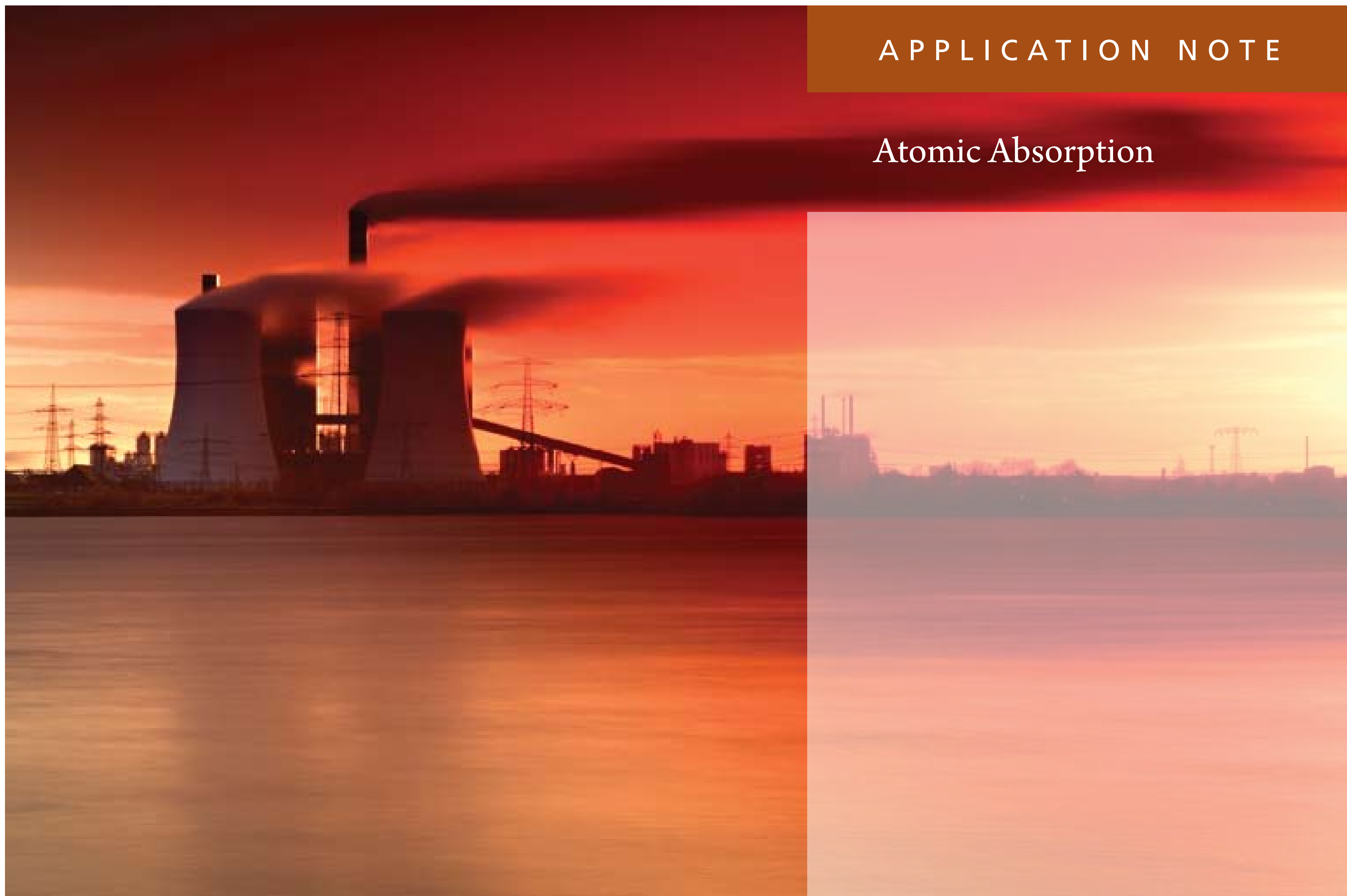

The Determination of Total Mercury in Whole Blood Using Thermal Decomposition and Amalgamation Coupled with Atomic Absorption

