

Dried Urine Mercury Stability and Quantification Using a Direct Mercury Analyzer



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Introduction

Mercury is a bio-accumulative heavy metal that is detrimental to human health. Urine mercury testing is useful for determining short or long term exposure to inorganic or elemental mercury¹, but is subject to analytical challenges such as sample stability and instrument memory effects. Urine samples often require the addition of a preservative such as hydrochloric or sulfamic acid to prevent the adsorption or transpiration of mercury in collection vessels, which in turn may dilute, contaminate, or interfere with analysis². Without the use of preservatives, urine mercury may be unstable, especially at high temperatures or low concentrations²⁻³. Another major problem is that shipment of urine for mercury analysis often recommends or requires refrigerated or frozen sample transportation to the testing laboratory, which is expensive and not always an available option².

Our study investigated the use of urine dried on filter paper as an alternate to liquid urine samples for mercury analysis. We tested the stability of urine mercury once dried on filter paper and the loss of mercury from both liquid and dried samples left at room temperature using a DMA-80 direct mercury analyzer. Urine reference controls with established stability information and verified mercury concentrations were compared in both liquid and dried forms, and dried human urine samples were further tested to see if dried urine mercury stability applied to human research samples.

Methods and Materials

Mercury Reference Control and Human Urine Selection

Four lyophilized urine controls with certified mercury concentrations were chosen. Included in their package inserts were certified values/ranges with information on sample stability [Table 1]. Six human urine samples collected during an internal study at ZRT Laboratory were selected for this study.

Dried Sample Preparation

Liquid standards [0, 0.3906, 1.5625, 6.25, 25 and 100 µg/L made from Inorganic Ventures 100 ppm mercury stock solution in 10% HCl], reference controls, and human urine samples were saturated on Ahlstrom/Perkin Elmer grade 226 filter paper (6.5x5 cm), and dried for 4 hours at room temperature. Once dried, the samples remained at room temperature throughout the study.

Dried Urine Mercury Analysis

Mercury analysis was completed on Milestone's DMA-80 Direct Mercury Analyzer [Image 1]. Six 6-mm punches were taken from each sample using a Perkin Elmer automatic dried blood spot puncher and were placed in individual quartz boats. Sample weights were all set to 1 g, as all sample sizes were equivalent at six punches. The mercury standard curve was run once and stored in the system for all future analyses. The standard curve was linear through zero with an R2 value of 0.9998. The dried urine mercury assay was validated prior to beginning the study.

Liquid Urine Mercury Analysis

Mercury analysis of liquid samples was completed using the same methodology as dried urine, except 100µl of sample was used in place of six dried urine punches. Liquid reference controls were analyzed immediately after reconstitution. The remaining liquid was transferred to polypropylene vials and sealed, kept at room temperature for the remainder of the study. The liquid urine mercury assay was validated prior to beginning the study.

Image 1.



Table 1.

Mercury Reference Control	Stability (Reconstituted)			Certified Mercury Value (µg/L)		
	At 15-30C	At 2-8C	At -20C	Expected	Low Range	High Range
BioRad Lyphocheck 376 Normal Quantitative Urine Control	NA	5 Days	NA	15	10.4	19.7
ClinChek Level I Urine Control	8 Hours	7 Days	1 Month	2.48	1.74	3.22
ClinChek Level II Urine Control	8 Hours	7 Days	1 Month	18.1	13.6	22.6
Seronorm Trace Elements Urine L-1	NA	7 Days	1 Month	0.036	0.015	0.057

Figure 1.

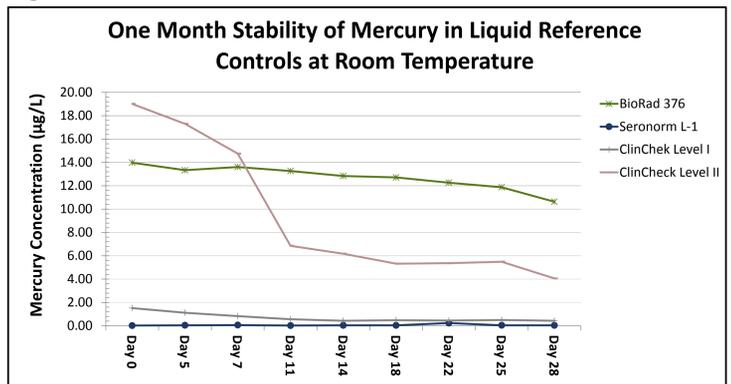


Figure 2.

