



Good Practice Guide on the Storage and Stability of HgCl_2 Solutions and Salts for Use with Mercury Gas Generators

19NRM03 SI-Hg

Sophie Page¹, Philip Dunn¹, John Entwisle¹, Sarah Hill¹, Heidi Goenaga-Infante¹, Jan Koesters², Carsten Röllig²

¹ LGC Ltd, National Measurement Laboratory, Queens Road, Teddington, Middlesex, TW11 0LY, UK

² TÜV Rheinland, AM Grauen Stein, 51105 Köln, Germany

Lead partner: LGC

Version No.: 3

Due date deliverable: M26

Actual submission date: 30/09/2023



This project has received funding from the EMPIR programme co-financed by the Participating States and from the European Union's Horizon 2020 research and innovation programme.



The EMPIR initiative is co-funded by the European Union's Horizon 2020 research and innovation programme and the EMPIR Participating States

Table of Contents

Table of Contents.....	3
1. Introduction	4
1.1 SI-Hg Project Background.....	4
1.2 Scope.....	4
2. Methods.....	5
2.1 Six Month Stability Study	5
2.1.1 Ambient and Shock Heat – Reagents and Storage Bottles	5
2.1.2 Ambient and Shock Heat – Instrumental Analysis.....	5
2.1.3 Fridge Temperature – Reagents and Storage Bottles.....	6
2.1.4 Fridge Temperature – Instrumental Analysis.....	7
2.2 Chromatographic Stability Study	8
2.2.1 Reagents and Storage Bottles	8
2.2.2 Instrumental Analysis.....	9
2.3 Storage of HgCl ₂ Salts.....	10
2.3.1 Reagents, Sample Preparation, and Instrumental Analysis.....	10
3. Results.....	11
3.1 Storage of HgCl ₂ Solutions Results.....	11
3.2 Storage of HgCl ₂ Salts Results	16
4. Recommendations	16
4.1 Recommendations for the Storage of HgCl ₂ Solutions	16
4.2 Recommendations for the Storage of HgCl ₂ Salts.....	17
References	17
Appendices.....	18

1. Introduction

1.1 SI-Hg Project Background

Mercury (Hg) is a global pollutant that can be carried by air around the world, sequestered into soils, water, and plants, and re-volatilised back into the air. Mercury and in particular elemental Hg vapour is toxic to neurological and developmental systems. Therefore, acute and chronic exposure to Hg can cause damage to the brain, kidneys, cardiovascular function, and foetuses ¹.

Current atmospheric Hg levels are 500 % above natural levels, and anthropogenic sources are responsible for 50 % of atmospheric Hg emissions, 65 % of which is from stationary combustion.² It is therefore important to monitor atmospheric Hg, and stationary combustion sources such as coal-fired power plants are required by EU and UK legislation to monitor and limit their Hg emissions. Mercury measurement systems and detectors that are used in the field to monitor Hg emissions are typically calibrated using Hg gas generators that produce a calculable mass flow of Hg. However, the Hg gas generators are not certified against primary standards, and therefore lack traceability. Although great efforts have been made in developing primary Hg standards and SI-traceable calibration methods for different Hg species, there are no standardised procedures that ensure the uptake of the developed metrological traceability by calibration and testing laboratories, and in the field. Certification protocols to determine the output of elemental mercury (Hg⁰) and oxidised mercury (Hg^{II}) gas generators, are of fundamental importance to guarantee the accuracy and comparability of the mercury measurement data in Europe and globally.

1.2 Scope

In the field, HgCl₂ solutions are used in liquid evaporative Hg gas generators such as the Optoseven (Optoseven Ltd & VTT Ltd, Espoo, Finland) and HovaCal (IAS GmbH, Oberusel, Germany) to calibrate Hg measurement systems and detectors for Hg^{II}. The generators vaporise a HgCl₂ solution of known concentration and mix the vapour with a carrier gas to produce a Hg^{II} vapour of calculable mass flow that can be varied to create a multi-point calibration curve. The HgCl₂ solutions are typically diluted from pure salts to create stock solutions that are stored long-term and diluted to create working solutions for the generators.

In [EMPIR 16ENV01 MercOx](#) a traceable calibration methodology for liquid evaporative HgCl₂ generators was developed. However, some essential performance characteristics such as the storage and stability of the stock HgCl₂ solutions and salts used in the generators were not assessed. Feedback from industry and research partners such as VDZ (Verein Deutscher Zementwerke e.V.), VTT (Technical Research Centre of Finland), CNR (National Research Council, Italy), and TÜV Rheinland revealed that a wide variety of storage conditions and protocols are used throughout industry including different storage temperatures, bottle types, matrices, and concentrations. Solution stability under these different storage conditions is important to constrain because it is a major control on the concentration of Hg^{II} gas produced by liquid evaporative generators. If the HgCl₂ solution stability changes over time, uncertainty is introduced to the output of liquid evaporative HgCl₂ generators, and therefore the calibration of Hg detectors used in the field. This guide therefore aims to advise on the best practice

for the storage of HgCl₂ salts and solutions derived from such salts based on findings from elemental and chromatographic stability experiments.

2. Methods

2.1 Six Month Stability Study

The storage conditions tested in this study were chosen to closely reflect the range of conditions used in the field by facilities that use Hg^{II} liquid evaporative generators for Hg detector calibration. To determine suitable test conditions, SI-Hg industry and research partners including VDZ, TÜV Rheinland, VTT, and CNR were consulted. A balance was struck between the range of conditions tested and experiment feasibility. The finalised storage conditions tested were storage temperature, bottle material, Hg concentration, solution matrix, and storage duration (**Appendix A**). Three different storage temperatures were tested; ambient temperatures (stored on open shelving in a sample preparation laboratory at LGC (Teddington, UK), exposed to indirect sunlight), fridge temperatures (in a fridge at TÜV Rheinland (Cologne, Germany)), and heating to 60 °C for 24 hours in an oven at LGC to simulate transport to hot climates in a “shock heat” experiment. For each temperature a different set of individual HgCl₂ solutions of 50, 200, and 1000 µg kg⁻¹ of Hg plus blanks were prepared from EMPLURA HgCl₂ salt (Lot: A0583617714) purchased from Merck Life Science UK Ltd (Gillingham, UK) in each of the three matrices (0.1 % (v/v) HCl, 0.024 % (v/v) HNO₃ + 0.0144 % (v/v) HCl, and 0.125 µg g⁻¹ HNO₃ + 0.125 µg g⁻¹ HCl) and were stored in each of the three bottle types (FEP (fluorinated ethylene propylene), FLPE (fluorinated high density polyethylene), and borosilicate), resulting in 36 different solutions per storage temperature. Over 6 months, the Hg concentration of each solution was measured monthly to monitor long-term stability.

2.1.1 Ambient and Shock Heat – Reagents and Storage Bottles

Twenty-four 500 mL low particulate/low metals FEP Nalgene™ bottles with ETFE (ethylene tetrafluoroethylene) screw caps were obtained from Thermo Scientific. Upon purchase the FEP bottles contained Romil UpA HNO₃ or HCl acids from Romil Ltd (Waterbeach, Cambridge, UK) and were used for this project after the acids had been used. Twenty-four 250 mL borosilicate bottles with PTFE capped lids were purchased from Sigma. Twenty-four 500 mL Nalgene narrow-mouth FLPE bottles from Thermo Scientific were donated by CNR.

Before the HgCl₂ solutions were prepared, all bottles were thoroughly cleaned. Firstly, the bottles were filled, shaken, and rinsed with ultrapure 18.2 MΩ cm Elga water (Elga, Veolia, High Wycombe, UK) three times. Then, shaken with 50 % SpA HNO₃, then 50 % SpA HCl, both purchased from Romil Ltd. Then the bottles were filled, shaken, and rinsed with 18.2 MΩ cm Elga water three more times. Finally, the bottles were filled with 18.2 MΩ cm Elga water and stored at room temperature until use.

2.1.2 Ambient and Shock Heat – Instrumental Analysis

Prior to measurement, all standards and all HgCl₂ solutions apart from the blank solutions were diluted with 10 % (v/v) HCl prepared from concentrated UpA grade hydrochloric acid (HCl) purchased from Romil Ltd and 18.2 MΩ cm Elga water. The blank HgCl₂ solutions were not diluted, and the 50, 200, and 1000 µg kg⁻¹ solutions were diluted to 2.0, 2.5, and 4 µg kg⁻¹ respectively. Four calibration standards were prepared from a 985 mg kg⁻¹ Hg standard stock solution purchased from Romil Ltd. NIST SRM

3133 (National Institute of Standards and Technology) was used as a quality control material and diluted to $3.0 \mu\text{g kg}^{-1}$. A 10 % (v/v) HNO_3 + $250 \mu\text{g kg}^{-1}$ Au solution was prepared from a 921 mg kg^{-1} Au standard stock solution purchased from Romil Ltd as an ICP-MS rinse between samples during measurement.

