Published in final edited form as:

Anal Chim Acta. 2018 December 24; 1041: 68–77. doi:10.1016/j.aca.2018.08.049.

Removal of inorganic mercury by selective extraction and coprecipitation for determination of methylmercury in mercury-contaminated soils by chemical vapor generation inductively coupled plasma mass spectrometry (CVG-ICP-MS)

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Abstract

A procedure is developed for selective extraction of methylmercury (CH_3Hg^+) from heavily Hg-contaminated soils and sediments for determination by chemical vapor generation inductively coupled plasma mass spectrometry (CVG-ICP-MS). Soils artificially contaminated with 40 μg g^{-1} inorganic mercury (Hg^{2+}) or methylmercury chloride (CH_3HgCl) were agitated by shaking or exposing to ultrasounds in dilute hydrochloric acid (HCl) or nitric acid (HNO_3) solutions at room temperature. Extractions in HCl (5 or 10% v/v) resulted in substantial leaching of Hg^{2+} from soils, whereas 5% (v/v) HNO_3 provided selectivity for quantitative extraction of CH_3Hg^+ with minimum Hg^{2+} leaching. Agitation with ultrasounds in 5% (v/v) HNO_3 for about 3 min was sufficient for extraction of all CH_3Hg^+ from soils. Coprecipitations with $Fe(OH)_3$, $Bi(OH)_3$ and HgS were investigated for removal of residual Hg^{2+} in soil extracts. Hydroxide precipitations were not effective. Thiourea or L-cysteine added to soil extracts prior to hydroxide precipitation improved precipitation of Hg^{2+}, but also resulted in removal of CH_3Hg^+. HgS precipitation was made with dilute ammonium sulfide solution, $(NH_4)_2S$. Adding 30 μL of 0.35 mole L^{-1} to soil extracts in 5% (v/v) HNO_3 resulted in removal of all residual Hg^{2+} without impacting CH_3Hg^+ levels. Vapor generation was carried out by reacting Hg^{2+}-free soil extracts with 1% (m/v) $NaBH_4$. No significant interferences were observed from $(NH_4)_2S$ on the vapor generation from CH_3Hg^+. The slopes of the calibration curves for CH_3HgCl standard solutions in 5% (v/v) HNO_3 with and without $(NH_4)_2S$ were similar. Limits of detection (LOD, 3s method) were around 0.08 μg L^{-1} for 5% (v/v) HNO_3 blanks (n = 10) and 0.10 μg L^{-1} for 5% (v/v) HNO_3 + 0.005 mol L^{-1} $(NH_4)_2S$ blanks (n = 10). Percent relative standard deviation (%RSD) for five replicate measurements varied between 3.1% and 6.4% at 1.0 CH_3HgCl level. The method is validated by analysis of two certified reference materials (CRM); purely Methylmercury sediment (SQC1238, 10.00 ± 0.291 ng g^{-1} CH_3Hg^+) and Hg-contaminated Estuarine sediment (ERM – CC580, 75 ± 4 ng g^{-1} CH_3Hg^+

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and 132 ± 3 μg g^{-1} total Hg). CH_3Hg^+ values for SQC1238 were between 13.0 and 13.2 ng g^{-1}, and 79 and 81 ng g^{-1} for ERM – CC580. Hg-contaminated soils (57 to 96 μg g^{-1} total Hg) collected from the floodplains of Oak Ridge, TN were analyzed for CH_3Hg^+ using the procedure by CVG-ICPMS. CH_3Hg^+ levels ranged from 30 to 51 ng g^{-1} and did not correlate with total Hg levels (R^2 =0.01).

Keywords

Methylmercury; extraction; coprecipitation; soil/sediment; chemical vapor generation; ICP-MS

1. Introduction

Mercury is a ubiquitous element that is present in various forms in the environment, including elemental (Hg^0), mercurous (Hg_2^{2+}), mercuric (Hg^{2+}), and alkylated mercury compounds (e.g., Methylmercury, CH_3Hg^+) [1,2]. The determination and speciation of Hg are important to understand the mobility, bioavailability in soils, sediments and biota, and potential toxicity on human and environmental health. Hg^{2+} is the predominant form in soil and waters. Hg^0 are the major species in the atmosphere, while CH_3Hg^+ is present mostly in biota and foodchain. These mercury forms also exhibit different solubilities and toxicities. Elemental Hg and mercurous chloride (Hg_2Cl_2) have a water solubility of 5.6×10^{-5} g L^{-1} and 2.0×10^{-3} g L^{-1} at 25 °C, respectively [3,4]. Mercuric compounds, namely HgO and $HgCl_2$, are more soluble in water for which solubilities are 0.051 g L^{-1} at 25 °C and 69 g L^{-1} at 20 °C, respectively [3,4]. In contrast, methylmercury chloride (CH_3HgCl) is considered lipid soluble due to its lower solubility in water (0.10 g L^{-1} at 21 °C).

During the 1950s and early 1960s, elemental mercury (Hg) was used to produce enriched ^6Li isotope at the Y-12 National Security Complex (Y-12 Plant) at the Oak Ridge Reservation of the Department of Energy (DOE) for manufacturing components of various nuclear weapons systems. It is estimated that 350,000 kg of Hg was released to the environment contaminating facilities, soil, sediment, surface water, and groundwater within the boundaries of the Y-12 Plant and the downstream environment along the East Fork Poplar Creek (EFPC) [5]. The EFPC floodplain soils were reported to contain predominantly cinnabar (HgS) form [5,6], but other inorganic and organic Hg species, including Hg^0, $HgCl_2$, Hg_2Cl_2, HgO and CH_3Hg^+ have been found within the Y-12 Facility boundaries and the 23-km long contaminated EFPC. Over the last 25 years, Hg fluxes from the Y-12 Plant have been reduced by various remediation efforts, yet, the Hg concentration in water continue to exceed both the regulatory limit (51 ng L^{-1}) and the remediation goal (200 μg L^{-1}) [5].

Methylmercury (CH_3Hg^+) is a known neurotoxin [3,7]. Not only is it more toxic than other Hg species, but also bio-accumulates in aquatic food chain posing reproductive and neurobehavioral health disparities to biota and humans [3,7]. Especially most Hg in fish and seafood is CH_3Hg^+. The Environmental Protection Agency (EPA) estimated a reference dose (RfD) of 0.1 μg kg^{-1} body weight based on daily oral exposure for methylmercury [8,9] while World Health Organization (WHO) recommends a maximum weekly intake of 1.6 μg

kg^{-1} for the protection of public health [9,10]. In the environment, CH_3Hg^+ is produced by a series of biogeochemical transformations involving microorganisms (e.g., sulfate- and iron-reducing bacteria) [11–13]. These microbial-methylation processes are influenced by the organic matter, pH and redox potential of the environment [14–16]. Anaerobic conditions that occur in soils and wetlands also favor the transformation of Hg^{2+} to CH_3Hg^+ [17,18]. Nonetheless, most microorganisms responsible for methylation of Hg are also capable of degrading CH_3Hg^+, consequently its concentration rarely correlates with total Hg concentration [19,20]. Additionally, reductive and oxidative demethylation processes convert CH_3Hg^+ to Hg^0 and Hg^{2+}, respectively [21]. As a result, CH_3Hg^+ is often present in soils and sediments at much lower concentrations, typically 1.5% of total Hg [22,23], and thus its determination requires sensitive detection techniques and suitable separation methods, especially in the presence of elevated Hg^{2+} [15,24].

