Cover Sheet for

ENVIRONMENTAL CHEMISTRY METHOD

Pestcide Name: Endosulfan I

MRID #: 411641-01

Matrix: Water

Analysis: GC/ECD

OFFICIAL COPY

STANDARD OPERATING PROCEDURE FOR DETERMINATION OF ENDOSULFANS IN POND AND RUN-OFF WATERS

1. SCOPE

1.1. This standard operating procedure (SOP) describes the analysis of α-endosulfan, β-endosulfan, and endosulfan sulfate (hereafter referred to as "endosulfans") in pond and run-off water samples.

2. SUMMARY

2.1. One liter of water is extracted with methylene chloride using a automatic separatory funnel shaker, and the extract is concentrated to 1 ml after solvent substitution with hexane. The extract is analyzed by capillary column gas chromatography (GC) using an electron capture detector (ECD). Optional Florisil and silica gel adsorption chromatography cleanup procedures are included in this SOP.

3. APPARATUS

3.1. Glassware. The required glassware must be solvent cleaned and heated at 400-500 C for at least 4 hours following SOP ASCC-50-019-01, or glassware may be cleaned as described in the SOP except for heating, then rinsed with methanol and methylene chloride.

- 3.1.1. Kimax separatory funnel -- 2-l, with a Teflon stopcock and stopper
- 3.1.2. Graduated cylinders -- 1-l, 500-ml, and 50-ml
- 3.1.3. Kuderna-Danish (K-D) equipment
  - 3.1.3.1. Concentration tube -- 25-ml
  - 3.1.3.2. Flask -- 500-ml
  - 3.1.3.3. Macro-Snyder column -- 3-ball chambers
  - 3.1.3.4. Micro-Snyder column -- 3-ball chambers
- 3.1.4. Collection flask -- 500-ml Erlenmeyer or round-bottom
- 3.1.5. Serological pipet -- 5-ml disposable
- 3.1.6. Vials -- 4-dram Teflon-lined screw caps

3.2. Miscellaneous materials

- 3.2.1. Pyrex glass wool -- heated at 400-500 C for at least 4 hours
- 3.2.2. Carborundum boiling chips -- heated at 400-500 C for at least 4 hours
- 3.2.3. Water bath -- Blue M Magniwhirl or equivalent.
- 3.2.4. Analytical balance -- capable of weighing with an accuracy of ±0.0001 g

3.2.5. Nitrogen evaporation device -- N-Evap Organomation Associates, or equivalent, with constant temperature water bath. The nitrogen gas must be filtered through activated charcoal.

3.2.6. Separatory funnel shaker -- capable of holding eight 2-l separatory funnels and shaking them with a rocking motion to achieve thorough mixing of separatory funnel contents (available from Eberbach Co., Ann Arbor, Michigan)

3.2.7. Gas Chromatograph -- analytical system complete with GC suitable for use with capillary columns and all required accessories including syringes, analytical columns, gases, and electron capture detector

3.2.7.1. Capillary column -- 30 meters long x 0.25 mm I.D. SPB-5 bonded fused silica column, 0.25 μm film thickness (available from Supelco). Alternative columns may be used in accordance with the provisions described in Section 5.2.

3.2.7.2. Detector -- ECD. Alternative detectors, including a mass spectrometer, may be used in accordance with the provisions described in Section 5.2.

3.3. Reagents

3.3.1. Solvents -- Burdick and Jackson distilled-in-glass grade methylene chloride, hexane, unpreserved ethyl ether, and ethyl acetate

3.3.2. Reagent water -- Millipore water or distilled water from Magnetic Springs Water Company (Columbus, Ohio) or equivalent

3.3.3. Sodium sulfate -- granular, anhydrous, heated at 400-500 C for at least 4 hours

3.3.4. Sodium chloride -- heated at 400-500 C for at least 4 hours

3.3.5. 0.1 N Hydrochloric acid solution -- dilute 8 ml of concentrated HCl to one liter with distilled water

3.3.6. 0.1 M Dipotassium phosphate solution -- dissolve 17.4 g of dipotassium phosphate in one liter of distilled water

