DETERMINATION OF MOLINATE SULFOXIDE RESIDUES IN SOIL BY ON COLUMN CAPILLARY GAS CHROMATOGRAPHY

1. SUMMARY/INTRODUCTION

This method is intended for determining molinate sulfoxide, a metabolite of molinate, in soil at levels of 0.05 ppm to 0.5 ppm. Molinate is the active ingredient in ORDRAM® Selective Herbicide. The chemical structures are as follows:

Molinate

Molinate Sulfoxide

Molinate sulfoxide is extracted directly from soil with water and dichloromethane. The dichloromethane extract is analyzed for molinate sulfoxide by capillary gas chromatography with nitrogen-specific detection.

2. MATERIALS/METHODS

2.1 Apparatus

The equipment and reagents described below were used to generate the data and chromatograms presented in this report. Equipment with equivalent performance specifications and reagents of comparable purity can be used.

2.1.1 Gas Chromatograph. Hewlett-Packard Model 5880A, equipped with nitrogen-phosphorus detector, on-column capillary inlet, Hewlett-Packard Model 7673A automatic sampler, and electronic integrator or data acquisition system.

2.1.2 Chromatographic Column. 8 m x 0.53 mm i.d., DB-1, 1.5 µm film thickness fused silica column. J & W Scientific, Inc., Cat. No. 125-1012 or equivalent.

2.1.3 Syringe. 250 microliter capacity, Hamilton 1725N or equivalent.

2.1.4 Glass Bottles. Four-ounce, wide mouth bottles with aluminum foil-lined caps. Two-dram vials with poly seal-lined caps.

2.1.5 Reciprocating Shaker. Eberbach Corporation, model 6010 or equivalent.

2.1.6 Centrifuge. IEC International, model C1582 or equivalent.

2.2 Reagents

2.2.1 Solvents. Acetone, Dichloromethane, Nanograde® or equivalent.

2.2.2 Sodium Sulfate. Anhydrous. Reagent Grade.

2.2.3 Molinate Sulfoxide. Analytical reference-standard molinate sulfoxide. Available from ICI Americas Inc., 1200 So. 47th Street, Box 4023, Richmond, CA 94804-0023, Attention: Environmental Sciences Department Manager.

2.2.4 Calibration and Fortification Solutions. To prepare a stock solution of molinate sulfoxide, weigh to the 4th decimal place a convenient quantity, e.g. approximately 50 mg, of primary standard of known purity into a suitably sized bottle. Calculate the weight of solvent to add, based on the weight of primary standard taken, the purity of primary standard, the density of the solvent, and the desired solution concentration, typically 1000 µg/mL, as follows:

$$S = \frac{W \times P \times D}{A}$$

where S = the weight of solvent to add (g),

W = the weight of primary standard taken (mg std),

P = the purity of the primary standard (mg a.i./mg std),

D = the density of the solvent (g/mL),

and A = the desired solution concentration (mg a.i./mL solvent)

Add the calculated weight of the appropriate solvent to the bottle, close the bottle with a polyseal cap, and mix thoroughly to dissolve the primary standard. Use dichloromethane (D = 1.326 g/mL) for calibration solutions, and acetone (D = 0.792 g/mL) for fortification solutions.

To prepare working calibration solutions, dilute the stock calibration solution by weight with dichloromethane to give solutions that contain 1.0, 0.1, and 0.01 µg/mL molinate sulfoxide or other concentrations as required.

Dilute the stock fortification solution by weight with acetone to give solutions that contain 10 µg/mL molinate sulfoxide or other concentrations as required.

2.3 Analytical Procedure

2.3.1 Extraction

Weigh 40 g of thoroughly-mixed soil sample into a 4-oz wide-mouth bottle. Add 40 mL of distilled water and 40 mL of dichloromethane. Cap the bottle with an aluminum foil-lined lid and shake it on the reciprocating shaker for 1 hour. Centrifuge for 10-20 minutes at 2000 rpm to aid in separating the phases. Filter an aliquot of the dichloromethane (lower) phase through anhydrous sodium sulfate for analysis.