Various separation methods have been developed for speciation of Hg in environmental and biological samples, which include cloud-point extraction (CPE) [15,24–26], solid phase microextraction (SPME) [22,24,27–30], high performance liquid chromatography (HPLC) [24,31–36], and gas chromatography (GC) [15,22,24,31,37–40]. Chemical vapor generation (CVG) has been utilized as a popular approach to further improve the sensitivity and selectivity for CH_3Hg^+ determinations by inductively coupled plasma mass spectrometry (ICP-MS) and atomic spectroscopy, namely fluorescence (AFS) and absorption (AAS) [15,25,31,32,41–43]. In recent applications, ICP-MS has been a dominant technique due to its high sensitivity, isotopic measurement capability and ease of coupling to CVG, HPLC and GC systems. A great deal of the studies to date, however, have undertaken the speciation of Hg in samples like seawater, wastewater seafood, blood, urine and hair that possess relatively low Hg levels and hence selectivity among Hg species could be achieved with chromatographic approaches or chemical vapor generation [22–43].

Unlike water and biological samples of low total Hg content, Hg-contaminated soil and sediments, such as that from Oak Ridge Reservation of the DOE, possess significantly high Hg levels (e.g., as high as 200 $\mu g\ g^{-1}$) which is predominantly Hg^{2+} [5,6]. Chromatographic separation methods cannot tolerate such high levels of Hg as it could cause contamination and costly damages to analytical column and instrumentation nor highly sensitive CVG approaches employed for Hg speciation could provide desired selectivity for CH_3Hg^+ under heavy Hg^{2+} matrix. To alleviate the hurdles associated with Hg^{2+} matrix, separation of CH_3Hg^+ from soil matrix or removal of Hg^{2+} matrix is imperative besides the use of sensitive methods for accurate determinations [22,31,44–46]. Acid leaching followed by extraction into benzene or toluene (i.e., Westöö method) is perhaps by far the most popular approach for extraction of CH_3Hg^+ and other organomercury species from the solid samples [47,48]. Over the years, various extraction procedures, such as alkaline digestion with KOH-methanol mixture [49] and Universol® (an alkaline cocktail of reagents) [45], microwave- and/or ultrasound-assisted extraction in HCl-methanol [46] and HNO_3 [50] have been developed as part of continued efforts to utilize robust, aqueous and less laborious methods. More recently, Carrasco and Vassileva [40] investigated various acidic and alkaline digestion/extraction methods for determination of CH_3Hg^+ from estuarine sediments, and reported that extraction with mixture of $HNO_3/CuSO_4$ provided the highest recoveries. Despite the improvements, nevertheless, acidic or alkaline digestion/extraction procedures

reported so far require additional extractions with organic solvents followed by back-extraction into aqueous solution, neutralization and derivatization or alkylation steps prior to chromatographic separation [22,24,39,40,44]. CVG approaches in contrast offer less laborious sample preparation and sufficient selectivity due to the capability of differential chemical reduction of CH_3Hg^+ and Hg^{2+} to Hg^0 vapor [24,43,46]. Hg^{2+} is more readily reduced to Hg^0 than CH_3Hg^+, and thus removal of elevated Hg^{2+} matrix is desirable to achieve interference-free determination of CH_3Hg^+ from Hg-contaminated soil and sediments.

In this work, we have developed a method for selective and sensitive determination of CH_3Hg^+ from highly Hg-contaminated soils and sediments in an attempt to characterize CH_3Hg^+ distribution from the soils and sediments collected from the Oak Ridge, TN Reservation of DOE that were impacted by the legacy Hg-contamination of 1960's. The objectives of the study were two-fold: (1) to determine the optimal conditions for selective extraction of CH_3Hg^+ from the soil matrix with minimal Hg^{2+}, (2) to remove the residual Hg^{2+} from solutions to improve selectivity and accuracy for CH_3Hg^+ for determination by chemical vapor generation. Extractions in dilute hydrochloric acid (HCl) and nitric acid (HNO_3) were performed via shaking and ultrasonic agitation at room temperature to determine the extraction profiles of CH_3Hg^+ and Hg^{2+} from the soils artificially contaminated with Hg^{2+} or CH_3Hg^+. For optimal extraction conditions, hydroxide and sulfide coprecipitation schemes were investigated to eliminate the residual Hg^{2+} from soil extracts. Chemical vapor generation (CVG) conditions were optimized for the optimum extraction conditions. The method was validated with determination of CH_3Hg^+ in estuarine sediment certified reference material (ERM - CC580) and methylmercury sediment reference material (SQC1238) by CVG-ICP-MS and then applied to the determination of CH_3Hg^+ in Hg-contaminated soils from Oak Ridge, TN.

2. Experimental

2.1. Reagents and solutions

Ultra-pure deionized water was used throughout. Tap water fed through MaxCap® reverse-osmosis deionization (RO/DI) unit (SpectraPure Inc., Tempe, AZ) was gravity-fed into a 4-stage Barnstead™ E-Pure deionization system producing ultra-pure deionized water with minimum resistivity of 18.0 MΩ cm resistivity. A 10 μg mL^{-1} Hg^{2+} solution was prepared from 1000 μg L^{-1} Hg^{2+} stock standard solution (Spex CertiPrep, Metuchen NJ) in 5% HNO_3. All working solutions of Hg^{2+} were prepared from a 10 μg L^{-1} Hg^{2+} stock. The stock solution of CH_3HgCl (1000 μg L^{-1}) was prepared by dissolving the CH_3HgCl salt (Aldrich, 99.9% pure, Lot# SBLQ9540V) in 0.5% (v/v) HCl and stored in plastic bottle. A secondary stock solution of 10 μg L^{-1} CH_3HgCl was prepared and used for preparing working solutions of CH_3Hg^+. The standard solutions of Hg^{2+} and CH_3Hg^+ were prepared freshly before each run and the acidity was matched with that of the sample solutions (HCl or HNO_3) used for preparing the samples. Sodium borohydride solutions ($NaBH_4$, 99.8%, Sigma Aldrich) were prepared in 0.1% (m/v) NaOH (99.9%, Sigma Aldrich). Stannous chloride solution ($SnCl_2$, Sigma, 98% Lot# S73836–039) were prepared in 0.5% (v/v) HCl. Trace metal grade hydrochloric acid (HCl, BDH Chemicals) and nitric acid (HNO_3, BDH

Chemicals) were used in all extraction and preparations. Hydrofluoric acid (HF, 48% m/v, Sigma Aldrich, 99.99%, Lot# 16595ME) was used for digesting soils for total Hg determination. Other reagents used for preparing stock solutions for coprecipitation studies include iron(III) nitrate nonahydrate, $Fe(NO_3)_3{\cdot}9H_2O$ (Sigma Aldrich, 99.99%, Lot# 09007BD), bismuth(III) nitrate pentahydrate, $Bi(NO_3)_3{\cdot}5H_2O$ (Alfa Aesar, +98%, Lot# I24Y019), ammonium sulfide, $(NH_4)_2S$ (Sigma Aldrich, 47.2% m/v, Lot#00819DH), thiourea (Sigma Aldrich), L-cysteine (97%, Sigma Aldrich), and triethylamine, TEA (trace metal grade, Acros Organics, Lot# A0374495).

