



## Introduction

Recent intercomparison studies<sup>1,2</sup> have highlighted the various approaches laboratories have taken to prepare water samples for methylmercury (MeHg) analysis. Although U.S. EPA Method 1630<sup>3</sup> recommends distilling samples after addition of ammonium pyrrolidine dithiocarbamate (APDC) to modify the sample matrix to improve MeHg recovery, in practice, a variety of reagents are used.

Preparation Method	2014	2015
Non-Distillation	11	8
L-cysteine	3	0
APDC	10	14
CuSO <sub>4</sub>	2	4
No Reagents	0	3
Distillation, Other	2	4

Table 1. Sample preparation methods used by the participants in the 2014 and 2015 BPI Interlaboratory Comparison Study for THg and MeHg in Waters

In the 2014 and 2015 interlaboratory comparison studies conducted by Brooks Rand Instruments (BRI), the majority of participants used distillation to prepare the aqueous samples for subsequent analysis of MeHg. In these studies, of those laboratories that utilized distillation, the primary reagents used were L-cysteine, APDC, and copper sulfate (CuSO<sub>4</sub>), while other laboratories used no additional reagents (Table 1).

The interlaboratory comparison study allows inferences to be made about the effectiveness of these sample preparation methods based on the performance scores received by the labs using them. However, because there are many other factors that contribute to a performance score, such as the analytical method, cleanliness of the labware, and detector sensitivity, it is not possible to determine from the study results if there is a significant difference in the effectiveness of the different distillation reagents.

In order to eliminate the lab-to-lab variability, a comparison was conducted to test the four distillation reagents most commonly reported by the labs in the interlaboratory comparison study. Method blanks, blank spikes (laboratory fortified blanks), natural water samples, and matrix spikes (laboratory fortified samples) were prepared on the same day with each of the reagents tested. Measured concentrations of the natural samples and recoveries of the blank spikes and matrix spikes were compared. Additionally, a method detection limit (MDL) was calculated for each reagent tested. To our knowledge, this is the first reported comparison of these four distillation methods in a single study.

## Methods

All samples were prepared using a Brooks Rand Instruments Methyl Mercury Distillation System with the aluminum heating block set to 138 °C. The selected reagent was added to a 50-mL aliquot of sample in the distillation vial (Table 2). The distillate was brought up to a final volume of 58 mL and approximately 30 mL of distillate was analyzed for MeHg. Due to the number of samples, the samples were distilled over two consecutive days. Distillates were analyzed by aqueous phase ethylation, Tenax trap preconcentration, gas chromatographic separation, isothermal deposition, and cold vapor atomic fluorescence spectrometry (CVAFS) using a Brooks Rand Instruments MERX-M Automated Methyl Mercury Analytical System (Figure 1). The analysis was performed on the third consecutive day. Due to the large capacity and high throughput of the MERX-M, all samples were analyzed in the same analytical run, preventing any day-to-day variability.

Table 2. Volume of each distillation reagent added to a 50-mL aliquot of sample

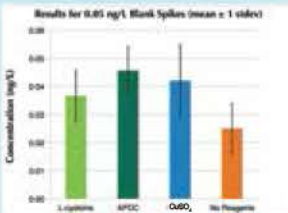
Distillation Reagent	Concentration	Volume Added (mL)
L-cysteine	0.2%	0.2
APDC	1%	0.22
CuSO <sub>4</sub>	20%	0.828



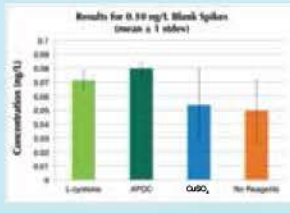
Figure 1. Brooks Rand Instruments MERX-M Automated Methyl Mercury Analytical System

## Results

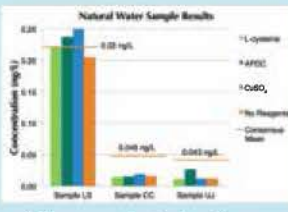
The method blanks for all experimental distillation reagents tested produced mean values for MeHg of  $\leq 0.012$  ng/L.



Graph A. The average results for 8 replicates of 0.05 ng/L blank spikes, with error bars representative of one standard deviation. For each distillation reagent tested, replicates were prepared to 0.5% (v/v) with concentrated hydrochloric acid (HCl).



Graph B. The average results for 4 replicates of 0.10 ng/L blank spikes, with error bars representative of one standard deviation. For each distillation reagent tested, replicates were prepared to 0.5% (v/v) with concentrated hydrochloric acid (HCl).



