



## The Determination of Mercury in Urine by Cold Vapor Atomic Fluorescence Spectroscopy

### Introduction

Mercury levels in urine can be used to help diagnose recent mercury exposure and to evaluate patient response to chelation therapy. This method was developed to minimize sample pretreatment without loss in accuracy and utilizes bromine monochloride as an oxidant.

### Reagents

**Bromine monochloride (BrCl)**—In a fume hood, dissolve 27 g of reagent grade KBr in 2.5 L of low-Hg HCl. Place a clean magnetic stir bar in the bottle and stir for approximately 1 h in the fume hood. Slowly add 38 g reagent grade KBrO<sub>3</sub> to the acid while stirring. When all of the KBrO<sub>3</sub> has been added, the solution color should change from yellow to red to orange. Loosely cap the bottle, and allow to stir another hour before tightening the lid.

**Hydroxylamine hydrochloride**—Dissolve 300 g of NH<sub>2</sub>OH·HCl in reagent water and bring to 1.0 L. This solution may be purified by the addition of 1.0 ml of SnCl<sub>2</sub> solution and purging overnight at 500 ml/min with Hg-free N<sub>2</sub>.

**Stannous chloride** – Add 100 ml of concentrated (12N) hydrochloric acid to about 500 ml of de-ionized water. Next, add 20 grams of anhydrous stannous chloride and swirl to dissolve. Add de-ionized water to 1.0 L.

**Rinse solution** – Add 20 ml of concentrated (12N) hydrochloric acid to about 500 ml of de-ionized water. Next, add de-ionized water to 1.0L.



## Procedures

### Sample Pretreatment

Dispense 0.5 ml of urine sample into 15 ml polypropylene test tubes. Add to each sample 9.9 ml of de-ionized water (18 MOhm) and 0.1 ml of bromine monochloride (BrCl) solution. Let mixture stand for two hours and immediately before analysis add 0.010 ml 30% hydroxylamine hydrochloride to remove excess bromine.

### Analysis

Calibrate the spectrometer with aqueous standards in the range of 0-10 ug/L. We used standards of 0, 0.2, 0.5, 1.0, 5.0, and 10.0 ug/L. Table I shows the operational parameters for the Hydra AF.

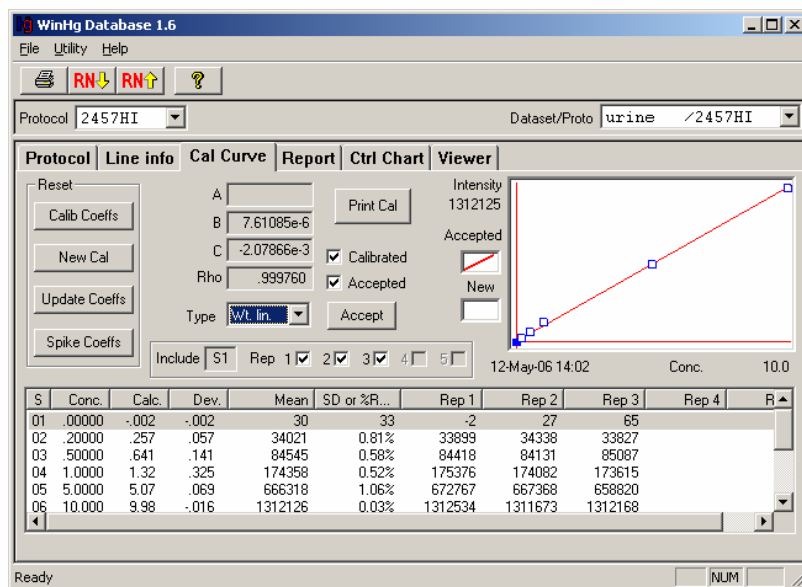
Parameter	Value
Carrier Gas	0.3 LPM
Pump Speed	7 ml/min
Rinse Time	60 sec.
Uptake Time	20 sec.
Integration Time	45 sec.
Fit Type	Wt. Linear
Method	CVAFS

*Table I: Operational Parameters*

*NOTE* - Both the Hydra AF and the Hydra AF Gold Plus are capable of determining mercury by the direct fluorescence method employed here while only the Gold Plus offers the trap and purge capability required with EPA Method 1631.

## Results

Figure I shows the calibration that was obtained using aqueous mercury standards stabilized with 2% hydrochloric acid (same as rinse solution). The correlation coefficient (Rho) for the curve using a weighted linear fit was 0.99976 and replicate readings exhibited a precision typically better than 1%.



**Figure 1: Calibration**

Reference urine samples were obtained from Bio-Rad Laboratories, Irvine, CA. Mercury levels for each sample appear below in Table II together with the values obtained with this method.

Sample	Lot	Mean	Range	Measured
Lyphochek 1	69091	47ug/L	38-57ug/L	45.2 ug/L
Lyphochek 2	69092	147	117-176	140.5

**Table II: Reference Samples**

## Conclusions

The Hydra AF has the sensitivity and precision needed to determine mercury at the typical levels found in healthy individuals (10-20ug/L) as well as the accuracy to determine mercury at elevated levels found in patients recently exposed to mercury or on chelation therapy.

Bromine monochloride digestion effectively releases the mercury in urine for determination with calculated recoveries of 96.2% (Lyphochek1) and 95.6% (Lyphochek2) without the need for heating or extended reaction times.