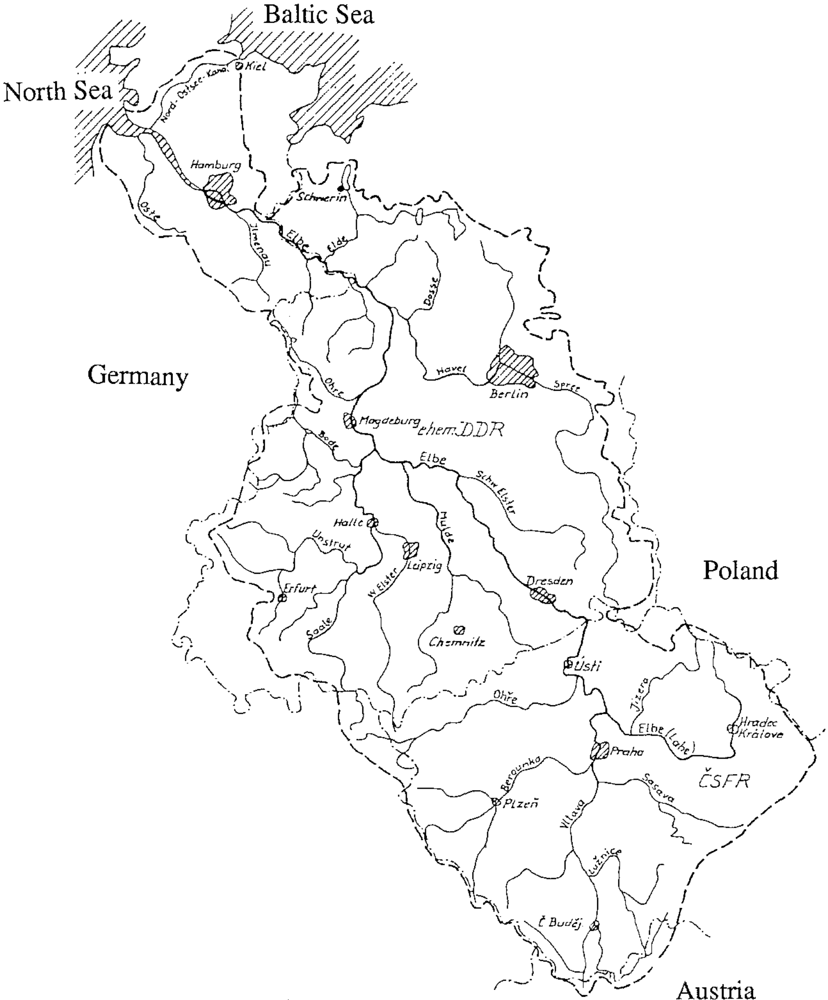

Figure 1. The catchment area of the river Elbe

Samples were also taken in 1984 - 1986 using an automated device in the tidal area of the Elbe upstream Hamburg Harbor. At this measuring station SPM samples appropriate to the cross section of the river for ebb and flood tides were taken. Mixed SPM samples were analyzed weekly for their Hg content. The device and the measuring concept has been described in more detail by Wilken et al. (1986b, 1989).

2.2 COLLECTION AND ANALYSIS FOR METHYLMERCURY SAMPLES

The harbor samples for the MeHg determination were collected during a dredging period from

June to July in 1988 at 0 to 1 m depth. The sample from the groyne field (July 1988) and the river Mulde (March 1990) were collected at the surface (0 to 10 cm) using a PTFE spoon. All samples were stored in acid washed glass bottles at 4°C and further handling was carried out with PTFE tools.

The MeHg determinations were performed with an HPLC procedure with photometric detection. The HPLC method is based on the so called "charge neutralization chromatography" which has been developed for organic Hg compounds (Mac Crehan et al., 1977; Krull et al., 1986; Evans and McKee, 1988). In this method 2-mercaptoethanol is added to the eluent in order to achieve an in situ complexation of the organomercurials on the column during elution. The reaction can be described as follows:

$$RHgX + HS(CH_2)_2OH \Leftrightarrow RHgS(CH_2)_2OH + H^+ + X^- \qquad (1)$$

The resulting mercaptoethanol complex is retained on the reverse phase column and separation and detection is possible. Although the UV-extinction maxima of the organomercurials are below 210 nm (Frimmel and Winkler, 1975a), the detection at 230 nm is also sufficient.

