National Human Exposure Assessment Survey (NHEXAS)

Arizona Study

Quality Systems and Implementation Plan for Human Exposure Assessment

The University of Arizona
Tucson, Arizona 85721

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Standard Operating Procedure **SOP-BCO-L-13.0**

Title: Extraction of Pesticides from Surface Wipe Samples

Source: The University of Arizona

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

Extraction of Pesticides from Surface Wipe Samples

1.0 Purpose and Applicability

This standard operating procedure (SOP) describes the procedures for extracting and preparing a sill surface wipe sample for GC/MS analysis of pesticides. This procedure is also used for the pre-shipment analysis of sill wipe samples for materials suitability. Samples are collected per SOP UA-F-8.0; sample extracts are analyzed by GC/MS per SOP BCO-L-15.0.

2.0 Definitions

2.1 Surrogate Recovery Standard (Surrogate or SRS): The compound that is used for QA/QC purposes to assess the extraction and recovery efficiency obtained for individual samples. A known amount of this compound is spiked into the collected sample (dust, soil, dermal wash, PUF air cartridge, etc.) prior to extraction. The "surrogate" is quantified at the time of analysis and its recovery indicates the probable extraction and recovery efficiency for native analytes that are structurally similar. The surrogate recovery standard is chosen to be as similar as possible to the native analytes of interest. It must not, however, interfere in the analysis.

2.2 Internal Standard (IS): The compound that is added to sample extracts just prior to GC/MS analysis. The ratio of the detection signal of the native analyte to the detection signal of the IS is compared to ratios obtained for calibration curve solutions where the IS level remains fixed and the native analyte levels vary. The IS is used to correct for minor run-to-run differences in GC injection, chromatographic behavior, and MS ionization efficiency.

3.0 References

3.1 Roinestad, K.S., Louis, J.B., and Rosen, J.D., "Determination of Pesticides in Indoor Air and Dust," J. AOAC Intl., **76**, 1121-1126 (1993).

4.0 Discussion

4.1 Each of two half SOF-WICK wipes is used to wipe a 30 cm (1 ft) linear section of a window sill (see field protocol that describes the selection of the two window sills, division of the window sill for pesticide and metal wipes, etc). The two wipes are composited after sample collection and are extracted and analyzed

together. This extraction procedure involves spiking the surface of one wipe with a surrogate recovery standard, Sohxlet extraction, C18 SPE cleanup, and GC/MS analysis. SOP BCO-L-15.0 covers the GC/MS analysis and quantification of the extract; SOP UA-F-8.0 covers the preparation of the wipe medium and the field collection of the samples.

4.2 The procedure outlined here provides for the addition of a structurally similar surrogate recovery standard (fenchlorphos). This surrogate recovery compound is a pesticide itself, but it is not used in residential applications and it is rarely used in agricultural applications. This information suggests that it is not likely to be encountered in samples as a native analyte. This structurally similar surrogate recovery standard provides essential QA/QC data on extraction efficiency and recovery for each sample. The use of a structurally similar IS for GC/MS quantification corrects for minor run-to-run variation in injection, chromatography, and ionization.

5.0 Responsibilities

5.1 The sample extractions will be performed by analysts of Battelle's pesticide extraction laboratory who are completely familiar with the methods and procedures listed here. The analyst will be responsible for obtaining samples from the Sample Coordinator and ensuring the chain-of-custody forms are properly documented, entering relevant information in the extraction/preparation log books, and sending final extracts for analysis.

5.2 After receipt of the analysis results, the Project Laboratory Director in the pesticide extraction laboratory will review the data. Once verified, the analyst will be responsible for filing analyte concentration values with the Data Coordinator.

5.3 The analyst will be responsible for following this SOP, for reporting deviations and changes to the supervisory scientist, for making sure that the materials and reagents used are of sufficient purity (as indicated by manufacturer's labels), and ensuring that the holding times for solutions used have not expired.

6.0 Materials and Reagents

6.1 Materials

6.1.1 Small Soxhlet extraction apparatus consisting of condenser, extractor (31 mm id x 135 mm length), and flask (250 mL round bottom); (Kontes 585000-0021).

6.1.2 Heating mantle for 250 mL round-bottom flask.

6.1.3 Variac controller.

6.1.4 Analytical syringes.

6.1.5 Latex gloves.

6.1.6 Tweezers and tongs.

6.1.7 1 dram glass vials with Teflon-lined screw cap; muffled and vacuum silylated.

6.1.8 1.8 mL glass GC vials with Teflon-lined screw-caps; muffled and vacuum silylated.

