National Human Exposure Assessment Survey (NHEXAS)

Maryland Study

Quality Systems and Implementation Plan for Human Exposure Assessment

Emory University
Atlanta, GA 30322

Cooperative Agreement CR 822038

Standard Operating Procedure **NHX/SOP-L06**

Title: Extraction of Metals from Sampling Media

Source: Harvard University/Johns Hopkins University

U.S. Environmental Protection Agency
Office of Research and Development
Human Exposure & Atmospheric Sciences Division
Human Exposure Research Branch

Notice: *The U.S. Environmental Protection Agency (EPA), through its Office of Research and Development (ORD), partially funded and collaborated in the research described here. This protocol is part of the Quality Systems Implementation Plan (QSIP) that was reviewed by the EPA and approved for use in this demonstration/scoping study. Mention of trade names or commercial products does not constitute endorsement or recommendation by EPA for use.*

1. Title of Standard Operating Procedure

   Harvard University/Johns Hopkins University Standard Operating Procedures:
   L06 Extraction of Metals from Sampling Media, Rev. 1.0

2. Overview and Purpose

   This standard operating procedure (SOP) describes the procedure for the extraction of metals from a variety of sampling media: air filters, dermal wipes, dust, and soil. Of the environmental and biological samples collected in the NHEXAS Phase I Study, these are the ones that will be analyzed by the Trace Metals Laboratory at the Harvard School of Public Health (HSPH).

   Unless otherwise specified, the procedures may be used for any of the metals of interest, namely lead (Pb), arsenic (As), cadmium (Cd), and chromium (Cr); and the samples may be analyzed by graphite furnace atomic absorption spectrometry (GF-AAS) or by inductively coupled plasma-mass spectrometry (ICP-MS).

   The procedure for extraction of air filter, soil, and dust samples is based on method 3050 by U.S. EPA (1986). The method for extraction of dermal wipe samples is based on ASTM (see references). The analyses by GF-AAS and ICP-MS are described in separate SOPs: L07 "Analysis of Metals by Graphite Furnace-Atomic Absorption Spectrometry (GF-AAS)," and L08 "Analysis of Metals by Inductively Coupled Plasma-Mass Spectrometry (ICP-MS)."

3. Discussion

   The soil, dust, and air particulate samples collected are an integral part of the exposure assessment component of NHEXAS because they will provide data on the exposure to toxic metals from different media. These samples will be collected during each Cycle, either from the target individual (personal air and dermal wipe), or from his/her home (indoor and outdoor air, house dust, and soil). The samples will be analyzed at HSPH.

4. Personnel Responsibilities

   Field staff are responsible for proper collection of samples, and for appropriate handling until samples are delivered to the Field Coordination Center (FCC) and custody is transferred to the Field Coordinator (FC). The FC or his designate is responsible for storage of samples and shipping to HSPH (air filters and dermal wipes for metals) or Southwest Research Institute (SwRI) (soil and dust). SwRI is responsible for sieving and dividing soil and dust samples and shipping the portion to be analyzed for metals to HSPH.

   Analytical laboratory personnel at HSPH will be responsible for all aspects of the air filter, soil, dust, and dermal wipe extraction and subsequent analysis. This includes completion of chain-of-custody forms, and all paperwork associated with the analysis. Laboratory personnel are also required to adhere to strict quality assurance and quality control procedures.