At this wavelength we found a shoulder in the UV-absorption spectrum which is shown in Figure 2.

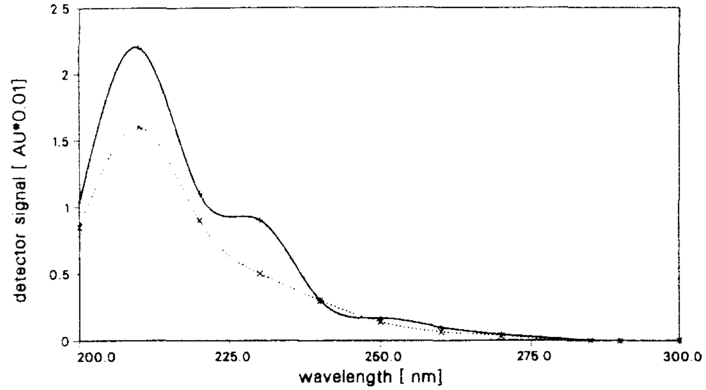

Figure 2: UV-absorption spectrum of CH_3HgCl and C_2H_5HgCl (in 35 % methanol / 65 % water mixture with 6.5 10^{-5} mol L^{-1} $HS(CH_2)_2OH$, -- methylmercury, ... ethylmercury)

This graph was recorded under chromatographic conditions. Two hundred ng of each component were injected and the resulting peak heights were plotted against the wavelength. The shoulder at 230 nm allows measurements with good reproducibility. The standard deviation of nine consecutive injections was 2.9%. The detection limit for MeHg was evaluated to be 0.5 ng absolute (ng Hg as CH_3HgCl) or 5 μg kg^{-1} (dw).

For the determination of MeHg in sediments a suitable strategy for its isolation is needed

(Wilken and Hintelmann, 1990). It must be taken in consideration, that this compound is a strong thiophilic reagent. Many compounds, which contain sulfhydryl (-SH) end groups, are present in sediments. Proteins, in particular, contain this functional group because of their content of cysteine as a component amino acid. These end groups may also be found in humic acids. The actual situation is even more complex, because Hg also has an affinity for other complexing agents, which occur in organic matrices, namely organic acids and the amino group ($-NH_2$). The sediments were therefore acidified before any further treatment. The use of halogenic acids has two advantages: with a high acid concentration the complexing groups such as $-S^-$, $-NH_2$ and $-COO^-$ are protonated and the MeHg cation CH_3Hg^+ is set free; the halogen anion then forms the covalent unpolar MeHg halogenide with this cation, which can be extracted from the acidic solution with toluene.

Finally a back extraction into an aqueous solution is necessary in order to obtain a sample in a solution which is miscible with the eluent of the HPLC. For this purpose cysteine and thiosulphate solutions were tested. But after several injections of cysteine extracts the column was blocked, possibly of crystallization of cysteine. Therefore sodium thiosulphate was the solution of choice for the back extraction. It was compatible with the chromatographic conditions at every concentration tested (10^{-2} to 10^{-4} mol L^{-1}).

For the determination of MeHg, 10 g of the sample were leached with 50 mL of 6 M HCl in a stoppered centrifuge tube and shaken overnight. The suspension was centrifuged at 2000 • g and the supernatant solution was extracted with 100 mL of toluene for 3 min. Occasionally at this point an emulsion was formed and had to be broken with an additional centrifugation. The organic phase was concentrated by means of a rotary evaporator at 50°C and a water jet vacuum. The toluene solution was pipetted in a graduated test-tube and extracted with a 0.1 mmol L^{-1} sodium thiosulphate solution. Centrifugation was sometimes also necessary in order to hasten phase separation. This final solution was injected directly onto the HPLC column. All steps were executed in darkness to avoid decomposition of MeHg by UV-radiation. The method and the chromatographic conditions are summarized in Table I.