2.2. Instrumentation

All measurements were performed with a Varian 820MS ICP-MS instrument (Varian, Australia). The instrument was equipped with a peltier-cooled double-pass glass spray chamber, all teflon Ari-mist nebulizer (SCP Science, Champlain NY), quartz torch, CRI-type Pt sampler and skimmer cones and all-digital detector (DDEM, Model AF250, ETP Australia). Samples were introduced manually. The instrument was optimized daily with 5 μg L^{-1} ^{138}Ba, ^{25}Mg, ^{115}In, ^{140}Ce, ^{208}Pb solution for optimal sensitivity, oxides ($^{156}CeO^+/^{140}Ce^+ < 3\%$) and doubly charged ions ($^{138}Ba^{2+}/^{138}Ba^+ < 2\%$). Data collection was achieved by ICP-MS Expert software package (version 2.2 b126). Measurements were made with solution nebulization mode for total Hg determinations, and determining the recoveries in extraction and coprecipitation studies. Washout was performed by pumping 5% (v/v) HCl for 30 s using fast-pump mode (6 mL min^{-1}) to clean up residual Hg before next measurement. Chemical vapor generation (CVG) was used for method validation and determination of CH_3Hg^+ from soil samples. A solution of 0.5% (m/v) potassium ferricyanide in 5% (v/v) HCl was used for Hg-washout as it was found effective in reducing memory effects on CVG-ICP-MS determinations [43]. The operating parameters of the ICP-MS instrument are summarized in Table 1 for nebulization and CVG modes. An internal standard (IS) solution of 5 μg L^{-1} germanium (Ge), rhodium (Rh), rhenium (Re) was used to correct for possible instrumental drift and matrix-related signal fluctuations during measurements with solution nebulization. The internal standard solution was mixed on-line with the calibration standard or sample solution. ^{185}Re was used as IS for Hg. Data were collected for ^{200}Hg and ^{202}Hg isotopes in both nebulization and CVG modes.

2.3. Soil samples

Method development studies were conducted with Hg-free soils that were ground and sieved through 0.25-mm apertures. Sub-samples were digested in HNO_3 and HF as described below (see section 2.4.1) and were analyzed by ICP-MS for Hg content before utilizing in any experimental work. Then, sub-samples of the soil were intentionally contaminated with Hg^{2+} or CH_3Hg^+ for investigating the acid extraction conditions. Briefly, about 0.5 g sub-samples (n = 4) were wetted (e.g., spiked) with 0.2 mL of 100 μg mL^{-1} Hg^{2+} or CH_3HgCl standard solution in 15-mL conical test tubes. This introduced 20 μg or 40 μg g^{-1} Hg^{2+} or CH_3HgCl to the soil matrix. Care was given to ensure the spiked soil was uniformly wetted in the tube. Then, all soil samples were air-dried for 48 to 72 h at room temperature.

Hg-contaminated soils were collected from a site located in a floodplain field of Lower East Fork Poplar Creek (LEFPC) of Oak Ridge, TN, USA. A total of 10 surface soils (0–10 cm)

were randomly sampled from both woodland and wetland/grassland areas. Fresh field samples were stored in a refrigerator for total Hg and speciation measurements. Sub-samples were air-dried, ground and passed through 0.25-mm sieves. The soil at the study site was Armuchee soil (clay, mixed, thermic Ochreptic Hapludults) that is formed in residuum of shale and river alluvia.

2.4. Procedures

2.4.1. Soil digestion for total Hg determination in floodplain soils—For total Hg determination, approximately 0.1 g (n = 3) of floodplain soil samples was weighed into teflon vessels (70 mL inner volume) (Savillex) and digested in 5 mL HNO_3 and 1 mL HF for 2 h at 120 °C on a graphite block digestion unit (SCP Science, Champlain NY). Then, the digestion temperature was increased to 140 °C and samples were digested for additional 3 h. After digestion, contents were cooled to room temperature, then transferred to 15-mL tubes and diluted to 10 mL with deionized water. All samples were centrifuged for 15 min at 5000 rpm to remove undissolved components. After centrifugation, 0.2 mL aliquots were taken and diluted to 2 mL with 0.1% (v/v) HCl and 5% (v/v) HNO_3 in 2-mL graduated micro-centrifuge tubes (Fisher Scientific) for ICP-MS analysis.

Effectiveness of the digestion procedure and accuracy of instrumental measurements were verified with analysis of Montana soil (SRM 2710) and Domestic sludge (SRM 2781) certified reference materials from National Institute of Standards and Technology (NIST, Gaithersburg, MD). Sub-samples (0.1 g, n = 5) of SRM 2710 and SRM 2781 were digested along with the floodplain soils using the procedure above. Similarly, 0.2 mL SRM solution (10 mL) were taken and diluted to 2 mL with 0.1% (v/v) HCl and 5% (v/v) HNO_3 and analyzed by ICP-MS for total Hg. Preliminary experiments indicated that Hg^{2+} was relatively unstable in 5% (v/v) HNO_3 solution. Within 3 to 4 days of preparation, signals for Hg^{2+} standard solutions declined about 20% in comparison to freshly prepared solutions. Addition of trace amounts of HCl improved the stability of Hg^{2+} standards for up to two weeks, and thus calibration standards ranging from 0 to 50 μg L^{-1} Hg^{2+} were prepared in 0.1% (v/v) HCl and 5% (v/v) HNO_3 for total Hg measurements. A solution of 5% (v/v) HCl was ran through for 30 s at fast pump-mode (6 mL min^{-1}) to washout Hg before introducing sample or standard solutions during measurements with nebulization mode.

2.4.2. Extraction of Hg^{2+} and CH_3Hg^+ from artificially contaminated soils via shaking—Treatment of soil and sediments with HCl and HNO_3 have been an effective means for extracting Hg species [40,46,50]. Here, we attempted to determine the suitability of HCl and HNO_3 extractions for selective separation of CH_3Hg^+ and Hg^{2+} from soil matrix. For this purpose, air-dried soils (0.5 g) contaminated with 20 μg Hg^{2+} or CH_3HgCl were suspended in 10 mL of water, dilute HCl or HNO_3 solutions (5 and 10% (v/v) concentrations) and shaken on a rocker shaker for 0, 1, 2, 6 and 24 h. For control set, unspiked soils (0.5 g, n = 4) were suspended similarly in water, HCl and HNO_3 solutions. There were 24 soil suspensions (12 treatments and 12 controls) for each acid medium. Overall, 48 soil suspensions were processed concurrently.

For sampling, shaking was stopped and suspensions were allowed to rest for about 2–3 min for settling of suspending soils particles. Then, 0.5 mL aliquots from the top of the suspension was pipetted into 2-mL micro-centrifuge tubes and spun 10 min at 12, 000 rpm on an Eppendorf Model 5415D centrifuge to settle the residual suspending particles. Of the 0.5 mL supernatant solution, 0.1 mL was taken and diluted to 2 mL with 5% (v/v) HCl for HCl treatments and with 5% (v/v) HNO_3 for HNO_3 treatments. Samples were then analyzed by ICP-MS using Hg^{2+} or CH_3HgCl standards (0 to 20 μg L^{-1}) in 5% (v/v) HCl or 5% (v/v) HNO_3 for each set. Washings were made between samples with 5% (v/v) HCl as described in section 2.4.1.

2.4.3. Extraction of Hg^{2+} and CH_3Hg^+ from artificially contaminated soils via ultrasonic agitation—

In another series of experiments, the effect of ultrasound-assisted agitation was examined for the extraction of Hg^{2+} or CH_3HgCl from soil matrix. Here, soils (n = 4) contaminated with Hg^{2+} or CH_3HgCl were suspended similarly in 10 mL water, dilute HCl or dilute HNO_3 along with the uncontaminated controls. Each suspension was then agitated by ultrasounds using a Fisher Scientific Model 100 Sonic Dismembrator equipped with a 2-mm diameter titanium microprobe. Sonication was implemented for 0, 3 and 6 min at 50% power setting. After each sonication, suspensions were allowed for 2 to 3 min for settling of soil particles. Then, 0.5 mL aliquot from the top layer was taken and spun as described above, and finally 0.1 mL of the spun suspension was diluted to 2 mL with either 5% (v/v) HCl or 5% (v/v) HNO_3 and analyzed by ICP-MS as above to determine the extraction efficiency (e.g., recovery) of Hg^{2+} or CH_3Hg^+.