2.3.2 Fortification

Analyze unfortified and fortified control samples with each set of treated samples to demonstrate method recovery according to the Quality Assurance SOP. For example, for 40-g samples, weigh 40 g of untreated control soil into a 4-oz wide-mouth bottle. Add 0.200 mL of 10 ug/mL acetone fortification solution to produce a fortification level of 0.50 ppm, or add 0.200 mL of 100 µg/mL acetone fortification solution to produce a fortification level of 0.50 ppm. Add 40 mL of water and 40 mL of dichloromethane and extract as above. If a different weight of soil is analyzed, use that weight and adjust the volume or concentration of fortification solution to give the desired analyte concentration. Extract using the same volumes of water and dichloromethane as for the treated samples.

2.4 Instrumentation

2.4.1 Operating Conditions

Follow the manufacturer's instructions for operation of the gas chromatograph and nitrogen-selective detector. Use these parameters for the analyses or other operating conditions that achieve equivalent sensitivity, reproducibility, and resolution.

Oven initial temp.	113°C
Initial time	0.05 min
Temp. programming rate	15°C/min
Oven final time	6 min
Oven final temperature	260°C
Detector temperature	300°C
Carrier gas	Helium
Carrier gas pressure	2.5 psi
Carrier gas flow	11.7 mL/min through column, 53.4 mL/min vented
Injection size	4.0 µL
Quantitation	Peak height (external standard)

Under the above conditions the elution time of molinate sulfoxide is 3.55 minutes. See Appendix A for typical chromatograms.

2.4.2 Calibration

The gas chromatograph is calibrated using the analyte calibration solutions specified in section 2.2.4. Chromatographic sensitivity is established by analysis of the 0.05 µg/mL calibration solution.

Quantitation of residues at levels above the detection limit is done by an external standard procedure in which peak heights or areas of analyte peaks in sample extracts are compared to corresponding peak heights or areas of analyte peaks in calibration solutions. See Section 2.7 below for details of calculational methods.

2.4.3 Analysis of Extracts

Inject the sample extracts using the same conditions used for calibration. The identity of the analyte peak in the sample chromatogram is assigned based upon the coincidence of retention times (within 0.03 minutes) with those of the calibration chromatograms. If the response of a peak identified as an analyte exceeds that of the highest calibration solution, dilute the sample extract until its response is within the calibrated range. Reinject the calibration solution after every two to four sample injections and recalibrate as needed. Reinject the calibration solution at completion of the sample analysis.

2.5 Interferences

No clean-up is required when this procedure is utilized as described. However, extractives from soil may occasionally contribute peaks with retention times near that of molinate sulfoxide. Satisfactory resolution can usually be achieved with appropriate oven temperature manipulations or column choice. Appendix A shows typical chromatograms. Analyze extracts of samples from untreated plots to demonstrate the absence of interferences from sample matrices, solvents, or labware.

2.6 Confirmatory Techniques

Unexpected positive results, as in untreated control or pre-application samples, should be confirmed by other means, preferably by GC/MS, mass selective detection, or use of a second capillary column of different polarity.

2.7 Calculations

Calculations are done in one of two ways. If the response is linear, a factor can be calculated as described in 2.7.1 below. If the response is non-linear, or if the analyst prefers, the analyte responses over a range of calibration solution concentrations can be fit to a linear or an exponential curve, and a factor can then be calculated as in 2.7.2 below for each point on the curve that

corresponds to an analyte response in an injection of sample extract.

2.7.1 Linear Response, Direct Calculation of Factor

a. Calibration Factors for Linear Response

F = the response factor for the analyte (ppm per electronic unit), calculated as follows:

$$F = \frac{C}{P \times S}$$

where C = the concentration of analyte in the calibration solution (µg/mL)

S = the amount of initial sample represented by each milliliter of final extract solution injected (g/mL)

P = the peak area or height (electronic units) of the analyte peak in the chromatogram of the calibration solution

Averaged response factors for multiple injections of calibration solutions and for more than one concentration of calibration solution can be used as appropriate in the calculation of the concentration of the analyte in the sample, as described below.

b. Analyte in Sample

The concentration of the analyte in the original sample is calculated using an external standard method as follows:

$$ppm = F \times R$$

where ppm = the amount of analyte in the soil in parts per million

R = the peak area or height (electronic units) of the analyte peak in the chromatogram of the sample extract

and F = the response factor for the analyte (ppm per electronic unit), calculated as described above.