Table I. HPLC-procedure for the determination of MeHg

1.) Weigh 10 g wet sediment, add 50 mL 6 M HCl, shake overnight, centrifuge at 2000 g

2.) Extract supernatant solution with 100 mL toluene, reduce to about 5 mL (rotary evaporator, 50 °C)

3.) Extraction with 1 mL 0.1 mM $Na_2S_2O_3$, 0.05 M CH_3COONH_4 (adjust to pH = 5.5 with acetic acid), centrifuge to hasten phase separation

4.) HPLC - chromatography of the aqueous phase. Eluent: 20 % methanol, 1mM CH_3COONH_4, pH=5.5, 0.5 mM 2-mercaptoethanol. Column: LiChrospher RP-18, 150-3 (5µm) glass cartridge

5.) Detection: UV-photometric at 230 nm

The measurements were performed with a Millipore-Waters system equipped with a Model 490 pump, an U6K injector and a 481 UV-detector. Injected volumes were 20 µL. Volatile organics (VO) were determined by loss on ignition (Greiser, 1988).

2.3. QUALITY CONTROL MEASUREMENTS FOR THE METHOD

Additional experiments with $CH_3{}^{203}HgCl$ as a radioactive tracer were carried out to optimize the extraction procedure and to evaluate the overall recovery of the method. Sediment samples were spiked in parallel with a known amount of $CH_3{}^{203}HgCl$ (mercury content: 0.7 µg CH_3HgCl as Hg or 15 kBq ^{203}Hg) prior to the addition of HCl. The samples were then treated as described in the section above. The activity in the remaining solutions was measured after each step of the extraction.

The 279 keV line of ^{203}Hg was measured on a NaI-detector and quantified with a Canberra Model Series 85 Multichannel Analyzer. The $CH_3{}^{203}HgCl$ solution was synthesized from $^{203}HgCl_2$ (Amersham) and tetraethyltin as described elsewhere (Cappon and Smith, 1977; Zamgar and Mushak, 1974).

It was subsequently proven that inorganic Hg^{2+} is not extractable from HCl into toluene (Hg^{2+} could be an artefact from MeHg decomposition). 50 mL of the aqueous and acidic solutions were spiked with $^{203}HgCl_2$ (activity of the basic solution: 103.6 MBq mL^{-1}) and were extracted with 10 mL of toluene in a stoppered centrifuge tube. 5 mL of the organic phase were pipetted into a vial and the activity was measured. The results are listed in Table II:

Table II. Extraction of $^{203}HgCl_2$.

Solution	Initial activity [cps]	Activity of toluene [cps]	Recovery [%]
H_2O	1025 ± 21	192 ± 4	19 ± 1
2 M HCl	1024 ± 20	< 20	< 1
6 M HCl	1091 ± 22	< 20	< 1
Synthesized $Me^{203}Hg$	890 ± 18	828 ± 17	93 ± 3

At pH 7 most of the Hg^{2+} in the solution is present as unpolar $HgCl_2$ which could partly be extracted into toluene; this was already observed for other Hg salts (Schucknecht and König, 1988). But in the presence of HCl no inorganic Hg species could be extracted.

The accuracy of the HPLC-method used for the MeHg determination was evaluated in a course of an intercalibration organized by the Community Bureau of Reference (BCR). The basic solution contained 2.5 mL 10 M HCl in 100 mL (3% NaCl) solution. One mL was diluted in 100 mL gravimatically. 50 mL of this solution was extracted three times with 5 mL of toluene. The combined organic phases were back extracted with an 1 mM solution of $Na_2S_2O_3$, buffered with 0.05 M NH_4CH_3COO. 20 µL of this aqueous solution was injected into the HPLC. The values determined (1005 ± 100 ng CH_3HgCl mL^{-1}) were in good agreement with the overall mean of the participant laboratories.

