METHOD 515.3 DETERMINATION OF CHLORINATED ACIDS IN DRINKING WATER BY LIQUID-LIQUID EXTRACTION, DERIVATIZATION AND GAS CHROMATOGRAPHY WITH ELECTRON CAPTURE DETECTION

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J.W. Hodgeson - Method 515, Revision 2.0 (1986)

T. Engels (Battelle Columbus Laboratory) and D.Munch (U.S.EPA, Office of Water) -National Pesticide Survey Method 3, Revision 3.0 (1987)

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Anne M. Pawlecki-Vonderheide (International Consultants, Inc.) and David J. Munch U.S.EPA, Office of Water) - Method 515.3, Revision 1.0 (1996)

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1. SCOPE AND APPLICATION

1.1 This is a gas chromatographic (GC) method (1-12) applicable to the determination of the listed chlorinated acids in drinking water, ground water, raw source water and water at any intermediate treatment stage.

Analyte	Chemical Abstract Services Registry Number
Acifluorfen[a]	50594-66-6
Bentazon	25057-89-0
Chloramben	133-90-4
2,4-D	94-75-7
Dalapon	75-99-0
2,4-DB	94-82-6
Dacthal acid metabolites[b]	
Dicamba	1918-00-9
3,5-Dichlorobenzoic acid	51-36-5
Diclorprop	120-36-5
Dinoseb	88-85-7
5-Hydroxydicamba	7600-50-2
4-Nitrophenol	100-02-7
Pentachlorophenol	87-86-5
Picloram	1918-02-1
2,4,5-T	93-76-5
2,4,5-TP (Silvex)	93-72-1

[a] The herbicide Lactofen will be quantitated as Acifluorfen as their structures represent different esters of the same carboxylate moiety.
[b] Dacthal monoacid and diacid metabolites as well as the parent di-ester included in method scope; Dacthal diacid used for validation studies.

1.2 This method is also applicable to the determination of salts and esters of analyte acids. The form of each acid is not distinguished by this method. Results are calculated and reported for each listed analyte as the total free acid.

1.3 Experimentally determined method detection limits (MDLs) (Section 9.4) for the above listed analytes are provided in Tables 2 and 3. Actual MDLs may vary according to the particular matrix analyzed and the specific instrumentation employed.

1.4 This method is designed for analysts skilled in liquid-liquid extractions, derivatization procedures and the use of GC and interpretation of gas chromatograms. Each analyst must demonstrate the ability to generate acceptable results with this method using the procedure described in Section 9.3.

1.5 When this method is used for the analyses of waters from unfamiliar sources, it is strongly recommended that analyte identifications be confirmed by GC using a dissimilar column or by GC/MS if concentrations are sufficient.

1.6 When using the diazomethane derivatization procedure, it is recommended that only qualitative identification be performed for 4-nitrophenol and 5-hydroxydicamba. Examination of supporting data presented in Tables 2, 4, 6, 8, 10 and 12 shows control over precision has not been achieved for these method analytes, and quantitative identification is therefore not recommended.

1.7 When using the base-promoted methylation procedure, it should be noted that the esterification efficiences of dinoseb and picloram were found to be less than 50%. Although the supporting data presented in Tables 3, 5, 7, 9, 11 and 13 demonstrates that accurate and precise data can be obtained through the use of procedural standards, care should be exercised.

1.8 5-Hydroxydicamba was not recovered from chlorinated waters. The exact interaction between this compound and the free chlorine is not known. The extremely low recoveries of 5-hydroxydicamba found in Tables 8 and 9 serve to illustrate this. As noted, the matrix used to obtain these results was local chlorinated tap water that was first fortified and then dechlorinated. (Note that in further experiments, 5-hydroxydicamba was recovered in waters that were dechlorinated prior to fortification.)

2. SUMMARY OF METHOD

2.1 A 40-mL volume of sample is adjusted to pH 12 with 4N sodium hydroxide for one hour to hydrolyze derivatives. (NOTE: Since many of the analytes contained in this method are applied as a variety of esters and salts, it is imperative to hydrolyze them to the parent acid prior to extraction). The aqueous sample is then acidified and extracted with 4-mL of methyl-tert-butyl-ether (MtBE). The chlorinated acids that have been partitioned into the organic phase are then converted to their methyl esters by one of two derivatization techniques. The first uses diazomethane as the methylating agent; the second is a base-promoted

esterification procedure and involves the addition of tetramethylammonium hydroxide followed by the addition of methyl iodide. The target esters are then identified and measured by capillary column gas chromatography using an electron capture detector (GC/ECD). Analytes are quantitated using procedural standard calibration.

3. DEFINITIONS

3.1 INTERNAL STANDARD (IS) -- A pure analyte(s) added to a sample, extract, or standard solution in known amount(s) and used to measure the relative responses of other method analytes and surrogates that are components of the same sample or solution. The internal standard must be an analyte that is not a sample component.

3.2 SURROGATE ANALYTE (SA) -- A pure analyte(s), which is extremely unlikely to be found in any sample, and which is added to a sample aliquot in known amount(s) before extraction or other processing and is measured with the same procedures used to measure other sample components. The purpose of the SA is to monitor method performance with each sample.

3.3 LABORATORY DUPLICATES (LD1 AND LD2) -- Two aliquots of the same sample designated as such in the laboratory. Each aliquot is extracted, derivatized and analyzed separately with identical procedures. Analyses of LD1 and LD2 indicate the precision associated with laboratory procedures, but not with sample collection, preservation, or storage procedures.

3.4 FIELD DUPLICATES (FD1 AND FD2) -- Two separate samples collected at the same time and place under identical circumstances and treated exactly the same throughout field and laboratory procedures. Analyses of FD1 and FD2 give a measure of the precision associated with sample collection, preservation and storage, as well as with laboratory procedures.

3.5 LABORATORY REAGENT BLANK (LRB) -- An aliquot of reagent water or other blank matrix that are treated exactly as a sample including exposure to all glassware, equipment, solvents, preservation and other reagents, internal standards, and surrogates that are used with other samples. The LRB is used to determine if method analytes or other interferences are present in the laboratory environment, the reagents, or the apparatus.

3.6 FIELD REAGENT BLANK (FRB) -- An aliquot of reagent water or other blank matrix that is placed in a sample container in the laboratory and treated as a sample in all respects, including shipment to the sampling site, exposure to sampling site conditions, storage, preservation and all analytical procedures. The purpose of the

FRB is to determine if method analytes or other interferences are present in the field environment.

3.7 LABORATORY FORTIFIED BLANK (LFB) -- An aliquot of reagent water or other blank matrix to which known quantities of the method analytes are added in the laboratory. The LFB should be treated like a sample including the addition of all preservation and other reagents. The LFB is analyzed exactly like a sample, and its purpose is to determine whether the methodology is in control, and whether the laboratory is capable of making accurate and precise measurements.

3.8 LABORATORY FORTIFIED SAMPLE MATRIX (LFM) -- An aliquot of an environmental sample to which known quantities of the method analytes are added in the laboratory. The LFM is analyzed exactly like a sample, including the addition of all preservation and other reagents, and its purpose is to determine whether the sample matrix contributes bias to the analytical results. The background concentrations of the analytes in the sample matrix must be determined in a separate aliquot and the measured values in the LFM corrected for background concentrations.

3.9 STOCK STANDARD SOLUTION (SSS) -- A concentrated solution containing one or more method analytes prepared in the laboratory using assayed reference materials or purchased from a reputable commercial source.

3.10 PRIMARY DILUTION STANDARD SOLUTION (PDS) -- A solution of several analytes prepared in the laboratory from stock standard solutions and diluted as needed to prepare calibration solutions and other needed analyte solutions.

3.11 CALIBRATION STANDARD (CAL) -- A solution prepared from the primary dilution standard solution and stock standard solutions of the internal standards and surrogate analytes. The CAL solutions are used to calibrate the instrument response with respect to analyte concentration.

3.12 QUALITY CONTROL SAMPLE (QCS) -- A solution of method analytes of known concentration which is used to fortify an aliquot of reagent water or sample matrix. The QCS is obtained from a source external to the laboratory and different from the source of calibration standards. It is used to check laboratory performance with externally prepared test materials.

3.13 LABORATORY PERFORMANCE CHECK SOLUTION (LPC) -- A solution of selected method analytes used to evaluate the performance of the instrumental system with respect to a defined set of method criteria.

3.14 METHOD DETECTION LIMIT (MDL) -- The minimum concentration of an analyte that can be detected, identified, measured and reported with 99% confidence that the analyte concentration is greater than zero.

3.15 MATERIAL SAFETY DATA SHEET (MSDS) -- Written information provided by vendors concerning a chemical's toxicity, health hazards, physical properties, fire and reactivity data including storage, spill, and handling precautions.

3.16 ESTIMATED DETECTION LIMIT (EDL) - Defined as either the MDL or a level of a compound in a sample yielding a peak in the final extract with a signal to noise (S/N) ratio of approximately 5, whichever is greater.

3.17 PROCEDURAL STANDARD QUANTITATION -- A quantitation method where aqueous calibration standards are prepared and processed (e.g. purged, extracted and/or derivatized) in exactly the same manner as a sample. All steps in the process from addition of sampling preservatives through instrumental analyses are included in the calibration. Using procedural standard calibration compensates for any inefficiencies in the processing procedure.

3.18 CONTINUING CALIBRATION CHECK (CCC) -- A procedural calibration standard containing the method analytes, which is extracted, derivatized and analyzed to verify the accuracy of the existing calibration curve or response factors for those analytes.

4. INTERFERENCES

4.1 Method interferences may be caused by contaminants in solvents, reagents, glassware and other sample processing apparatus that lead to discrete artifacts or elevated baselines in chromatograms. All reagents and apparatus must be routinely demonstrated to be free from interferences under the conditions of the analysis by analyzing laboratory reagent blanks as described in Section 9.5. Subtracting blank values from sample results is not permitted.

4.1.1 Glassware must be scrupulously cleaned. (1) Clean all glassware as soon as possible after use by thoroughly rinsing with the last solvent used in it. Follow by washing with hot water and detergent and thorough rinsing with tap water and reagent water. Drain and heat in an oven or muffle furnace at 400°C for 1 hr. Do not heat volumetric ware. (Thorough rinsing with reagent grade acetone may be substituted for the heating. Thermally stable materials such as PCBs may not be eliminated by this treatment.) After drying and cooling, store glassware in a clean environment to prevent any accumulation of dust or other contaminants. Store inverted or capped with aluminum foil.

4.1.2 The use of high purity reagents and solvents helps to minimize interference problems. Solvent blanks should be analyzed for each new bottle of solvent before use. An interference free solvent is a solvent containing no peaks yielding data at ≥ MDL (Tables 2 and 3) at the retention times of the analytes of interest. Purification of solvents by distillation in all-glass systems may be required.

4.2 Interfering contamination may occur when a sample containing low concentrations of analytes is analyzed immediately following a sample containing relatively high concentrations of analytes. Routine between-sample rinsing of the sample syringe and associated equipment with MTBE can minimize sample cross-contamination. After analysis of a sample containing high concentrations of analytes, one or more injections of MTBE should be made to ensure that accurate values are obtained for the next sample.

4.3 Matrix interferences may be caused by contaminants that are coextracted from the sample. The extent of matrix interferences will vary considerably from source to source, depending upon the water sampled. Analyte identifications should be confirmed using the confirmation column specified in Table 1 or another column that is dissimilar to the primary column or by GC/MS if the concentrations are sufficient.

4.4 Interferences by phthalate esters can pose a major problem in pesticide analysis when using an electron-capture detector. These compounds generally appear in the chromatogram as large peaks. Common flexible plastics contain varying amounts of phthalates that are easily extracted or leached during laboratory operations. Cross contamination of clean glassware routinely occurs when plastics are handled during extraction steps, especially when solvent-wetted surfaces are handled. Interferences from phthalates can best be minimized by avoiding the use of plastics in the laboratory. Exhaustive purification of reagents and glassware may be required to eliminate background phthalate contamination. (2,3)

4.5 The presence of water may cause incomplete methylation when using the base-promoted esterification procedure. It is imperative to ensure that all reagents and glassware are completely free of water.

4.6 5-Hydroxydicamba was not recovered from chlorinated waters. The exact interaction between this compound and the free chlorine is not known. The extremely low recoveries of 5-hydroxydicamba found in Tables 8 and 9 serve to illustrate this. As noted, the matrix used to obtain these results was local chlorinated tap water that was first fortified and then dechlorinated. (Note that in further experiments, 5-hydroxydicamba was recovered in waters that were dechlorinated prior to fortification.)