2.5. Removal of residual Hg^{2+} via hydroxide coprecipitation

Extraction studies performed with artificially contaminated soils indicated that a great deal of Hg^{2+} leached into solution. Unless removed, Hg^{2+} would confound, if not, preclude the accurate determination of CH_3Hg^+ by CVG-ICP-MS besides contaminating the sample introduction system. To avoid these hurdles, attempts were made to remove Hg^{2+} from the soil extracts by coprecipitation approaches. Hg shows high affinity to sulfur containing groups, such as thiourea and L-cysteine. It is also reported that Hg^{2+} coprecipitates with hydroxides of iron(III) [51] and bismuth (III) [52]. In a series of experiments, the coprecipitation of $Hg^{2+}$and CH_3Hg^+ was examined with Fe(III) and Bi(III). A volume of 20 μL of 10 μg mL^{-1} Hg^{2+} or CH_3HgCl was added into 2-mL microcentrifuge tubes (n = 5). To simulate elemental composition of soil extracts, 20 μL of 10 μg mL^{-1} multi-element solution (Ag, Al, As, Ba, Be, Cd, Cr, Co, Cu, Li, Mn, Mo, Ni, Pb, Sb, Se, Sr, Tl, V, Zn) and 100 μL of 100 μg mL^{-1} of major element solution (Ca, Mg, Fe, Na, K, P, and Ti) were also added. Then, 0.2 mL of 10 mg mL^{-1} of Fe(III) (as nitrate) or Bi(III) (as nitrate) were added to each tube. The volume was completed to 2 mL with 5% (v/v) HNO_3. The solution contained 100 μg L^{-1} Hg^{2+} or CH_3HgCl in a matrix of 0.1 μg mL^{-1} of trace metals, 5 μg mL^{-1} of the major elements and 1000 μg mL^{-1} of Fe(III) or Bi(III) prior to precipitation. Precipitation of $Fe(OH)_3$ and $Bi(OH)_3$ was made by adding 0.2 mL of TEA. Upon adding TEA, nucleation of hydroxides occurred rapidly. For effective coprecipitation, solutions were allowed for 10 min for complete precipitation and then centrifuged for 15 min at 12,000 rpm. The supernatant solution was discarded. Pellets were washed gently, dissolved in 1 mL of 10% (v/v) HNO_3, and then completed to 2 mL with water. All solutions were then analyzed by

ICP-MS in nebulization mode to determine the recoveries (e.g., coprecipitation efficiency) for Hg^{2+} or CH_3Hg^+.

In another experiment, the effects of thiourea and L-cysteine were examined in combination with Fe(III) and Bi(III). Test solutions of Hg^{2+} or CH_3HgCl (n = 5) were prepared similarly as above. A volume of 0.2 mL of 1% (m/v) thiourea or 1% (m/v) L-cysteine were added to the microcentrifuge tubes before adding other solutions, including Fe(III) and Bi(III). Precipitation, centrifugation and dissolution were performed as described above. The pellet solutions were analyzed for Hg^{2+} or CH_3Hg^+ content.

2.6. Removal of Hg^{2+} via sulfide coprecipitation

Mercury also forms strong sulfide precipitates, such as HgS which is highly insoluble even in acidic media [53,54]. Thus, as an alternative approach for removal of Hg^{2+}, the precipitation of HgS was examined in simulated soil solutions using ammonium sulfide as coprecipitation agent. The test solutions of 100 μg L^{-1} Hg^{2+} or CH_3HgCl (n = 5) were prepared similarly as described above in a matrix of trace metals and major elements. The volume was completed to 2 mL with 5% (v/v) HNO_3. Precipitation was made by adding 30 μL of 0.35 mole L^{-1} ammonium sulfide solution, $(NH_4)_2S$. Contents were allowed to precipitate for about 10 min, and then centrifuged at 12,000 rpm for 15 min. The supernatant solutions were analyzed by ICP-MS to determine the concentration of Hg^{2+} or CH_3Hg^+ remained in solution.

2.7. Optimization of chemical vapor generation conditions

Chemical vapor generation (CVG) as a sensitive approach offers selective reduction of Hg^{2+} and CH_3Hg^+ species to gaseous Hg^0 under optimal conditions [24,43,45]. When coupled to ICP-MS, CVG provides unsurpassed sensitivity for detection of trace levels of Hg species. CVG measurements were carried using the setup illustrated in Fig. 1. The quartz spray chamber ICP-MS instrument was used as gas liquid separator. A polypropylene T-piece (1/8” i.d., 1/4” o.d.) purchased from a local hardware store was inserted through the nebulizer housing on the Teflon end-cap of the spray chamber. A 12-cm long PTFE transfer line (1.6 mm. i.d. & 1.8 mm o.d.) was inserted through the T-piece extending into the spay chamber. Outer end of the T-piece was sealed tightly to prevent gas leak. Carrier gas was supplied from the nebulizer argon port of the instrument through the lower arm of T-piece. Color-coded tygon pump tubings were used with the following diameters; sample and $NaBH_4$: red-red stop (1.14 mm i.d.), waste: purple-white stop (2.79 mm i.d.). The mixing coil or reaction coil was 15-cm long PTFE tubing (1.0 mm i.d.) designated to mix the sample and $NaBH_4$ solutions.

Experiments were performed with 10 μg L^{-1} Hg^{2+} or CH_3HgCl solutions prepared in 5% (v/v) HNO_3. Solutions were introduced via CVG manifold and reacted with 1% (m/v) $NaBH_4$ or 1% (m/v) $SnCl_2$ solution. At first, the acidity of medium was examined from 0 to 6% (v/v) HNO_3. Then, the effects of reducing agents were examined on the generation of Hg vapor from Hg^{2+} or CH_3HgCl. In the last step, interferences from $(NH_4)_2S$ were investigated by measuring CVG signal profiles for 10 μg L^{-1} CH_3Hg^+ solutions that contained 30 μL of 0.35 M $(NH_4)_2S$ against those in 5% (v/v) HNO_3 (e.g., control).

Calibration curves were constructed using 0, 0.5, 2, 5 and 10 μg L^{-1} CH_3HgCl solutions prepared in 5% (v/v) HNO_3 with and without $(NH_4)_2S$ matrix. The slopes and limits of detection were compared to determine the most optimal setting for CVG-ICP-MS determinations. Washout was performed by running 0.5% (m/v) potassium ferricyanide solution in 5% (v/v) HCl before each test or sample solution to reduce memory effects.

2.8. Method validation and applications

The procedure was applied to the determination of CH_3Hg^+ in soils and sediments. Two soil and sediment certified reference materials (CRM) were used for method validation; methylmercury in sediment (SQC1238, Lot# LRAA7422)) and estuarine sediment (ERM CC580, Lot# 0650) that were obtained from Sigma Aldrich (St. Louis, MO). The SQC1238 contained trace levels of CH_3HgCl in soil, whereas ERM - CC580 was inorganic Hg sediment with trace levels of CH_3Hg^+. About 0.2 g sub-samples of these CRMs (n = 5) were suspended in 5 mL of 5% (v/v) HNO_3 and subjected to ultrasounds using Fisher Scientific model 100 dismembrator for 2 to 3 min. After sonication, contents were centrifuged at 5000 rpm for 10 min. Then, 2 mL of extracts were taken into 2-mL centrifuge tubes. A volume of 30 μL of 0.35 M $(NH_4)_2S$ was added to precipitate Hg^{2+} as HgS. Contents were centrifuged at 12,000 rpm for 15 min to remove precipitated HgS. The supernatant solutions were transferred to new 2-mL tubes and analyzed by CVG-ICP-MS. To match the matrix, 30 μL of 0.35 M $(NH_4)_2S$ solution were added to CH_3HgCl calibration standards. Once the method was deemed accurate, floodplain soil samples were subjected to the optimized procedures as for the CRMs. About 0.2 sub-samples (n = 5) were placed in 15-mL tubes and suspended in 5% (v/v) HNO_3 and exposed to ultrasounds. Coprecipitation in extracts was made as for the CRMs. The soil extracts were then analyzed by CVG-ICP-MS along with freshly prepared CRM samples and CH_3HgCl calibration standards. Hg-washout was made with 0.5% (m/v) potassium ferricyanide solution in 5% (v/v) HCl as described above.