5. Equipment and Reagents

Procedure	Equipment	Reagents
Inspection (section 6.1)	chain-of-custody forms computer with bar code reader	(none)
Weighing (section 6.2)	100-mL beaker balance (0.001 g precision)	(none)
Acid digestion of air filters, soil, and dust (section 6.3)	disposable latex gloves, unpowdered 100-mL beaker, graduated tweezers, Millipore (for air filter) hot plate, large thermometer, 0° to 110°C ribbed watch glass micropipettors, 1-10 mL, with disposable plastic tips storage bottle: LDPE, 60 mL (2 oz.), Nalgene or equivalent	ultra-high-purity water (> 18 megohm) such as Milli-Q or Nanopure HNO_3 -- trace metal grade, 1:1 with ultra-high-purity water HNO_3 -- trace metal grade, concentrated 30% H_2O_2
Acid digestion of dermal wipes (section 6.4)	disposable latex gloves, unpowdered plastic forceps 250-mL beaker, graduated watch glass, ribbed, for 250-mL beaker hot plate, large thermometer, 0° to 110°C micropipettors, 1-50 mL, with disposable plastic tips storage bottle: LDPE, 60 mL (2 oz.), Nalgene or equivalent	ultra-high-purity water HNO_3 -- trace metal grade, 1:1 with ultra-high-purity water HNO_3 -- trace metal grade, concentrated 30% H_2O_2
Preparation for GF-AAS analysis (section 6.5)	disposable latex gloves, unpowdered sample in beaker with acid-peroxide solution (from section 6.2 or 6.3) ribbed watch glass, 73 mm diameter hot plate, large filter paper -- Whatman No. 41 or equivalent 50-mL graduated cylinder with lid or stopper, or 50-mL volumetric flask (Class A, with ground glass stopper) storage bottle: LDPE, 60 mL (2 oz.), Nalgene or equivalent	ultra-high-purity water
Preparation for ICP-MS analysis (section 6.6)	disposable latex gloves, unpowdered sample from section 6.2 or 6.3 (10 mL) hot plate, large filter paper -- Whatman No. 41 or equivalent 25-mL volumetric flask (Class A, with ground glass stopper) storage bottle: LDPE, 60 mL (2 oz.), Nalgene or equivalent	HCl -- trace metal grade, concentrated Milli-Q water

6. Procedure

Summary table of contents:

6.1 Sample Inspection

Inspect and log in samples, using the computer database. The laboratory will use the ID numbers that are on the field samples or fractions that are received.

- Ø Air filters and dermal wipe samples will be received from the Field Coordination Center (FCC).
- Ø Dust and soil fractions will be received from the SwRI laboratory, where the samples are sieved and divided.
- Ø Sign the chain-of-custody form for each sample. Photocopy the forms, and file the originals in file folders in the designated filing cabinet. File them by sample type, in ascending order by ID number. Send the photocopies to the FCC. [RAW -- correct?]

Table 1 -- Sample Characteristics

Sample	Sample Types	Mass Required	Mass Received	Container etc.
Air filter	11 (outdoor) 13 (indoor) 15 (personal)	N/A	N/A	Indoor, outdoor, and personal filters from one household in Petri slides in a plastic bag.
Dust	22 (metals fraction)	0.1 - 0.2 g	0.1 - 1.0 g	Jar in bag with ID labels
Soil	32 (metals fraction)	0.1 - 0.2 g	usually 1.0 g	Jar in bag with ID labels
Dermal wipe	41 (metals sample)	N/A	N/A	One sample consists of 2 gauze pads in a jar.

- Ø Prepare the computer and bar code reader. Scan the ID numbers with the bar code reader. ID numbers should match those on the inventory list. Merge the scanned data with the data on the disk sent from the FCC.
- Ø If the ID numbers do not match or if there is any other problem, the source of the error must be determined and documented before proceeding.
- Ø Record the date and your initials in the database (Table 2 below).
- Ø Check containers for damage.
- Ø Inspect filters for:
  - ® holes, tears, or weak spots
  - ® uneven discoloration or spots that are (or have been) wet
  - ® white edge not all the way around (indicating that the filter was installed off-center and may have had an air leak at the edge)

Ø If you find any problems, enter the appropriate flag code into the database.

Table 2 -- Database Headers for Check-in and Inspection

Sample ID	Date received	Initials of inspector	Flag code(s)

6.2 Weighing

When ready to do acid digestion of dust or soil:

Ø Check the database for the mass of sample fraction that was sent after division.

