National Human Exposure Assessment Survey (NHEXAS)

Arizona Study

Quality Systems and Implementation Plan for Human Exposure Assessment

The University of Arizona
Tucson, Arizona 85721

Cooperative Agreement CR 821560

Standard Operating Procedure **SOP-BCO-L-24.0**

Title: Analysis of Pesticide Samples by GC/ECD

Source: The University of Arizona

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

Analysis of Pesticide Samples by GC/ECD

1.0 Purpose and Applicability

This standard operating procedure (SOP) describes the methods used for detection and quantification by gas chromatography/electron capture detection (GC/ECD) of pesticides in a variety of matrices, including air, house dust, soil, handwipes, and surface wipes. Other SOPs (BCO-L-11 through BCO-L-14) detail the extraction of these samples. This procedure involves automated GC/ECD analysis using a high-resolution capillary column and electron capture detection.

2.0 Definitions

2.1 Extract: the 1.0-mL sample volume that contains native analytes, surrogate recovery standard, and internal standard. Each sample (dust, soil, wipe, air) is reduced to this type of extract for analysis.

3.0 References

3.1 J.P. Hsu, H.G. Wheeler, Jr., D.E. Camann, H.J. Schattenberg III, R.G. Lewis, and A.E. Bond, "Analytical Methods for Detection of Nonoccupational Exposure to Pesticides," J. Chromatogr. Sci., **26**, 181-189 (1988).

3.2 Hewlett Packard 5890A Gas Chromatograph Reference Manual, Vol. 1 and 2.

3.3 Hewlett Packard 7673A Automatic Sampler Operating and Service Manual.

3.4 Chrom Perfect for Windows, User's Manual, 1994, Justice Innovations, Inc.

4.0 Discussion

4.1 For analysis, a small aliquot of the extract (2 µL) is injected splitless into a heated GC injector (250 °C). Analytes and solvent vapors are swept onto the GC column by the helium carrier gas. The GC column temperature is then increased (computer-programmed), the components elute from the column, and are identified and quantified. Component identification is normally accomplished on the basis of the GC retention time. For the present study, the pesticides of interest are chlorpyrifos, diazinon, carbaryl, and malathion.

4.2 Both the range and limit of detection depend strongly on the properties of the individual compounds of interest. Absolute limits of detection may vary from tens of picograms (pg) to a few nanograms (ng). Precision also depends greatly on the chemical; for GC introduction, repeatability is typically ± 20% at a 1 ng level.

5.0 Responsibilities

5.1 The GC/ECD analyst will have been trained in the use and maintenance of the GC/ECD instrument, and this will be verified either through the use of training records or through years of service at Battelle with known participation in programs where GC/ECD operating skill was required.

5.2 It will be the responsibility of the Gas Chromatography Laboratory Director to certify that GC/ECD operators are fully trained.

5.3 It will be the responsibility of the operator to verify the correct operation of the instrument, through standard calibration curve analyses, prior to analysis of samples. It will be his/her responsibility to promptly report any deviations in instrument performance to the Laboratory Director, to seek guidance in the correction of the problems that go beyond routine repairs and maintenance (e.g., detector replacement, autosampler repair, etc.), and to reanalyze samples which were acquired under poor instrument operating conditions.

5.4 The Laboratory Director will review data before submission, especially high end and low end sample analyses for correct interpretation of the data (e.g., was signal saturated, was signal-to-noise ratio sufficient for quantification, etc.).

6.0 Materials and Reagents

6.1 Materials

6.1.1 60 m x 0.25 mm id fused silica column coated with nonpolar 5% phenyl methylsilicone stationary phase (DB-5 or equivalent).

6.1.2 Automated Hewlett-Packard Model 5890 gas chromatograph equipped with an electron capture detector, and Hewlett-Packard Model 7673A autosampler (or equivalent).

6.1.3 Liquid microliter syringes, 5-10 µL, for injection of standards and sample extracts into GC/ECD system.

6.2 Reagents

6.2.1 Helium carrier gas (purity > 99.9%).

6.2.2 P-10 Gas Mixture (10% methane in argon, purity > 99.9%).

6.2.3 Calibration solutions as detailed in SOP BCO-L-21.0.

7.0 Procedure

7.1 Initial Preparations

7.1.1 GC/ECD Instrument Set-Up

7.1.1.1. The helium sweep flow (across the GC injector septum) and carrier gas flow are set at approximately 3-5 mL/min and 1-2 mL/min, respectively.

7.1.1.2 The P-10 flow is set at approximately 30-45 mL/min.

7.1.1.3 The GC/ECD and Chrom Perfect data system are set according to the manufacturer's instructions. Once the GC data acquisition system has been set up, the system is calibrated as described in Section 7.1.7.

7.1.2 Initial Calibration of the GC/ECD System

7.1.2.1 Before analyzing a sample set on a new column, or after the instrument has been vented for cleaning or maintenance, calibration runs are performed with the Calibration Standards, under the same conditions used to analyze the field samples.

7.1.2.2 Calibration standards encompass five levels, plus a zero level, that bracket the expected concentration range of interest.

7.1.2.3 For the present purposes, a linear response corresponds to a correlation coefficient >0.98 for a linear least squares fit of the concentration versus relative response (peak area of the target ion of the analyte divided by the peak area of the target ion of the IS; A_S/A_{IS}) data.

7.1.2.4 Once response linearity has been demonstrated, samples can be analyzed as described below.

7.2 Preparation of Samples for Analysis

7.2.1 Arrange the sample vials in sets of 12 samples with six standards (one standard vial from each concentration level; see BCO-L-21.0) in the following order: standard, 2 samples, standard, 2 samples, etc, until all are used.