3.3.7. Phosphate buffer solution -- prepare by mixing 29.6 ml 0.1 N HCl and 50 ml 0.1 M dipotassium phosphate

3.3.8. Florisil -- 60 to 100 mesh, J. T. Baker or equivalent, activated by heating at 140 C overnight

3.3.9. Silica gel -- 100-200 mesh, chromatographic grade, Sigma Chemical Company or equivalent, activated by heating at 140 C overnight

3.3.10. 2,3,4,5,2'-Pentachlorobiphenyl -- >95% purity, for use as internal standard (Note: another polychlorinated biphenyl isomer, 2,3,4,2',5'-pentachlorobiphenyl, can also be successfully substituted as the internal standard)

3.3.11. Stock standard solution of endosulfans (1 mg/ml) -- The stock standard solution is prepared from pure standard materials using the following procedures:

3.3.11.1. Prepare stock standard solution by accurately weighing 10 mg each of α-endosulfan, β-endosulfan, and endosulfan sulfate, dissolving the materials in ethyl acetate and diluting to volume with ethyl acetate in a 10-ml volumetric flask. Larger quantities may be prepared if necessary. If compound purity is certified at 95% or greater, the weight may be used without correction to calculate the concentration of the stock standard. Addition of 25 µl of the stock standard solution to one liter of water results in a concentration of 25 µg/l for each endosulfan.

3.3.11.2. Transfer the stock standard solution into a screw-cap vial with a Teflon-lined cap. Store at 4±2 C and protect from light.

3.3.11.3. The stock standard solution must be replaced after three months or sooner if GC-ECD analyses indicate a problem.

3.3.12. Pond water spike solution (0.5 µg/ml) -- Add 50 µL of the stock standard solution (Section 3.3.11.) into a 100-ml volumetric flask, and dilute to volume with ethyl acetate. Addition of 100 µl of the pond water spike solution to one liter of water results in a concentration of 50 ng/l for each endosulfan. Transfer the pond water spike solution into a screw-cap vial with a Teflon-lined cap. Store at 4±2 C and protect from light. The pond water spike solution must be replaced after three months or sooner if comparison GC-ECD analyses indicate a problem.

3.3.13. Internal standard spike solution (0.5 mg/ml) -- Prepare the internal standard spiking solution by accurately weighing 25 mg of pure 2,3,4,5,2'-pentachlorobiphenyl, dissolving the compound in hexane, and diluting to volume with hexane in a 50-ml volumetric flask. Transfer the internal standard spiking solution to a Teflon-lined screw-top bottle and store at room temperature. Addition of 5 µl of the internal standard spiking solution to 1 ml of sample extract yields an internal standard concentration of 2.5 μg/ml.

4. CALIBRATION

4.1. Establish retention times of each analyte and the internal standard. Calibrate the GC-ECD using the internal standard method.

4.2. Internal standard calibration procedure --

4.2.1. Prepare calibration solutions containing 2.5, 5, 10, 25, 50, 100, 250, 500, and 1000 ng/ml each of α-endosulfan, β-endosulfan, and endosulfan sulfate and 2.5 μg/ml of internal standard. Prepare calibration solutions by adding volumes of the spike solutions (Sections 3.3.11. and 3.3.12.) to a volumetric flask. Add the appropriate constant amount of the internal standard solution (Section 3.3.13.) to each calibration standard, and dilute to volume with hexane.

4.2.2. One of the calibration standards should represent the analyte concentration near, but above, the estimated detection limit. The other concentrations should correspond to the range of concentrations expected in the samples or should define the linear range of the instrument.

4.2.3. Inject 2 μl of each calibration standard and tabulate the relative response for each analyte (RR_a) to an internal standard using the equation:

$$RR_a = A_a/A_{is}$$

where: A_a = analyte peak area, and
A_{is} = internal standard peak area.

Generate a calibration curve of analyte concentration response, RR_a, versus analyte concentration in the extract in ng/ml.

4.2.4. The working calibration curve must be verified on each working shift by the measurement of one or more calibration standards. If the response for any analyte varies from the predicted response by more than ±20%, the test must be repeated using a fresh calibration standard. Alternatively, a new calibration curve must be prepared for that analyte.