Summary

Mercury has long been recognized as a serious global pollutant that has a significant impact upon our ecosystem. Unlike most other pollutants, it is highly mobile, non-biodegradable, and bio-accumulative and as a result has to be closely monitored to ensure its harmful effects on local populations are minimized. Approximately 50 tons of mercury particulates are emitted into the atmosphere every year by a variety of different man-made and natural sources including coal-fired power plants, solid waste incineration plants, volcanoes and forest fires. When the mercury falls back to earth it is deposited on the land and gets into the soil, river sediments and water ecosystems, where it is converted into the highly toxic organo mercury compound, methyl mercury (CH_3Hg^+). This toxicant enters both the plant and aquatic system food chain, and eventually ends up in the crops, vegetables and seafood we consume. In addition to being ingested via the food we eat, mercury can also enter the human body through contact with the skin and by inhalation into the lungs, where it can eventually end up in the bloodstream.

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This application note will focus on a rapid test method for determining mercury directly in whole blood using the principles of thermal decomposition, amalgamation and detection by atomic absorption described in EPA Method 7473 and ASTM Method 6722-01. Because there is very little sample preparation required, this novel approach can determine the total mercury content in whole blood samples in less than five minutes, which offers significant time-saving over traditional methods, which use dilution and/or acid digestion/oxidation followed by conventional chemical reduction using cold vapor atomic absorption spectrometry (CVAA).

Introduction

Mercury is distributed throughout the environment in a number of different forms. It exists mainly as elemental mercury vapor in the atmosphere, while most of the mercury found in water, sediments, soil, plants, and animals is in the inorganic and organic forms of the element. Natural sources of mercury come from volcanoes, forest fires and the weathering of mercury-bearing rocks. However, this is small compared to the vast amount of mercury which is generated from anthropogenic sources (human activities), such as fossil fuel combustion, solid waste incineration, mining and smelting, manufacture of cement and the use of mercury cells in the commercial production of chlorine.

Of all the anthropogenic activities, by far the largest polluters are coal-fired power plants, which release approximately 50 tons of elemental mercury into the atmosphere each year via the effluent generated by the combustion process.[1] Once released, the mercury particulates fall back down to the ground and get absorbed by soils, where it eventually gets into agricultural crops and vegetables. It also enters surface waters, such as lakes, rivers, wetlands, estuaries and the open ocean, where it is converted to organic mercury (mainly methyl mercury – CH_3Hg^+) by the action of anaerobic organisms. The methyl mercury bio-magnifies up the aquatic food chain as it is passed from a lower food chain to a subsequently higher food chain level through feeding and eventually finds its way into the fish we eat.

In addition to being ingested through the food we consume, mercury can also enter the body via the lungs and absorption through the skin. Repeated exposure to mercury has adverse health effects whose symptoms are well-documented. Individuals at high risk of exposure or those who are suspected of mercury intoxication are typically monitored through analysis of blood and urine samples. The mercury blood test will detect all types of mercury but because mercury remains in the bloodstream for only a few days, the test should be performed as soon after exposure as possible. It is important to emphasize that the only way that exposure to total mercury can be assessed, is by analyzing whole blood. The urine test only measures inorganic and elemental mercury, because organic forms of the element, particularly methyl mercury are not excreted from the human body.

As a result, the EPA considers there is sufficient evidence for methyl mercury to be considered a developmental toxicant that can potentially change the genetic material of an organism and thus increases the frequency of mutations above the natural background level.[2] At particular risk are women of childbearing age because the developing fetus is the most sensitive to the toxic effects of methyl mercury. It has been proved that children who are exposed to methyl mercury before birth may be at increased risk of poor performance on neuro-behavioral tasks, such as those measuring attention, fine motor function, language skills, visual-spatial abilities and verbal memory. For that reason, the EPA has initiated the Clean Air Interstate Rule (CAIR)[3] and the Clean Air Mercury Rule (CAMR)[4] in March 2005, which is a two-phase plan to reduce the amount of mercury emission from coal-fired power stations from 48 tons to 15 tons by the year 2018, requiring new and improved mercury-specific control technology for power utilities.