7.2.2 Inspect each vial. If any defects, such as low volume with respect to the marked volume line are observed, note them in the "Comments" column of the Data Sheet.

7.2.3 For samples with low volume, dilute to a 1 mL volume with methyl t-butyl ether (mtbe). Recap and analyze.

7.3 Sample Extract Analysis

7.3.1 Sample analysis is accomplished using a 60 m x 0.25 mm id, 0.25 Tm film thickness, DB-5 fused silica capillary column (or equivalent). Optimum analytical results are achieved with this column by temperature-programming the GC oven from 160 °C to 220 °C at 2 °C/min, 220 °C to 300 °C at 20 °C/min, then hold at 300 °C for 10 min. The injection port is held at 250 °C, and the detector is held at 300 °C.

7.3.2 Quantification of the target pesticides present is based on the peak area response using calibrated concentrations of the target compounds as the references.

7.3.3 After the final target compound elutes from the column, terminate the acquisition.

7.3.4 Once a stable baseline has been achieved, the system may be readied for the next analysis.

7.3.5 Initial data processing generally involves (1) qualitatively determining the presence or absence of each target compound on the basis of retention times, and (2) quantification of each identified component by integrating the area of the response peak and comparing the value to that of the calibration standard.

7.3.6 For sample extracts where pesticide levels exceed the calibration range of standards, prepare dilutions at 1:10 and 1:100 for reanalysis.

7.3.6.1 For 1:10 dilutions, add 900 µL of mtbe to a muffled/silylated GC vial, add 10 µL of the IS spiking solution, and 100 µL of the sample extract. Cap, invert several times to mix, label, and analyze as per 7.3.1-7.3.6.

7.3.6.2 For 1:100 dilutions, add 990 µL of mtbe to a muffled/silylated GC vial, add 10 µL of the IS spiking solution, and 10 µL of the sample extract. Cap, invert to mix, label and analyze as per 7.3.1-7.3.6

7.4 Calculations

7.4.1 Extract Concentrations

7.4.1.1 The relative area (A_S/A_{IS}) of the analyte response peaks and analyte concentration values (C_s) are analyzed using a standard linear regression analysis routine to calculate the slope (m) and the intercept (b) from the equation $y = mx+b$, where y is the relative area (A_S/A_{IS}) and x is the analyte concentration in µg/mL. These data are taken from the standards that were analyzed concurrently with a given sample set.

7.4.1.2 Once the slope and intercept have been established for each analyte, these values can be input into the same equation ($y = mx+b$), together with the A_S/A_{IS} (y) value from a sample, to thus obtain the corresponding concentration value (x).

7.4.1.3 For diluted samples, multiply the solution concentration obtained by the appropriate factor (10 or 100) to obtain the concentration of the original sample extract.

7.4.2 Calculation of Target Compound Content of Field Samples

7.4.2.1 For dermal wipe samples, multiply the analyte concentration (µg/mL) by the volume (1.0 mL) to obtain the amount (µg) of analyte in the hand wipe.

7.4.2.2 For surface wipe samples, multiply the analyte concentration (μg/mL) by the volume (1.0 mL), and divide by the surface area wiped (m^2) to obtain the surface loading of the analyte (μg/m^2).

7.4.2.3 For dust or soil samples, multiply the analyte concentration (μg/mL) by the volume (1.0 mL); then divide by the amount of material extracted (g) and the factor 0.8 (4 of total 5 mL extract volume used). This gives the dust or soil concentration (μg/g or ppm). To obtain the surface loading of dust samples (μg/m^2), multiply the analyte concentration (μg/mL) by the extract volume (1.0 mL); then divide by the area vacuumed (m^2) and the 0.8 factor.

7.4.2.4 For air samples, multiply the analyte concentration (μg/mL) by the extract volume (1.0 mL) and divide by the air volume sampled (typically 5.76 m^3 for sampling 24 h at 4 L/min) to obtain the air concentration (μg/m^3). Multiply this air concentration by 1,000 to obtain air concentrations as ng/m^3.

7.4.2.5 To determine the percent recovery of the surrogate recovery compound, multiply its concentration in the extract (μg/mL) by the volume of the extract (1.0 mL); divide this value by the amount spiked (μg) and multiply by 100. For the dust and soil samples, divide by the factor 0.8 (see 7.4.2.3 above.)

7.4.3 Limit of Detection

The limit of detection LOD for a target compound is obtained from the above data. It is defined as:

$$LOD = A + 3.3\sigma$$

where A = intercept (coefficient) from the least squares fit to the calibration curve; and s = standard deviation of the lowest concentration measurements.

7.5 Quality Control

7.5.1 Refer to the quantitative analysis calibration procedures in Sections 7.1.6 and 7.1.7. If quantitative responses (in area counts) of the lowest level standard mixture fall below the detection limits, the instrument and/or GC column and injector must be checked for performance degradation. The injector should be cleaned or the first 0.5 m of the column should be removed.

7.5.2 Those samples which were analyzed during the period when low level standards were not detected will be reanalyzed.

8.0 Records

8.1 All operations, maintenance and performance calibration data are stored in each instrument log book.

8.2 All analytical results are logged in specific project books.

8.3 Hardcopy output of chromatograms and data reports are available after each run.

8.4 All data files are stored on floppy disks for permanent record.

8.5 Hard copies of the data will be stored with the laboratory notebook and will be sent to UA after one-over-one review of the data.