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Speciation Analysis of Mercury in Natural Water

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Abstract: Separation techniques (extraction, adsorption, electrochemical separation and chromatography) and detection techniques (Spectrophotometry, Atomic Spectrometry, Mass Spectrometry and Electrochemical Analysis) for mercury species in natural water are briefly reviewed in this paper from 22 references. New approaches for trace and ultra-trace mercury speciation analysis in the future should focus on pretreatment system or exploiting a portable instrument so as to realize the species analysis *in situ*. Moreover, it is worthy mentioning that Liquid Chromatography-Hydrate Generation-Atomic Fluorescence Spectrometry (LC-HG-AFS) with low cost and good performance will have a bright prospect for trace and ultra trace mercury species analysis.

Key words: mercury; speciation analysis; natural water; pretreatment

天然水体中汞形态分析

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摘要: 简要归纳了天然水体中汞形态分析的分离(萃取、吸附、电化学分离、色谱)及检测技术(分光光度法、原子光谱法、质谱法、电化学分析法),指出了未来针对痕量超痕量汞形态分析新技术主要集中在样品前处理和便携式在线现场分析,着重推荐高效、经济并具有广阔应用前景的液相色谱 - 氢化物发生原子荧光光谱(LC - HG - AFS)用于痕量超痕量汞形态分析。

关键词: 汞; 形态分析; 天然水体; 前处理
中图分类号: O656.5; O614.243　　**文献标识码**: A

Mercury (Hg) is a highly toxic element and has seriously threatened human health for its cumulative effect in the environment. Mercury and its compounds are listed in third place on the 'Priority List of Hazardous Substances' and one of the thirty 'Precarious Dangerous Pollutants' by The United States Comprehensive Environmental Response, Compensation, and Liability Act (CERCLA) and the European Water

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Framework Directive. The toxicity of mercury strongly depends on its concentration and chemical species. Natural water is a complicated system composed of a water phase, solid phase and biological phase. Mercury exists in many species in these phases. For example, Hg^{2+}, CH_3Hg^+, $CH_3Hg(OH)$, CH_3HgCl and $C_6H_5Hg^+$ are prominent species for the aqueous phase; Hg^0, Hg^{2+}, HgO, HgS, $CH_3Hg(SR)$, $(CH_3Hg)_2S$ for the solid phase and Hg^0, Hg^{2+}, HgO, HgS, $CH_3Hg(SR)$, $(CH_3Hg)_2S$ for the biological phase. Meanwhile, mercury species will convert with ambient conditions. Accurate determination of mercury and its compounds is a great challenge due to its relatively low concentration in natural water. In this paper, the pretreatment and determination techniques for mercury speciation analysis in natural water are reviewed.

1 Separation Techniques

Due to the concentration of mercury in natural water being at trace and ultra trace levels, the separation and enrichment method for mercury speciation analysis is of great importance. The common separation and enrichment methods used for mercury speciation analysis include extraction, adsorption, electrochemical separation and chromatography are summarized in Table 1.

1.1 Extraction

Based on the difference of solubility or distribution coefficient of solutes, this extraction method is widely used in speciation analysis with the advantages of convenience, speediness, safety and easily automated. Liquid-liquid and liquid-solid extraction are usually used for mercury speciation analysis. In order to avoid contamination of the organic solvent used during the extraction process, a new technique free of organic solvent named cloud point extraction (CPE)[8-9] was proposed on the basis of the solubility of surfactant micelle aqueous solution and cloud point phenomenon. Hydrophobic and hydrophilic substances were successfully separated by changing experimental parameters to generate phase separation. For example, Chen et al[8] used CPE to separation Hg^{2+} and $MeHg^+$ in different water samples with recoveries for both between 95% -112%.

Solid phase extraction (SPE) has been developed based on the liquid-solid extraction and combined with the liquid chromatographic column technique to separate different compounds. Selective absorption and elution are often employed by SPE[1-2] to enrich, separate and purify the target component from samples and have the advantages of safety, convenience, high efficiency and using less organic agent.