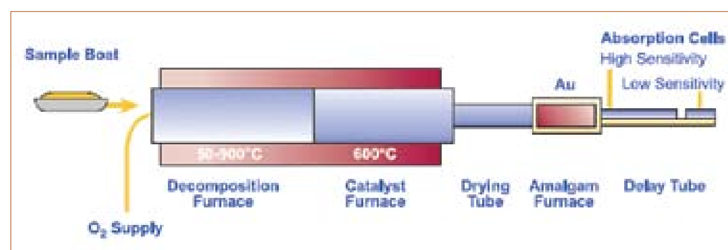

Figure 1. A schematic of the SMS 100 mercury analyzer.

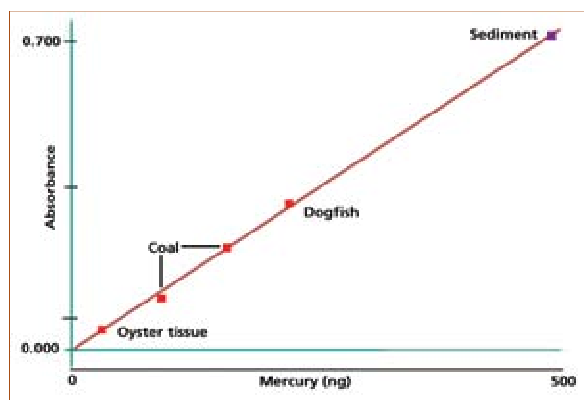

Figure 2. Under similar operating conditions, different sample matrices generate similar absorbance values as shown by the straight line calibration graph obtained from different concentrations of mercury in widely different samples.

The Study

Traditional methods for the determination of total mercury in environmental and biological samples typically involve dilution with a suitable surfactant and/or hotplate/microwave digestion using a highly corrosive acid, followed by oxidation with a strong oxidizing agent and finished off by the addition of a reducing agent to generate mercury vapor, which is then measured using cold vapor atomic absorption spectrometry.[5] Besides being extremely labor intensive and time consuming, all the sample preparation steps are a potential source of contamination and error. Even though on-line methods have been developed over the years using flow injection techniques,[6] it's still difficult to fully automate, and as a result sample throughput is significantly affected. An additional disadvantage to this method is the need to dispose of all the hazardous chemicals used for the analysis.

The goal of this study was to therefore evaluate a novel approach using the principles of thermal decomposition, amalgamation and detection by atomic absorption to determine total mercury directly in a series of freeze-dried whole blood standard reference materials (Lypho 1, 2 and 3 SRMs) using aqueous calibration standards. The benefit of this approach is that very little sample preparation/pretreatment is required, which translates into an analysis time of less than five minutes per sample.

Instrumentation

The SMS™ 100 mercury analyzer (PerkinElmer, Inc., Shelton, CT) was used for the study. This is a dedicated mercury analyzer for the determination of total mercury in solid and liquid samples using the principles of thermal decomposition, amalgamation and atomic absorption described in EPA Method 7473[7] and ASTM Method 6722-01.[8] The SMS 100 uses a decomposition furnace to release mercury vapor instead of the chemical reduction step used in traditional liquid-based analyzers. Both solid and liquid matrices can be loaded onto the instrument's autosampler and analyzed without acid digestion or sample preparation prior to analysis. Because this approach does not require the conversion of mercury to mercuric ions, the lengthy sample pretreatment and digestion steps mentioned earlier, are unnecessary. As a result, there is no need for reagents such as strong acids, oxidizing chemicals or reducing agents, which means there is no hazardous waste to be disposed of.