Ø Transfer an ID label from the sample container to a 100-mL beaker.

Ø Weigh the empty beaker (nearest 0.001 g) and record its mass in the database (Table 3 below).

Table 3 -- Database Headers for Weighing

Sample ID	Date weighed	Initials of technician	Mass of empty beaker (g)	Mass of beaker & sample (g)	Mass of sample (g)

Ø Shake the jar to mix the sample. Use a Teflon-coated spatula to transfer 0.1-0.2 g of sample into the beaker. If the sample is no more than 0.2 g, use all of it. Record the mass of the beaker and sample. The computer will calculate the mass of the sample.

Ø If not all of the sample was used, reseal the jar and store it. Write the date and the storage location on the chain-of-custody form.

6.3 Acid Digestion -- Air Filters, Dust, and Soil (to be carried out in a hood; gloves must be worn)

1 Dust or soil: Leave the sample in the 100-mL beaker in which it was weighed.

2 Air filter: Use Millipore tweezers to transfer the filter carefully into a clean 100-mL beaker. Transfer the ID label from the bag to the beaker.

3 Add 10 mL of 1:1 HNO_3. Ensure that the sample is well covered by the acid, and cover with a watch glass. (If digesting dust or soil, swirl the beaker to mix the slurry.)

4 Use a hot plate to heat the sample to 95°C and digest for 10 to 15 minutes without boiling. (Prior experiments will have shown the time and hot plate setting needed to reach this temperature, depending on the number of beakers.)

5 Remove the beaker from the hot plate and place it on the hood surface. Allow it to cool for five minutes.

6 Add 5 mL of concentrated HNO_3, replace the watch glass, and place the beaker on the hot plate. Let it digest for 30 minutes, under continuous visual monitoring.

7 Repeat steps 4-6 to ensure complete oxidation.

8 Using a ribbed watch glass on top of the beaker, allow the solution to evaporate to approximately 5 mL without boiling, while maintaining a covering of solution over the bottom of the beaker.

9 Remove the beaker from the hot plate and place it on the hood surface. Allow it to cool for five minutes.

10 Add 2 mL of ultra-high-purity water and 3 mL of 30% H_2O_2.

11 Cover the beaker with a watch glass and return the covered beaker to the hot plate to start the peroxide reaction. Adjust the hot plate to ensure that losses do not occur due to effervescence.

12 Heat until effervescence subsides.

13 Remove the beaker from the hot plate and place it on the hood surface. Allow it to cool for five minutes.

14 Continue to add 30% H_2O_2 in 1-mL aliquots with warming until you have added the amount shown in Table 4. By the time the last H_2O_2 is added, the effervescence should be minimal and the general sample appearance is unchanged.

Table 4 -- Total Amount of 30% H_2O_2 to Add

Medium	Air Filter	Dust	Soil
Total amount of 30% H_2O_2 to add			

15 Add ultra-high-purity water to bring the sample to a volume of 50 mL.

16 If the sample is to be stored before preparation for analysis, transfer the solution to a 60 mL Nalgene HDPE/LDPE bottle. Transfer the label from the beaker to the bottle. Store the solution in the refrigerator.

17 If the sample is being prepared for GF-AAS analysis of As, Cd, Cr, and Pb, follow the procedure in section 6.5.

18 If the sample is being prepared for ICP-MS analysis of Cd, Cr, and Pb, and GF-AAS analysis of As, follow the procedure in section 6.6.

6.4 Acid Digestion -- Dermal Wipes (to be carried out in a hood)

1 Carefully open the container and remove the wipes using plastic forceps and a new pair of gloves. Place the wipes in a clean 250-mL beaker.

2 Add 50 mL of 1:1 HNO_3.

3 Gently swirl to mix, and cover with a watch glass.

4 Use a hot plate to gently heat the sample to 85° to 100°C and digest for 10-15 minutes without boiling (prior experiments will have shown the time and hot plate setting needed for sample to reach this temperature).