3. Results and discussion

3.1. Total Hg levels of Hg-contaminated floodplain soil samples

Total Hg contents of Hg-contaminated floodplain soils varied between 57.4 ± 6.0 μg g^{-1} and 95.7 ± 3.4 μg g^{-1}. Concentrations for individual soil samples are summarized in Table 6 (see section 3.7) along with CH_3Hg^+ levels. The results indicated persistence of Hg contamination from the catastrophic spill occurred more than a half century ago. Earlier reports have shown that these soils contain various inorganic and organic Hg species, but are mostly insoluble cinnabar, HgS [5,6]. Total Hg concentrations determined from SRM 2710 (Montana Soil Elevated Traces) and SRM 2781 (Domestic Sludge) digests were 34.1 ± 0.81 μg g^{-1} and 4.01 ± 0.65 μg g^{-1}, respectively. The results were within the 95% confidence interval of the certified concentrations of SRM 2710 (32.6 ± 1.8 μg g^{-1}) and SRM 2781 (3.68 ± 0.14 μg g^{-1}). It should be noted that soil and sludge samples were not totally dissolved by the HNO_3/HF digestion method; rather a partial dissolution was performed to extract Hg species from the alumino-silicate skeleton of the soil. Within this context, the results demonstrated that partial digestive dissolution with HNO_3/HF mixture provide quantitative extraction of Hg from soil and sludge matrices. Various studies have also

reported that Hg could also be extracted from soils even at mild acidic extractions with HCl or HNO_3 [46,55].

3.2. Extraction of Hg^{2+} and CH_3Hg^+ from soils via shaking and ultrasonic agitation

The recoveries for the extraction of CH_3HgCl or Hg^{2+} via shaking from the artificially contaminated soils in water, and dilute HCl and HNO_3 are summarized in Table 2. Expected concentrations in 10-mL extracts were 200 μg L^{-1} for quantitative extraction of 20 μg CH_3HgCl or Hg^{2+} from 0.5 g soil samples. Neither Hg^{2+} nor CH_3Hg^+ did show any significant leaching from soils in water in 24 h. Extractions with HCl resulted in leaching of more Hg^{2+} to solution in comparison to that with HNO_3. Maximum extraction was about 71% in 10% (v/v) HCl after 24-h shaking, while that in 10 (v/v) HNO_3 was about 18%. CH_3Hg^+ was extracted from the soils both in HCl and HNO_3. The latter provided faster extraction. Within 1 h, more than 90% of CH_3Hg^+ extracted from the soil matrix in 5% (v/v) HNO_3. In 10% (v/v) HNO_3, CH_3Hg^+ extracted rapidly and quantitatively when soil suspension was inverted briefly for mixing (see Table 2 last row).

The results for ultrasounds-assisted extraction are summarized in Table 3. Extraction patterns were similar to that with shaking, but occurred faster in minutes in contrast to hours. Leaching of Hg^{2+} to solution was also substantial in HCl. All Hg^{2+} leached into solution in 10% (v/v) HCl upon 6 min sonication. This result was consistent with that reported by Park et al. [46] who achieved extraction of Hg^{2+} and CH_3Hg^+ from an estuarine sediment reference material (BCR 580) in a (1:1 v/v) methanol and HCl mixture via sonication. Sonication in HNO_3 also performed similarly for Hg^{2+} with that observed for shaking. Despite vigorous agitation, extraction of Hg^{2+} from the soils was very low; 4.7% and 8.6% for 6 min sonication in 5% and 10 (v/v) HNO_3, respectively. Likewise, CH_3Hg^+ was extracted completely in 5% and 10 (v/v) HNO_3 (Table 3). It was evident that 3 min agitation in 5% (v/v) HNO_3 was optimal for fast and effective extraction of CH_3Hg^+ with minimal Hg^{2+} (e.g., 4.3% extraction). The Hg^{2+} concentration in soil extracts was about 8.6 μg L^{-1} (e.g, 4.3% of 200 μg L^{-1} Hg^{2+}). HCl performed similarly to HNO_3 on CH_3Hg^+, but a major limitation of HCl extraction was that a great deal of Hg^{2+}, as high as 20% (ca. 40 μg L^{-1} Hg^{2+}) leached into the solution under brief sonication (see Table 3). This was likely due to the solubilization of Hg^{2+} as soluble $HgCl_2$ species. Though 10% (v/v) HNO_3 afforded instantaneous extraction of CH_3Hg^+, it was not suitable for CVG measurements owing to vigorous reaction with $NaBH_4$ that was essential for reduction of CH_3Hg^+ to Hg^0.

3.3. Selective coprecipitation of Hg^{2+} from soil extracts

It should be noted that Hg-contaminated floodplain soils contained about 57.4 and 95.7 μg g^{-1} Hg, and accordingly 5% (v/v) HNO_3 extracts of these soils are expected to contain at least 100 to 160 μg L^{-1} Hg^{2+} in 5 mL volume when 0.2 g samples are processed (e.g., 4.3% of total Hg). In the presence of high Hg^{2+} matrix, accurate determination of sub-μg L^{-1} levels of CH_3Hg^+ is hardly feasible by using traditional Hg^{2+} and total Hg CVG approaches. Besides, memory effects and contamination to CVG manifold and ICP-MS instrument are inevitable unless the residual Hg^{2+} is eliminated.

In order to remove the residual Hg^{2+} matrix in soil extracts, first attempt was made to coprecipitate Hg^{2+} with $Bi(OH)_3$ and $Fe(OH)_3$ in alkaline solutions as described in section 2.5. Nevertheless, neither $Bi(OH)_3$ nor $Fe(OH)_3$ coprecipitation was effective for removal of Hg^{2+} (Table 4). Though $Fe(OH)_3$ coprecipitation removed as high as 23% of Hg^{2+}, CH_3Hg^+ levels were also reduced (ca. 8.8%). The pH dependence of hydroxide precipitation could be a limitation for the lack of removal of Hg^{2+} since $Fe(OH)_3$ and $Bi(OH)_3$ precipitate within a narrow pH window of pH 8.5 to 9.3 [51,52]. The pH of the solutions after addition of TEA, on the other hand, was around pH 10 and 10.5. In the next attempt, combinations of thiourea and L-cysteine were examined in conjunction with Bi(III) and Fe(III) to improve coprecipitation of sulfuroly Hg^{2+} species onto $Bi(OH)_3$ and $Fe(OH)_3$. Both thiourea and L-cysteine increased removal of Hg^{2+} with $Fe(OH)_3$ coprecipitation, but the results were far from being satisfactory; removal efficiency was 56% with L-cysteine + Fe(III) and 64% with thiourea + Fe(III). Obviously, any scenarios of the hydroxide coprecipitation were not suitable for coprecipitation of Hg^{2+} as they did impact CH_3Hg^+ in solution; especially thiourea significantly decreased CH_3Hg^+ levels with and without Fe(III) or Bi(III) (see Table 4). Thiourea and L-cysteine are known to form sulfomercurial complexes with organic mercury species [45]. It appears that these sulfomercurial complexes lead to coprecipitation of CH_3Hg^+ in alkaline conditions.