5. SAFETY

5.1 The toxicity or carcinogenicity of each reagent used in this method has not been precisely defined; however, each chemical compound must be treated as a potential health hazard. From this viewpoint, exposure to these chemicals must be minimized. The laboratory is responsible for maintaining a current awareness file of OSHA regulations regarding the safe handling of the chemicals specified in this method. A reference file of material safety data sheets should also be made available to all personnel involved in the chemical analysis. Additional references to laboratory safety are available and have been identified (4-6) for the information of the analyst.

5.2 The toxicity of the extraction solvent, MTBE, has not been well defined. Susceptible individuals may experience adverse affects upon skin contact or inhalation of vapors. Therefore protective clothing and gloves should be used and MTBE should be used only in a chemical fume hood or glove box. The same precaution applies to pure standard materials.

5.3 Diazomethane is a toxic carcinogen which can explode under certain conditions. The following precautions must be followed:

5.3.1 Use the diazomethane generator behind a safety shield in a well ventilated fume hood. Under no circumstances can the generator be heated above 90°C, and all grinding surfaces such as ground glass joints, sleeve bearings and glass stirrers must be avoided. To minimize safety hazards, the diazomethane generator apparatus used in the esterification procedure (Section 11.2) produces micromolar amounts of diazomethane in solution. If the procedure is followed exactly, no possibility for explosion exists.

5.4 Methyl iodide is a toxic carcinogen. When handling, protective clothing and gloves should be worn, and this reagent should only be used in a fume hood or glove box.

6. APPARATUS AND EQUIPMENT

6.1 SAMPLE CONTAINERS -- Amber glass bottles, approximately 50 mL, fitted with teflon-lined screw caps.

6.2 EXTRACTION VIALS -- 60 mL clear glass vials with teflon-lined screw caps.

6.3 VIALS -- Autosampler, 2.0 mL vials with screw or crimp cap and a teflon-faced seal.

6.4 STANDARD SOLUTION STORAGE CONTAINERS -- 10-20 ml amber glass vials with teflon-lined screw caps.

6.5 GRADUATED CONICAL CENTRIFUGE TUBES WITH TEFLON-LINED SCREW CAPS -- 15-mL with 1 mL graduation markings.

6.6 BLOCK HEATER (or SAND BATH) -- Capable of holding screw cap conical centrifuge tubes in Section 6.5.

6.7 PASTEUR PIPETS -- Glass, disposable.

6.8 PIPETS -- 2.0 mL and 4.0 mL, type A, TD, glass.

6.9 VOLUMETRIC FLASKS -- 5 ml, 10 mL, 100 mL.

6.10 MICRO SYRINGES -- 10 µL, 25 µL, 50 µL, 100 µL, 250 µL, 500 µL and 1000 µL.

6.11 BALANCE -- analytical, capable of weighing to 0.0001 g.

6.12 DIAZOMETHANE GENERATOR -- See Figure 1 for a diagram of an all glass system custom made for these validation studies. A micromolar generator is also available from the Aldrich Chemical Company.

6.13 GAS CHROMATOGRAPH -- Analytical system complete with gas chromatograph equipped for electron-capture detection, split/splitless capillary or direct injection, temperature programming, differential flow control, and with all required accessories including syringes, analytical columns, gases and strip-chart recorder. A data system is recommended for measuring peak areas. An autoinjector is recommended for improved precision of analyses. The gases flowing through the electron-capture detector should be vented through the laboratory fume hood system.

6.14 PRIMARY GC COLUMN -- DB-1701 [fused silica capillary with chemically bonded (14% cyanopropylphenyl)-methylpolysiloxane)] or equivalent bonded, fused silica column, 30 m x 0.25 mm ID, 0.25 µm film thickness.

6.15 CONFIRMATORY GC COLUMN -- DB-5.625 [fused silica capillary with chemically bonded (5% phenyl)-methylpolysiloxane)] or equivalent bonded, fused silica column, 30m x 0.25mm ID, 0.25 µm film thickness.

7. REAGENTS AND STANDARDS

7.1 REAGENT WATER -- Reagent water is defined as a water in which an interference is not observed ≥ to the MDL of each analyte of interest.

7.1.1 A Millipore Super-Q water system or its equivalent may be used to generate deionized reagent water. Distilled water that has been passed through granular charcoal may also be suitable.

7.1.2 Reagent water is monitored through analysis of the laboratory reagent blank (Section 9.5).

7.2 SOLVENTS

7.2.1 METHYL tert-BUTYL ETHER (MtBE) -- High purity, demonstrated to be free from analytes and interferences, redistilled in glass if necessary.

7.2.2 ACETONE -- High purity, demonstrated to be free from analytes and interferences.

7.2.3 CARBITOL (DIETHYLENE GLYCOL MONOETHYL ETHER) -- High purity, demonstrated to be free from analytes and interferences.

7.2.4 ETHYL ETHER -- High purity, unpreserved, demonstrated to be free from analytes and interferences.

7.3 REAGENTS

7.3.1 SODIUM SULFATE, Na_2SO_4 -- (ACS) granular, anhydrous. If interferences are observed, it may be necessary to heat the sodium sulfate in a shallow tray at 400°C for up to 4 hr. to remove phthalates and other interfering organic substances. Alternatively, it can be extracted with methylene chloride in a Soxhlet apparatus for 48 hr. Store in a capped glass bottle rather than a plastic container.

7.3.2 ACIDIFIED SODIUM SULFATE -- Acidify by slurrying 500g of muffled sodium sulfate with enough ethyl ether to just cover the solid. Add 0.7 mL concentrated sulfuric acid dropwise while mixing thoroughly. Remove the ether under vacuum. Mix 1g of the resulting solid with 5 mL of reagent water and measure the pH of the mixture. The pH must be below pH 4. Store at 100°C.

7.3.3 COPPER II SULFATE PENTAHYDRATE, $CuSO_4 \cdot 5H_2O$ -- ACS reagent grade.

7.3.4 SODIUM HYDROXIDE, pellets -- ACS reagent grade.

7.3.5 POTASSIUM HYDROXIDE, pellets -- ACS reagent grade

7.3.6 SODIUM THIOSULFATE, $Na_2S_2O_3$ -- ACS reagent grade, used as a dechlorinating agent in this method.

7.3.7 DIAZALD -- ACS reagent grade.

7.3.8 SULFURIC ACID, CONCENTRATED -- ACS reagent grade

7.3.9 METHYL IODIDE -- ACS reagent grade

7.3.10 TETRABUTYLAMMONIUM HYDROXIDE -- ACS reagent grade. This reagent can be purchased as a 1.0M solution in methanol. It is important that it contain no water as moisture may result in incomplete methylation.

7.3.11 SILICA GEL -- ACS reagent grade. If interferences are observed, it may be necessary to heat this reagent at 100°C for 1 hour.

7.3.12 FLORISIL -- 60-100/PR mesh. Activate by heating in a shallow container at 150°C for at least 24 hours and not more than 48 hours.

7.4 SOLUTIONS

7.4.1 4N NaOH SOLUTION -- Dissolve 16g NaOH pellets in reagent water and dilute to 100 mL.

7.4.2 37% (w/v) KOH SOLUTION -- Dissolve 37g KOH pellets in reagent water and dilute to 100 mL.

7.4.3 DIAZALD SOLUTION -- Prepare a solution containing 5g Diazald in 50 mL of a 50:50 by volume mixture of ethyl ether and carbitol. This solution is stable for one month or longer when stored at 4°C in an amber bottle with a Teflon-lined screw cap.

7.5 STANDARDS

7.5.1 4,4'-DIBROMOOCTAFLUOROBIPHENYL, 99+% -- For use as the internal standard. Prepare a stock internal standard solution of 4,4'-Dibromooctafluorobiphenyl by accurately weighing approximately 0.0200 g of neat material. Dissolve the neat material in MtBE and dilute to

volume in a 10 mL volumetric flask. Transfer the solution to an amber glass vial with a teflon-lined screw cap and store at 4°C. The resulting concentration of the stock internal standard solution will be approximately 2.0 mg/mL. Prepare a primary dilution standard at approximately 2.5 µg/mL by the addition of 12.5 µL of the stock standard to 10 mL of MtBE. Transfer the primary dilution to an amber glass vial with a teflon-lined screw cap and store at 4°C. Addition of 10 µL of the primary dilution standard to the final 1 mL extract results in a final internal standard concentration of 25 ng/mL. The solution should be replaced when ongoing QC indicates a problem. This compound has been shown to be an effective internal standard for the method analytes, but other compounds may be used if the QC requirements in Section 9 are met.

7.5.2 2,4-DICHLOROPHENYLACETIC ACID, 99+% -- For use as a surrogate compound. Prepare a surrogate stock standard solution of 2,4-Dichlorophenylacetic acid by accurately weighing approximately 0.0100 g of neat material. Dissolve the neat material in acetone and dilute to volume in a 10 mL volumetric flask. Transfer the solution to an amber glass vial with a teflon-lined screw cap and store at 4°C. The resulting concentration of the stock surrogate solution will be approximately 1.0 mg/mL. Prepare a primary dilution standard at approximately 100 µg/mL by the addition of 1 mL of the stock standard to 10 mL of acetone. Transfer the primary dilution to an amber glass vial with a teflon-lined screw cap and store at 4°C. Addition of 10 µL of the primary dilution standard to the 40 mL aqueous sample results in a surrogate concentration of 25 µg/L. The solution should be replaced when ongoing QC indicates a problem. This compound has been shown to be an effective surrogate for the method analytes, but other compounds may be used if the QC requirements in Section 9 are met.

7.5.3 STOCK STANDARD SOLUTION (SSS)

Prepare separate stock standard solutions for each analyte of interest at a concentration of 1-5 mg/mL in acetone. Method analytes may be obtained as neat materials or ampulized solutions (> 99% purity) from a number of commercial suppliers but ampulized solutions should not be used if the solvent is methanol. (7) These stock standard solutions should be stored at 4°C. They are stable for at least one month but should be checked frequently for signs of evaporation.

7.5.3.1 For analytes which are solids in their pure form, prepare stock standard solutions by accurately weighing approximately 0.01 to 0.05 grams of pure material in a 10.0 mL volumetric flask.

Dilute to volume with acetone. Each compound's purity must be assayed to be 96% or greater.

7.5.3.2 Stock standard solutions for analytes which are liquid in their pure form at room temperature can be accurately prepared in the following manner.

7.5.3.3 Place about 9.8 mL of acetone into a 10.0 mL volumetric flask. Allow the flask to stand, unstoppered, for about 10 minutes to allow solvent film to evaporate from the inner walls of the volumetric, and weigh to the nearest 0.1 mg.

7.5.3.4 Use a 10 µL syringe and immediately add 10.0 µL of standard material to the flask by keeping the syringe needle just above the surface of the acetone. Be sure that the standard material falls dropwise directly into the acetone without contacting the inner wall of the volumetric.

7.5.3.5 Reweigh, dilute to volume, stopper, then mix by inverting the flask several times. Calculate the concentration in milligrams per milliliter from the net gain in weight.

7.5.4 PRIMARY DILUTION STANDARD (PDS)

Prepare the primary dilution standard solution by combining and diluting stock standard solutions with acetone. This primary dilution standard solution should be stored at 4°C. It is stable for at least one month but should be checked before use for signs of evaporation. As a guideline to the analyst, the primary dilution standard solution used in the validation of this method is described below.

	Concentration, µg/mL
Acifluorfen	5.0
Bentazon	10.
Chloramben	5.0
2,4-D	10.
Dalapon	10.
2,4-DB	10.
Dacthal acid metabolites	5.0
Dicamba	5.0
3,5-Dichlorobenzoic acid	5.0
Diclorprop	10.

Dinoseb	10.
5-Hydroxydicamba	5.0
4-Nitrophenol	10.
Pentachlorophenol	1.0
Picloram	10.
2,4,5-T	2.5
2,4,5-TP (Silvex)	2.5

This primary dilution standard is used to prepare calibration standards, which comprise five concentration levels of each analyte with the lowest standard being at or near the MDL of each analyte. The concentrations of the other standards should define a range containing the expected sample concentrations or the working range of the detector.

7.5.5 CALIBRATION STANDARDS (CAL)

A five-point calibration curve is to be prepared by diluting the primary dilution standard into acetone at the appropriate levels. A designated amount of each acetone calibration standard is then spiked into separate 40 mL aliquots of reagent water to produce a calibration curve ranging from near the detection limit to approximately 10-20 times the lowest calibration level. These aqueous calibration standards should be treated like samples and therefore require the addition of all preservation and other reagents. They are extracted by the procedure set forth in Section 11. (The calibration standard solutions in acetone should be stored at 4°C. They are stable for at least one month but should be checked frequently for signs of evaporation.)