At the start of each month from July 2022 until January 2023 the concentration of Hg in each of the ambient temperature Hg^{II} solutions was determined at LGC using an Agilent 8800 ICP-MS/MS using a four-point external calibration. The typical measurement parameters used for Hg measurement over the six month study are outlined in **Fout! Verwijzingsbron niet gevonden..** Mercury mass-to-charge ratios 199, 200, 201, and 202 were measured in spectrum mode using the single quadrupole with no transition, and 100 replicates of 50 sweeps, with a 0.1 second integration time per mass. Each sample had an uptake time of 160 seconds at 0.1 rps, then a stabilisation time of 30 seconds at 0.1 rps followed by 1 minute of acquisition. A solution of 10 % (v/v) HNO_3 + 250 ppb Au was used to rinse between samples for 150 seconds at 0.5 rps. All samples were analysed in a matrix of 10 % (v/v) HCl with an internal standard mix of 10 ppb Tl and 20 ppb Pt in 10 % (v/v) HCl. The total acquisition time for each sample was three minutes. NIST SRM 3133 was used as an independent standard to monitor quality. NIST SRM 3133 recoveries over the 6 months of analysis varied between 96.4 % and 104.3 % with an average recovery of 99.8 %.

The shock heat HgCl_2 solutions were measured before and after heating to 60 °C for 24 hours in February 2023 using the same method as the ambient temperature solutions.

Table 1: Summary of typical measurement parameters used for each of the Hg concentration measurement sessions.

Parameter	Agilent 8800 ICP-MS/MS Setting
Nebuliser	Micromist
Spray chamber	Scott double pass
Spray chamber temperature	20 °C
Autosampler	Cetac ASX-520
RF Power	1450 W
Plasma Gas	15 L min ⁻¹
Carrier Gas	0.20 L min ⁻¹
Makeup Gas	0.95 mL min ⁻¹
Nebuliser Pump Speed	0.10 rps
Measurement Mode	Single Quad
Isotope product ions	²⁰⁰ Hg
Integration Time/Mass	0.1 sec

2.1.3 Fridge Temperature – Reagents and Storage Bottles

For the fridge temperature experiments, FEP, FLPE, and borosilicate bottles from the same sources as the bottles used in the ambient and shock-heat experiments were filled, shaken, and rinsed with ultrapure 18.2 MΩ cm Elga water three times. Then, shaken with 50 % SpA HNO_3 , then 50 % SpA HCl. Then the bottles were filled, shaken, and rinsed with 18.2 MΩ cm Elga water three more times. Finally, the bottles were filled with 18.2 MΩ cm Elga water and stored at room temperature until use.

All standard calibration solutions used in the fridge experiments were stored in borosilicate bottles. Each bottle has only ever contained one type of calibration solution that is always the same Hg concentration. The Hg concentration of the calibration standards follow the fixed standard concentrations used for testing at TÜV Rheinland.

2.1.4 Fridge Temperature – Instrumental Analysis

The fridge temperature analyses used two different measurement techniques; cold vapour coupled to atomic absorption spectroscopy (CV-AAS) and a pyrolyzer coupled to AAS (Pyr-AAS). CV-AAS reduces divalent Hg to the elemental form in an acidic medium by a reducing agent. In absorption solutions, this is carried out with SnCl₂ after pre-reduction with hydroxylamine hydrochloride. The elemental mercury is then expelled using a stream of inert gas and transported as an atomic gas into a cuvette. The absorbances are determined at a wavelength of 253.65 nm in the beam path of an atomic absorption spectrometer. The concentrations can be determined with the aid of a reference curve. All CV-AAS analyses were performed using MLS DMA 80 L cold vapour mercury analyser (Mikrowellen und Laborsysteme Leutkirch, Germany). Pyr-AAS reduces monovalent or divalent Hg to elemental Hg in a thermocatalytic reaction. Aqueous samples are weighed into a vessel filled with quartz sand and the weight automatically transferred from the analytical balance to the analyser. Drying and thermal decomposition then take place in an oxygen atmosphere. Mercury and other combustion products are released and passed through the catalyst, eliminating any interfering substances such as halogens and nitrogen or sulphur oxides. Then the Hg is selectively retained by amalgamation, while other combustion by-products are flushed out. The amalgamation furnace is heated, and the Hg rapidly released again. The Hg is finally measured quantitatively by atomic absorption at 253.65 nm in several measuring cells of the optical spectrometer. All Pyr-AAS were conducted using a MLS DMA 80 evo III direct mercury analyser (Mikrowellen und Laborsysteme Leutkirch, Germany).

Table 2 and **Table 3** outline the typical operating parameters for the CV-AAS and Pyr-AAS methods respectively.

Table 2: Summary of typical CV-AAS measurement parameters used for the fridge temperature stability measurements.

Parameter	Instrument Setting
Amalgamation time H	15s
Recording time A	14 s
Follow-up time N	30 s
Follow-up time N0	70 s
Cleaning time R	28 s
Permissible start temperature	350° C
Maximum heating temperature	850°C
Maximum heating time	720s
Catalyst temperature	600°C
Amalgam heating temperature	850°C
Amalgam heating time H	12s
Rest temp. amalgam heating	200°C
Cuvette temperature	100°C
Rinsing time S	60s
Recording time A	30s
Minimum temperature rise of drying	0°C/s
Maximum temperature rise of drying	15°C/s

Flushing pump	
Cleaning time	8s
Waiting time	30s
Target volume	4.9943 ml,
Pump capacity	0.247 ml/s

Table 3: Summary of typical Pyr-AAS measurement parameters used for the fridge temperature stability measurements.

Parameter	Instrument Setting
Absorbance/emission limit cuvette 0	42
Permissible starting temperature	250°C
Maximum heating temperature	1000°C
Maximum heating time	720s
Catalyst temperature	600°C
Purge time S	60s
Amalgam heating temperature	850°C
Amalgam heating time H	12s
Rest temp. amalgam heating	175°C
Recording time, A	30s
Cuvette temperature	120°C
Detector	
Spectrometer Signal source	AAS 3-fold (analogue)
Reference channel	X18
Lamp temperature maximum	28.0°C
Lamp-temperature control	deactivated
Lamp-temperature span	15.0°C
Aperture time	0.70s

Estimation of the measurement uncertainty for the determination of Hg (component Hg) in aqueous solution by CV-AAS (with amalgam trap) and Pyr-AAS for the concentration range: 0.01 - 5 µg/L was based on DIN ISO 11352:2013 Water quality - Estimation of measurement uncertainty based on validation and control data. The relative expanded uncertainty was 6.16 % ($k = 2$) and 3.74 % ($k = 2$) for all CV-AAS and Pyr-AAS results respectively.

2.2 Chromatographic Stability Study

The 36 HgCl₂ solutions stored for 6 months at ambient temperatures were retained and used for a comparative study of the Hg species present in HgCl₂ solutions after storage under different conditions. The stored HgCl₂ solutions were analysed after one year of storage and compared to freshly prepared solutions.

2.2.1 Reagents and Storage Bottles

Prior to measurement via HPLC-ICP-MS, all HgCl₂ solutions were diluted to 40 µg kg⁻¹ by the addition of a 0.1 % (m/v) cysteine, 1 mM acetic acid mobile phase adjusted to pH 4 with ammonia solution.² The ammonia solution and acetic acid were purchased from Sigma-Aldrich (Poole, Dorset, UK), the cysteine HCl (from non-animal sources) was purchased from Merck Life Science UK Ltd. Species-specific Hg standards were also prepared including a methylmercury (LGC Standards Ltd, Teddington, UK), Hg^I (Hg₂Cl₂ Alfa Asears, Ward Hill, Massachusetts, USA), Hg^{II} (new bottle of EMPLURA HgCl₂) and a Hg⁰ (from

a liquid Hg thermometer). Standards were diluted to approximately $4 \mu\text{g kg}^{-1}$ in the mobile phase, apart from the new EMPLURA HgCl_2 solution which was diluted to $40 \mu\text{g kg}^{-1}$ to match the concentration of the stored HgCl_2 solutions.