Note for the above external standard calculations, equal volumes of both the extract and the calibration solutions are injected.

2.7.2 Curve Fit for Linear or Non-Linear Response

If the instrumental response to injections of calibration solutions is reproducible and either linear or exponentially non-linear, a concentration-response curve can be used for sample quantitation. Any valid curve-fitting program can be used. Input the concentration and response for each injection of calibration solution. The program will generate the formula for the corresponding linear or exponential curve. From the formula, determine the calculated concentration for each injection of calibration solution as described below. The calculated and actual concentrations should agree within 10 % relative; that is, the ratio of the actual to the calculated concentration should be between 0.9 and 1.1. If the agreement is adequate, calculate the concentration of analyte in the sample, and corresponding response factor as follows:

a. Linear Response:

The formula will be of form $Y = mX + b$, where

Y = the concentration of the analyte, ppm,

X = the analyte response, peak height or area units,

and

m and b = constants calculated by the curve-fit program.

Since the analyte concentration should be zero if the response is zero, the constant b should be zero if there are no systematic errors in the analysis. However, it is not necessary for b to be zero for the calculational method to be valid, as long as calibration solution responses are reproducible and the calculated concentrations of the calibration solutions are within 10 % of the actual concentrations.

For each sample injection, determine Y by using the response, X, in the formula.

Calculate the response factor, F, from the formula:

$F = Y/X$

Note that this factor should be the same for any point on a linear curve which passes through the intercept; b = 0.

b. Exponential non-linear response:

The curve will be of form $Y = aX^b$, where

Y = the concentration of the analyte, ppm,

X = the analyte response, peak height or area units,

and
a and b = constants calculated by the curve-fit program.

For each sample injection, determine Y by using the response, X, in the formula.

Calculate the response factor, F, from the formula:

$F = Y/X$

The response factor will be different for each point on the curve.

3.3 Notes

This method was developed using columns from 3 to 8 meters in length. Unsatisfactory chromatography resulted when using shorter or longer columns.

The technique of cool on-column injection (wherein the syringe needle deposits the sample extract directly into the fused silica

column) [illegible]cerioration of the upper portion of the column resulting [illegible]cline of sensitivity and peak shape; trimming the injector end of the column by 1/2 to 1 meter will restore chromatography.

3.4 Safety Precautions

Personnel untrained in the routine safe-handling of chemicals and good laboratory practices should not attempt to use this procedure. Information on any specific chemical regarding physical properties, hazards, toxicity, and first-aid procedures can be found on the Material Safety Data Sheets accompanying the chemical or available from the chemical supplier. In general, always wear safety glasses, work in a well ventilated area, avoid inhaling vapors, and avoid contact of any chemical with skin and clothing. Flammable solvents should be kept away from potential sources of ignition.

3.4.1 Dichloromethane

Vapor is harmful.
Use only with adequate ventilation, avoid prolonged breathing of vapor.
Avoid prolonged or repeated contact with skin.

3.4.2 Acetone

Acetone is flammable.
Use in well-ventilated area; avoid prolonged breathing of vapor.
Avoid prolonged or repeated contact with skin.

3.4.3 Molinate Sulfoxide

Avoid contact with skin and eyes.
Use in well-ventilated area.
Wash with soap and water after any accidental contact.

4. CONCLUSIONS

The method is specific for the analysis of molinate sulfoxide in soil. Only readily available laboratory equipment and reagents are required. The analysis can be completed by one person in an 8-hour period. Untreated and fortified untreated samples should be extracted and analyzed with each set of samples to demonstrate absence of interferences and adequate recovery. If determination of molinate sulfoxide at a concentration other than 0.05 ppm to 0.5 ppm is required, suitably fortified samples must be analyzed to validate the method at that concentration.