7.5.6 LABORATORY PERFORMANCE CHECK STANDARD (LPC)

The low level standard of the calibration curve can serve as the LPC standard. (Section 9.2)

8. SAMPLE COLLECTION, PRESERVATION AND STORAGE

8.1 SAMPLE VIAL PREPARATION

8.1.1 Grab samples must be collected in accordance with conventional sampling practices (8) using amber glass containers with TFE-lined screw-caps and capacities of at least 50 ml.

8.1.2 If residual chlorine is present, add 4 mg of sodium thiosulfate per 50 mL of sample to the sample bottle prior to collecting the sample.

8.2. SAMPLE COLLECTION

8.2.1 Fill sample bottles but take care not to flush out the sodium thiosulate. Because the target analytes of this method are not volatile, it is not necessary to ensure that the sample bottles are completely headspace free.

8.2.2 When sampling from a water tap, open the tap and allow the system to flush until the water temperature has stabilized (usually about 3-5 minutes). Remove the aerator so that no air bubbles can be visibly detected and collect samples from the flowing system.

8.2.3 When sampling from an open body of water, fill a 1-quart wide-mouth bottle or 1-liter beaker with sample from a representative area, and carefully fill sample vials from the container.

8.2.4 After collecting the sample in the bottle containing the sodium thiosulfate, seal the bottle and agitate by hand for 15 seconds.

8.2.5 Because of the several pH adjustments made to the samples in the course of this method, the addition of hydrochloric acid to the samples to retard biological activity has been omitted. However, the analyst should be aware of the potential for the biological degradation of the analytes.

8.3 SAMPLE STORAGE/HOLDING TIMES

8.3.1 Samples must be iced or refrigerated at 4°C and maintained at these conditions away from light until extraction. Synthetic ice (i.e. blue ice) is not recommended. Holding studies performed to date have shown that, in samples preserved with sodium thiosulfate, the analytes are stable for up to 14 days. (See Tables 16-19) Thus, once extracted, samples must be analyzed within 14 days. Since stability may be matrix dependent, the analyst should verify that the prescribed preservation technique is suitable for the samples under study.

8.3.2 Extracts (Sections 11.2.7 and 11.3.11) must be stored at 4°C or less away from light in glass vials with Teflon-lined caps. Holding time studies indicate that the analytes are stable for up to 14 days in the extracts. (Tables 20-23)

9. QUALITY CONTROL

9.1 Each laboratory that uses this method is required to operate a formal quality control (QC) program. Minimum quality control requirements are monitoring the laboratory performance check standard, initial demonstration of laboratory capability, performance of the method detection limit study, analysis of laboratory reagent blanks and laboratory fortified sample matrices, determination of surrogate compound recoveries in each sample and blank, monitoring internal standard peak area or height in each sample, blank and CCC, and analysis of QC samples. Additional QC practices may be added.

9.2 LABORATORY PERFORMANCE CHECK STANDARD (LPC)

At the beginning of an analysis batch, prior to any calibration standard or sample analysis and after an initial solvent blank, a laboratory performance check standard must be analyzed. It is not necessary that a new check standard be extracted each day. This check standard ensures proper performance of the GC by evaluation of the instrument parameters of detector sensitivity, peak symmetry, and peak resolution. It also demonstrates that instrument sensitivity has not changed drastically since the analysis of the MDL study. Inability to demonstrate acceptable instrument performance indicates the need for re-evaluation of the instrument system. Criteria are listed in Table 14.

9.2.1 The sensitivity requirement is based on the EDLs published in this method. If laboratory EDLs differ from those listed in Tables 2 and 3, concentrations of the LPC standard may be adjusted to be compatible with the laboratory EDLs.

9.2.2 The compounds listed in Table 15 for the LPC may not be included in the analyses of a particular laboratory. Therefore, other analytes may be chosen as long as each of the parameters (detector sensitivity, peak symmetry and peak resolution) can be sufficiently evaluated.

9.2.3 If column or chromatographic performance cannot be met, a new column may need to be installed, column flows corrected or modifications made to the oven temperature program.

9.3. INITIAL DEMONSTRATION OF CAPABILITY (IDC)

9.3.1 Select a representative fortification concentration for each of the target analytes. Concentrations near those in Tables 6 and 7 are recommended. Prepare 4-7 replicate laboratory fortified blanks by adding an appropriate aliquot of the primary dilution standard or another certified quality control sample to reagent water. Analyze the LFBs according to the method beginning in Section 11.

9.3.2 Calculate the mean percent recovery and the standard deviation of the recoveries. For each analyte, the mean recovery value, expressed as a percentage of the true value, must fall in the range of 80-120% and the relative standard deviation should be less than 20%. For those compounds that meet these criteria, performance is considered acceptable and sample analysis may begin. For those compounds that fail these criteria, this procedure must be repeated using 4-7 fresh samples until satisfactory performance has been demonstrated. Maintain these data on file to demonstrate initial capabilities.

9.3.3 Furthermore, before processing any samples, the analyst must analyze at least one laboratory reagent blank to demonstrate that all glassware and reagent interferences are under control.

9.3.4 The initial demonstration of capability is used primarily to preclude a laboratory from analyzing unknown samples via a new, unfamiliar method prior to obtaining some experience with it. As laboratory personnel gain experience with this method, the quality of data should improve beyond those required here.

9.3.5 The analyst is permitted to modify GC columns, GC conditions, internal standard or surrogate compounds. Each time such method modifications are made, the analyst must repeat the procedures in Section 9.3.1 through Section 9.3.4.

9.4 METHOD DETECTION LIMIT STUDY (MDL)

9.4.1 Prior to the analysis of any field samples, the method detection limits must be determined. Initially, estimate the concentration of an analyte which would yield a peak equal to 5 times the baseline noise and drift. Prepare seven replicate laboratory fortified blanks at this estimated concentration. Analyze the LFB's according to the method beginning in Section 11.

9.4.2 Calculate the mean recovery and the standard deviation for each analyte. Multiply the Student's t value at 99% confidence and n-1 degrees of freedom (3.143 for seven replicates) by this standard deviation to yield a statistical estimate of the detection limit. This estimate is the MDL.

9.4.3 MDL's should be recalculated after major changes in the chromatographic temperature program or stationary phase or after a change in instrument or detector.

9.5 LABORATORY REAGENT BLANKS (LRB) -- Each time a set of samples is extracted or reagents are changed, a LRB must be analyzed. The LRB must contain the preservation and other reagents added to the sample. If the LRB produces an interferant peak within the retention time window (Section 12.3) of any analyte that would prevent the determination of that analyte or a peak of concentration greater than the MDL for that analyte, the analyst must determine the source of contamination and eliminate the interference before processing samples. For the analyte(s) that failed to meet this criteria, concentrations in field samples are considered suspect.

9.6 LABORATORY FORTIFIED BLANK (LFB) Since this method utilizes procedural calibration standards, which are fortified reagent water, there is no difference between the LFB and the continuing calibration check standard. Consequently, the analysis of an LFB is not required (Section 10.2).

9.7 LABORATORY FORTIFIED SAMPLE MATRIX (LFM)

9.7.1 The concentrations of the analytes in a given sample may be equal to or greater than the fortified concentrations. Subsequently, relatively poor accuracy and precision may be anticipated when a large background must be subtracted. For many samples, the concentrations may be so high that fortification may lead to a final extract with instrumental responses exceeding the working range of the electron capture detector. If this occurs, the extract must be diluted. In spite of these problems, sample sources should be fortified and analyzed as described below. By fortifying sample matrices and calculating analyte recoveries, any matrix induced analyte bias is evaluated.

9.7.2 The laboratory must add known concentrations of analytes to one sample per extraction set or a minimum of 10% of the samples, whichever is greater. The concentrations should be equal to or greater than the back-ground concentrations in the sample selected for fortification. If the fortification level is less than the background concentration, recoveries are not reported. Over time, samples from all routine sample sources should be fortified.

9.7.3 Calculate the mean percent recovery, R, of the concentration for each analyte, after correcting the total mean measured concentration, A, from the fortified sample for the back-ground concentration, B, measured in the unfortified sample, i.e.:

$$R = 100 (A - B) / C,$$

where C is the fortifying concentration. In order for the recoveries to be considered acceptable, they must fall between 70% and 130% for all the target analytes.

9.7.4 If a recovery falls outside of this acceptance range, a matrix induced bias can be assumed for the respective analyte and the data for that analyte must be reported to the data user as suspect due to matrix effects.

9.8 ASSESSING SURROGATE RECOVERY

The surrogate standard is fortified into the aqueous portion of all calibration standards, samples, QC samples and laboratory reagent blanks. The surrogate is a means of assessing method performance from extraction to final chromatographic performance.

9.8.1 When surrogate recovery from a sample, blank, QC sample or CCC is < 70% or > 130%, check (1) calculations to locate possible errors, (2) standard solutions for degradation, (3) contamination, and (4) instrument performance. If those steps do not reveal the cause of the problem, reanalyze the extract.

9.8.2 If the extract reanalysis meets the surrogate recovery criterion, report only data for the reanalyzed extract.

9.8.3 If the extract reanalysis fails the 70-130% recovery criterion, the analyst should check the calibration by analyzing the most recently acceptable continuing calibration check standard. If the CCC fails the criteria of Section 10.2.1, recalibration is in order per Section 10.1. If the CCC is acceptable, it may be necessary to extract another aliquot of sample. If the sample re-extract also fails the recovery criterion, report all data for that sample as suspect.

9.9 ASSESSING THE INTERNAL STANDARD

9.9.1 The analyst must monitor the IS response (peak area or peak height) of all injections during each analysis day. A mean IS response should be determined from the five point calibration curve. The IS response for any run should not deviate from this mean IS response by more than 30%.

9.9.2 If a greater deviation than this occurs with an individual extract, optimize instrument performance and inject a second aliquot of that extract.

9.9.2.1 If the reinjected aliquot produces an acceptable internal standard response, report results for that aliquot.

9.9.2.2 If a deviation of greater than 30% is obtained for the reinjected extract, the analyst should check the calibration by analyzing the most recently acceptable CCC. If the CCC fails the criteria of Section 10.2.1, recalibration is in order per Section 10.1. If the CCC is acceptable, analysis of the sample should be repeated beginning with Section 11, provided the sample is still available. Otherwise, report results obtained from the reinjected extract, but annotate as suspect.

9.10 QUALITY CONTROL SAMPLE (QCS) -- At least quarterly, analyze a QCS from an external source. If measured analyte concentrations are not of acceptable accuracy, check the entire analytical procedure to locate and correct the problem source.

9.11 The laboratory may adapt additional QC practices for use with this method. The specific practices that are most productive depend upon the needs of the laboratory and the nature of the samples. For example, field or laboratory duplicates may be analyzed to assess the precision of the environmental measurements or field reagent blanks may be used to assess contamination of samples under site conditions, transportation and storage.

10. CALIBRATION AND STANDARDIZATION

10.1 INITIAL CALIBRATION CURVE

10.1.1 Establish GC operating parameters equivalent to the suggested specifications in Table 1. The GC system must be calibrated using the internal standard (IS) technique. Other columns or conditions may be used if equivalent or better performance can be demonstrated.

10.1.2 Calibration standards at five concentrations are required. The lowest should contain the analytes at a concentration near to but greater than the EDL (Tables 2 and 3) for each compound. The others should be evenly distributed throughout the concentration range expected in the samples.

10.1.3 Inject 2 µL of each calibration standard extract and tabulate peak height or area response and concentration for each analyte and the internal standard.

10.1.4 Generate a calibration curve by plotting the area ratios (A_a/A_{is}) against the concentration ratios (C_a/C_{is}) of the five calibration standards where

A_a is the peak area of the analyte.
A_{is} is the peak area of the internal standard.
C_a is the concentration of the analyte.
C_{is} is the concentration of the internal standard.

This curve can be defined as either first or second order; the correlation coefficients must be greater than 0.95. Also, the working calibration curve must be verified daily by measurement of one or more calibration standards (Section 10.2). If the response for any analyte falls outside the predicted response by more than 30%, the calibration check must be repeated using a freshly prepared calibration standard. Should the retest fail, a new calibration curve must be generated.