6.1.9 Kuderna-Danish concentrators (large 24/40 3-ball Snyder condenser, 125 mL reservoir flask and 25 mL tube); (Kontes 570000).

6.1.10 Small 19/22 3-ball Snyder condensers.

6.1.11 Disposable glass pipettes (muffled and stored in clean glass jar).

6.1.12 Vortex mixer (American Scientific Products).

6.1.13 C18 SPE cartridges (Baker, 500 mg, 6mL).

6.1.14 60 mL SPE reservoirs (Supelco).

6.1.15 Multi-port SPE manifold (Supelco).

6.1.16 Glass funnels (muffled).

6.1.17 Graduated cylinders (muffled).

6.1.18 25 mL volumetric flasks (muffled).

6.1.19 Heated water bath.

6.1.20 Nitrogen evaporator (N-Evap).

6.2 Reagents

6.2.1 Boiling chips (Hengar crystals).

6.2.2 Acetone (high purity).

6.2.3 Methyl-t-butyl ether (mtbe; high purity).

6.2.4 Methanol (high purity).

6.2.5 Surrogate Recovery Standard Spiking Solution (see SOP BCO-L-21.0).

6.2.6 Internal Standard Spiking Solution (see SOP BCO-L-21.0).

6.2.7 Distilled, deionized water (DI water)

6.2.8 4% acetone in DI water (4 mL acetone diluted to 100 mL with DI water)

7.0 Procedure

7.1 Extraction of Surface Wipe Sample

7.1.1 To the extent possible, retrieve at least 10 samples for simultaneous processing from the Sample Custodian, and sign and date the chain-of-custody form. For the pre-shipment materials suitability test, obtain two wipes from a batch that has been cleaned per SOP UA-F-9.0.

7.1.2 Assemble the Soxhlet extractor in a hood, add 100 mL of high purity acetone to the flask with 3 boiling chips. (see SOP BCO-L-2.0 for details).

7.1.3 Put on clean latex gloves.

7.1.4 Spike 50 µL of the Surrogate Recovery Standard spiking solution (fenchlorphos at 5 µg/mL) onto one wipe using a 50 µL syringe.

7.1.5 Using acetone-rinsed tweezers, place the two wipes into the Soxhlet body, and place the condenser over the Sohxlet body. Turn on the water flow through the condenser.

7.1.6 Adjust the temperature of the flask with the Variac so that the acetone boils smoothly and drips at a constant rate from the condenser into the

Soxhlet body. Solvent should fill the extractor body and dump into the flask in approximately 15-20 min. Continue the extraction overnight (14h).

7.1.7 Turn off the Variac, and remove the heating mantle. Allow the Soxhlet apparatus to cool for 15 min, then remove the condenser.

7.1.8 Tilt the Soxhlet body to pour the remaining solvent from the extractor body into the round-bottom flask.

7.1.9 Using a wide-neck funnel, pour the extract from the round-bottom flask into a Kuderna-Danish concentrator (KD) which has a 125 mL reservoir flask. Rinse the round-bottom flask twice with 2 mL acetone and add the rinsate to the KD flask.

7.1.10 Add 3-5 boiling chips to the KD tube/flask. Add the large Snyder condenser to the flask.

7.1.11 Concentrate the extract in a heated (65 °C) water bath to 5 mL. Remove the KD from the water bath and let it stand in the hood to cool.

7.1.12 Remove the flask from the tube.

7.1.13 Vortex the tube for 2-3 s to mix.

7.1.14 Place SPE cartridges on the SPE manifold, and condition each cartridge in sequence with 6 mL of methanol, DI water, and 4% acetone in DI water. Close the stem valve on the SPE manifold between solvents so that the cartridge does not go dry.

7.1.15 Add an SPE reservoir to the top of each cartridge.

7.1.16 Using a clean Pasteur pipette, transfer and dilute each 5 mL sample extract to 25 mL with DI water in a 25 mL volumetric flask. Invert several times to mix.

7.1.17 Using a Pasteur pipette, transfer a sample extract to the SPE reservoir.

7.1.18 Load the extract onto the cartridge under reduced pressure (20 in Hg on manifold gauge)

7.1.19 When all of the extract has run through the cartridge, remove the reservoir and dry the cartridge for 20 min.

7.1.20 Elute the cartridge into a pre-marked (1 mL) muffled/silanized 1 dram vial with 2 mL of mtbe followed by 1 mL of mtbe.

7.1.21 Use N_2 evaporation (with N-Evap in a hood) to concentrate the extract to 1 mL. Adjust the water temperature to 40 - 50 °C and the nitrogen stream about 0.5 - 1 inch above the solvent. The nitrogen stream should not perturb the solvent surface.