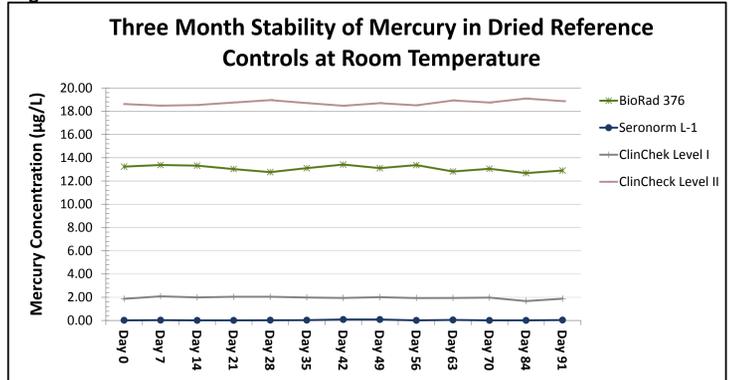
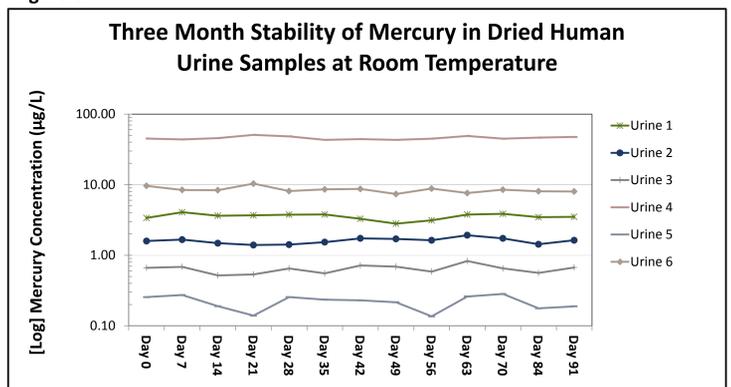


Figure 3.



Results

Liquid and Dried Reference Control Stability

Over a 1-month period, liquid reference controls kept at room temperature were analyzed [Figure 1]. When stored at room temperature, liquid reference controls were not stable for longer than a couple of days in sealed polypropylene vials.

Over a 3-month period, dried reference controls kept at room temperature were analyzed [Figure 2]. The dried reference controls proved to be stable once dried on filter paper, with no significant loss of analyte concentration. The reference controls, two of which list stability as 8 hours at room temperature [Table 1], showed no drop in mercury concentration throughout the 3-month testing period.

Dried Human Urine Stability

Over a 3-month period, human urine samples dried on filter paper and kept at room temperature were analyzed [Figure 3]. Similar to the dried reference controls, mercury in human urine samples remained stable for a period of 3 months at room temperature.

Discussion and Conclusions

The Problem

- Liquid urine sample collection often requires refrigerated/frozen transport of the sample and the addition of a preservative to stabilize mercury².
- Liquid urine reference material will only remain stable for hours or days without proper storage conditions.
- Memory effect are a common problem encountered during mercury analysis by ICP-MS (inductively coupled plasma mass spectrometry), resulting in long washout times, analyte loss, and the need for mercury stabilizers such as gold, hydrochloric or sulfamic acid in the diluent and rinse solutions⁴⁻⁵.

Our Solution

- Our study shows no loss of mercury in urine and reference control material dried on filter paper kept at room temperature without preservatives for at least 3 months, extending the time in which the sample can be transported and analyzed.
- The direct combustion of filter paper without extraction on the DMA-80 eliminates the possibility of mercury adhering to tubing or the sample introduction system.
- Dried urine technology could also be applied to dried blood spots on filter paper for the determination of putative organic mercury¹. The combination of dried urine and dried blood spot testing would cover exposure analysis for inorganic, organic and elemental mercury.
- Simple sample collection, minimal processing, small storage requirements, quick analysis and indefinite stability makes urine dried on filter paper for mercury analysis cost effective, reliable and convenient.

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