10.1.5 Alternately, an average relative response factor can be calculated and used for quantitation. Relative response factors are calculated for each analyte at the five concentration levels using the equation below:

$$RRF = \frac{(A_a)(C_{is})}{(A_{is})(C_a)}$$

If the RRF value over the working range is constant (<20% RSD), the RRF can be assumed to be invariant and the average RRF used for calculations. Also, the average RRF must be verified daily by measurement of one or more calibration standards (Section 10.2). If the RRF for the continuing calibration standard deviates from the average RRF by more than 30%, the calibration check must be repeated using a freshly prepared calibration standard. Should the retest fail, a new calibration curve must be generated.

10.1.6 A data system may be used to collect the chromatographic data, calculate relative response factors, or calculate linear or second order calibration curves.

10.2 CONTINUING CALIBRATION CHECK (CCC)

10.2.1 At least one CCC must be extracted with each set of samples. A CCC must be analyzed at the beginning of each analysis set, after every tenth sample analysis and after the final sample analysis, to ensure that the instrument is still within calibration. These checks should be at two different concentration levels. Calculate analyte recoveries for all target analytes. In order for the calibration check to be considered valid and subsequently for the preceding ten samples to be considered acceptable with respect to calibration, recoveries must fall between 70% and 130% for

all the target analytes. Additionally, the internal standard area must be within 30% of the mean IS response. (Section 9.9.1)

NOTE: Continuing calibration check standards need not all be different extracts but can be injections from the same extract as long as the holding time requirements for extracts (Sect. 8.3.2) are met. However, at least one must be extracted with each batch of samples.

10.2.2 If this criterion cannot be met, the continuing calibration check standard extract is re-injected in order to determine if the response deviations observed from the initial analysis are repeated. If this criterion still cannot be met, a CCC that has already been analyzed and has been found to be acceptable should be run. If this second CCC fails, then the instrument is considered out of calibration and needs to be recalibrated. Should all CCC's associated with a particular set of samples fail, the set of samples must be re-extracted.

11. PROCEDURE

11.1 SAMPLE EXTRACTION AND HYDROLYSIS

11.1.1 Remove the samples from storage (Sect. 8.3.1) and allow them to equilibrate to room temperature.

11.1.2 Place 40 mL of the water sample into a precleaned 60 mL glass vial with a teflon-lined screw cap using a graduated cylinder.

11.1.3 Add 10 µL of surrogate standard (100 µg/mL 2,4-Dichlorophenylacetic acid in acetone per Section 7.5.2) to the aqueous sample.

NOTE: After injection, cap the sample and invert once. This ensures that the fortification volume is mixed well with the water.

11.1.4 Add 1 mL of the 4N NaOH solution prepared in Section 7.4.1 to each glass vial. Check the pH of the sample with pH paper or a pH meter; if the sample does not have a pH greater than or equal to 12, adjust the pH by adding more 4N NaOH solution. Let the sample sit at room temperature for 1 hour, shaking the contents periodically.

NOTE: Since many of the herbicides contained in this method are applied as a variety of esters and salts, it is vital to hydrolyze them to the parent acid prior to extraction. This step must be included in the analysis of all extracted field samples, LRBs, LFMs and calibration standards.

11.1.5 Adjust the pH to less than 0.5 by adding at least 2 mL of concentrated sulfuric acid. Cap, shake and then check the pH with a pH meter or narrow range pH paper.

11.1.6 Quickly add approximately 2 g of copper II sulfate pentahydrate and shake until dissolved. This colors the aqueous phase blue and therefore allows the analyst to better distinguish between the aqueous phase and the organic phase in this micro extraction.

11.1.7 Quickly add approximately 16 g of muffled sodium sulfate and shake for 3 to 5 minutes until almost all is dissolved. Sodium sulfate is added to increase the ionic strength of the aqueous phase and thus further drive the chlorophenoxy acids into the organic phase. The addition of this salt and the copper II sulfate pentahydrate should be done quickly so that the heat generated from the addition of the acid (Section 11.1.5) will help dissolve the salts.

11.1.8 Add exactly 4.0 mL MtBE and place on the mechanical shaker for 30 minutes. (If hand-shaken, two minutes is sufficient if performed vigorously).

11.1.9 Allow the phases to separate for approximately 5 minutes.

11.2 METHYLATION - DIAZOMETHANE

NOTE: It is not recommended that this method of derivatization be used if 4-nitrophenol and 5-hydroxydicamba are included in the target list.

11.2.1 Generation of Diazomethane.

11.2.1.1 Assemble the diazomethane generator (Figure 1) in a hood. The collection vessel is a 10 or 15-mL glass vial equipped with a teflon-lined screw cap and maintained at 0-5°C.

11.2.1.2 Add a sufficient amount of ethyl ether (approximately 7 ml) to tube 1 to cover the first impinger. Add 5 ml of MtBE to the collection vial. Set the nitrogen flow at 5-10 mL/min. Add 4 mL Diazald solution (Section 7.4.3) and 3 mL of 37% KOH solution (Section 7.4.2) to the second impinger. Connect the tubing as shown and allow the nitrogen flow to purge the diazomethane from the reaction vessel into the collection vial for 30 minutes. Cap the vial when collection is complete and maintain at 0-5°C. When stored at 0-5°C, this diazomethane solution may be used over a period of 48 hours.

11.2.2 Using a Pasteur pipet, transfer the sample extract (upper MtBE layer) to a 10 mL screw cap vial. Add 0.1 g acidified sodium sulfate and shake. This step is included to ensure the MtBE extract contains no water.

11.2.3 Using a Pasteur pipet, transfer exactly 3 mL of the dried MtBE extract to a 15 mL graduated conical centrifuge tube.

11.2.4 Add 250 µL of the diazomethane solution prepared in Section 11.2.1.2. to each centrifuge tube. The contents of the centrifuge tube should remain slightly yellow in color. If this is not the case, more diazomethane solution may be added, making sure to add the exact additional amount to every calibration standard, blank, QC sample and field sample. Let the esterification reaction proceed for 30 minutes.

11.2.5 Remove any unreacted diazomethane by the addition of 0.1 g silica gel. Effervescence (evolution of nitrogen) is an indication that excess diazomethane was present. Allow the extracts to sit for 0.5 hour.

11.2.6 Place a small plug of glass wool into a disposable Pasteur pipet. Fill the pipet with approximately 2 inches of florisil. (Section 7.3.12) (This step is the preparation of clean-up columns for the methylated extracts. One column should be prepared for each extract.)

11.2.7 Apply the methylated extract to the prepared clean-up column and collect the eluate in a 5 mL vial.

11.2.8 Transfer exactly 1.0 ml of the MtBE extract to an autosampler vial. A duplicate vial should be filled using the excess extract.

11.2.9 Add 10 µL of internal standard to the vial to be analyzed. (2.5 µg/mL 4,4'-Dibromooctafluorobiphenyl in MtBE per Section 7.5.1). Internal standard should be added to the duplicate vial before analysis.

11.2.10 Analyze the sample extracts as soon as possible. The sample extract may be stored up to 14 days if kept at 4°C or less. Keep the extracts away from light in amber glass vials with Teflon-lined caps.

11.3. METHYLATION - BASE-PROMOTED ESTERIFICATION

11.3.1 Using a Pasteur pipet, transfer approximately 3 mL of the sample extract (upper MtBE layer) to a 10 mL screw cap vial. Add 0.1 g acidified sodium sulfate and shake. This step is included to ensure that the MtBE extract contains no water.

11.3.2 Using a Pasteur pipet, transfer exactly 3 mL of the dried MtBE extract to a 15 mL graduated conical centrifuge tube.

11.3.3 Add 80 µL of the 1.0M solution of tetrabutylammonium hydroxide.

11.3.4 Add 40 µL of methyl iodide.

11.3.5 Cap the centrifuge tubes and place in the heating block (or sand bath) at 50°C and maintain for 1.5 hr. The vials must fit snugly into the heating block to ensure proper heat transfer. At this stage, methylation of the method analytes is attained and the tetrabutylammonium iodide by-product may be viewed as a precipitate.

11.3.6 Remove the centrifuge tubes from the heating block (or sand bath) and allow them to cool before removing the caps.

11.3.7 Place a small plug of glass wool into a disposable Pasteur pipet. Fill the pipet with approximately 2 inches of florisil. (Section 7.3.12) (This step is the preparation of clean-up columns for the methylated extracts. One column should be prepared for each extract.)

11.3.8 Apply the methylated extract to the prepared clean-up column and collect the eluate in a 5 mL vial.

11.3.9 Transfer exactly 1.0 ml of the MtBE extract to an autosampler vial. A duplicate vial should be filled using the excess extract.

11.3.10 Add 10 µL of internal standard to the vial to be analyzed. (2.5 µg/mL 4,4'-Dibromooctafluorobiphenyl in MTBE per Section 7.5.1). Internal standard should be added to the duplicate vial before analysis.

11.3.11 Analyze the samples as soon as possible. The sample extract may be stored up to 14 days if kept at 4°C or less. Keep the extracts away from light in amber glass vials with Teflon-lined caps.

11.4 GAS CHROMATOGRAPHY

11.4.1 Table 1 summarizes recommended GC operating conditions and retention times observed using this method. Figure 2 illustrates the performance of the recommended primary column with the method analytes. Figure 3 illustrates the performance of the recommended confirmation column with the method analytes. Other GC columns or chromatographic conditions may be used if the requirements of Section 9 are met.

11.4.2 Calibrate the system daily by either the analysis of a calibration curve (Section 10.1) or a continuing calibration check as described in Section 10.2.

11.4.3 Inject 2 µL of the sample extract. Record the resulting peak sizes in area or height units.

11.4.4 If the response for the peak exceeds the working range of the system, dilute the extract, add an appropriate additional amount of internal standard and reanalyze. The analyst must not extrapolate above or below the calibration range established.

12. DATA ANALYSIS AND CALCULATIONS

12.1 Identify sample components by comparison of retention times to retention data from the calibration standard analysis. If the retention time of an unknown peak corresponds, within limits (Section 12.3), to the retention time of a standard compound, then the identification is considered positive. Calculate analyte concentrations in the samples and reagent blanks from the calibration curves generated in Section 10.1.

12.2 If an average relative response factor has been calculated, analyte concentrations in the samples and reagent blanks are calculated using the following equation:

$$C_a = \frac{(A_a)(C_{is})}{(A_{is})(RRF)}$$

12.3 The width of the retention time window used to make identifications should be based upon measurements of actual retention time variations of standards over the course of a day. Three times the standard deviation of a retention time can be used to calculate a suggested window size for a compound. However, the experience of the analyst should weigh heavily in the interpretation of chromatogram.

13. METHOD PERFORMANCE

13.1 In a single laboratory, accuracy and precision data were obtained at three concentrations in reagent water (Tables 5-8). The MDL and EDL data are given in Tables 2 and 3. In addition, recovery and precision data were obtained at a medium concentration for dechlorinated tap water (Tables 9 and 10), high ionic strength ground water (Tables 11 and 12) and high humectant reagent water (Tables 13 and 14).

14. POLLUTION PREVENTION

14.1 This method utilizes a micro-extraction procedure which requires the use of very small quantities of organic solvents. This feature reduces the hazards involved with the use of large volumes of potentially harmful organic solvents needed for conventional liquid-liquid extractions.

14.2 For information about pollution prevention that may be applicable to laboratory operations consult "Less is Better: Laboratory Chemical Management for Waste Reduction" available from the American Chemical Society's Department of Government Relations and Science Policy, 1155 16th Street N.W., Washington, D.C. 20036.

15. WASTE MANAGEMENT

15.1 Due to the nature of this method there is little need for waste management. No large volumes of solvents or hazardous chemicals are used. The matrices of concern are finished drinking water or source water. However, the Agency requires that laboratory waste management practices be conducted consistent with all applicable rules and regulations, and that laboratories protect the air, water, and land by minimizing and controlling all releases from fume hoods and bench operations. Also compliance is required with any sewage discharge permits and regulations, particularly the hazardous waste identification rules and land disposal restrictions. For further information on waste management, consult "The Waste Management Manual for Laboratory Personnel" also available from the American Chemical Society at the address in Sect. 14.2.