2.2.2 Instrumental Analysis

The chromatographic stability study employed an Agilent 1100 HPLC coupled to an Agilent 7700 ICP-MS/MS. **Table 4** outlines the instrument parameters used for the analysis. Three main peaks were observed from the four Hg standards (**Figure 1**). However, these standards were not used for quantification, and instead only for context about possible peak identities given the potential for Hg disproportionation.

Table 4: Summary of typical HPLC and ICP-MS/MS measurement parameters used for the Hg chromatography measurements.

Parameter	Instrument Setting
HPLC	Isocratic
Column	ProteCol™ C18G 120 Å 3 μm particle size, length 150 mm 4.6 mm id manufactured by Trajan (Victoria, Australia)
Mobile phase	0.1% (m/v) Cysteine (HCl), 1 mM acetic acid, adjusted to pH 4 with ammonia
Column Oven temperature [°C]	20
Autosampler temperature [°C]	5
Injection volume [μL]	100
Eluent flow rate [mL/min]	0.9
Column back pressure [bar]	140
Run time per sample [mins]	17
ICP-MS	Agilent 7700 ICP-MS/MS
Forward Power [W]	1590
Carrier gas flow (L/min)	1.20
Spray chamber temperature [°C]	2
Sampling depth [mm]	6.9
Extraction lens 1 [V]	0
Extraction lens 2 [V]	-160
Octopole bias [V]	-8
KED [V]	5
Acquisition mode	Single quad
Monitored mass m/z	199,200,201,202
Integration time/Mass [sec]	0.23

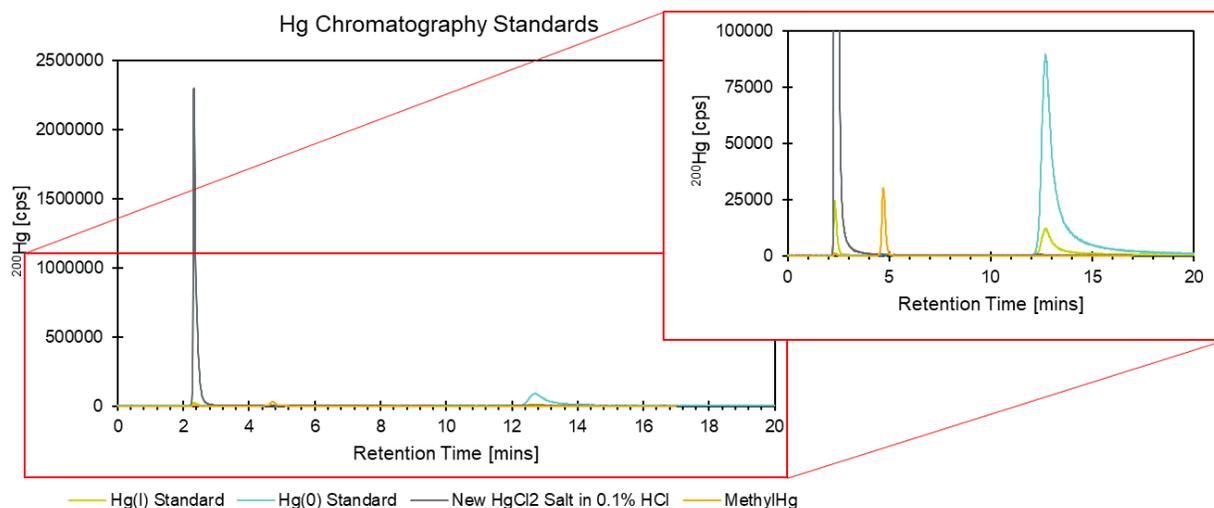


Figure 1: Chromatogram of the four Hg standards run during the chromatographic stability study. The green line relates to the ^{202}Hg counts per second of the Hg^{I} standard, the blue line relates to the Hg^{0} standard, the grey line relates to the new HgCl_2 solution, and the orange line relates to the methylmercury standard. The Hg^{0} , Hg^{I} and methylmercury standards were diluted to approximately $4 \mu\text{g kg}^{-1}$ in the mobile phase, and the HgCl_2 solution is $40 \mu\text{g kg}^{-1}$ diluted first in 0.1 % HCl and then in the mobile phase.

2.3 Storage of HgCl_2 Salts

The best practice for storing pure HgCl_2 salts that are diluted to prepare working and storage HgCl_2 solution was also investigated. The EMLURA HgCl_2 salt is hygroscopic and therefore repeat exposure to atmosphere and improper storage may affect the moisture content and ultimately the purity and Hg concentration of the salt. Traditionally the best practice for storage of hygroscopic solids is in a desiccator. However, the effects of repeated exposure of the salt to atmosphere during repeated opening to produce stock and working solutions should be assessed. Therefore, two new EMLURA HgCl_2 salts (batch number: K50571117) were purchased from Merck Life Science Ltd. Both were stored in the same desiccator. Over three months one bottle was opened weekly for two minutes, and the salt stirred with a spatula, the other was left sealed. The total Hg concentration of both salts was determined by isotope dilution mass spectrometry (IDMS). IDMS is a high precision, low uncertainty quantification technique and therefore gave the best resolution to detect any differences in Hg mass fraction between the bottles.

2.3.1 Reagents, Sample Preparation, and Instrumental Analysis

The IDMS protocol used the double matched method (**Appendix B**) in which both the sample and a primary Hg standard are spiked with isotopically enriched material (^{199}Hg) to give a gravimetric ratio close to 1 for $^{200}\text{Hg} : ^{199}\text{Hg}$ was used. This method minimises the effects of instrument detector linearity, mass bias effects, and isotope spike calibration. The blends were diluted to a target concentration of $10 \mu\text{g kg}^{-1}$ prior to direct nebulisation to a quadrupole ICP-MS.

A ^{199}Hg enriched isotopic standard from Oak Ridge National Laboratory, USA (Lot: 168490) was used as the isotopic spike for the IDMS blends, the concentration of which was determined by reverse IDMS in 2022. NIST SRM 3133 was used as the primary calibration standard, thereby providing traceability, and was diluted in 10 % (v/v) HCl (prepared with UpA hydrochloric acid (Romil Ltd) and 18.2 MΩ cm Elga

water) and subsequently in 1 % (v/v) HCl. 0.2 g aliquots of the HgCl₂ salts were diluted using 1% (v/v) HCl serial dilutions to a gravimetrically accurate concentration of 1300 ± 13 µg kg⁻¹. Six replicate blends were prepared for each HgCl₂ salt from the 1300 µg kg⁻¹ salt dilutions and the ¹⁹⁹Hg isotopic spike to give a ²⁰⁰Hg : ¹⁹⁹Hg ratio of close to 1. Blends were diluted to a target concentration of 10 µg kg⁻¹. The calibration blend was prepared from NIST SRM 3133 and the ¹⁹⁹Hg isotopic spike in the same manner as the samples.

Analyses used an Agilent 7700 ICP-MS operated in standard mode. Samples were introduced into the plasma via a micro-flow quartz concentric nebuliser and a Scott double pass spray chamber cooled to 2 °C. All samples and standard were spiked with 1000 mg kg⁻¹ Au (Romil Ltd) to obtain a concentration of 5 mg kg⁻¹ and reduce Hg memory effects and instrument wash out times.³ For quality control purposes, an independent replicate of the unopened HgCl₂ salt was gravimetrically spiked with NIST SRM 3133. Mercury recoveries were 102 %. A secondary independent Hg standard (Romil Ltd) and achieved recoveries of 101 %, and an additional secondary independent Hg standard (expired NIST SRM 3133) achieved recoveries of 103 % and 100 %.

3. Results

3.1 Storage of HgCl₂ Solutions Results

The ambient temperature stability experiments show that all solutions except for those stored in FLPE bottles in 0.125 µg g⁻¹ HNO₃ + 0.125 µg g⁻¹ HCl experienced no significant change in Hg concentration considering the associated expanded measurement uncertainty ($k = 2$) over the six month study (**Appendix C**). Of the three solutions in which the Hg concentration decreased, the 50 ng g⁻¹ solution experienced an 89 % decrease over the six months, the 200 ng g⁻¹ solution a 74 % decrease, and the 1000 ng g⁻¹ solution a 30 % decrease (**Figure 2**). The percentage change over the six month study for all other solutions varied between -3 % and 5 % (**Appendix C**). All blank solutions had Hg concentrations below the limit of detection (0.11 µg kg⁻¹).