Table 1 Selected application of different methods for the determination of Hg species in natural water

Matrix	Hg species	Pre-treatment	Detection method	Detection limit/ ($ng \cdot L^{-1}$)	Reference
Sea water	Hg^{2+} $MeHg^+$ $EtHg^+$	SPE-HPLC	ICP-MS	Hg^{2+}: 3 $MeHg^+$: 3 $EtHg^+$: 3	[1]
River water	Hg^{2+} $MeHg^+$	SPE, HPLC	CV-AAS	Hg^{2+}: 0.25 $MeHg^+$: 0.96	[2]
River water	Hg^{2+} $MeHg^+$ $EtHg^+$ $PhHg^+$	SPME	AFS	Hg^{2+}: 800 $MeHg^+$: 4300 $EtHg^+$: 1400 $PhHg^+$: 800	[3]
Sea water	Hg^{2+} $MeHg^+$	HPLC	CV-AAS	Hg^{2+}: 0.5 $MeHg^+$: 0.5	[4]
River water, Sea water, Artic snow	$MeHg^+$	GC	ICP-MS	$MeHg^+$: 0.03	[5]
Aquatic environmental samples	Hg^{2+} $MeHg^+$ $EtHg^+$	Capillary GC	AES	Hg^{2+}: 6500 $MeHg^+$: 1300 $EtHg^+$: 210	[6]
Drinking water	Hg^{2+} $MeHg^+$	HPLC	ICP-MS	$MeHg^+$: 500	[7]

Yin et al[1] separated and enriched Hg^{2+}, $MeHg^+$ and $EtHg^+$ in seawater with a commercially available C18 SPE column pretreated with dithizone. Recoveries of 95.4%, 86.0%, and 79.1% were obtained for Hg^{2+}, $MeHg^+$ and $EtHg^+$ respectively after elution with $Na_2S_2O_3$ solution. During the last decade, Solid Phase Micro-Extraction (SPME)[3,10], which was used for the extraction of analytes, has been established as a valuable alternative to traditional SPE. SPME coupled with gas chromatography has been widely used for mercury compound separation and determination. After extracting $MeHg^+$ and $EtHg^+$ by SPME, Carrasco et al[10] successfully detected them through GC coupled to pyrolysis-atomic fluorescence spectrometry (Py-AFS). The detection limits and quantification limits were in 0.04 ng/g, 0.13 ng/g for $MeHg^+$ and $EtHg^+$ at the optimum conditions.

1.2 Adsorption

Adsorption methods include static adsorption and dynamic adsorption, and the adsorption mechanism varies with the structure and property of adsorbent. Recently, with the development of nanotechnology, a number of papers reported that nano-materials were used to separate different speciation of metal elements based on their adsorption capability of metal ions[11-12]. Zierhut et al[11] used a metal collector of nanometer surface structure to separate and enrich Hg^0, Hg^{2+} and $MeHg^+$ in natural water, and carried out the adsorption mechanism. The mechanism of Hg^{2+} and $MeHg^+$ adsorption can be explained by the catalytic activity of the nanometer surface structure on the metal collector and the Hg^0 adsorption being an amalgamation process. Meanwhile, the surface morphology of the metal collector is a key factor for recoveries. After a series of experiments, including the selection of metal material and pyrolysis temperature, the recoveries for Hg^0, Hg^{2+} and $MeHg^+$ were all greater than 90%, and the relative standard deviation (RSD) for Hg^{2+} and $MeHg^+$ were within 3% -5%.

1.3 Electrochemical Separation

High Performance Capillary Electrophoresis (CE) is one of the most rapidly developing analysis methods with the advantages of high efficiency, speediness and trace sample usage. It is used to separate different ions with different charge and properties based on the different migration rate of ions in an electric field. Hong et al[13] used CE to separate Hg^{2+} and $MeHg^+$ after optimizing the buffer solution, separation voltage and content of methanol. The RSDs of migration time, peak area and peak height were 1.7% -3.1%, 3.8% -4.7% and 1.6% -2.8%, respectively.