16. REFERENCES

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17. TABLES, DIAGRAMS, FLOWCHARTS, AND VALIDATION DATA

TABLE 1. RETENTION DATA AND CHROMATOGRAPHIC CONDITIONS OF ANALYTE METHYL DERIVATIVES

Analyte	Retention Time, min. Primary column	Confirmatory column
Dalapon	15.43	19.73
3,5-Dichlorobenzoic Acid	34.62	39.98
4-Nitrophenol	38.45	40.31
2,4-Dichlorophenylacetic Acid[(b)]	40.62	48.50
Dicamba	41.88	49.21
Dichlorprop	47.67	53.59
4,4'Dibromooctafluorobiphenyl[(a)]	49.13	55.88
2,4-D	49.83	54.60
Pentachlorophenol	50.18	58.13
Silvex	53.58	59.13
5-Hydroxydicamba	55.32	59.59
2,4,5-T	55.60	60.21
Chloramben	57.17	59.59
2,4-DB	57.38	62.77
Dinoseb	59.68	62.61
Bentazon	61.55	63.64
Dacthal acid metabolites	62.78	67.82
Picloram	65.77	66.70
Acifluorfen	72.57	75.60

DB-1701, 30 m x 0.25 mm i.d., 0.25 µm film thickness, Injector Temp. = column: 200°C, Detector Temp. = 290°C, Helium Linear Velocity = 24 cm/sec at 35°C, Splitless injection with 30 s delay

Program: Hold at 35°C for 10 min, ramp to 150°C at 5C°/min. and hold 10 min., ramp to 222°C at 4C°/min. and hold 5 min, ramp to 260°C at 5C°/min. and hold 6 min.

Confirmatory DB-5.625, 30 m x 0.25 mm i.d., 0.25 µm film thickness, Injector Temp. = column: 200°C, Detector Temp. = 290°C, Linear Helium Velocity = 25 cm/sec at 35°C, splitless injection with 30 s delay.

Program: Hold at 35°C for 10 min, ramp to 150°C at 5C°/min. and hold 10 min., ramp to 222°C at 4C°/min. and hold 5 min, ramp to 260°C at 5C°/min. and hold 6 min.

[(a)] Internal Standard

[(b)] Surrogate Compound

TABLE 2. ANALYTE ACCURACY AND PRECISION DATA AND METHOD DETECTION LIMITS[a]

DERIVATIZATION BY DIAZOMETHANE

LEVEL 1 IN REAGENT WATER

Analyte	Fortified Conc., µg/L	Mean Measured Conc., µg/L	Std. Dev., µg/L	Relative Std. Dev., %	Method Detection Limit[b], µg/L	Estimated Detection Limit[c], µg/L
Dalapon	1.25	1.60	0.31	19	0.97	1.25
3,5-Dichlorobenzoic Acid	0.625	0.804	0.060	7.5	0.19	0.625
4-Nitrophenol[d]	1.25	1.24	0.29	23	0.91	1.25
Dicamba	0.625	0.844	0.095	11	0.30	0.30
Dichlorprop	1.25	1.66	0.16	9.6	0.51	0.51
2,4-D	1.25	1.71	0.11	6.4	0.35	0.35
Pentachlorophenol	0.125	0.176	0.027	15	0.085	0.085
Silvex	0.314	0.409	0.046	11	0.14	0.14
5-Hydroxydicamba[d]	0.625	0.822	0.19	23	0.60	0.625
2,4,5-T	0.314	0.307	0.064	21	0.20	0.20
Chloramben	0.625	0.857	0.079	9.2	0.25	0.25
2,4-DB	1.25	1.61	0.21	13	0.66	1.25
Dinoseb	1.25	1.56	0.26	17	0.82	0.82
Bentazon	1.25	1.46	0.28	19	0.88	1.25
Picloram	1.25	1.31	0.33	25	1.0	1.0
Acifluorfen	0.625	0.720	0.15	21	0.47	0.47
Dacthal Acid Me-tabolites	1.25	1.47	0.20	14	0.63	0.63

[a] Produced by analysis of seven aliquots of fortified reagent water.

[b] The MDL is a statistical estimate of the detection limit. To determine the MDL for each analyte, the standard deviation of the mean concentration of the seven replicates is calculated. This standard deviation is then multiplied by the Student's t-value at 99% confidence and n-1 degrees of freedom (3.143 for seven replicates). The result is the MDL.

[c] The EDL is defined as either the MDL or a level of a compound in a sample yielding a peak in the final extract with a signal to noise (S/N) ratio of approximately 5, whichever is greater.

[d] Quantitation not recommended due to poor precision.

TABLE 3. ANALYTE ACCURACY AND PRECISION DATA AND METHOD DETECTION LIMITS[a]

DERIVATIZATION BY TETRABUTYLAMMONIUM HYDROXIDE AND METHYL IODIDE

LEVEL 1 IN REAGENT WATER

Analyte	Fortified Conc., µg/L	Mean Measured Conc., µg/L	Std. Dev., µg/L	Relative Std. Dev., %	Method Detection Limit[b], µg/L	Estimated Detection Limit[c], µg/L
Dalapon	1.25	1.37	0.17	12	0.53	1.25
3,5-Dichlorobenzoic Acid	0.625	0.781	0.041	5.2	0.13	0.625
4-Nitrophenol	1.25	1.07	0.21	20	0.66	1.25
Dicamba	0.625	0.689	0.065	9.4	0.20	0.20
Dichlorprop	1.25	1.55	0.13	8.4	0.41	0.41
2,4-D	1.25	1.39	0.11	7.9	0.36	0.36
Pentachlorophenol	0.125	0.108	0.0068	6.3	0.021	0.021
Silvex	0.314	0.326	0.023	7.1	0.072	0.072
5-Hydroxydicamba	0.625	0.708	0.068	9.6	0.21	0.21
2,4,5-T	0.314	0.255	0.052	20	0.16	0.16
Chloramben	0.625	0.644	0.044	6.8	0.14	0.14
2,4-DB	1.25	1.47	0.19	13	0.60	0.60
Dinoseb	1.25	1.06	0.24	23	0.75	1.25
Bentazon	1.25	0.927	0.16	17	0.50	1.25
Picloram	1.25	1.12	0.15	13	0.47	0.47
Acifluorfen	0.625	0.639	0.12	21	0.38	0.38
Dacthal Acid Metabolites	1.25	1.52	0.12	7.9	0.38	0.38

[a] Produced by analysis of seven aliquots of fortified reagent water.

[b] The MDL is a statistical estimate of the detection limit. To determine the MDL for each analyte, the standard deviation of the mean concentration of the seven replicates is calculated. This standard deviation is then multiplied by the Student's t-value at 99% confidence and n-1 degrees of freedom (3.143 for seven replicates). The result is the MDL.

[c] The EDL is defined as either the MDL or a level of a compound in a sample yielding a peak in the final extract with a signal to noise (S/N) ratio of approximately 5, whichever is greater.

TABLE 4. ANALYTE ACCURACY AND PRECISION DATA[a]

DERIVATIZATION BY DIAZOMETHANE

LEVEL 2 IN REAGENT WATER

Analyte	Fortified Conc., µg/L	Mean Measured Conc., µg/L	Std. Dev., µg/L	Relative Std. Dev., %	Percent Recovery, %
Dalapon	5.00	5.26	0.34	6.5	105
3,5-Dichlorobenzoic Acid	2.50	3.04	0.27	8.9	122
4-Nitrophenol[b]	5.00	5.69	0.96	17	114
Dicamba	2.50	2.62	0.068	2.6	105
Dichlorprop	5.00	5.97	0.18	3.0	119
2,4-D	5.00	6.34	0.20	3.2	127
Pentachlorophenol	0.500	0.524	0.012	2.3	105
Silvex	1.25	1.26	0.084	6.7	101
5-Hydroxydicamba[b]	2.50	2.32	0.45	19	93
2,4,5-T	1.25	1.30	0.065	5.0	104
Chloramben	2.50	2.51	0.13	5.2	100
2,4-DB	5.00	5.88	0.37	6.3	118
Dinoseb	5.00	5.55	0.21	3.8	111
Bentazon	5.00	5.09	0.18	3.6	102
Picloram	5.00	5.12	0.35	6.8	102
Acifluorfen	2.50	2.82	0.24	8.3	113
Dacthal Acid Metabolites	5.00	5.53	0.38	6.8	111

[a] Produced by the analysis of seven aliquots of fortified reagent water.

[b] Quantitation not recommended due to poor precision.

TABLE 5. ANALYTE ACCURACY AND PRECISION DATA[a]

DERIVATIZATION BY TETRABUTYLAMMONIUM HYDROXIDE AND METHYL IODIDE

LEVEL 2 IN REAGENT WATER

Analyte	Fortified Conc., µg/L	Mean Measured Conc., µg/L	Std. Dev., µg/L	Relative Std. Dev., %	Percent Recovery, %
Dalapon	5.00	6.37	0.67	10	127
3,5-Dichlorobenzoic Acid	2.50	3.26	0.18	5.4	131
4-Nitrophenol	5.00	5.24	0.29	5.5	105
Dicamba	2.50	2.75	0.22	8.2	110
Dichlorprop	5.00	5.70	0.29	5.1	114
2,4-D	5.00	5.59	0.33	5.9	112
Pentachlorophenol	0.500	0.520	0.065	12	104
Silvex	1.25	1.31	0.081	6.2	104
5-Hydroxydicamba	2.50	2.75	0.17	6.0	110
2,4,5-T	1.25	1.20	0.19	16	96
Chloramben	2.50	2.34	0.25	11	94
2,4-DB	5.00	5.46	0.24	4.4	109
Dinoseb	5.00	5.42	0.69	13	108
Bentazon	5.00	4.54	0.33	7.3	91
Picloram	5.00	4.28	0.52	12	86
Acifluorfen	2.50	2.58	0.50	19	103
Dacthal Acid Metabolites	5.00	5.16	0.66	13	103

[a] Produced by the analysis of seven aliquots of fortified reagent water.

TABLE 6. ANALYTE ACCURACY AND PRECISION DATA[a]

DERIVATIZATION BY DIAZOMETHANE

LEVEL 4 IN REAGENT WATER

Analyte	Fortified Conc., µg/L	Mean Measured Conc., µg/L	Std. Dev., µg/L	Relative Std. Dev., %	Percent Recovery, %
Dalapon	10.0	10.3	0.27	2.6	103
3,5-Dichlorobenzoic Acid	5.00	4.63	0.24	5.2	93
4-Nitrophenol[b]	10.0	14.3	3.1	22	143
Dicamba	5.00	4.80	0.083	1.7	96
Dichlorprop	10.0	9.26	0.28	3.1	93
2,4-D	10.0	9.67	0.19	1.9	97
Pentachlorophenol	1.00	0.967	0.015	1.5	97
Silvex	2.50	2.43	0.032	1.3	97
5-Hydroxydicamba[b]	5.00	6.70	2.4	36	134
2,4,5-T	2.50	2.46	0.088	3.6	98
Chloramben	5.00	4.95	0.081	1.6	99
2,4-DB	10.0	9.67	0.22	2.3	97
Dinoseb	10.0	9.78	0.19	2.0	98
Bentazon	10.0	9.78	0.19	1.9	98
Picloram	10.0	10.2	0.25	2.4	102
Acifluorfen	5.00	5.15	0.40	7.8	103
Dacthal Acid Metabolites	10.0	8.85	0.31	3.5	88

[a] Produced by the analysis of seven aliquots of fortified reagent water.

[b] Quantitation not recommended due to poor precision.

TABLE 7. ANALYTE ACCURACY AND PRECISION DATA[a]

DERIVATIZATION BY TETRABUTYLAMMONIUM HYDROXIDE AND METHYL IODIDE

LEVEL 4 IN REAGENT WATER

Analyte	Fortified Conc., µg/L	Mean Measured Conc., µg/L	Std. Dev., µg/L	Relative Std. Dev., %	Percent Recovery, %
Dalapon	10.0	8.37	0.88	10	84
3,5-Dichlorobenzoic Acid	5.00	4.55	0.16	3.5	91
4-Nitrophenol	10.0	9.52	1.0	11	95
Dicamba	5.00	4.55	0.14	3.0	91
Dichlorprop	10.0	8.87	0.18	2.0	89
2,4-D	10.0	9.09	0.38	4.2	91
Pentachlorophenol	1.00	0.870	0.16	19	87
Silvex	2.50	2.29	0.050	2.2	92
5-Hydroxydicamba	5.00	4.23	0.30	7.1	85
2,4,5-T	2.50	2.26	0.12	5.2	90
Chloramben	5.00	4.76	0.34	7.2	95
2,4-DB	10.0	10.2	0.34	3.3	102
Dinoseb	10.0	10.7	3.1	29	107
Bentazon	10.0	9.54	0.84	8.8	95
Picloram	10.0	9.60	0.44	4.6	96
Acifluorfen	5.00	5.27	0.46	8.7	105
Dacthal Acid Metabolites	10.0	8.17	1.2	15	82

[a] Produced by the analysis of seven aliquots of fortified reagent water.