2.5. QUALITY CONTROL MEASUREMENTS FOR SEDIMENT SAMPLES

The losses of $CH_3{}^{203}HgCl$ during the extraction procedure are shown in Table III. Approximately 7% were still bound to the sediment and could not be removed from the binding sites even with 6 M HCl, and about 13% were not extracted from the acidic solution into toluene. One reason for these losses could be an incomplete leaching and extraction. But on the other hand it seems possible that the decomposition of the radioactive MeHg in inorganic Hg^{2+} occurred. The Hg^{2+}-ions would react immediately with sulphides present in the sediment to form insoluble HgS. Besides, Hg^{2+} forms the tetrachloro complex $HgCl_4{}^{2-}$ (Bartholome et al., 1980) in concentrated HCl which cannot be extracted into toluene.

The concentration step was also a source of CH_3HgCl losses. Because of its volatility it was likely that a small amount of MeHg was evaporated during the concentration in the rotary evaporator. The back extraction with $Na_2S_2O_3$ was almost complete. Losses of about 10% were mainly due to difficulties with the phase separation in this step. The investigated sediments were rich in organic matter and we always observed protein precipitation in the extractions. The overall recovery of the method was approximately 50% for these organic rich sediments.

Table III. Losses of $CH_3{}^{203}HgCl$ in spiked sediments

	Groyne field	Guanofleet Hamburg Harbor	Reiherstieg Hamburg Harbor
weighed sediment [g]	9.0	10.34	10.65
CH_3HgCl-spike	584 ± 13 cps[1)]	592 ± 13 cps	582 ± 13 cps
sediment after leaching	31 ± 2cps 5 ± 1%[2)]	60 ± 2cps 10 ± 1%	42 ± 2cps 7 ± 1%
HCl (after toluene-extraction)	75 ± 2cps 13 ± 1%	77 ± 2cps 13 ± 1%	71 ± 2cps 12 ± 1%
toluene (distillate)	90 ± 2cps 15 ± 1%	90 ± 2cps 15 ± 1%	62 ± 2cps 11 ± 1%
toluene (after thiosulphate extraction)	88 ± 2cps 15 ± 1%	43 ± 1cps 9 ± 1%	28± 1cps 5 ± 1%
final thiosulphate solution	266 ± 5cps 46 ± 2%	293 ± 6cps 49 ± 2%	294 ± 6cps 51 ± 2%

[1)] cps= counts per second for ^{203}Hg [2)] percentages refer to initial activity

3. Results

3.1. TOTAL MERCURY CONCENTRATIONS

Figure 3 shows new results for total Hg concentrations which were measured during a time series. Typical values range between 5 and 100 mg Hg kg^{-1} SPM (dw), as were found also by others (ARGE Elbe, 1988) for many years. The results for SPM from1984 - 1986 are shown in Table IV.