5 Remove the beaker from hot plate. Allow the sample to cool to near room temperature.

6 Add 20 mL of concentrated HNO_3, replace the watch glass, and reflux for 30 minutes without boiling.

7 Repeat steps 5 and 6 to ensure complete oxidation.

8 Using a ribbed watch glass, allow the solution to evaporate to approximately 10 mL without boiling, while maintaining a covering of solution (and undissolved wipe material) over the bottom of the beaker.

9 Allow the sample to cool to near room temperature after evaporation.

10 Add 5 mL of water and 5 mL of 30% H_2O_2.

11 Cover the beaker with a ribbed watch glass and return the covered beaker to the hot plate for warming and to start the peroxide reaction. Note: care must be taken during heating to ensure that losses do not occur due to excessively vigorous effervescence. Heat until effervescence subsides, then cool the beaker to near room temperature.

12 Continue to add 30% H_2O_2 in 1 mL aliquots with warming until the effervescence is minimal or until the general sample appearance is unchanged. In general, it is not recommended to add more than a total of 10 mL of 30% H_2O_2 even if effervescence has not been reduced to a minimal level. However, if the severity of the reaction continues to be high, then more 30% H_2O_2 may be added.

13 Add ultra-high-purity water to bring the sample to a volume of 50 mL.

14 If the sample is to be stored before preparation for analysis, transfer the solution to a 60 mL Nalgene HDPE/LDPE bottle. Transfer the label from the beaker to the bottle. Store the solution in the refrigerator.

15 If the sample is being prepared for GF-AAS analysis of As, Cd, Cr, and Pb, follow the procedure in section 6.4.

16 If the sample is being prepared for ICP-MS analysis of Cd, Cr, and Pb, and GF-AAS

analysis of As, follow the procedure in section 6.5.

6.5 Preparing Samples for GF-AAS analysis of As, Cd, Cr, and Pb

1 If the sample has been stored, transfer the label to a beaker and transfer the sample solution into the beaker. If proceeding directly from digestion, leave the sample in its beaker.

2 Cover the sample with a ribbed watch glass and continue heating the acid-peroxide digestate until the volume has been reduced to approximately 5 mL.

3 Remove the beaker from the hot plate. Let the sample cool to room temperature (2-5 minutes).

4 Let the particulates settle. Quantitatively transfer the digestate into a 50-mL graduated cylinder with T/S stopper or PE lid. Add 1 mL of 2% HNO_3 to the residue. Swirl gently and transfer the liquid as above. Repeat the 2% HNO_3 treatment two more times.

5 Dilute to volume (50 mL) with ultra-high-purity water. The diluted digestate solution contains approximately 10% (v/v) HNO_3.

6 Transfer the solution to a 60 mL Nalgene HDPE/LDPE bottle. Transfer the label from the beaker to the bottle. Store the solution in the refrigerator.

7 Carry out the analysis according to SOP L07 "Analysis of Metals by GF-AAS."

6.6 Preparing Samples for ICP-MS analysis of Cd, Cr, and Pb, and GF-AAS analysis of As

6.6.1 GF-AAS aliquot

1 Make sure that the sample solution from section 6.3 or 6.4 has a volume of 50 mL.

2 Transfer one 5-mL aliquot to a 100-mL beaker. If the sample is not already in a 60-mL PE bottle, transfer the rest of the sample solution to a bottle with the correct ID label.

3 Cover the sample with a ribbed watch glass and continue heating the acid-peroxide digestate until the volume has been reduced to approximately 5 mL.

3 Let the sample cool.

4 Let the particulates settle. Quantitatively transfer the digestate into a 50-mL graduated cylinder with T/S stopper or PE lid. Add 1 mL of 2% HNO_3 to the residue. Swirl gently and transfer the liquid as above. Repeat the 2% HNO_3 treatment two more times.