TABLE 8. ANALYTE ACCURACY AND PRECISION DATA[a]

DERIVATIZATION BY DIAZOMETHANE

LEVEL 3 IN DECHLORINATED TAP WATER[b]

Analyte	Background Conc., µg/L	Fortified Conc., µg/L	Mean Measured Conc., µg/L	Std. Dev., µg/L	Relative Std. Dev., %	Percent Recovery, %
Dalapon	<1.25	7.50	8.18	0.93	11	109
3,5-Dichlorobenzoic Acid	<0.625	3.75	4.07	0.38	9.3	108
4-Nitrophenol[c]	<1.25	7.50	5.76	1.0	18	77
Dicamba	<0.30	3.75	3.91	0.16	4.0	104
Dichlorprop	<0.51	7.50	7.29	0.40	5.4	97
2,4-D	<0.35	7.50	7.00	0.38	5.4	93
Pentachlorophenol	<0.085	0.750	0.754	0.016	2.2	101
Silvex	<0.14	1.87	1.70	0.077	4.5	90
5-Hydroxydicamba[c]	<0.625	3.75	0.233	0.12	51	6[d]
2,4,5-T	<0.20	1.87	1.66	0.046	2.8	88
Chloramben	<0.25	3.75	3.93	0.25	6.4	105
2,4-DB	<1.25	7.50	7.51	0.70	9.3	100
Dinoseb	<0.82	7.50	8.02	0.32	4.0	107
Bentazon	<1.25	7.50	7.64	0.44	5.8	102
Picloram	<1.0	7.50	7.91	1.0	13	105
Acifluorfen	<0.47	3.75	3.97	0.38	9.7	106
Dacthal Acid Metabolites	<0.63	7.50	7.87	0.52	6.6	105

[a] Produced by the analysis of seven aliquots of fortified reagent water.

[b] Chlorinated surface water from a local utility to which sodium thiosulfate was added as the dechlorinating agent.

[c] Quantitation not recommended due to poor precision.

[d] As noted in Section 4.6, 5-Hydroxydicamba cannot be recovered from chlorinated waters.

TABLE 9. ANALYTE ACCURACY AND PRECISION DATA[a]

DERIVATIZATION BY TETRABUTYLAMMONIUM HYDROXIDE AND METHYL IODIDE

LEVEL 3 IN DECHLORINATED TAP WATER[b]

Analyte	Background Conc., µg/L	Fortified Conc., µg/L	Mean Measured Conc., µg/L	Std. Dev., µg/L	Relative Std. Dev., %	Percent Recovery, %
Dalapon	<1.25	7.50	8.06	0.39	4.8	108
3,5-Dichlorobenzoic Acid	<0.625	3.75	4.18	0.54	13	111
4-Nitrophenol	<1.25	7.50	6.63	0.78	12	88
Dicamba	<0.20	3.75	3.69	0.14	3.6	98
Dichlorprop	<0.41	7.50	6.70	0.24	3.6	89
2,4-D	<0.36	7.50	6.85	0.55	8.0	91
Pentachlorophenol	<0.021	0.750	0.771	0.071	9.2	103
Silvex	<0.072	1.87	1.69	0.13	7.6	90
5-Hydroxydicamba	<0.21	3.75	0.180	0.048	26	5[c]
2,4,5-T	<0.16	1.87	1.55	0.19	12	83
Chloramben	<0.14	3.75	3.22	0.26	8.1	86
2,4-DB	<0.60	7.50	7.31	0.48	6.6	97
Dinoseb	<1.25	7.50	8.81	1.8	20	117
Bentazon	<1.25	7.50	6.98	0.58	8.3	93
Picloram	<0.47	7.50	7.11	0.74	10	95
Acifluorfen	<0.38	3.75	4.08	0.63	15	109
Dacthal Acid Metabolites	<0.38	7.50	6.71	0.70	10	90

[a] Produced by the analysis of seven aliquots of fortified reagent water.
[b] Chlorinated surface water from a local utility to which sodium thiosulfate was added as the dechlorinating agent.
[c] As noted in section 4.6, 5-Hydroxydicamba cannot be recovered from chlorinated waters.

TABLE 10. ANALYTE ACCURACY AND PRECISION DATA[a]

DERIVATIZATION BY DIAZOMETHANE

LEVEL 3 IN HIGH IONIC STRENGTH WATER[b]

Analyte	Background Conc., µg/L	Fortified Conc., µg/L	Mean Measured Conc., µg/L	Std. Dev., µg/L	Relative Std. Dev., %	Percent Recovery, %
Dalapon	<1.25	7.50	5.98	0.47	7.8	80
3,5-Dichlorobenzoic Acid	<0.625	3.75	3.86	0.14	3.7	103
4-Nitrophenol[c]	<1.25	7.50	10.44	2.1	20	139
Dicamba	<0.30	3.75	3.80	0.098	2.6	101
Dichlorprop	<0.51	7.50	7.12	0.16	2.2	95
2,4-D	<0.35	7.50	6.98	0.14	2.0	93
Pentachlorophenol	<0.085	0.750	0.787	0.053	6.7	105
Silvex	<0.14	1.87	1.76	0.066	3.7	94
5-Hydroxydicamba[c]	<0.625	3.75	5.43	2.0	36	145
2,4,5-T	<0.20	1.87	1.72	0.044	2.6	92
Chloramben	<0.25	3.75	4.16	0.24	5.8	111
2,4-DB	<1.25	7.50	8.84	0.38	4.3	118
Dinoseb	<0.82	7.50	7.74	0.21	2.7	103
Bentazon	<1.25	7.50	7.83	0.44	5.6	104
Picloram	<1.0	7.50	7.29	0.39	5.4	97
Acifluorfen	<0.47	3.75	3.82	0.27	7.2	102
Dacthal Acid Metabolites	<0.63	7.50	7.73	0.28	3.7	103

[a] Produced by the analysis of seven aliquots of fortified reagent water.

[b] Chlorinated ground water from a water source displaying a hardness of 460 mg/L as $CaCO_3$.

[c] Quantitation not recommended due to poor precision.

TABLE 11. ANALYTE ACCURACY AND PRECISION DATA[a]

DERIVATIZATION BY TETRABUTYLAMMONIUM HYDROXIDE AND METHYL IODIDE

LEVEL 3 IN HIGH IONIC STRENGTH WATER[b]

Analyte	Background Conc., µg/L	Fortified Conc., µg/L	Mean Measured Conc., µg/L	Std. Dev., µg/L	Relative Std. Dev., %	Percent Recovery, %
Dalapon	<1.25	7.50	7.33	0.65	8.9	98
3,5-Dichlorobenzoic Acid	<0.625	3.75	4.25	0.29	6.8	113
4-Nitrophenol	<1.25	7.50	7.90	0.51	6.4	105
Dicamba	<0.20	3.75	3.96	0.12	3.0	106
Dichlorprop	<0.41	7.50	7.11	0.74	10	95
2,4-D	<0.36	7.50	6.67	0.17	2.6	89
Pentachlorophenol	<0.021	0.750	1.00	0.068	6.8	134
Silvex	<0.072	1.87	1.80	0.081	4.5	96
5-Hydroxydicamba	<0.21	3.75	3.62	0.12	3.4	96
2,4,5-T	<0.16	1.87	1.65	0.074	4.5	88
Chloramben	<0.14	3.75	3.84	0.30	7.8	102
2,4-DB	<0.60	7.50	7.74	0.54	7.0	103
Dinoseb	<1.25	7.50	7.45	0.72	9.7	99
Bentazon	<1.25	7.50	7.88	0.46	5.8	105
Picloram	<0.47	7.50	6.15	0.63	10	82
Acifluorfen	<0.38	3.75	4.17	0.46	11	111
Dacthal Acid Metabolites	<0.38	7.50	6.82	0.39	5.7	91

[a] Produced by the analysis of seven aliquots of fortified reagent water.

[b] Chlorinated ground water from a water source displaying a hardness of 460 mg/L as $CaCO_3$.

TABLE 12. ANALYTE ACCURACY AND PRECISION DATA[a]

DERIVATIZATION BY DIAZOMETHANE

LEVEL 3 IN HIGH HUMIC CONTENT WATER[b]

Analyte	Background Conc., µg/L	Fortified Conc., µg/L	Mean Measured Conc., µg/L	Std. Dev., µg/L	Relative Std. Dev., %	Percent Recovery, %
Dalapon	<1.25	7.50	8.41	1.92	23	112
3,5-Dichlorobenzoic Acid	<0.625	3.75	4.46	0.38	8.6	119
4-Nitrophenol[c]	<1.25	7.50	6.21	2.4	39	83
Dicamba	<0.30	3.75	4.36	0.19	4.4	116
Dichlorprop	<0.51	7.50	9.22	0.69	7.5	123
2,4-D	<0.35	7.50	9.28	0.77	8.3	124
Pentachlorophenol	<0.085	0.750	0.797	0.020	2.5	106
Silvex	<0.14	1.87	1.96	0.048	2.5	104
5-Hydroxydicamba[c]	<0.625	3.75	2.52	1.4	55	67
2,4,5-T	<0.20	1.87	2.06	0.19	9.1	110
Chloramben	<0.25	3.75	3.86	0.29	7.6	103
2,4-DB	<1.25	7.50	9.10	0.36	3.9	121
Dinoseb	<0.82	7.50	8.66	1.0	12	115
Bentazon	<1.25	7.50	7.89	0.30	3.7	105
Picloram	<1.0	7.50	6.79	1.9	29	91
Acifluorfen	<0.47	3.75	3.67	0.53	14	98
Dacthal Acid Me-tabolites	<0.63	7.50	9.23	1.0	11	123

[a] Produced by the analysis of seven aliquots of fortified reagent water.

[b] Reagent water fortified at 1.0 mg/L with fulvic acid extracted from Ohio River water. Sample simulates high TOC matrix.

[c] Quantitation not recommended due to poor precision.

TABLE 13. ANALYTE ACCURACY AND PRECISION DATA[a]

DERIVATIZATION BY TETRABUTYLAMMONIUM HYDROXIDE AND METHYL IODIDE

LEVEL 3 IN HIGH HUMIC CONTENT STRENGTH WATER[b]

Analyte	Background Conc., µg/L	Fortified Conc., µg/L	Mean Measured Conc., µg/L	Std. Dev., µg/L	Relative Std. Dev., %	Percent Recovery, %
Dalapon	<1.25	7.50	6.82	1.1	16	91
3,5-Dichlorobenzoic Acid	<0.625	3.75	3.86	0.33	8.5	103
4-Nitrophenol	<1.25	7.50	7.04	0.49	7.0	94
Dicamba	<0.20	3.75	3.67	0.13	3.4	98
Dichlorprop	<0.41	7.50	7.24	0.52	7.2	97
2,4-D	<0.36	7.50	6.37	0.93	15	85
Pentachlorophenol	<0.021	0.750	0.736	0.12	16	98
Silvex	<0.072	1.87	1.77	0.19	11	95
5-Hydroxydicamba	<0.21	3.75	3.17	0.34	11	84
2,4,5-T	<0.16	1.87	1.49	0.29	20	79
Chloramben	<0.14	3.75	3.68	0.24	6.6	98
2,4-DB	<0.60	7.50	8.01	0.42	5.2	107
Dinoseb	<1.25	7.50	7.08	0.81	11	94
Bentazon	<1.25	7.50	7.50	0.44	5.9	100
Picloram	<0.47	7.50	5.66	1.0	18	75
Acifluorfen	<0.38	3.75	3.89	0.45	12	104
Dacthal Acid Metabolites	<0.38	7.50	6.93	0.53	7.6	92

[a] Produced by the analysis of seven aliquots of fortified reagent water.

[b] Reagent water fortified at 1.0 mg/L with fulvic acid extracted from Ohio River water. Sample simulates high TOC matrix.

TABLE 14. LABORATORY PERFORMANCE CHECK SOLUTION

PARAMETER	ANALYTE	CONC., µg/L IN WATER SAMPLE	ACCEPTANCE CRITERIA
INSTRUMENT SENSITIVITY	DINOSEB	2.50	DETECTION OF ANALYTE; S/N > 3[a]
CHROMATOGRAPHIC PERFORMANCE	4-NITROPHENOL	2.50	PGF BETWEEN 0.80 AND 1.15[b]
COLUMN PERFORMANCE	CHLORAMBEN 2,4-DB	1.25 2.50	RESOLUTION > 0.50[c]

[a] S/N, a ratio of peak signal to baseline noise.

peak signal - measured as height of peak.
baseline noise - measured as maximum deviation in baseline (in units of height) over a width equal to the width of the base of the peak.

[b] PGF = Peak Gaussian Factor

$$PGF = \frac{1.83 \times W_{1/2}}{W_{1/10}}$$

where $W_{1/2}$ = the peak width at half height.
$W_{1/10}$ = the peak width at one-tenth height.