Third attempt was made to precipitate Hg^{2+} as insoluble HgS by adding 30 μL of 0.35 M aqueous $(NH_4)_2S$ solution to 2-mL simulated soil solutions in 5% (v/v) HNO_3. Precipitation occurred instantaneously after adding $(NH_4)_2S$. Solutions were briefly shaken, and then centrifuged. The acidic supernatant solutions were analyzed by ICP-MS. The results for CH_3Hg^+ and Hg^{2+} are shown in Table 4 (last row). The recoveries are for the CH_3Hg^+ and Hg^{2+} concentration remained in the solution after precipitation. It was clear that Hg^{2+} was effectively removed from the solution (from 100 μg L^{-1} to around 2 μg L^{-1}). Unlike hydroxide coprecipitation, $(NH_4)_2S$ did not interfere with stability or retention of CH_3Hg^+ in solution. Even increasing the volume up to 50 μL had no symptoms of precipitation, yet, the volume was kept at 30 μL to avoid any interferences from excessive sulfide (S^{2-}) in vapor generation studies.

3.4. Optimization chemical vapor generation conditions

The concentrations of HNO_3 and reducing agents (e.g., $SnCl_2$ and $NaBH_4$) were optimized for chemical vapor generation of 10 μg L^{-1} CH_3Hg^+ or Hg^{2+} solutions using the manifold shown in Fig. 1. The acidity was examined up to 6% (v/v) HNO_3 for 1% (m/v) $SnCl_2$ and 1% (m/v) $NaBH_4$. The CVG profiles for CH_3Hg^+ and Hg^{2+} are shown in Fig. 2A. Signals for Hg^{2+} steadily increased up to 3% (v/v) HNO_3 and leveled off afterwards, while that for CH_3Hg^+ reached a plateau at and above 4% (v/v) HNO_3. The acidity of soils extracts - 5% (v/v) HNO_3 - was within the operational acid range of the CVG. $NaBH_4$ is essential for total Hg determination (Hg^{2+} + CH_3Hg^+) while $SnCl_2$ is used for selective reduction of Hg^{2+} [31,45]. The signals for CVG of CH_3Hg^+ were at the baseline levels (ca. 3000–3500 cps) when reducing agent was $SnCl_2$, which verified that $SnCl_2$ did not affect CH_3Hg^+, but performed similarly to $NaBH_4$ in reduction of Hg^{2+}.

The concentrations of $NaBH_4$ and $SnCl_2$ were examined from 0 to 2% (m/v) for both reagents and the results are illustrated in Fig. 2B. A concentration of 0.5% (m/v) $SnCl_2$ was sufficient for complete reduction of Hg^{2+} to Hg^0, while for CH_3HgCl no significant signal was observed up to 2% (m/v) $SnCl_2$. Higher concentrations of $SnCl_2$ (e.g., 1 to 2%) caused white deposition along the mixing coil due to the oxidation of $SnCl_2$ to SnO_2 when mixed with HNO_3. With $NaBH_4$, CVG signals for Hg^{2+} and CH_3HgCl showed maxima between 0.5 and 1% (m/v) $NaBH_4$. However, reduction of CH_3HgCl occurred consistently and more effectively with 1% (m/v) $NaBH_4$. Signals declined at 2% (m/v) $NaBH_4$ because of vigorous reaction between $NaBH_4$ and HNO_3 generating excessive H_2 that consequently changed the optimal the carrier gas flow rate.

3.5. Interferences and analytical merits

Potential chemical interferences from excess sulfide were investigated on vapor generation for 10 μg L^{-1} CH_3HgCl (n = 5, treatment) in 5% (v/v) HNO_3 that contained 30 μL of 0.35 mole L^{-1} $(NH_4)_2S$ in 2 mL volume (e.g., 0.005 mole L^{-1}). A 10 μg L^{-1} CH_3HgCl in 5% (v/v) HNO_3 (n = 5, control) was used as control solution. No significant suppression or enhancement occurred in vapor generation in comparison to the control solutions. The ratio between treatment and control signals (cps) (e.g., treatment/control) was 0.94 ± 0.04 (e.g., 94 ± 4%). This result was further verified by constructing calibration curves for CH_3HgCl (0 to 10 μg L^{-1}) using 1% (m/v) $NaBH_4$ as reducing agent. For ^{202}Hg isotope, the slopes for 5% (v/v) HNO_3 and 5% (v/v) HNO_3 + 0.005 mole L^{-1} $(NH_4)_2S$ were 84929 (R^2 = 0.998) and 82450 (R^2 = 0.999), respectively, demonstrating that $(NH_4)_2S$ matrix had no effect on vapor generation of CH_3HgCl in HNO_3 solutions. Using the same calibration curves, limits of detection (LODs) were calculated for blank solutions (n = 10) of 5% (v/v) HNO_3 and 5% (v/v) HNO_3 + 0.005 mole L^{-1} $(NH_4)_2S$. The LODs (concentration equivalent to 3s of the blank standard deviation) were 0.12 and 0.10 μg L^{-1} for ^{200}Hg and ^{202}Hg, respectively in the presence of $(NH_4)_2S$. For 5% (v/v) HNO_3 blanks, LODs were slightly lower but were not significantly different; 0.082and 0.085 μg L^{-1} for ^{200}Hg and ^{202}Hg, respectively. Eventually, the LODs were limited due to the persistent Hg background (ca. 4,000 cps) in the CVG system. Despite repetitive washings with 0.5% (m/v) potassium ferricyanide solution in 5% (v/v) HCl, background signals could not be fully eliminated. Still though, these LODs were sufficiently low to achieve determination of CH_3Hg^+ in the contaminated soils. Precision expressed as per cent relative standard deviation (%RSD) for 10 consecutive scans/readings (see Table 1) of 1.0 μg L^{-1} CH_3HgCl solution was around 2.1%. For 5 separate replicate measurements, precision varied between 3.1% and 6.4% for 1.0 μg L^{-1} CH_3HgCl standard solution.

3.6. Method validation and analysis of floodplain soils

For validation of the procedure, 0.2 g sub-samples (n = 5) of methylmercury sediment (SQC1238) and Estuarine sediment (ERM – CC580) were processed as described in section 2.8. CH_3Hg^+ determination was carried out using the optimized $NaBH_4$ method (5% HNO_3 vs 1% $NaBH_4$) by CVG-ICP-MS. The remaining of the CRM solutions were reanalyzed using $SnCl_2$ reduction method (e.g., 5% HNO_3 vs 0.5% $SnCl_2$) to determine the residual Hg^{2+}, if any, remained after $(NH_4)_2S$ precipitation. All calibration standards and blanks were prepared in 2-mL micro-centrifuge tubes and contained 30 μL of 0.35 mole L^{-1} of

$(NH_4)_2S$ solution to match with samples. The results for CRMs are summarized in Table 5 for ^{200}Hg and ^{202}Hg isotopes. The certified concentration for CH_3Hg^+ in SQC1238 was 10.00 ± 0.291 ng g^{-1}. ERM – CC580 was heavily Hg-contaminated estuarine sediment with trace amounts of CH_3Hg^+; certified values were 132 ± 3 μg g^{-1} for total Hg and 75 ± 4 ng g^{-1} for CH_3Hg^+. The experimental values were between 13.0 ± 3 and 13.2 ± 3 ng g^{-1} for SQC1238 and 79 ± 8 and 81 ± 7 ng g^{-1} for ERM – CC580, where uncertainties are expressed as standard deviation for five replicate samples (Table 5). It should be noted that acceptance limits of CH_3Hg^+ for SQC1238 ranged from 5.13 to as high as 14.9 ng g^{-1}, indicating inhomogeneity within sample. To improve precision, a sample size of 1.0 g is recommended for SCQ1238. In this work, measurements were made with 0.2 g samples. Despite higher mean CH_3Hg^+ levels, the experimental results agreed with the certified values; high uncertainty (e.g. variation) were likely related to small sample size and inhomogeneity. The analysis of the extracts with $SnCl_2$ reduction method showed about 0.15 μg L^{-1} Hg^{2+} in solution though SQC1238 was fully methylmercury chloride, which was indicative of the fact that background Hg signals could have also contributed to uncertainty due to very low CH_3Hg^+ levels. For ERM-CC580, better accuracy was achieved with the certified values. Mean CH_3Hg^+ levels were slightly higher; 79 to 81 ng g^{-1} that were equivalent to about 3.2 μg L^{-1} CH_3Hg^+ in 5 mL extracts (0.2 g sample). This concentration was about 0.2 μg L^{-1} higher (ca. 6.5%) than that for the certificate value (3.0 μg L^{-1} CH_3Hg^+ at 75 ng g^{-1}). Further, the concentration of residual Hg^{2+} in ERM-CC580 extracts varied from below LOD values (< LOD) to 0.180 μg L^{-1} when the remaining solutions were analyzed by CVG-ICP-MS using $SnCl_2$ reduction. These results indicated that residual Hg^{2+} was effectively eliminated by HgS precipitation. Eventually, the concentrations for Hg^{2+} from both CRM extracts were within the vicinity of LODs and hence were likely affected from the persistent background signals.

