Arsenic and Mercury Speciation: Chromatographic ICP-MS Methods for the U.S. National Biomonitoring Program, Centers for Disease Control & Prevention (CDC)

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As a member laboratory of the CDC's National Biomonitoring Program (www.cdc.gov/ exposurereport), we recently reported (Caldwell KL et al., 2009. J Expos Sci Env Epidem 19:59-68) the measurement of speciated arsenic concentrations in 2,568 human urine specimens from the National Health and Nutrition Examination Survey (www.cdc.gov/ NHANES), multi-year study whose purpose is to follow changes in the health of the U.S. population by monitoring hundreds of health indices including many blood and urine analytes. Our arsenic speciation method (Verdon CP et al., 2009. Anal Bioanal Chem 393:939-947) measures seven species of arsenic commonly found in human urine (arsenate, arsenite, dimethylarsenite, monomethylarsonate, arsenocholine, trimethylarsine oxide, and arsenobetaine). This high performance liquid chromatography (HPLC) method uses a gradient elution anion-exchange column (Hamilton PRP-X100®) coupled to a PerkinElmer "ELAN DRCII" inductively-couple plasma mass spectrometer (ICP-MS) with a Dynamic Reaction Cell® (DRC) using 10% H_2/Ar to eliminate polyatomic interference by ArCl at m/z 75. This method has been shown to be accurate per comparisons with SRM target values, and exhibits long term reproducibility by our QA/QC program criteria (Caudill SP et al., 2009. Statistics in Medicine 27: 4094-4106). The method incorporates an external 6-port, 2-position programmatically-activated switching valve (Rheodyne) with an inline sample loop which is used for post-column injection of the arsenic-containing internal standard. This configuration allows the internal standard peak to be placed anywhere in the chromatogram. Urinary speciated arsenic data obtained from the analysis of NHANES samples showed a direct relationship between total urine arsenic and arsenobetaine concentrations. Additionally, our laboratory has two methods for speciation of inorganic mercury (InHg), methyl mercury (MeHg) and ethyl mercury (EtHg) in human whole blood. The first is a HPLC-ICP-MS method using a cation-exchange column (Phenomenex "Luna® SCX", 4.6x150 mm). The HCl digested blood sample is diluted with L-cysteine and 2-mercaptoethanol, centrifuged and the supernatant analyzed by HPLC-ICP-MS. An external switching valve introduces the internal standard (methyl mercury) in the same manner as is done with arsenic speciation. The spray chamber is replaced with a desolvation device (ESI APEX Q) to improve analyte signal intensity. Our second method uses a species-specific isotope dilution (SSID) technique with gas chromatography (GC) coupled to ICP-MS to analyze whole blood for InHg, MeHg and EtHg species. Blood spiked with enriched $CH_3{}^{200}Hg$, $C_2H_5{}^{201}Hg$, and ^{199}Hg is solubilized in tetramethylammonium hydroxide (TMAH) then derivatized using sodium tetra(n-propyl)borate (Na-TPB) at pH 5. Volatile propylated Hg species are adsorbed onto a solid-phase microextraction (SPME) fiber which is injected into a splitless 200°C sample port of a PerkinElmer "Claris" GC. Species are separated on a PerkinElmer "Elite-5" (crossbond 5% diphenyl/95% PDMS) 30 m column using a 2.0 mL/min He flow rate and a 75–250°C temperature ramp. A PerkinElmer "ELAN DRCII" ICP-MS connected to the GC exit port by way of a 150°C transfer line (Hyphenated Solutions) detects Hg masses. Mass bias and dead-time corrected isotope peak ratios are used in a isotope ratio deconvolution algorithm (Applied Isotope Technologies) to mathematically correct for in situ species interconversion. Various problems were solved, most notable were 1) the idiosyncratic appearance of a large Hg^0 peak apparently coming from thermal depropylation of the derivatized inorganic Hg species, and 2) large amount of contaminating mercury in reagent blanks. Switching buffers (sodium acetate to ammonium citrate) and SPME fibers (carboxy-PDMS to PDMS) solved these problems. We intend to use this method to study the potential problem of Hg species interconversion during sample collection, transport and extended storage.