This is a measure of the symmetry of the peak.

[c] Resolution between two peaks is defined by the equation:

$$R = \frac{t}{W_{ave}}$$

where t = the difference in elution times between the two peaks.
W_{ave} = the average peak width of the two peaks (measurements taken at baseline).

This a measure of the degree of separation of two peaks under specific chromatographic conditions.

FIGURE 1. APPARATUS FOR GENERATION OF DIAZOMETHANE

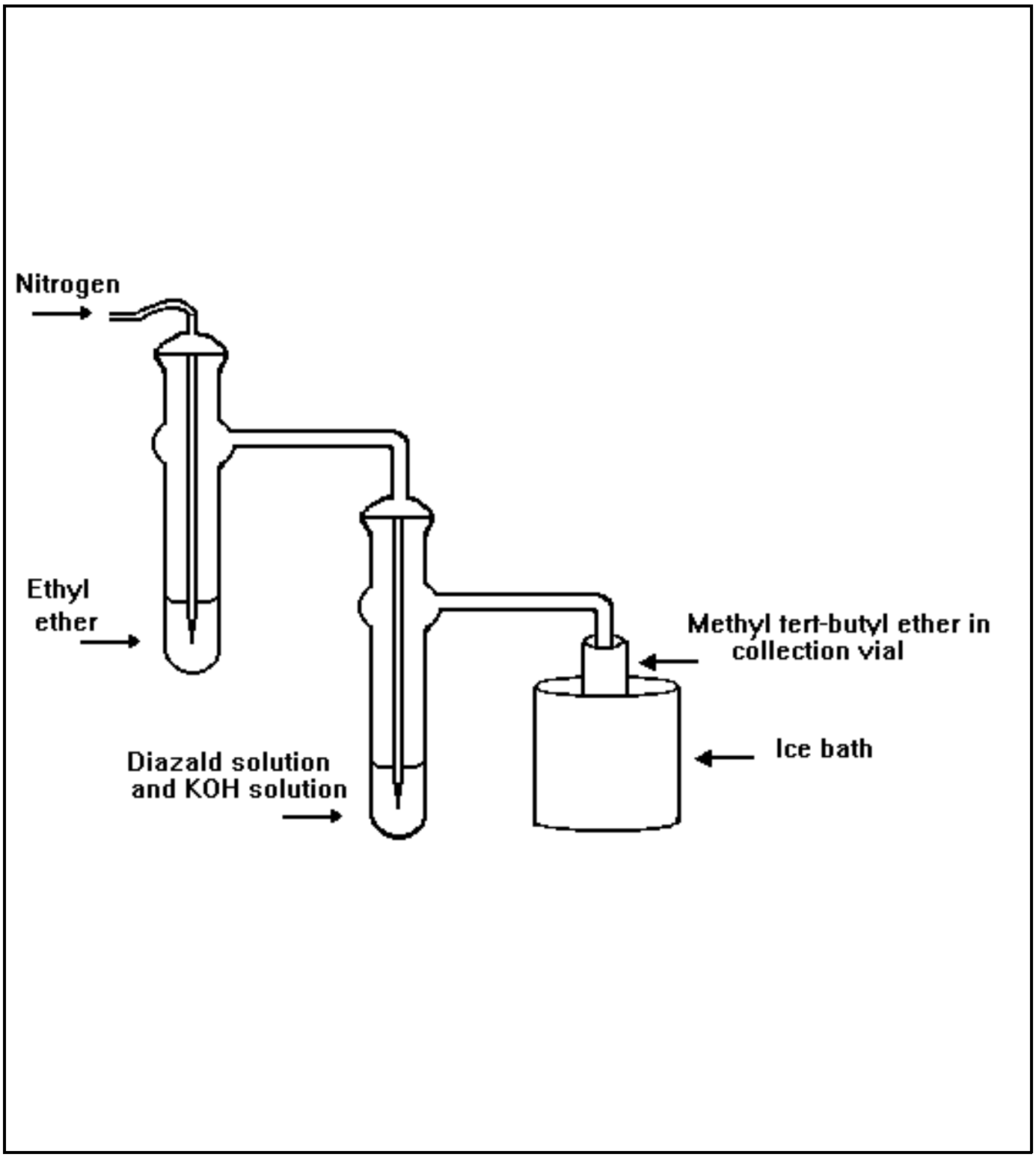

FIGURE 2. CHROMATOGRAM OF CHLOROPHENOXY HERBICIDES ON DB-1701. DERIVATIZATION BY DIAZOMETHANE (LEVEL 4 CALIBRATION)

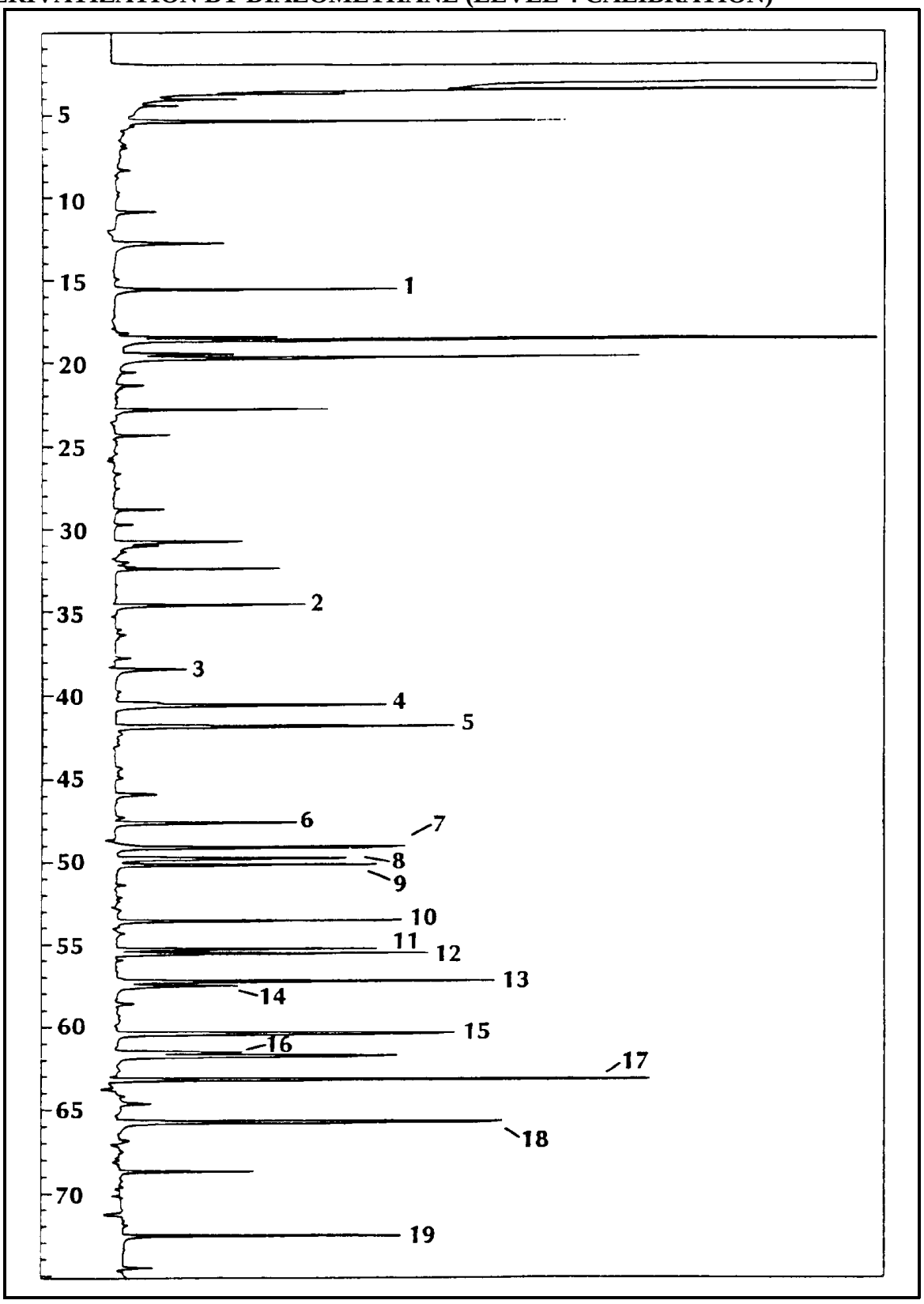

FIGURE 3. CHROMATOGRAM OF CHLOROPHENOXY HERBICIDES ON DB-5.625. BASE-PROMOTED DERIVATIZATION (LEVEL 2 CALIBRATION)

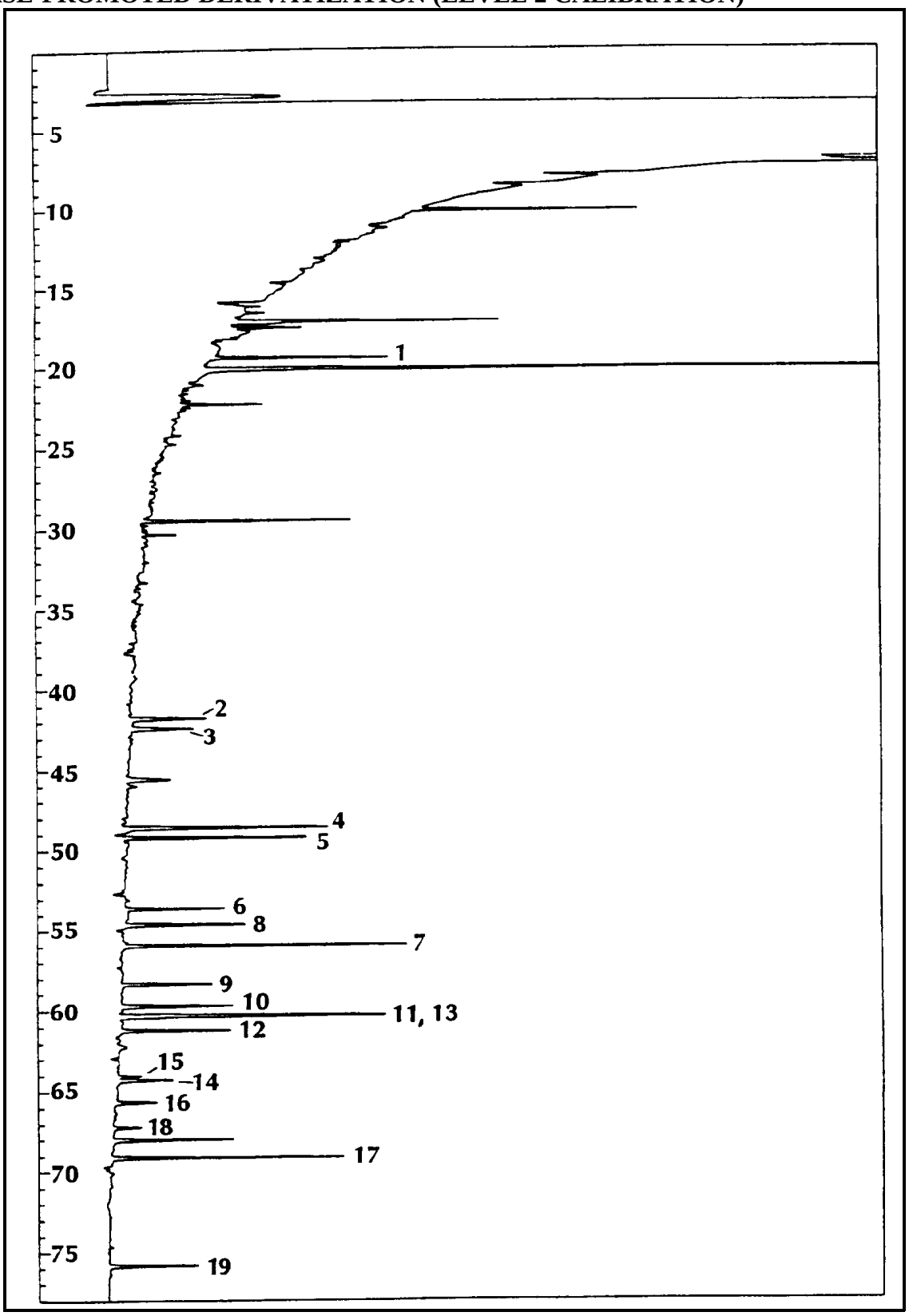

TABLE 15. KEY FOR PEAK NUMBERS DISPLAYED IN FIGURES 2 AND 3

Peak Number	Method 515.3 compound
1	Dalapon
2	3,5-Dichlorobenzoic Acid
3	4-Nitrophenol
4	2,4-Dichlorophenylacetic Acid (Surrogate)
5	Dicamba
6	Dichlorprop
7	4,4'-Dibromooctafluorobiphenyl (Internal Standard)
8	2,4-D
9	Pentachlorophenol
10	Silvex (2,4,5-TP)
11	5-Hydroxydicamba
12	2,4,5-T
13	Chloramben
14	2,4-DB
15	Dinoseb
16	Bentazon
17	Dacthal Acid Metabolites
18	Picloram
19	Acifluorfen