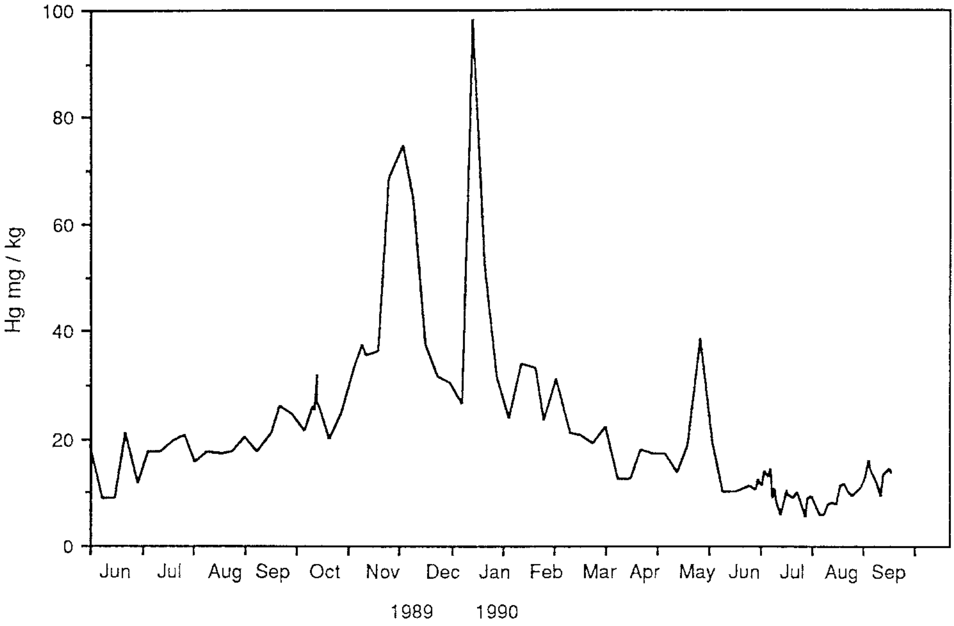

Figure 3. Mercury bound to suspended particulate matter (weekly sampling from June 1989 to September 1990).

In Table V the calculated transported amounts are given. Compared to the rivers Rhine, the Weser and the Ems, the Elbe contributes most of the riverine input of Hg to the North Sea.

Table IV. Results for Hg-concentrations in SPM, 1984 - 1986
(weekly mixed ebb tide samples)

max. value [mg kg^{-1}]	average [mg kg^{-1}]	natural conc. [mg kg^{-1}]	factor of pollution
47	29	≈ 0.4	110

Table V. Transported amounts of Hg, 1987

	1987 t a^{-1}	
Elbe	17.3	(Wilken et al., 1989)
Rhine	9.5	((1986), order of magnitude, IKSR, 1986)
Weser	0.8	(Umweltbundesamt, 1989)
Ems	0.2	(Umweltbundesamt, 1989)

3.2. METHYLMERCURY CONCENTRATIONS

Some typical chromatograms of our determination are shown in Figure 4. Figure 4a illustrates the separation of MeHg chloride, ethylmercury chloride and phenylmercury chloride. In order to get reasonable retention times for phenylmercury this chromatogram was recorded with 35% methanol in the eluent. The other determinations were run with 20% methanol. This content gave the best separation in real environmental samples. Figure 4b shows a MeHg standard in 0.1 mM sodium thiosulphate solution, and it can be seen that the $Na_2S_2O_3$ does not interfere with the MeHg peak.

The result of a sediment measurement is presented in Figure 4c. A sample of the river Mulde (a tributary of the Elbe) was analyzed, the MeHg peak represents 24 ng absolute (10 µL injected volume) and the MeHg content of the sediment was estimated to be 2.7 mg kg^{-1}(dw). Some of our results are listed in Table VI.

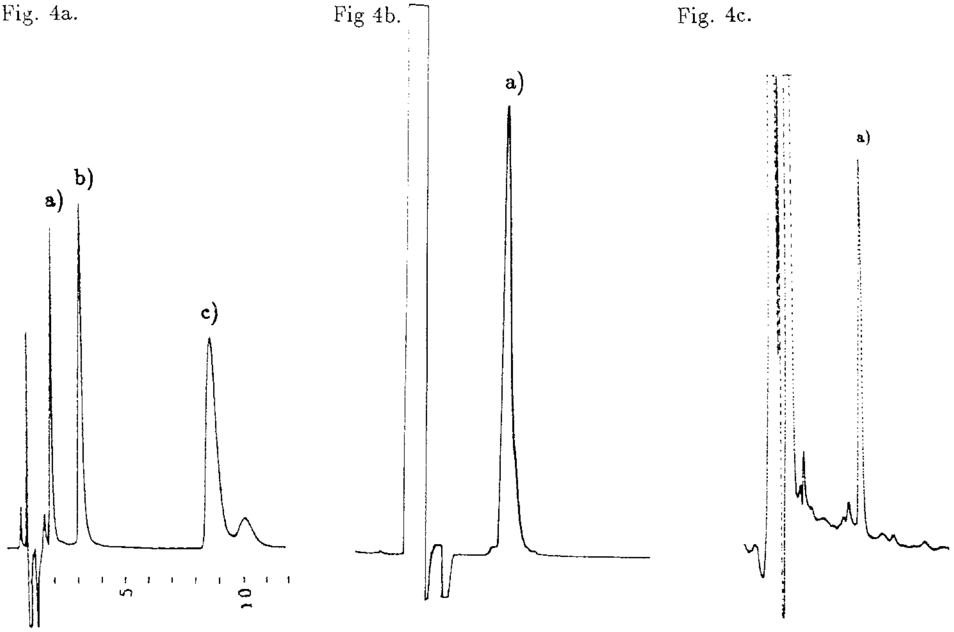
Fig. 4a.