1.4 Chromatography

Usually, the chromatography method can be classified into Gas Chromatography (GC) and Liquid Chromatography (LC). As GC is suitable for volatile compounds, a derivatization[14] step is necessary for $MeHg^+$ and $EtHg^+$ prior to separation by GC. When compared with GC, LC can separate mercury species in aqueous solution at room temperature, thus avoiding the error caused by the decomposition of compounds during the GC process and the toxicity of volatile organic solvents. Different compounds were separated by LC based on their slight difference in absorption capacity, distribution ratio, ion-exchange ability or molecular force in a chromatography column. The most common separating modes for mercury speciation include Reversed Phase Chromatography[2-3,7-8] and Ion Chromatography (IC)[15]. Certain organic modification agents (such as methanol, acetonitrile), complex agents or ion-pair agents were usually added to the mobile phase; the appropriate mobile phase and mobile phase ratio can efficiently shorten the separation time of mercury species. Gómez-Ariza et al[16] used the gradient elution [mobile phase A: 0.02% (V/V) 2-mercaptoethanol, 5 mmol/L TBABr, 5% (V/V) CH_3OH, 20 mmol/L ammonium acetate, pH 4.0; mobile phase B: 100% methanol] to analyze Hg^{2+} and $MeHg^+$ in river water. The analytical time was decreased by up to 12 min. In our work, reversed phase chromatography was used to separate Hg^{2+}, $MeHg^+$ and $EtHg^+$. After optimization of the mobile phase, 8% methanol, 60 mmol/L ammonium acetate and 9 mmol/L L-cysteine (pH = 6.4) were chosen, and a higher resolution ($R > 1.5$) and lower analytical time (< 7 min) were finally obtained, as shown in Fig. 1.

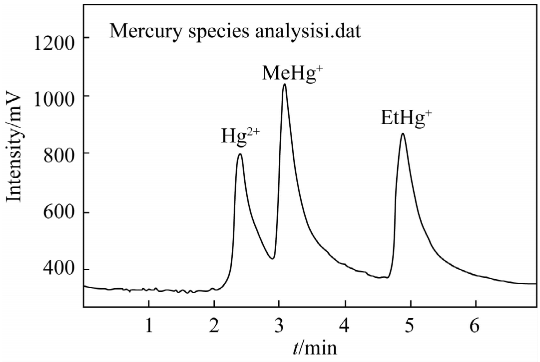

Fig. 1 Chromatogram of Hg species by LC-CV-AFS (10 μg/L for each Hg species)

2 Detection Techniques

A detector of high sensitivity and accuracy is crucial for detecting trace amounts of mercury speciation in natural water samples after separation and enrichment. Commonly used detectors include spectrophotometers [Ultraviolet-Visible Spectrophotometry (UV-VIS)], atomic spectrometry, such as Atomic Absorption Spectrometry (AAS), Atomic Emission Spectrometry (AES), Atomic Fluorescence Spectrometry (AFS), Mass Spectrometry (MS), and Electrochemical Analysis (ECD).

2.1 Spectrophotometry

Moghaddam et al[17] reported coupled reversed-phase column High-Performance Liquid Chromatography (RP-HPLC) with UV-VIS to detect Hg^{2+}, $MeHg^+$ and $PhHg^+$ in seawater. After the organic and inorganic mercury was enriched in SPE with a highly selective reagent of 6-mercaptopurine, it was then eluted into a HPLC column with dithizone in ammonia solution. The target compounds were then separated and eluted with methanol, the mercury species of Hg^{2+}, $MeHg^+$ and $PhHg^+$ were finally determined by UV-VIS. Recoveries for all of Hg^{2+}, $MeHg^+$ and $PhHg^+$ were greater than 90%, and the detection limits were 0.07, 0.06, 0.08 μg/L, respectively.

2.2 Atomic Spectrometry

AAS was applied for the determination of elements and species in early years, and especially for flame atomic absorption as it has higher selectivity and is suitable for liquid sample analysis [4,18]. Coupling AAS with HPLC is mature for speciation analysis. However, the sensitivity is still lower and needs further improvement. Válega et al[19] used acid leaching to extract organic mercury from the sample matrix; mercury speciation in extraction solution was pyrolyzed and collected in a metal amalgamator then determined by AAS. The detection limit reached 10.53 pg/mL after optimization of the thermal temperature and collection time.