TABLE 16. HOLDING TIME STUDY FOR AQUEOUS SAMPLES

DERIVATIZATION BY TETRABUTYLAMMONIUM HYDROXIDE AND METHYL IODIDE

DAY 0

Analyte	Fortified Conc., µg/L	Mean Measured Conc., µg/L	Std. Dev., µg/L	Relative Std. Dev., %	Percent Recovery, %
Dalapon	10.0	8.37	0.88	10	84
3,5-Dichlorobenzoic Acid	5.00	4.55	0.16	3.5	91
4-Nitrophenol	10.0	9.52	1.0	11	95
Dicamba	5.00	4.55	0.14	3.0	91
Dichlorprop	10.0	8.87	0.18	2.0	89
2,4-D	10.0	9.09	0.38	4.2	91
Pentachlorophenol	1.00	0.870	0.16	19	87
Silvex	2.50	2.29	0.050	2.2	92
5-Hydroxydicamba	5.00	4.23	0.30	7.1	85
2,4,5-T	2.50	2.26	0.12	5.2	90
Chloramben	5.00	4.76	0.34	7.2	95
2,4-DB	10.0	10.2	0.34	3.3	102
Dinoseb	10.0	10.7	3.1	29	107
Bentazon	10.0	9.54	0.84	8.8	95
Picloram	10.0	9.60	0.44	4.6	96
Acifluorfen	5.00	5.27	0.46	8.7	105
Dacthal Acid Metabolites	10.0	8.17	1.2	15	82

[a] Produced by the analysis of seven aliquots of fortified reagent water.

TABLE 17. HOLDING TIME STUDY FOR AQUEOUS SAMPLES

DERIVATIZATION BY TETRABUTYLAMMONIUM HYDROXIDE AND METHYL IODIDE

DAY 7

Analyte	Fortified Conc., µg/L	Mean Measured Conc., µg/L	Std. Dev., µg/L	Relative Std. Dev., %	Percent Recovery, %
Dalapon	10.0	9.48	1.4	15	95
3,5-Dichlorobenzoic Acid	5.00	4.98	0.19	3.8	100
4-Nitrophenol	10.0	9.65	0.38	4.0	97
Dicamba	5.00	4.99	0.052	1.0	100
Dichlorprop	10.0	9.86	0.16	1.6	99
2,4-D	10.0	9.76	0.19	2.0	98
Pentachlorophenol	1.00	1.10	0.15	14	110
Silvex	2.50	2.62	0.057	2.2	105
5-Hydroxydicamba	5.00	5.07	0.19	3.8	101
2,4,5-T	2.50	2.75	0.20	7.2	110
Chloramben	5.00	5.32	0.16	3.1	106
2,4-DB	10.0	10.3	0.22	2.1	103
Dinoseb	10.0	9.50	2.4	26	95
Bentazon	10.0	10.9	0.35	3.2	109
Picloram	10.0	11.5	2.4	21	115
Acifluorfen	5.00	5.92	0.38	6.4	118
Dacthal Acid Metabolites	10.0	10.0	0.57	5.7	100

[a] Produced by the analysis of seven aliquots of fortified reagent water.

TABLE 18. HOLDING TIME STUDY FOR AQUEOUS SAMPLES

DERIVATIZATION BY TETRABUTYLAMMONIUM HYDROXIDE AND METHYL IODIDE

DAY 14

Analyte	Fortified Conc., µg/L	Mean Measured Conc., µg/L	Std. Dev., µg/L	Relative Std. Dev., %	Percent Recovery, %
Dalapon	10.0	9.53	0.27	2.8	95
3,5-Dichlorobenzoic Acid	5.00	4.85	0.16	3.4	97
4-Nitrophenol	10.0	9.18	0.32	3.5	92
Dicamba	5.00	4.60	0.15	3.3	92
Dichlorprop	10.0	8.98	0.23	2.5	90
2,4-D	10.0	8.73	0.20	2.3	87
Pentachlorophenol	1.00	1.22	0.15	12	122
Silvex	2.50	2.42	0.066	2.7	97
5-Hydroxydicamba	5.00	4.26	0.066	1.6	85
2,4,5-T	2.50	2.28	0.12	5.2	91
Chloramben	5.00	4.65	0.12	2.7	93
2,4-DB	10.0	9.30	0.20	2.1	93
Dinoseb	10.0	10.1	1.2	12	101
Bentazon	10.0	9.92	0.33	3.3	99
Picloram	10.0	9.43	2.2	23	94
Acifluorfen	5.00	4.89	0.28	5.6	98
Dacthal Acid Metabolites	10.0	8.23	0.67	8.1	82

[a] Produced by the analysis of seven aliquots of fortified reagent water.

TABLE 19. HOLDING TIME STUDY FOR AQUEOUS SAMPLES

DERIVATIZATION BY TETRABUTYLAMMONIUM HYDROXIDE AND METHYL IODIDE

DAY 21

Analyte	Fortified Conc., µg/L	Mean Measured Conc., µg/L	Std. Dev., µg/L	Relative Std. Dev., %	Percent Recovery, %
Dalapon	10.0	9.65	1.3	13	97
3,5-Dichlorobenzoic Acid	5.00	5.32	0.49	9.2	106
4-Nitrophenol	10.0	9.32	0.81	8.7	93
Dicamba	5.00	4.64	0.27	5.8	93
Dichlorprop	10.0	9.34	0.46	4.9	93
2,4-D	10.0	8.35	0.13	1.6	84
Pentachlorophenol	1.00	1.10	0.071	6.5	110
Silvex	2.50	2.10	0.13	6.2	84
5-Hydroxydicamba	5.00	3.76	0.19	5.0	75
2,4,5-T	2.50	1.97	0.24	12	79
Chloramben	5.00	3.74	0.46	12	75
2,4-DB	10.0	8.32	0.38	4.6	83
Dinoseb	10.0	9.66	1.8	19	97
Bentazon	10.0	8.63	0.81	9.4	86
Picloram	10.0	11.4	2.9	25	114
Acifluorfen	5.00	4.20	0.43	10	84
Dacthal Acid Metabolites	10.0	8.22	0.78	9.5	82

[a] Produced by the analysis of seven aliquots of fortified reagent water.

TABLE 20. HOLDING TIME STUDY FOR MTBE EXTRACTS

DERIVATIZATION BY TETRABUTYLAMMONIUM HYDROXIDE AND METHYL IODIDE

DAY 0

Analyte	Fortified Conc., µg/L	Mean Measured Conc., µg/L	Std. Dev., µg/L	Relative Std. Dev., %	Percent Recovery, %
Dalapon	10.0	8.37	0.88	10	84
3,5-Dichlorobenzoic Acid	5.00	4.55	0.16	3.5	91
4-Nitrophenol	10.0	9.52	1.0	11	95
Dicamba	5.00	4.55	0.14	3.0	91
Dichlorprop	10.0	8.87	0.18	2.0	89
2,4-D	10.0	9.09	0.38	4.2	91
Pentachlorophenol	1.00	0.870	0.16	19	87
Silvex	2.50	2.29	0.050	2.2	92
5-Hydroxydicamba	5.00	4.23	0.30	7.1	85
2,4,5-T	2.50	2.26	0.12	5.2	90
Chloramben	5.00	4.76	0.34	7.2	95
2,4-DB	10.0	10.2	0.34	3.3	102
Dinoseb	10.0	10.7	3.1	29	107
Bentazon	10.0	9.54	0.84	8.8	95
Picloram	10.0	9.60	0.44	4.6	96
Acifluorfen	5.00	5.27	0.46	8.7	105
Dacthal Acid Metabolites	10.0	8.17	1.2	15	82

[a] Produced by the analysis of seven aliquots of fortified reagent water.

TABLE 21. HOLDING TIME STUDY FOR MTBE EXTRACTS

DERIVATIZATION BY TETRABUTYLAMMONIUM HYDROXIDE AND METHYL IODIDE

DAY 7

Analyte	Fortified Conc., µg/L	Mean Measured Conc., µg/L	Std. Dev., µg/L	Relative Std. Dev., %	Percent Recovery, %
Dalapon	10.0	9.67	1.6	17	97
3,5-Dichlorobenzoic Acid	5.00	5.50	0.60	11	110
4-Nitrophenol	10.0	10.7	1.0	9.7	107
Dicamba	5.00	5.31	0.18	3.4	106
Dichlorprop	10.0	10.4	0.50	4.8	104
2,4-D	10.0	10.4	0.66	6.3	104
Pentachlorophenol	1.00	1.21	0.23	19	121
Silvex	2.50	2.64	0.17	6.5	106
5-Hydroxydicamba	5.00	5.32	0.38	7.1	106
2,4,5-T	2.50	2.58	0.32	12	103
Chloramben	5.00	5.75	0.60	10	115
2,4-DB	10.0	11.0	0.24	2.1	110
Dinoseb	10.0	9.60	0.91	9.5	96
Bentazon	10.0	11.4	0.98	8.6	114
Picloram	10.0	13.8	1.3	9.6	138
Acifluorfen	5.00	6.27	0.88	14	125
Dacthal Acid Metabolites	10.0	10.8	1.1	10	108

[a] Produced by the analysis of seven aliquots of fortified reagent water.

TABLE 22. HOLDING TIME STUDY FOR MTBE EXTRACTS

DERIVATIZATION BY TETRABUTYLAMMONIUM HYDROXIDE AND METHYL IODIDE

DAY 14

Analyte	Fortified Conc., µg/L	Mean Measured Conc., µg/L	Std. Dev., µg/L	Relative Std. Dev., %	Percent Recovery, %
Dalapon	10.0	10.1	0.72	7.2	101
3,5-Dichlorobenzoic Acid	5.00	5.55	0.35	6.3	111
4-Nitrophenol	10.0	10.8	1.1	9.8	108
Dicamba	5.00	5.40	0.30	5.5	108
Dichlorprop	10.0	10.1	0.42	4.1	101
2,4-D	10.0	10.1	0.50	5.0	101
Pentachlorophenol	1.00	1.07	0.12	11	107
Silvex	2.50	2.57	0.16	6.2	103
5-Hydroxydicamba	5.00	5.13	0.40	7.8	103
2,4,5-T	2.50	2.43	0.21	8.6	97
Chloramben	5.00	5.40	0.68	12	108
2,4-DB	10.0	10.5	0.74	7.0	105
Dinoseb	10.0	9.68	2.0	21	97
Bentazon	10.0	10.8	1.2	11	108
Picloram	10.0	13.3	2.6	19	133
Acifluorfen	5.00	5.17	0.79	15	103
Dacthal Acid Metabolites	10.0	10.1	1.3	13	101

[a] Produced by the analysis of seven aliquots of fortified reagent water.

TABLE 23. HOLDING TIME STUDY FOR MTBE EXTRACTS

DERIVATIZATION BY TETRABUTYLAMMONIUM HYDROXIDE AND METHYL IODIDE

DAY 21

Analyte	Fortified Conc., µg/L	Mean Measured Conc., µg/L	Std. Dev., µg/L	Relative Std. Dev., %	Percent Recovery, %
Dalapon	10.0	10.8	1.8	16	108
3,5-Dichlorobenzoic Acid	5.00	5.72	0.88	15	114
4-Nitrophenol	10.0	10.1	1.7	16	101
Dicamba	5.00	5.13	0.50	9.7	103
Dichlorprop	10.0	10.4	0.71	6.8	104
2,4-D	10.0	9.51	0.85	9.0	95
Pentachlorophenol	1.00	1.06	0.27	26	106
Silvex	2.50	2.65	0.28	10	106
5-Hydroxydicamba	5.00	5.20	0.82	16	104
2,4,5-T	2.50	2.50	0.27	11	100
Chloramben	5.00	5.36	0.58	11	107
2,4-DB	10.0	11.1	1.2	11	111
Dinoseb	10.0	8.92	1.6	18	89
Bentazon	10.0	10.2	0.74	7.3	102
Picloram	10.0	12.6	1.0	8.1	126
Acifluorfen	5.00	5.67	0.71	12	113
Dacthal Acid Metabolites	10.0	14.0	1.1	7.9	140

[a] Produced by the analysis of seven aliquots of fortified reagent water.