In the shock heat experiment, the same three solutions stored in FLPE bottles with 0.125 µg g⁻¹ HNO₃ + 0.125 µg g⁻¹ HCl showed a significant decrease in Hg concentration after heating at 60 °C for 24 hours. The percentage change in Hg concentration for the 50 ng g⁻¹ solution was -85 %, -61 % for the 200 ng g⁻¹ solution, and -34 % for the 1000 ng g⁻¹ solution (**Figure 3** and **Appendix E**). All percentage change in Hg concentration for all other solutions did not exceed 4%.

The results of the fridge temperature experiments show that all solutions stored in 0.125 µg g⁻¹ HNO₃ + 0.125 µg g⁻¹ HCl irrespective of bottle type experienced a significant reduction in Hg concentration over the six month study period (**Figure 4** and **Appendix D**). The only HgCl₂ solution stored in 0.125 µg g⁻¹ HNO₃ + 0.125 µg g⁻¹ HCl that did not experience significant Hg loss was the 1000 ng g⁻¹ borosilicate solution, which experienced a 6 % loss over the six months. The borosilicate bottles experienced the least Hg loss of the 0.125 µg g⁻¹ HNO₃ + 0.125 µg g⁻¹ HCl solutions, and again higher Hg storage concentrations also experienced less Hg loss. The FLPE bottles containing 0.125 µg g⁻¹ HNO₃ + 0.125 µg g⁻¹ HCl experienced the most Hg loss. A 75 % reduction in Hg concentration was observed in the 50 ng g⁻¹ HgCl₂ solution, an 89 % reduction in the 200 ng g⁻¹ solution, and an 88 % reduction in the

1000 ng g⁻¹ solution. A less than 10 % reduction in Hg concentration was observed in the other solutions over the six month study period.

The chromatographic stability study results show that all solutions except for those stored in FLPE bottles and 0.125 µg g⁻¹ HNO₃ + 0.125 µg g⁻¹ HCl have very similar chromatograms that overlap with the new HgCl₂ salt chromatogram and the Hg^I, Hg^{II}, and Hg⁰ standard peak retention times at 3 and 13 minutes (**Figure 5** and **Figure 6**). The 0.125 µg g⁻¹ HNO₃ + 0.125 µg g⁻¹ HCl solutions stored in FLPE bottles have a series of small but unidentified peaks between the 4 and 10 minutes (**Figure 6**). These samples are the same samples that experienced significant Hg loss during the ambient, fridge and shock heat stability experiments. In addition, the recovery of total Hg in those three solutions compared to the last month of the ambient stability study results were 43 % for the 50 ng g⁻¹ solution, 54 % for the 200 ng g⁻¹ solution, and 85 % for the 1000 ng g⁻¹ solution (**Appendix F**), suggesting further Hg loss between the end of the elemental stability study and start of the chromatographic stability study.

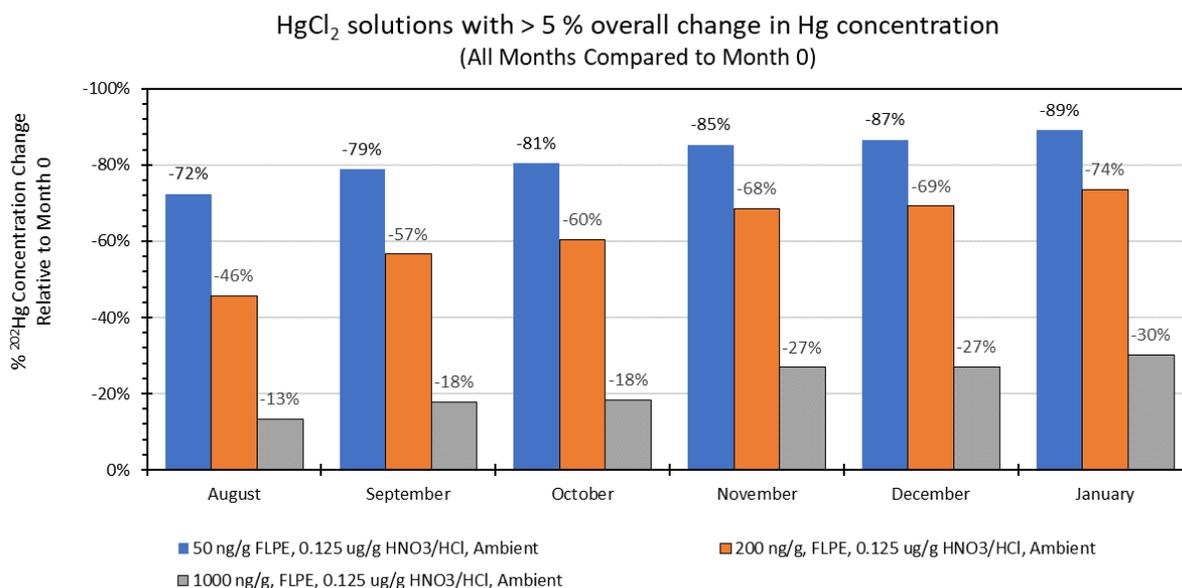


Figure 2: The percentage change in Hg concentration compared to month 0 for each month between August and January for the three samples with greater than a 5% reduction in concentration.

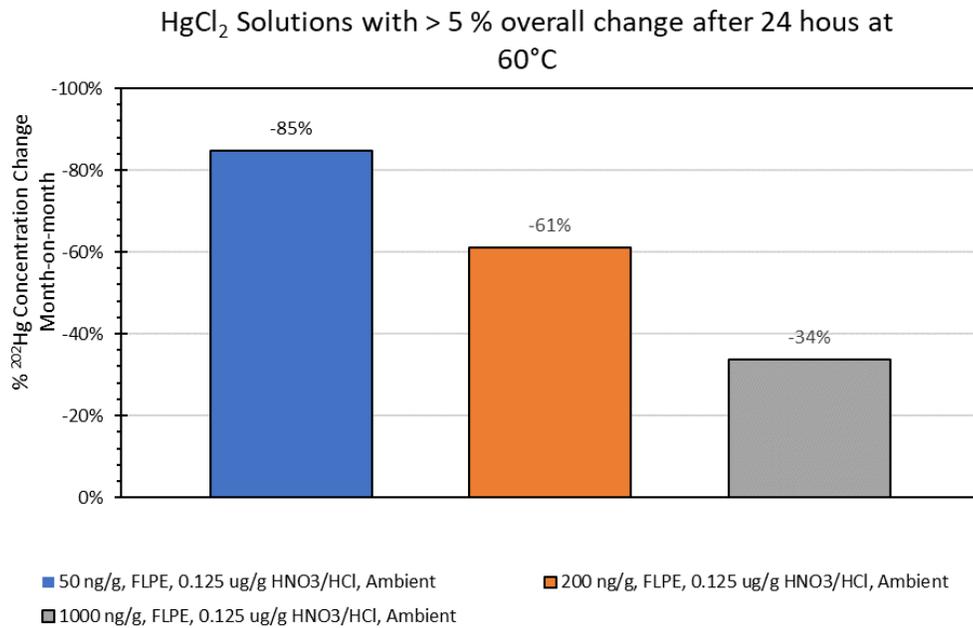


Figure 3: The percentage change in Hg concentration after heating the HgCl₂ solutions to 60 °C for 24 hours for the three samples with greater than a 5% reduction in concentration.

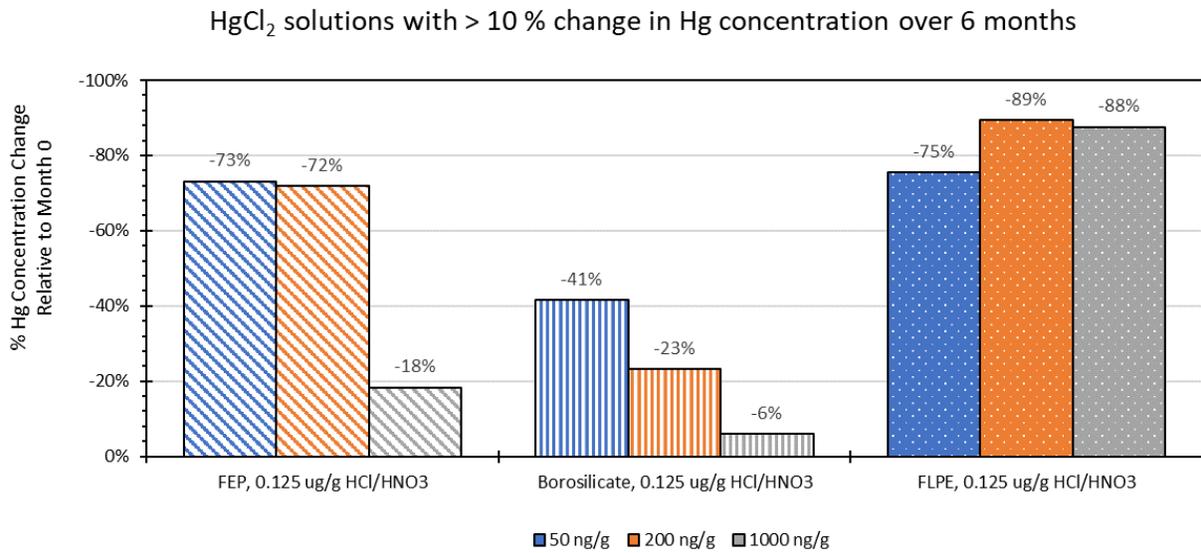


Figure 4: The percentage change in Hg concentration after storage of HgCl₂ solutions at fridge temperatures for six months. The solutions included are all of those that experienced more than a 10% change in Hg concentration, except for the 1000 ppb borosilicate solution which was included for context.