7.1.22 Spike the extract with 50 µL of the Internal Standard spiking solution (trichloronate at 5 µg/mL), and vortex for 3 s to mix.

7.1.23 Transfer the extract, using a muffled disposable glass Pasteur pipette, to a clean prelabeled 1.8 mL GC vial for GC/MS analysis. Mark the volume on the side of the vial and label the sample with the laboratory notebook number and field sample ID.

7.1.24 Store the extract in a -20 °C freezer until required for GC/MS analysis.

7.2 Calculations

None.

7.3 Quality Control

7.3.1 As discussed in SOP UA-F-8.0, the pre-shipment verification for residue levels of pesticides in sill wipe media is performed on two half-wipes that represent one field sample. This wipe material is not moistened with water (the moistening solution used in the field) so that the specific media that may be contaminated can be identified before use.

7.3.2 Those samples that are designated as field blank, field duplicate and field spike samples provide important field QA/QC information. The field blank analyses are performed to verify the contaminant background levels that arise through sample handling during shipping and field operations. The field spike analyses are performed to verify the retention of analytes through shipping and handling procedures. The surrogate recovery standard added to each sample is used to verify precision and accuracy in extraction and analysis. The field duplicate samples are used to assess the precision of the field sampling methods.

7.3.3 Laboratory blanks of the sill wipes (those that are analyzed as a sample concurrently with a field sample set) will not be analyzed unless significant pesticide levels (>0.050 μg) are found in the field blanks. In that case, the source of contamination must be identified, and laboratory blanks, together with additional field blanks, trip blanks, and storage blanks, will be analyzed.

7.3.4 A field blank or field spike sample consists of two half-wipes, each moistened with water, that are composited in the field as a single sample and are extracted and analyzed together.

7.3.5 If field blank levels exceed 0.05 μg/sample, the data from the corresponding 20 homes will be flagged and inspected for possible blank correction. Additional sampling media (filter and PUF) will be sent to the field within 3 days for additional field blank measurements. The field team responsible for the flagged data will be requested to process these field blanks under field conditions as quickly as possible; their analyses at Battelle will be caried out as soon as samples are received.

7.3.6 Field crews will be reminded to wear clean laboratory coats and shoes, to remove all pesticide products from their home/residence that contain these analytes and to refrain from using these materials while a member of the field crew. Field crews will also be reminded to obtain a clean laboratory coat after visiting a home where they know or suspect that these pesticides have been applied within the previous week.

7.3.7 Field spike recovery values of 70-130% of the true value will be acceptable. Recoveries less than 70% will require a review of field and analytical protocols to verify that the procedures are being correctly implemented, especially those having to do with storage at Blue Ice/freezer temperatures after field collection. For recoveries greater than 130%, the preparation date of the field spiking solution will be checked, and recoveries in a second field spike sample (any matrix) prepared by the same field crew will be checked. If the expiration date on the spike is imminent, and/or another field spike has a high recovery, a new field spiking solution will be prepared and shipped immediately.

7.3.8 Surrogate recovery values of 70-130% in blanks, field spikes, and actual samples will be deemed acceptable, and no correction to the data will be made. For recoveries less than 70%, the data will be flagged, and the analyte concentrations will be corrected (divided) by the percent recovery

of the surrogate. For recoveries greater than 130%, the concentration of the surrogate spiking solution will be checked against a calibration curve to determine whether inadvertent solvent loss has resulted in higher spike levels. If this has occurred, the surrogate spiking solution is re-prepared.

8.0 Records

8.1 Records of the field blank levels and field spike recovery values will be retained in a NHEXAS laboratory notebook that is kept in the pesticide extraction laboratory. This notebook will serve as a continuing file for reference on expected performance of the methods and likely contaminant levels that will arise as a result of field handling. These samples will be identified in the laboratory notebook by field sample ID and the assigned laboratory analysis number (a unique number that combines the 5 digit lab book number-2 digit page number- 2 digit line number), the date of extraction, the lot number of acetone used for extraction, the batch number of the wipe material used, and the surrogate recovery value. This notebook will be transferred to the Battelle co-PIs office at the conclusion of the program.

8.2 The record of the extraction of samples will be maintained in a NHEXAS laboratory notebook that is retained in the pesticide extraction laboratory. This notebook will contain the field sample ID, the assigned laboratory analysis number (see above), the date of extraction, and the lot number of acetone used for extraction. Check-off columns will be included for addition of the surrogate and IS to verify their addition to the extracts.