5 Dilute to volume (25 mL) with Milli-Q water. The diluted digestate solution contains approximately 10% (v/v) HNO_3.

6 Carry out the analysis according to SOP L07 "Analysis of Metals by GF-AAS."

6.6.2 ICP-MS aliquot

1 Transfer a 5-mL aliquot to a 100-mL beaker. Add 5 mL of concentrated HCl and 10 mL of Milli-Q water to the beaker.

2 Cover the beaker and return it to the hot plate. Heat it at 85-95°C for an additional 15 minutes without boiling.

3 Let the sample cool.

4 Let the particulates settle. Quantitatively transfer the digestate into a 50-mL graduated cylinder with T/S stopper or PE lid. Add 1 mL of 2% HNO_3 to the residue. Swirl gently and transfer the liquid as above. Repeat the 2% HNO_3 treatment two more times.

5 Dilute to volume with Milli-Q water (note: the diluted digestate solution contains approximately 5% (v/v) HNO_3 and 5% (v/v) HCl).

6 Carry out the analysis by ICP-MS according to SOP L08 "Analysis of Metals by Inductively Coupled Plasma-Mass Spectrometry (ICP-MS)."

7. Quality Assurance Procedures

7.1 Laboratory Blanks

1 per 20 samples for a minimum of 1 per batch of samples analyzed. Laboratory blanks will be prepared by carrying out the sample preparation on precleaned glass beads, an unused cellulose filter, dermal wipe, or just the reagents for the dust and soil samples. These blanks will reflect whether samples are being contaminated from laboratory activity.

7.2 Field Blanks

Field blanks, collected at a rate of 1 per 10 samples, will be analyzed identically to samples and laboratory blanks.

7.3 Duplicate Samples

1 per 10 samples or less, for a minimum of 1 per batch of samples analyzed. Two aliquots of each type of sample are carried through the extraction procedure.

7.4 Spiked Samples

1 per 20 samples, for a minimum of 1 per batch of samples analyzed. This will consist of a filter spiked with all the target analytes before extraction. The spike will be adjusted so that the resultant concentration is in the range being analyzed.

7.5 Standard Reference Materials (SRM)

1 per batch of samples. These can be obtained from the National Institute of Standards and Technology (NIST). SRM #2704 (Buffalo River sediment) and SRM #1648 (Urban Particulate Matter) can be used as standards for soil and dust, respectively.

8. References

American Society of Testing and Materials (ASTM). "(Proposed) Standard Practice for Sample Digestion of Dust Wipe Samples for the Determination of Lead by Atomic Spectrometry". Draft, May 1993.

Aschengrau, Ann, Robert Bornschein, Merrill Brophy, et al. *Three City Urban Soil-Lead Demonstration Project: Protocols for Sampling and Analysis of Soils, Dust, and Handwipes.* Internal EPA document, October 1991.

Harvard University/Johns Hopkins University Standard Operating Procedures:
G03 Identification Numbers for Samples and Forms
G04 Chain-of-Custody and Sample Tracking
G05 Storage and Shipping of Samples
L02 Cleaning of Glass and Plastic Containers
L05 Sieving and Division of Dust and Soil Samples
L07 Analysis of Metals by GF-AAS
L08 Analysis of Metals by ICP-MS

Que Hee, Shane S., Belinda Peace, C. Scott Clark, et al. "Evolution of Efficient Methods to Sample Lead Sources, Such as House Dust and Hand Dust, in the Homes of Children," *Environmental Research,* 38:77-95 (1985).

U.S. EPA. "Method 3050: Acid Digestion of Sediments, Sludges, and Soils", *Test Methods for Evaluating Solid Waste, Volume 1A: Laboratory Manual - Physical/Chemical Methods,* Washington, D.C., SW-846, 3rd edition, November 1986.