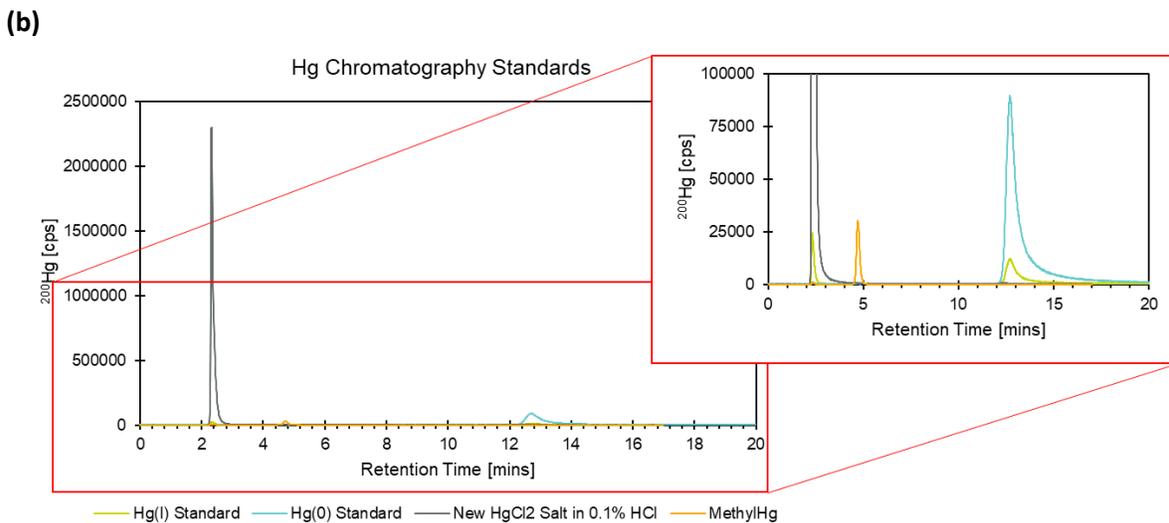
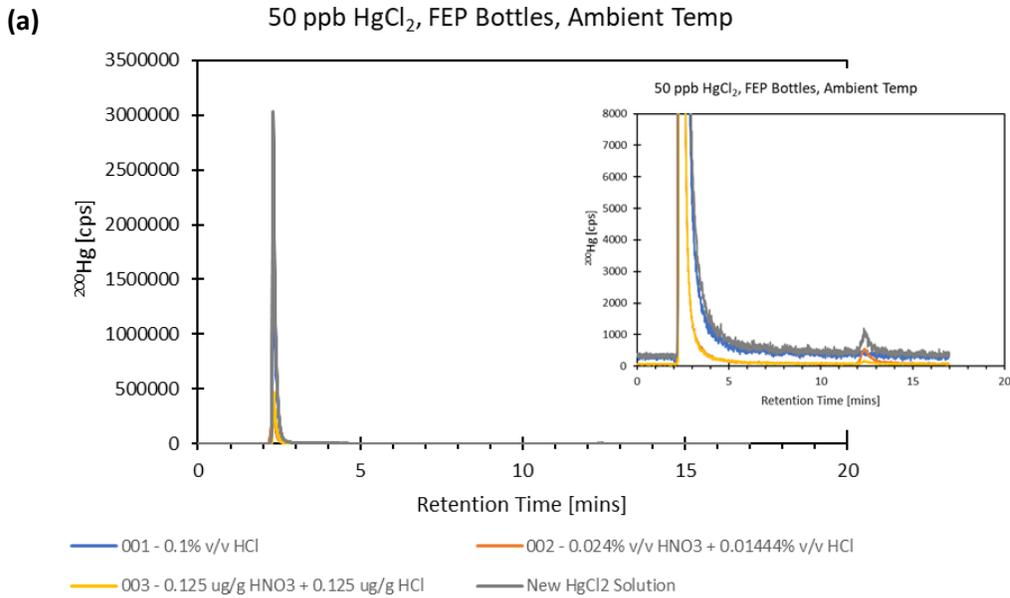


Figure 5: (a) Mercury species chromatogram for the three HgCl₂ solutions stored in FEP bottles at ambient temperatures and a newly prepared HgCl₂ solution. The x-axis is retention time in minutes and the y-axis is the ²⁰⁰Hg counts per second. These chromatograms are typical of all HgCl₂ solutions stored in FEP and borosilicate bottles, regardless of Hg concentration or storage matrix, and solutions stored in FLPE bottles with 0.1 % (v/v) HCl and 0.024 % (v/v) HNO₃ + 0.0144 % (v/v) HCl. (b) Chromatogram of the four Hg standards run during the chromatographic stability study for. The green line relates to the ²⁰²Hg counts per second of the Hg(I) standard, the blue line relates to the Hg⁰ standard, the grey line relates to the new HgCl₂ solution, and the orange line relates to the methylmercury standard. The 3 minute peak has the same retention time as the Hg^I and Hg^I standards, and the 13 minute peak has the same retention time as peaks for the Hg⁰ and Hg^I standards.