Graph C. Mean experimental results for duplicate analysis of the samples from the BRI 2015 Interlaboratory Comparison Study. The three samples were Lake Sammamish (LS), Cranberry Creek (CC), and Uncle John Creek at Chapman Cove (UJ). LS and CC were both freshwater samples, and UJ was an estuarine water sample. Site LS showed a significant sample discoloration. The consensus value is the mean value from the 39 reported results in the study.



Graph D. Recoveries for matrix spikes on the experimental samples. Sample LS was spiked at 0.5 ng/L, and Samples CC and UJ were each spiked at 0.2 ng/L. The matrix spike prepared with APDC yielded a 58% recovery for Sample LS, below the recovery criteria of 65-135% specified in EPA Method 1630. All other matrix spike recoveries were within the EPA Method 1630 acceptance criteria, though the matrix spike on site LS that was prepared without reagents recovered near the bottom of the acceptance range at 68%.

All reagents tested produced results that were consistent with the consensus mean for site LS. Sites CC and UJ yielded results that were significantly lower than the consensus mean; however, the results were consistent between the different distillation reagents tested.

that was prepared without reagents recovered near the bottom of the acceptance range at 68%.

## Discussion

Distillation Reagent	MDL (ng/L)
L-cysteine	0.027
APDC	0.029
CuSO <sub>4</sub>	0.040
No Reagents	0.027

Table 3. Method detection limits calculated from 8 replicates of 0.05 ng/L blank spikes

Method detection limits were calculated from 8 replicates of 0.05 ng/L spikes into a 0.5% (v/v) HCl solution as described in 40 CFR Part 136 Appendix B (Table 3). The distillations using L-cysteine, APDC, and no reagents produced MDLs that were consistent with one another; however the MDL for CuSO<sub>4</sub> was significantly higher. The MDL required by EPA Method 1630 is 0.02 ng/L.

Though the MDL achieved for the experimental samples with 'no reagents' was consistent with samples that had L-cysteine and APDC added, the recoveries of the spikes were consistently low for blank spikes with spiking levels of both 0.05 ng/L and 0.10 ng/L. Additionally, the 0.10 ng/L blank spikes for the 'no reagent' samples also showed a high variability in the results, with a standard deviation of 0.022 ng/L.

The samples distilled with CuSO<sub>4</sub> yielded results with a larger variability for blank spikes at both 0.05 ng/L and 0.10 ng/L, producing standard deviations of 0.015 ng/L and 0.026 ng/L, respectively.

Distillation Reagent	Median Score	n
L-cysteine	4.50	8
APDC	4.50	14
CuSO <sub>4</sub>	3.07	8
No Reagents	3.07	3

Table 4. Median scores by distillation reagent for methylmercury from the 2015 BPI Interlaboratory Comparison Study with n as the number of laboratories reporting

The distilled samples prepared with both L-cysteine and APDC yielded results within the EPA Method 1630 acceptance criteria for all analyzed parameters, with the exception of the matrix spike for site LS prepared with APDC. However, since this sample was not run in duplicate due to limited sample volume, and the low recovery could not be confirmed.

The results from the 2015 BPI Interlaboratory Comparison Study for Hg and MeHg in Waters show higher median scores for L-cysteine and APDC sample preparation reagents and lower scores resulting from preparation with CuSO<sub>4</sub> and no reagents (Table 4). The data from comparing the preparation reagents side-by-side in our study support this trend.

### Citations

- Creswell, J., J. Metz, A. Carter, and C. Davies (2014), 2014 Brooks Rand Instruments Interlaboratory Comparison Study for Total Mercury and Methylmercury (Intercomp 2014), Brooks Rand Instruments, Seattle, WA USA. <http://www.brooksrandinc.com/InstrumentManufacturing/intercomp/ILC2014%20Report.pdf>
- Creswell, J., E. Kimer, P.A., and C. A. Davies (2015), 2015 Brooks Rand Instruments Interlaboratory Comparison Study for Total Mercury and Methylmercury (Intercomp 2015), Brooks Rand Instruments, Seattle, WA USA. <http://www.brooksrandinc.com/InstrumentManufacturing/intercomp/ILC2015%20Report.pdf>
- U.S. Environmental Protection Agency (1998), Method 1630: Methyl Mercury in Water by Distillation, Aqueous Ethylation, Purge and Trap, and Cold Vapor Atomic Fluorescence Spectrometry, U.S. Environmental Protection Agency, Washington, D.C.