Principles of Operation

A small amount of the sample (0.05-1.00 gms, depending on the mercury content) is weighed into a nickel sample boat. The boat is heated in an oxygen rich furnace, to release all the decomposition products, including mercury. These products are then carried in a stream of oxygen to

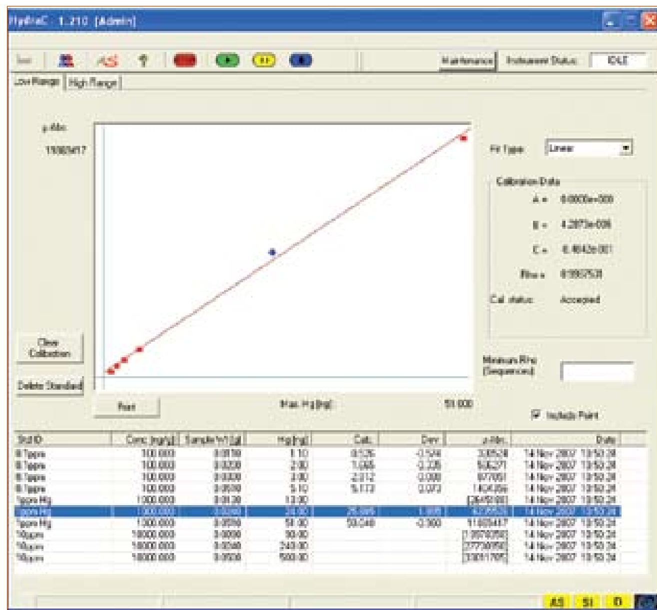

Figure 3. 0-50 ng mercury calibration plot in 10% nitric acid used for the analysis of Lypho 1 SRM.

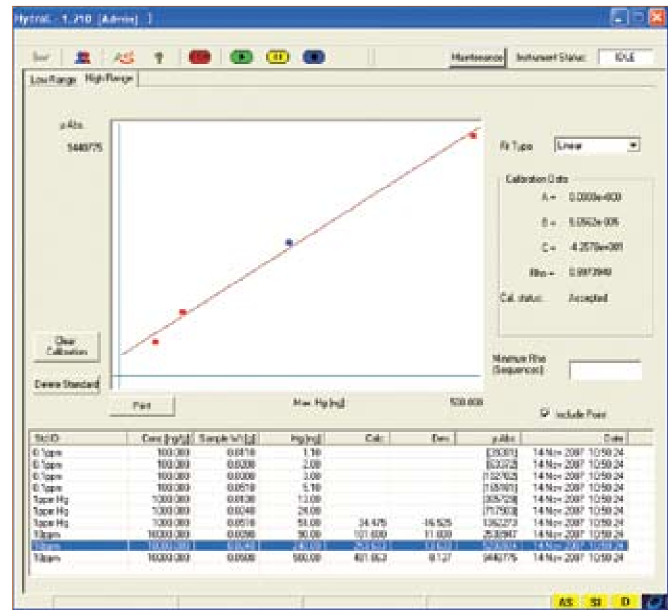

Figure 4. 50-500 ng mercury calibration plot in 10% nitric acid used for the analysis of Lypho 2 and 3 SRMs.

a catalytic section of the furnace. Any halogens or oxides of nitrogen and sulfur in the sample are trapped on the catalyst. The remaining vapor is then carried to an amalgamation cell that selectively traps mercury. After the system is flushed with oxygen to remove any remaining gases or decomposition products, the amalgamation cell is rapidly heated, releasing mercury vapor. Flowing oxygen carries the mercury vapor through an absorbance cell positioned in the light path of a single wavelength atomic absorption spectrophotometer. Absorbance is measured at the 253.7 nm wavelength as a function of the mercury concentration in the sample. A detection limit of 0.005 ng (nanogram) of mercury is achievable with a 25 cm path length cell, while a 2 cm cell allows a maximum concentration of 20 µg (microgram) of mercury. A schematic of the SMS 100 is shown in Figure 1.

Operating Conditions

Table 1 shows the instrumental operating conditions for all the Lypho SRMs.