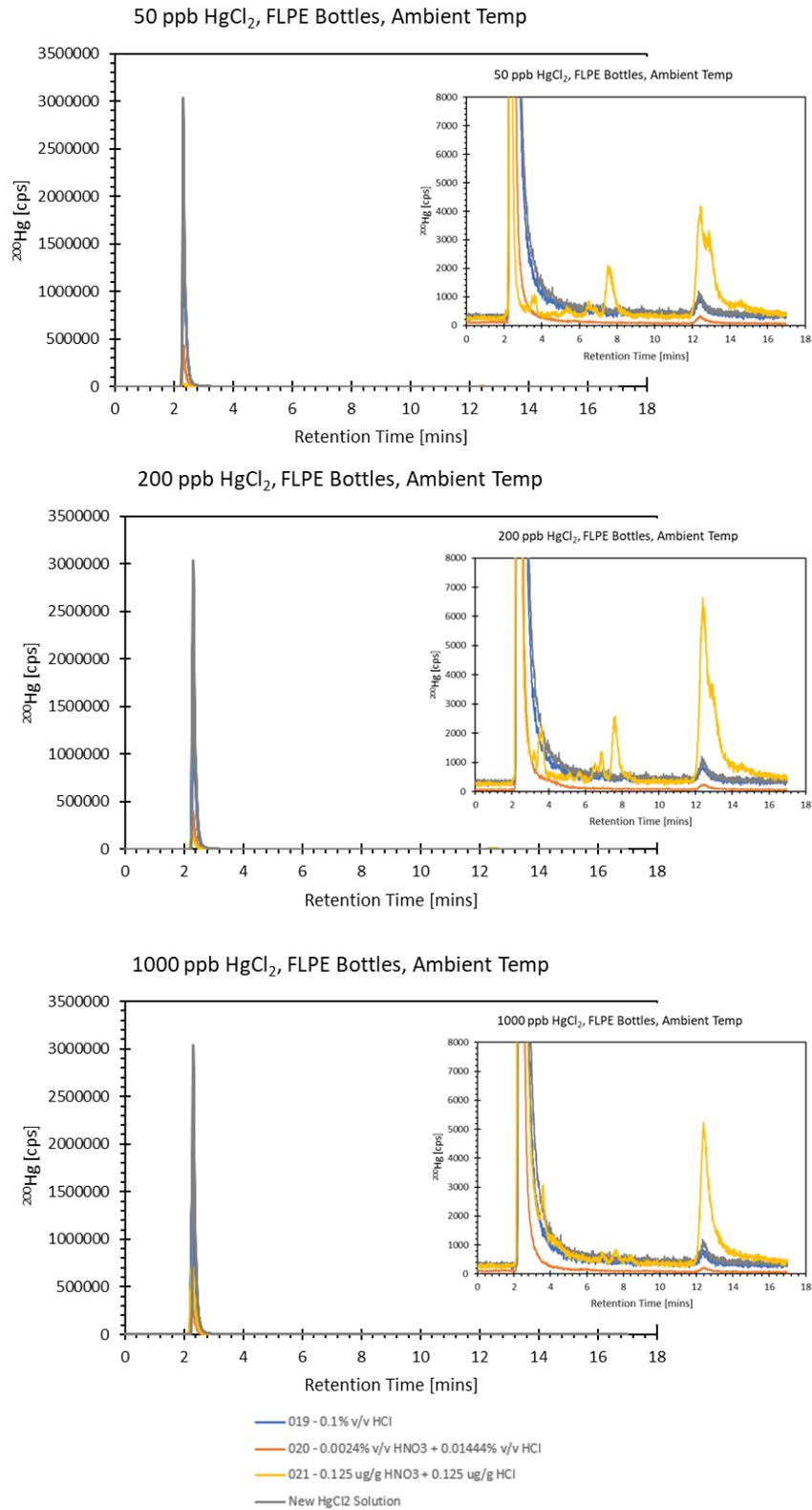


Figure 6: Mercury species chromatogram for the three HgCl₂ solutions stored in borosilicate bottles at ambient temperatures. The x-axis is retention time in minutes and the y-axis is the ²⁰⁰Hg counts per second. The peaks between 4 and 10 minutes are

unidentified, and only present in these three solutions. The 3 minute peak has the same retention time as the Hg^I and Hg^{II} standards, and the 13 minute peak has the same retention time as peaks for the Hg⁰ and Hg^I standards.

3.2 Storage of HgCl₂ Salts Results

The sealed HgCl₂ salt had an expected Hg mass fraction of 1299 µg kg⁻¹ (Uc = 8.9 µg kg⁻¹; k = 2) and an IDMS result of 1300 µg kg⁻¹ (Uc = 28 µg kg⁻¹; k = 2) (**Table 5**). The opened bottle, similarly, had an expected Hg mass fraction of 1298 µg kg⁻¹ (Uc = 9.0 µg kg⁻¹; k = 2) and an IDMS result of 1313 µg kg⁻¹ (Uc = 27 µg kg⁻¹; k = 2) (**Table 5**). Therefore, both bottles had expected Hg mass fractions that were within the expanded measurement uncertainty of their associated IDMS result, and when measurement uncertainty was account for, there was no observable difference in Hg mass fraction between HgCl₂ stored in a sealed bottle and a bottle that's been opened for two minutes once a week for three months.

Table 5: Mass fraction and expanded uncertainty (k = 2) for the expected and experimental mass fractions of Hg in the sealed and opened HgCl₂ salts stored in a desiccator for three months. The expected mass fraction was calculate using gravimetric data from the preparation of the sample blends for IDMS analysis. The experimental results are the measured Hg mass fractions from the IDMS analysis of the salts.

HgCl ₂ Bottle Status	Expected (Gravimetric)		Experimental Results		
	Hg Mass Fraction	Expanded Uncertainty (k = 2)	Hg Mass Fraction	Expanded Uncertainty (k = 2)	Relative Expanded Uncertainty
	(µg kg ⁻¹)	(µg kg ⁻¹)	(µg kg ⁻¹)	(µg kg ⁻¹)	(µg kg ⁻¹)
Sealed	1299	8.9	1300	28	2.10%
Opened	1298	9.0	1313	27	2.10%

4. Recommendations

4.1 Recommendations for the Storage of HgCl₂ Solutions

- Solutions prepared from HgCl₂ salts for use with Hg gas generators should only be stored in 0.1 % (v/v) HCl or 0.024 % (v/v) HNO₃ + 0.0144 % (v/v) HCl.
- Solutions prepared from HgCl₂ salts should only be stored in FEP or borosilicate bottles.
- Solutions can be stored at either fridge or ambient temperatures using the bottle types and matrices described in recommendations 4.1a and 4.1b.
- Temporary heating for up to 24 hours (e.g. during transport) does not affect the Hg mass fraction of the solutions.
- FLPE bottles are not suitable storage containers for HgCl₂ salts in solution with low concentration HCl/HNO₃ since considerable Hg loss has been observed when stored at both ambient and fridge temperatures.
- HgCl₂ solution matrix decisions should always consider the measurement technique, since for example HNO₃/HCl mixes may degrade gold traps due to the production of NOCl.

4.2 Recommendations for the Storage of HgCl₂ Salts

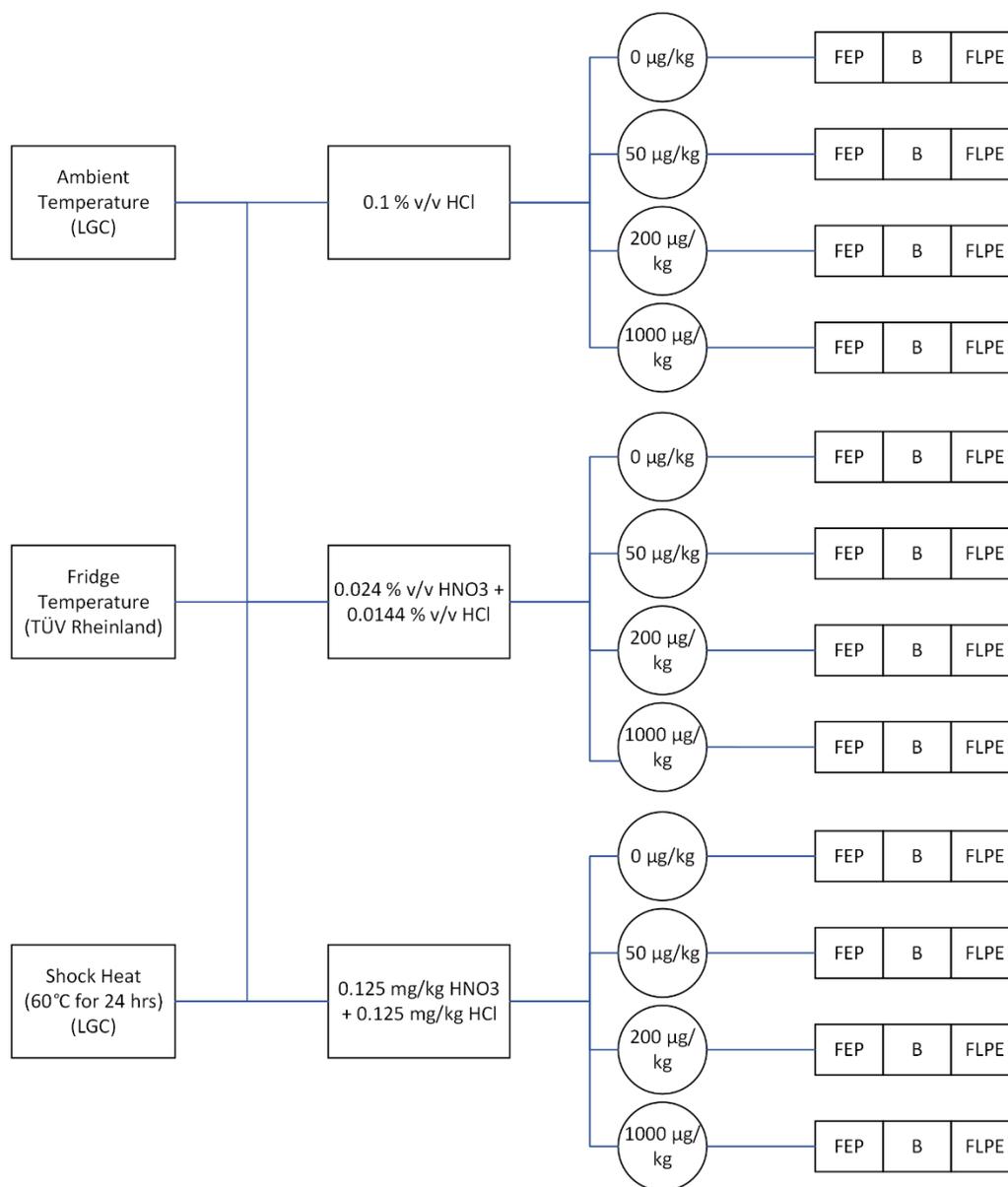
- a) HgCl₂ salts are hygroscopic and should always be stored in a desiccator.
- b) Stored HgCl₂ salts bottles can be safely opened for up to two minutes at a time for solution preparation for three months without observable effects on the mass fraction of Hg.
- c) No recommendations can be given for practices after three months from opening since it is out of scope.