For analysis of floodplain soils, the same strategy was followed as for the method validation with CRMs. About 0.2 g sub-samples (n = 5) of the soils were subjected to the procedure. A set of SQC1238 and ERM – CC580 (n = 5) were also prepared freshly and analyzed concurrently by CVG-ICP-MS using $NaBH_4$ reduction method. The results are summarized in Table 6 for ^{200}Hg and ^{202}Hg isotopes. CH_3Hg^+ levels ranged from 30 to 51 ng g^{-1} indicating that CH_3Hg^+ levels in the floodplain soils were much lower than the total Hg levels (ca. 0.05% of total Hg). Further, they did not correlate with total Hg levels (R^2 = 0.01). Despite low levels, however, continuous leaching of CH_3Hg^+ to creek and riverine water could be a significant source for the persistent Hg levels in the waters of the EFPC at Oak Ridge, TN.

4. Conclusion

In this study, a new method is developed for selective extraction and determination of CH_3Hg^+ in soil and sediment samples, specifically in sediments contaminated with Hg. Between HCl and HNO_3, the latter was most suitable for selective extraction of CH_3Hg^+ from soils. Ultrasonic agitation in 5% (v/v) HNO_3 at room temperature afforded fast extraction of CH_3Hg^+ with minimal dissolution of inorganic Hg species. Residual Hg^{2+} leached into solution during extraction was effectively eliminated via HgS precipitation prior to vapor generation determinations. Both HNO_3 extraction and HgS coprecipitation steps

were vital components of the procedure to achieve inorganic Hg-free determinations of CH_3Hg^+, and to eliminate the deleterious effects and contamination that would otherwise occur inevitably in the presence of high Hg^{2+} matrix. Additionally, presence of small amounts of sulfide in analysis solutions had no adverse effects on chemical vapor generation of CH_3Hg^+. The procedure is simple and fast in contrast to chromatographic separation and speciation approaches, and more importantly it offers unique advantages for selective and accurate determination of trace amounts of highly toxic CH_3Hg^+ from heavily Hg-contaminated sediments and other environmental materials.

Acknowledgements

This work is by supported by the Department of Energy Office of Environmental Management (DOE-EM) Minority Serving Institution Partnership Program (MSIPP) managed by the Savannah River National Laboratory (SRNL) under Savannah River Nuclear Solutions (SRNS) contract 291435. Partial funding is provided the National Institutes of Health (NIH) through Research Centers in Minority Institutions (RCMI) Program (Grant No: G12RR013459) at Jackson State University (Grant No: 1461143). The views expressed herein are those of authors and do not necessarily represent the official views of the funding agencies, and any of their sub-agencies.

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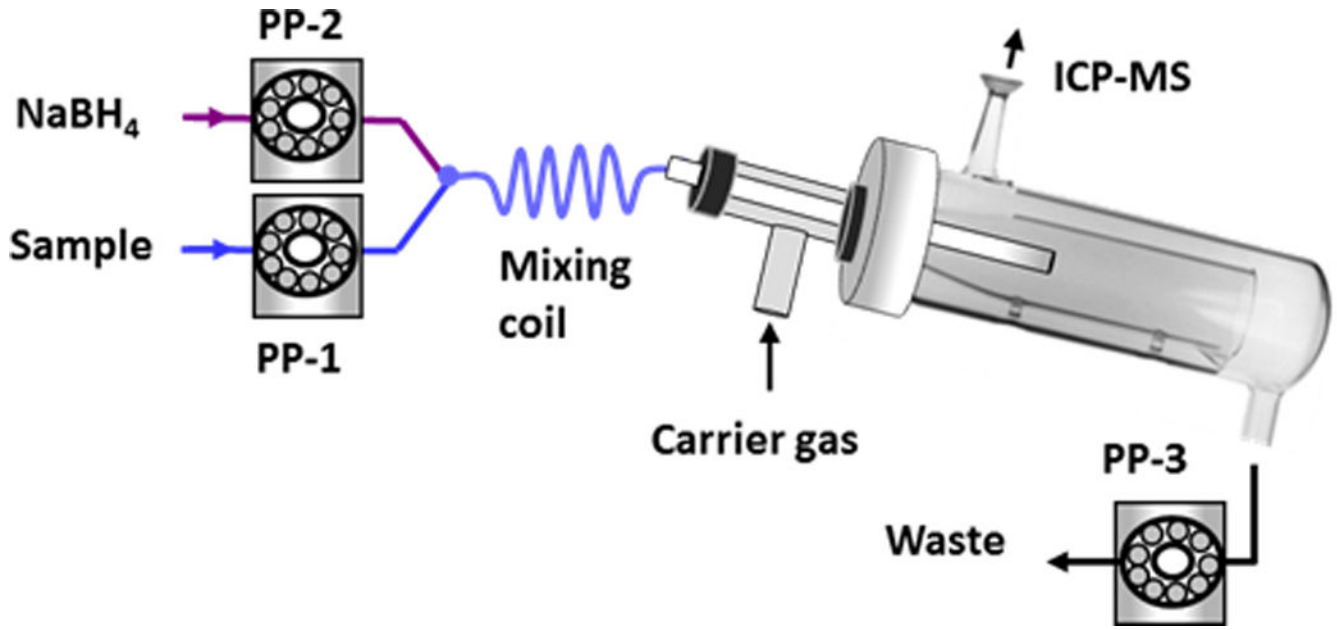

Fig. 1.
Schematic diagram of chemical vapor generation manifold for CH_3Hg^+ determination.

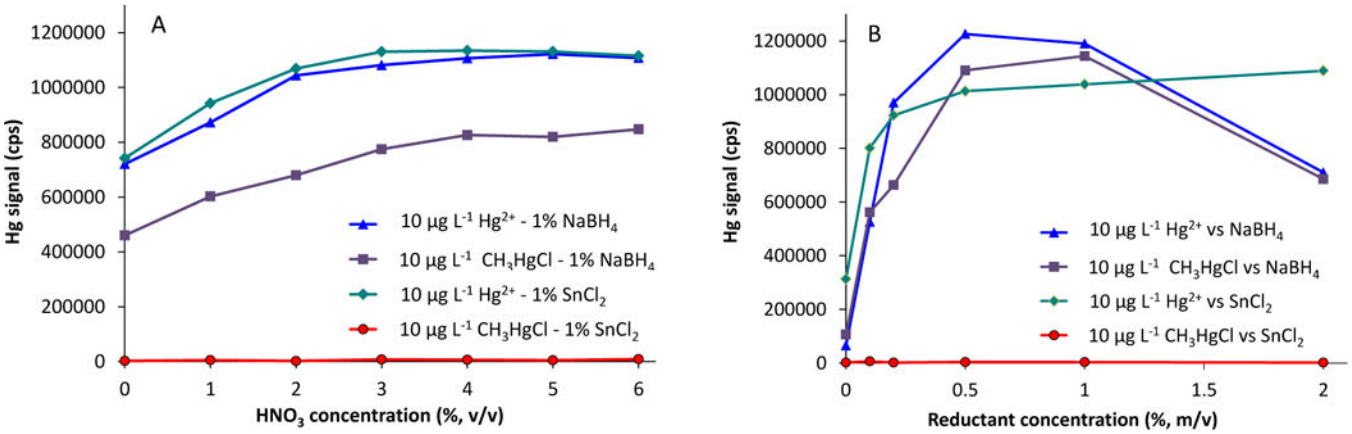

Fig. 2.