***Table 1.* The SMS 100 operating conditions for all three Lypho SRMs.**

Parameter	Setting
Sample Weight	0.500 gm (weighed accurately)
Sample Boat	Nickel
Drying Temp/Time	300 °C for 45 sec
Decomposition Temp/Time	800 °C for 150 sec
Catalyst Temp	600 °C
Catalyst Delay Time	60 sec
Gold Trap Temp	700 °C for 30 sec
Measurement Time	90 sec
Oxygen Flow Rate	300 mL/min

Calibration

The SMS 100 measurement process involves the thermal generation of mercury vapor from the sample, which means the instrument can either be calibrated with aqueous standards or directly with solid certified reference materials. However, it is not critical that the calibration standards are of a similar matrix to the sample, because under similar operating conditions, different sample matrices generate similar absorbance values. This is shown by the straight line calibration graph in Figure 2 obtained from different concentrations of mercury in widely different samples, such as coal[9] river sediment,[10] oyster tissue and dogfish samples.[11]

For this study, all the freeze-dried Lypho SRMs were calibrated against standards made up in dilute nitric acid. Calibration graphs of 0-50 ng and 50-500 ng of mercury were generated from 0.1 and 1.0 ppm aqueous standards in 10% nitric acid respectively, by injecting different weights into a nickel sampling boat. The 0-50 ng calibration was obtained using the high sensitivity 25 cm optical path length cell, while the optional 2 cm cell was used for the 50-500 ng. The 0-50 ng calibration plot is shown in Figure 3, and the 50-500 ng plot is shown in Figure 4. Both calibration plots are displayed as absorbance against total mercury injected. The low calibration plot was then used to determine mercury in Lypho 1, and the high calibration plot was used for the Lypho 2 and 3 SRM samples.

Results

The SMS 100 results for all the Lypho SRMs evaluated are shown in Table 2.

***Table 2.* Results of the direct determination of mercury in a series of freeze-dried blood SRMs using the SMS 100.**

Blood SRM	Certified Conc. (ppb)	Accepted Range (ppb)	Conc. Found (ppb)	Recovery (%)
Lypho 1	9.6	7.7-11.6	9.08	94.8
Lypho 2	39	31-47	35.5	90.9
Lypho 3	73	58-87	66.7	91.4

Conclusion

The study shows that the thermal decomposition, amalgamation and atomic absorption technique gives excellent correlation with standard reference materials for the determination of mercury in a series of freeze-dried blood SRMs. The fact that a sample can be analyzed in approximately 5 minutes using aqueous calibration standards, means the lengthy sample preparation steps associated with the analysis of whole blood by traditional wet chemical-based mercury analyzers, can be avoided.

References

1. Pollution from Fossil Fuel Power Plants: http://en.wikipedia.org/wiki/Fossil_fuel_power_plant

2. Effects of Methyl Mercury: http://en.wikipedia.org/wiki/Methylmercury

3. EPA Clean Air Interstate Rule (CAIR) – http://www.epa.gov/CAIR/

4. EPA Clean Air Mercury Rule (CAMR) – http://www.epa.gov/oar/mercuryrule/

5. Ultratrace Analysis of Mercury and Methylmercury in Rain Water Using Cold Vapor Atomic Absorption Spectrometry: R. Ahmed, K. May and M. Stoeppler; Fresenius' Anal. Chem., Vol. 326, pp. 510-516, (1987).

6. On-Line Microwave Sample Pretreatment for the Determination of Mercury in Blood by Flow Injection Cold Vapor Atomic Absorption: T. Guo and J. Baasner, Talanta, Vol. 40, No. 12, pp. 1927-1936, (1993)

7. EPA Method 7473: http://www.epa.gov/osw/hazard/testmethods/sw846/pdfs/7473.pdf

8. ASTM Method D6722-01: http://www.astm.org/Standards/D6722.html

9. Determination of Total Mercury in Coal and Fly Ash Using Thermal Decomposition and Amalgamation Coupled with Atomic Absorption. PerkinElmer, Inc. Application: http://las.perkinelmer.com/content/ApplicationNotes/APP_DeterminationMercuryCoalAmalgamationCoupledAtomicAbsorption.pdf

10. Determination of Total Mercury in Soils and River Sediments Using Thermal Decomposition and Amalgamation Coupled with Atomic Absorption. PerkinElmer, Inc.

11. Determination of Total Mercury in Fish and Agricultural Plant Materials Using Thermal Decomposition and Amalgamation Coupled with Atomic Absorption. PerkinElmer, Inc.

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