References

- (1) Karagas, M. R.; Choi, A. L.; Oken, E.; Horvat, M.; Schoeny, R.; Kamai, E.; Cowell, W.; Grandjean, P.; Korrick, S. Evidence on the Human Health Effects of Low-Level Methylmercury Exposure. *Environmental Health Perspectives*. June 2012, pp 799–806. <https://doi.org/10.1289/ehp.1104494>.
- (2) UN Environment. *GLOBAL MERCURY ASSESSMENT*; Geneva, Switzerland, 2019.
- (3) Entwisle, J. Determination of Mercury in Microwave Digests of Foodstuffs by ICP-MS. *Agilent Application Note* **2004**, 1–4.

Appendices

Appendix A: Tree diagram showing the experiment design for the HgCl_2 solution stability study. For each temperature condition (ambient, fridge, and shock heat) 1.5 L of each matrix solution was prepared. From each of these solutions, 300 mL of 0, 50, 200, and 1000 $\mu\text{g}\cdot\text{g}^{-1}$ Hg was prepared from a pure HgCl_2 salt. 100 mL of each solution was then decanted into either a fluorinated ethylene propylene (FEP), fluorinated high density polyethylene (FLPE), or borosilicate (B) bottle. Due to the number of variables, bottles, and measurements the preparation and analysis was split between LGC and TÜV. LGC prepared and analysed the ambient temperature and shock heat solutions according to the above diagram, and TÜV Rheinland prepared and analysed the fridge temperature solutions according to the above diagram.



Appendix B: Equation for double matched bracketing IDMS.

$$(1) \quad w'_X = \lambda \cdot w_Z \cdot \frac{m_Y}{m_X} \cdot \frac{m_{Zc}}{m_{Yc}} \cdot \frac{R_Y - R'_B \cdot \frac{R_{Bc}}{R'_{Bc}}}{R'_B \cdot \frac{R_{Bc}}{R'_{Bc}} - R_X} \cdot \frac{R_{Bc} - R_Z}{R_Y - R_{Bc}} \cdot \frac{\sum R_{iX}}{\sum R_{iZ}}$$

w'_X mass fraction of analyte in sample X obtained from one measurement

w_Z mass fraction of analyte in primary standard Z

m_Y mass of spike Y added to sample X to prepare the blend B (=X+Y)

m_X mass of sample X added to the spike Y to prepare the blend B (=X+Y)

m_{Zc} mass of primary standard solution Z added to the spike Y to make the calibration blend Bc (Bc=Y+Z)

m_{Yc} mass of spike Y added to the primary standard solution Z to make the calibration blend Bc

R'_B measured isotope amount ratio of the sample blend B

R'_{Bc} measured isotope amount ratio of the calibration blend Bc

R_{Bc} gravimetric value of the isotope amount ratio of the calibration blend

R_X isotope amount ratio of sample X

R_Z isotope amount ratio of primary standard Z

$\sum R_X$ sum of isotope amount ratios in sample X

$\sum R_Z$ sum of isotope amount ratios in primary standard Z

Appendix C: Results summary of the monthly Hg concentration measurements of the HgCl₂ solutions. Each concentration is based on the signal of ²⁰⁰Hg using either a ²⁰⁵Tl or ¹⁹⁵Pt internal standard since this routinely gave the best calibration curve and the best NIST SRM 3133 quality control recoveries. All concentrations are reported in µg.kg⁻¹ with an expanded measurement uncertainty of 15 % (95% C.I.). The matrices are (1) 0.1% (v/v) HCl; (2) 0.024% (v/v) HNO₃ + 0.0144% (v/v) HCl; (3) 0.125 µg.g⁻¹ HNO₃ + 0.125 µg.g⁻¹ HCl. The samples highlighted in red have an overall percentage change in Hg concentration over the study of more than 5 %. The expanded uncertainty for each sample is the standard deviation of the mean of the monthly concentration measurements x 2.

Bottle ID	Approx Hg Conc (µg kg ⁻¹)	Bottle Type	Matrix	Storage Temp	Dilution & Blank Corr Hg Concentration (µg kg ⁻¹) - Expanded Uncertainty ± 15 %							Relative U _c (%)	Change % (Month 0 vs Month 6)
					Month 0	Month 1	Month 2	Month 3	Month 4	Month 5	Month 6		
					05/07/22	03/08/22	05/09/22	03/10/22	03/11/22	06/12/22	03/01/23		
001	50	FEP	1	Ambient	54.0	51.4	45.0	51.4	52.1	53.2	54.2	5	0
002	50	FEP	2	Ambient	48.7	47.6	47.7	48.3	48.0	48.1	50.4	2	3
003	50	FEP	3	Ambient	52.9	49.7	52.9	51.8	51.2	51.3	54.3	2	3
010	50	Borosilicate	1	Ambient	53.8	51.7	48.3	54.4	52.6	52.5	54.5	3	1
011	50	Borosilicate	2	Ambient	50.0	47.6	47.3	50.3	48.1	47.6	49.3	2	-1
012	50	Borosilicate	3	Ambient	52.7	52.0	51.5	49.9	51.1	51.1	55.5	3	5
019	50	FLPE	1	Ambient	53.3	51.6	52.2	53.2	51.9	51.4	54.5	2	2
020	50	FLPE	2	Ambient	52.6	51.0	51.8	52.4	50.6	50.8	52.1	1	-1
021	50	FLPE	3	Ambient	48.7	13.4	10.3	9.5	7.1	6.5	5.3	81	-89

028	200	FEP	1	Ambient	218	212	215	218	209	212	219	1	0
029	200	FEP	2	Ambient	207	196	204	206	199	201	216	2	4
030	200	FEP	3	Ambient	195	192	194	197	188	195	199	1	2
037	200	Borosilicate	1	Ambient	217	210	208	215	208	220	218	2	0
038	200	Borosilicate	2	Ambient	208	201	203	204	199	210	206	1	-1
039	200	Borosilicate	3	Ambient	197	180	194	185	186	189	197	3	0
046	200	FLPE	1	Ambient	219	206	209	216	229	210	223	3	2
047	200	FLPE	2	Ambient	204	194	206	206	198	202	208	2	2
048	200	FLPE	3	Ambient	198	108	86.0	78.4	62.4	61.0	52.3	41	-74

055	1000	FEP	1	Ambient	968	912	961	966	954	1000	958	2	-1
-----	------	-----	---	---------	-----	-----	-----	-----	-----	------	-----	---	----

056	1000	FEP	2	Ambient	979	926	940	948	944	970	993	2	1
057	1000	FEP	3	Ambient	984	949	987	953	954	993	1002	2	2
064	1000	Borosilicate	1	Ambient	978	965	965	985	946	978	986	1	1
065	1000	Borosilicate	2	Ambient	981	959	967	956	940	1001	1008	2	3
066	1000	Borosilicate	3	Ambient	981	969	951	983	955	991	966	1	-2
073	1000	FLPE	1	Ambient	979	933	951	932	951	963	972	1	-1
074	1000	FLPE	2	Ambient	992	933	968	938	946	982	960	2	-3
075	1000	FLPE	3	Ambient	995	863	818	812	728	726	696	10	-30

Appendix D: Results summary of the monthly Hg concentration measurements of the HgCl₂ solutions stored at fridge temperatures at TÜV Rheinland. The Hg concentrations from months 0 to 3 were determined by cold vapour coupled to AAS and months 4 to 6 by pyrolysis-AAS. All concentrations are reported in µg.kg⁻¹ with an expanded measurement uncertainty of 6.16 % (95% C.I.) for the CV-AAS results, and 3.74 % (95 % C.I.) for the Pyr-AAS results. The matrices are (1) 0.1% (v/v) HCl; (2) 0.024% (v/v) HNO₃ + 0.0144% (v/v) HCl; (3) 0.125 µg.g⁻¹ HNO₃ + 0.125 µg.g⁻¹ HCl. The samples highlighted in red have an overall percentage change in Hg concentration over the study of more than 10%. The expanded uncertainty for each sample is the standard deviation of the mean of the monthly concentration measurements x 2.