Vapor generation profiles from 10 µg L^{-1} CH_3Hg^+ and Hg^{2+}. A = Effect of HNO_3 concentration on Hg signals with 1% (m/v) $NaBH_4$ and 1% (m/v) $SnCl_2$. B = Effect of concentrations of $NaBH_4$ and $SnCl_2$ on vapor generation against 5% (v/v) HNO_3.

Table 1

Operating conditions for Varian 820MS ICP-MS in nebulization and vapor generation modes.

ICP-MS	Nebulization	Vapor generation
RF Power (kW)	1.4	1.4
Plasma argon flow (L min^{-1})	17	17
Auxiliary argon flow (L min^{-1})	1.8	1.8
Nebulizer argon flow (L min^{-1})	1.1	1.5
Sheath argon flow (L min^{-1})	0.15	0.2
Sampling depth (mm)	6.5	6.5
Sample flow rate (mL min^{-1})	0.5	0.7
Stabilization time (s)	15	50
Spray chamber temperature (°C)	4	4
Scan mode	Peak hopping	Peak hopping
Dwell time (ms)	20	50
Points/peak	1	1
Scans/peak	4	10
Scans/replicate	4	10

Table 2

Recoveries for CH_3Hg^+ and Hg^{2+} for extraction from artificially contaminated soils with dilute HCl and HNO_3 via shaking at room temperature. Values are mean ± standard deviation of five replicate extractions (n = 5).

Analyte	Medium	Shaking time (h)				
		0	1	2	6	24
		Recovery (%)				
Hg^{2+}	Water	0.95 ± 0.6	1.2 ± 1.0	2.0 ± 1.0	2.1 ± 1.0	4.2 ± 2.0
	5% HCl	24 ± 3	32 ± 6	34 ± 6	33 ± 5	38 ± 4
	10% HCl	57 ± 5	65 ± 3	66 ± 4	66 ± 6	71 ± 6
	5% HNO_3	3.5 ± 1.1	4.4 ± 2.1	3.9 ± 1.8	5.8 ± 1.0	15 ± 4
	10% HNO_3	3.6 ± 1.2	5.2 ± 2.2	4.1 ± 2.1	7.4 ± 1.5	18 ± 3
CH_3Hg^+	Water	2.1 ± 1.3	2.4 ± 2.1	2.5 ± 1.5	3.2 ± 1.2	8.8 ± 3.0
	5% HCl	68 ± 7	79 ± 8	84 ± 7	82 ± 3	98 ± 7
	10% HCl	78 ± 4	74 ± 4	74 ± 5	90 ± 5	91 ± 6
	5% HNO_3	71 ± 7	91 ± 8	89 ± 7	92 ± 7	101 ± 7
	10% HNO_3	95 ± 4	102 ± 6	102 ± 4	101 ± 4	103 ± 6

Table 3

Recoveries for CH_3Hg^+ and Hg^{2+} for extraction from artificially contaminated soils with dilute HCl and HNO_3 via ultrasounds agitation at room temperature. Values are mean ± standard deviation of five replicate extractions (n = 5)

Analyte	Medium	Sonication time (min)		
		0	3	6
		Recovery (%)		
Hg^{2+}	Water	0.95 ± 0.5	8.5 ± 3.2	12 ± 4
	5% HCl	24 ± 3	20 ± 5	29 ± 6
	10% HCl	58 ± 5	84 ± 6	101 ± 6
	5% HNO_3	3.5 ± 1.1	4.3 ± 1.8	4.7 ± 3.0
	10% HNO_3	3.5 ± 1.2	5.5 ± 2.5	8.6 ± 2.0
CH_3Hg^+	Water	2.1 ± 1.3	33 ± 6	58 ± 5
	5% HCl	65 ± 3	86 ± 4	98 ± 2
	10% HCl	82 ± 3	102 ± 5	101 ± 3
	5% HNO_3	71 ± 6	100 ± 4	102 ± 5
	10% HNO_3	94 ± 5	99 ± 4	104 ± 4

Table 4

Recoveries for CH_3Hg^+ and Hg^{2+} from hydroxide and sulfide coprecipitation procedures implemented with Bi(III) and Fe(III) with and without thiourea and L-cysteine additives, and ammonium sulfide. Recoveries for ammonium sulfide precipitation are for the concentration of CH_3Hg^+ and Hg^{2+} remained in solution after precipitation of 100 μg L^{-1} CH_3Hg^+ or Hg^{2+}. Results are given as mean ± standard deviation of 5 separate replicates (n = 5).

Matrix/Precipitant	**Recovery (%)**	
	CH_3Hg^+	Hg^{2+}
1.0 mg mL^{-1} Bi(III)	4.3 ± 1.9	8.8 ± 2.0
1.0 mg mL^{-1} Fe(III)	8.8 ± 1.0	23 ± 5
0.1% L-cysteine	2.5 ± 0.7	3.1 ± 0.6
1.0 mg mL^{-1} Bi(III) + 0.1% L-cysteine	2.8 ± 0.7	12.4 ± 0.8
1.0 mg mL^{-1} Fe(III) + 0.1% L-cysteine	7.6 ± 3.5	56 ± 7
0.1% Thiourea	13 ± 2	3.2 ± 2.2
1.0 mg mL^{-1} Bi(III) + 0.1% Thiourea	67 ± 2.0	26 ± 4.2
1.0 mg mL^{-1} Fe(III) + 0.1% Thiourea	25 ± 8	64 ± 4.2
0.005 mol L^{-1} Ammonium sulfide	102 ± 3	2.1 ± 1.1

Table 5

Methylmercury concentrations measured from methlymercury sediment (SQC1238) and estuarine sediment (ERM – CC580) certified reference materials by the optimized CVG-ICP-MS. Values are given as mean ± standard deviation of 5 separate replicates (n = 5).

Sample	CH_3Hg^+ concentration ($ng\ g^{-1}$)	Certified value ($ng\ g^{-1}$)	
	^{200}Hg	^{202}Hg	
SQC1238	13.0 ± 3	13.2 ± 3	10.00 ± 0.291
ERM – CC580	81 ± 7	79 ± 8	75 ± 4

Table 6

Total mercury and methylmercury concentrations measured from the floodplain soils from Oak Ridge, TN. Total Hg levels measured via acid digestion of Hg-contaminated soils. Values are given as mean ± standard deviation of 3 replicates (n = 3) for total Hg and 5 separate replicates (n = 5) for methylmercury.

Sample	CH_3Hg^+ (ng g^{-1})		Total Hg (μg g^{-1})
	^{200}Hg	^{202}Hg	^{202}Hg
Soil 1	43 ± 6	45 ± 6	85.3 ± 1.6
Soil 2	40 ± 8	41 ± 8	68.6 ± 5.3
Soil 3	40 ± 12	40 ± 13	57.4 ± 6.0
Soil 4	32 ± 3	33 ± 4	72.5 ± 4.5
Soil 5	32 ± 9	33 ± 9	95.7 ± 3.4
Soil 6	50 ± 10	51 ± 15	66.5 ± 1.4
Soil 7	46 ± 5	47 ± 5	87.5 ± 6.6
Soil 8	30 ± 2	31 ± 3	67.7 ± 3.3
Soil 9	33 ± 9	33 ± 8	85.9 ± 6.5
Soil 10	42 ± 11	42 ± 12	83.5 ± 6.7