Similar to the way that AAS developed early on AES is also used for qualitative and quantitative analysis on the basis of the nature and intensity of characteristic spectral line of different elements or atoms excited by thermal energy or electrical energy. AES can be used to analyze dozens of elements in sample including gas, solid or liquid samples at a time, in a few minutes with good selectivity[6]. The detection limit can reach the μg/g or μg/mL level for common light source, and ng/mL level for inductively coupled plasma (ICP). For example, Rodil et al[20] analyzed Hg^{2+} and $MeHg^+$ by solid-phase micro-extraction (SPME) and GC coupled with microwave-induced plasma atomic emission spectrometry. The detection limits for Hg^{2+} and $MeHg^+$ were 0.86, 0.12 ng/mL for Hg^{2+} and $MeHg^+$, respectively.

AFS has developed rapidly and has been applied to speciation analysis in recent years. LC hyphenated to Cold Vapour-Atomic Fluorescence Spectrometry (LC/CV-AFS) has gradually become one of the principle techniques for the analysis and determination of mercury speciation. The separated organic mercury compounds should be converted into Hg^{2+} by UV irradiation[19,21] or chemical oxidization[8] before determination by AFS. The generated Hg^{2+} is then reduced by $NaBH_4 + NaOH$, $KBH_4 + KOH$ or $SnCl_2$ to generate metal mercury, and then delivered by argon flow into an AFS detector. Margetinová et al[3] analyzed mercury compounds using $KBr + KBrO_3 + HONH_3Cl + HCl$ as the oxidizing agent and $SnCl_2$ as the reducing agent. The detection limits for $MeHg^+$, $EtHg^+$, Hg^{2+} and $PhHg^+$ were 4.3, 1.4, 0.8 and 0.8 μg/L, respectively. In our work, HPLC/CV-AFS was used to detect Hg^{2+}, $MeHg^+$ and $EtHg^+$ under UV irradiation by 1% $K_2S_2O_8$ (*w/V*) in 0.5% KOH (*w/V*) as the oxidizing agent and KBH_4 0.5% (*w/V*) in 1% KOH (*w/V*) as the reducing agent. The detection limits for Hg^{2+}, $MeHg^+$ and $EtHg^+$ were 0.58, 0.31, 0.42 μg/L, respectively.

2.3 Mass Spectrometry

Inductively Coupled Plasma-Mass Spectrometry (ICP-MS) combines the characteristics of high atomization and ionization efficiency of ICP with the high sensitivity and isotope ratio determination of MS and is an excellent trace multi-element analysis and isotope analysis technique. At present, ICP-MS is widely used for element species analysis for its low detection limits, broad linear range and relatively few interferences, and is often coupled with LC or GC to analyze mercury compounds with only a simple pretreatment procedure, thus keeping species stability[5,7], Yin et al[1] developed a simple SPE-HPLC-ICP-MS method to analyze Hg^{2+}, $MeHg^+$ and $EtHg^+$ in water samples. The detection limit was 3 ng/L for each mercury speciation.

2.4 Electrochemical Analysis

Electrochemical Analysis has been established based on the change of electrical and electrochemical properties of sample solutions, and can simultaneously detect many organometallic compounds with high sensitivity. However, ECD analysis needs special electrodes with low selectivity, meanwhile removing reducing substances from solutions and is therefore not widely used in practical work.

3 Conclusions

Due to the toxicity and biological effects of mercury being strongly dependent on its chemical form and speciation, it is essential for the determination of mercury speciation analysis rather than total amount in water, so as to evaluate the toxicity and bioactivities of mercury. The hyphenated technique of HPLC/CV-HG-AFS and HPLC/CV-(HG)-ICP-MS are effective methods to separate and quantify mercury species in the aquatic environment with a relatively low detection limit and high accuracy and selectivity. It is worth mentioning that the instrument and running costs of HPLC/CV-AFS are about 1/30 and 1/15 when compared with the HPLC/CV-ICP-MS method. It is doubtless that LC/CV-HG-AFS will have a bright prospect for trace and ultra trace of mercury species analysis in the future. Besides, the determination of mercury species in natural water is still a challenge for the analyst due to the high risk of contamination, sample losses, and interspecies conversion during sampling, storage and pretreatment processes. Therefore, new approaches in the future should focus on the pretreatment system or in exploiting a portable instrument so as to realize species analysis *in situ*.

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