Bottle ID	Approx Hg Conc (µg kg ⁻¹)	Bottle Type	Matrix	Storage Temp	Dilution & Blank Corr Hg Concentration (µg kg ⁻¹)							Relative U _c (%)	Percentage Change (Month 0 vs Month 6)
					Month 0	Month 1	Month 2	Month 3	Month 4	Month 5	Month 6		
					CV-AAS	CV-AAS	CV-AAS	CV-AAS	Pyr-ASS	Pyr-AAS	Pyr-AAS		
004	50	FEP	1	Fridge	55	48	54	48	49	49	51	4	-7
005	50	FEP	2	Fridge	54	47	53	49	50	48	49	4	-10
006	50	FEP	3	Fridge	39	9	8	6	8	9	10	69	-73
013	50	Borosilicate	1	Fridge	48	48	48	51	51	50	50	2	4
014	50	Borosilicate	2	Fridge	48	48	48	50	50	*85	48	2	0
015	50	Borosilicate	3	Fridge	47	44	42	42	39	36	28	12	-41
022	50	FLPE	1	Fridge	48	48	48	50	51	50	50	2	4
023	50	FLPE	2	Fridge	48	48	47	49	49	46	46	2	-3
024	50	FLPE	3	Fridge	30	5	5	4	4	5	7	84	-75

031	200	FEP	1	Fridge	220	188	200	214	199	197	190	4	-14
032	200	FEP	2	Fridge	214	191	189	208	195	195	193	4	-10

033	200	FEP	3	Fridge	169	91	85	79	53	54	48	38	-72
040	200	Borosilicate	1	Fridge	206	189	192	200	199	196	198	2	-4
041	200	Borosilicate	2	Fridge	191	188	201	197	199	190	194	2	1
042	200	Borosilicate	3	Fridge	184	173	168	191	137	134	141	11	-23
049	200	FLPE	1	Fridge	190	188	204	200	206	195	192	3	1
050	200	FLPE	2	Fridge	191	188	191	201	195	190	191	2	0
051	200	FLPE	3	Fridge	121	8	9	10	10	11	13	122	-89

058	1000	FEP	1	Fridge	1026	950	950	999	992	1016	1020	2	-1
059	1000	FEP	2	Fridge	1002	962	884	1027	980	1005	1003	4	0
060	1000	FEP	3	Fridge	915	864	852	883	636	704	747	10	-18
067	1000	Borosilicate	1	Fridge	946	956	990	1009	1020	1006	1007	2	6
068	1000	Borosilicate	2	Fridge	955	958	958	1008	1016	1010	1005	2	5
069	1000	Borosilicate	3	Fridge	957	950	970	993	813	824	900	6	-6
076	1000	FLPE	1	Fridge	967	978	969	1032	1005	1009	1006	2	4
077	1000	FLPE	2	Fridge	968	978	978	1017	996	988	960	1	-1
078	1000	FLPE	3	Fridge	756	337	302	221	128	101	94	63	-88

Appendix E: Summary of results from the shock heat experiments. The Hg concentration of the HgCl₂ solutions was measured as soon as the solutions were made, and then again after 24 hours at 60 °C. Each concentration is based on the signal of ²⁰⁰Hg using a ¹⁹⁵Pt internal standard. All concentrations are reported in µg kg⁻¹ with an expanded measurement uncertainty of 15 % (95 % C.I.). The matrices are (1) 0.1 % (v/v) HCl; (2) 0.024 % (v/v) HNO₃ + HCl; (3) 0.125 µg g⁻¹ HNO₃ + 0.125 µg g⁻¹ HCl. The samples highlighted in red have an overall percentage change in Hg concentration over the study of more than 5 %. The expanded uncertainty associated with each sample is the standard deviation of the mean of the control and 60 °C concentration measurements multiplied by two.

Bottle ID	Approx Hg Conc (µg kg ⁻¹)	Bottle Type	Matrix	Dilution & Blank Corr ²⁰⁰ Hg Concentration (µg kg ⁻¹) – Expanded Uncertainty ± 15 % (95% C.I.)				
				Ambient Control	24 hrs at 60 °C	Percentage Change	Uc (k = 2)	Relative Uc (%)
				21/02/23	22/02/23			
007	50	FEP	1	48.6	50.4	4%	1.7	3%
008	50	FEP	2	49.3	49.9	1%	0.6	1%
009	50	FEP	3	49.6	50.7	2%	1.1	2%
016	50	Borosilicate	1	48.5	50.2	3%	1.7	3%
017	50	Borosilicate	2	49.1	50.1	2%	1.1	2%
018	50	Borosilicate	3	49.5	50.0	1%	0.5	1%
025	50	FLPE	1	48.5	48.9	1%	0.4	1%
026	50	FLPE	2	47.1	48.0	2%	0.9	2%
027	50	FLPE	3	29.0	4.4	-85%	24.6	147%

034	200	FEP	1	191	198	4%	6.9	4%
035	200	FEP	2	191	196	3%	4.9	3%
036	200	FEP	3	193	195	1%	1.9	1%
043	200	Borosilicate	1	193	197	2%	4.1	2%
044	200	Borosilicate	2	193	195	1%	1.9	1%
045	200	Borosilicate	3	191	194	2%	2.9	2%
052	200	FLPE	1	185	191	3%	5.9	3%
053	200	FLPE	2	185	192	4%	6.7	4%
054	200	FLPE	3	152	59.2	-61%	93.1	88%

061	1000	FEP	1	994	988	-1%	6.2	1%
062	1000	FEP	2	986	985	0%	0.4	0%
063	1000	FEP	3	968	986	2%	17.2	2%
070	1000	Borosilicate	1	984	997	1%	13.0	1%
071	1000	Borosilicate	2	995	986	-1%	9.1	1%
072	1000	Borosilicate	3	978	984	1%	6.9	1%
079	1000	FLPE	1	974	975	0%	1.7	0%
080	1000	FLPE	2	973	960	-1%	13.2	1%
081	1000	FLPE	3	836	553	-34%	283	41%

Appendix F: Semi-quantitative summed peak area data for the chromatographic stability study. The peak area data was roughly calculated using the peak area of the Hg^{II} standard as a reference. The peak area of the 13 minute peak was decay corrected using an exponential decay correction equation since peak area decay with increasing time between sample preparation and measurement was observed. The correction had a minimal effect on the semi-quantitative calculated Hg concentrations since in all cases it made up less than 0.5 % of the total Hg.

Bottle ID	Approx Hg Conc ($\mu\text{g kg}^{-1}$)	Bottle Type	Matrix	Storage Temp	Month 6 Total Hg Result ($\mu\text{g kg}^{-1}$)	Semi-quant Summed Chromatographic Peaks ²⁰² Hg ($\mu\text{g kg}^{-1}$)	Summed Peaks % Recovery Compared to Last Totals Result
001	50	FEP	1	Ambient	54.2	51.5	95%
002	50	FEP	2	Ambient	50.4	46.3	92%
003	50	FEP	3	Ambient	54.3	50.8	94%
010	50	Borosilicate	1	Ambient	54.5	51.7	95%
011	50	Borosilicate	2	Ambient	49.3	45.9	93%
012	50	Borosilicate	3	Ambient	55.5	50.2	90%
019	50	FLPE	1	Ambient	54.5	49.9	92%
020	50	FLPE	2	Ambient	52.1	48.1	92%
021	50	FLPE	3	Ambient	5.3	2.27	43%

028	200	FEP	1	Ambient	219	213	97%
029	200	FEP	2	Ambient	216	201	93%
030	200	FEP	3	Ambient	199	190	95%
037	200	Borosilicate	1	Ambient	218	211	97%
038	200	Borosilicate	2	Ambient	206	200	97%
039	200	Borosilicate	3	Ambient	197	190	97%
046	200	FLPE	1	Ambient	223	207	93%
047	200	FLPE	2	Ambient	208	197	94%
048	200	FLPE	3	Ambient	52.3	28.7	55%

055	1000	FEP	1	Ambient	958	958	100%
056	1000	FEP	2	Ambient	993	974	98%
057	1000	FEP	3	Ambient	1002	976	97%
064	1000	Borosilicate	1	Ambient	986	958	97%
065	1000	Borosilicate	2	Ambient	1008	971	96%
066	1000	Borosilicate	3	Ambient	966	967	100%
073	1000	FLPE	1	Ambient	972	956	98%
074	1000	FLPE	2	Ambient	960	957	100%
075	1000	FLPE	3	Ambient	696	593	85%