Fig 4b.

Fig. 4c.

Figure 4 . Chromatograms of organomercury determinations:

Figure 4a. Separation of methylmercury (a), ethylmercury (b) and phenylmercury (c, recorded with 35% methanol, 65% water and $6.5*10^{-5}$ mol L^{-1} $HS(CH_2)_2OH$);

Figure 4b. MeHg standard in sodium thiosulphate solution (0.1 mM);

Figure 4c. Sample from the river Mulde (recorded with 20% methanol/ 80% water and 10^{-4} mol L^{-1} $HS(CH_2)_2OH$).

The concentrations of MeHg in the Elbe are extremely high. The concentrations range between 0.05 and 2.7 mg kg^{-1}(dw). These are the highest values reported for sediments to our knowledge thus far. The percentage of MeHg of the total Hg concentrations is very high also. A proportion of up to 10% is unusual in sediments. Some authors estimated that the maximum of 1% (Robertson et al., 1987; Bartlett et al., 1978) or 2% (Frimmel and Winkler, 1975b) of the total Hg occurring in sediments was in the methylated form.

It was obvious that the highest concentrations were found in sediments with a high total Hg concentration. In comparison to the highly contaminated location at the Mulde, the Sandauhafen has the lowest MeHg concentration measured. Another relation might be between the organic matter content of the sediments and the concentration observed [Kudo et al., 1977]. Fifty four % of the Mulde sediment were volatile organics, and the observed MeHg proportion was also the highest measured. However, in order to perform reliable correlation analysis much more data are necessary.

Table VI. Methylmercury contents in the river Elbe

Location	Volatile organics [%]	Σ Hg [mg kg^{-1}]	CH_3-Hg-R [mg kg^{-1}]	% of Σ
Groyne field (Elbe, upstream Hamburg)	15 ± 1	18 ± 0.8	1.02 ± 0.1	5 %
Sandauhafen (Hamburg Harbor)	15 ± 2	2 ± 0.2	0.05 ± 0.004	2 %
Guanofleet (Hamburg Harbor)	18 ± 1	15 ± 0.8	1.22 ± 0.1	8 %
Reiherstieg (Hamburg Harbor)	21 ± 1	59 ± 3.1	1.50 ± 0.1	3 %
Mulde (tributary of the Elbe)	54 ± 3	28 ± 1.9	2.70 ± 0.3	10 %

Background: $\leq$ 0.4 mg Hg kg^{-1}

Usually MeHg is not a result of anthropogenic pollution, but is produced [Callister and Winfrey, 1986; Thayer, 1989] by microorganisms from inorganic Hg. Thus, one explanation for the high values reported could be an uncommon microbial activity in the sediments. Bacteria might be induced to produce volatile Hg species [Wilken et al., 1990] or to generate a high methylation rate. This could be because the methylation of inorganic Hg can be regarded as the detoxification mechanism for microorganisms [Wood, 1974; Robinson and Tuovien, 1984]. Our results are thus a hint that there are some specially adapted bacteria in the Elbe environment which are capable of methylating Hg^{2+} very efficiently.

4. Conclusion

The river Elbe is one of the most Hg polluted rivers in the world. Not only are the total Hg concentrations in sediments and suspended particulate matter very high compared to the natural background but also the MeHg concentrations are very high. An Hg program for the Elbe is therefore needed, to get the Hg pollution under control, which is in existance since big chlor-alkaline-plants-became operative between 1930 and 40.

5. References

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