5. QUALITY CONTROL

5.1. The minimum quality control requirements for this program consist of the following: an initial demonstration of laboratory capability; the analysis of spiked control samples (prepared in the field) as a continuing check on sample integrity; the analysis of background control samples (prepared in the field) as a continuing check on sample cross-contamination; the analysis of spiked process blanks (prepared in the analytical laboratory) as a continuing check on analytical method performance; and the analysis of process blanks (prepared in the analytical laboratory) as a continuing check on laboratory contamination.

5.2. In recognition of the rapid advances occurring in chromatography, the analyst is permitted to modify GC columns, GC conditions, or detectors to improve the separations or lower the cost of measurements. In addition, the analyst is also permitted to introduce a cleanup procedure to permit lower detection limits in a specific water sample.

5.2.1. Each time such modifications are made, the laboratory must demonstrate acceptable method performance by extracting four representative water samples, three spiked at 5-15 times the estimated method detection limit and one unspiked. The average recovery of each endosulfan must be between 70 and 130 percent, and the relative standard deviation of the three measurements must be equal to or less than 20 percent. Alternatively, the demonstration described in Section 5.2.2. can be substituted.

5.2.2. If a lower method detection limit is claimed, the laboratory must demonstrate acceptable method performance by extracting four representative water samples, three spiked at the estimated method detection limit and one unspiked. The average recovery of each endosulfan should be between 70 and 130 percent, and the relative standard deviation of the three measurements should be equal to or less than 20 percent. The level of interferences detected as one of the endosulfans in the unspiked water sample must be less than half of the claimed method detection limit.

5.3. Assessing Laboratory Performance -- The laboratory must, on an ongoing basis, analyze at least one spiked process blank per sample set. A sample set consists of 22 samples, one process blank, and one spiked process blank.

5.3.1. Pond water -- Spike a 1-l aliquot of reagent water with 100 µl of the pond water spike solution (3.3.12.) and process. Analyze the sample to determine the concentration of each analyte in the final extract (A). Calculate percent recovery for each analyte (Ri) as (100 x A)%/T, where T is the known true concentration of the spike.

5.3.2. Run-off water -- Spike a 1-l aliquot of reagent water with 25 µl of the stock standard solution (3.3.11.) and process and analyze the sample to determine the concentration of each analyte in the final extract (A). Calculate percent recovery for each analyte (Ri) as described in Section 5.3.2.

5.3.3. Monitor the percent recovery (R_i) for each analyte. The recoveries must be within ±30% of the true value. If the recovery of analyte falls outside the designated range, the laboratory performance for that analyte is judged to be out of control, and the source of the problem must be immediately identified and resolved before continuing analyses. The analytical results for that analyte in samples is suspect and must be so labelled. All results for that analyte in that sample set must also be labelled suspect.

5.4. Assessing Sample Integrity -- Spiked control samples, prepared in the field will be analyzed on a regular basis.

5.4.1. Monitor and report all data from the spiked samples.

5.4.2. If the recovery of any analyte falls outside the range specified in Section 5.3.3. and the laboratory performance for that analyte is judged to be in control, the recovery problem encountered with the dosed sample is judged to be matrix-related, not system-related. The result for that analyte in unspiked samples is labelled suspect/matrix to indicate that the results were suspect due to matrix effects.

5.5. Assessing Laboratory Contamination -- Before processing any samples, the analyst must demonstrate that all glassware and reagent interferences are under control. This is accomplished by the analysis of method blanks and unspiked control samples.

5.5.1. A method blank is a 1-l aliquot of reagent water analyzed as if it were a sample. A method blank is prepared and analyzed for each sample set (Section 5.3.) or when there is a change in reagents as a continuing check on laboratory contamination. The level of interferences detected as one of the endosulfans in the unspiked water sample must be less than half of the claimed method detection limit.

5.5.2. Unspiked control samples, prepared in the field, will be analyzed on a regular basis. Monitor and report all data from the unspiked samples. Detectable levels of any of the endosulfans may indicate sample cross-contamination during handling, shipping or storage.

6. PROCEDURE

6.1. Thoroughly thaw and mix the sample prior to analysis. Any suspended particulate is considered part of the sample. Measure approximately one liter of sample into a 1000-ml graduated cylinder. If the available sample volume is less than 800 ml, dilute to one liter with reagent water. Pour the sample into a 2-l separatory funnel. Record the sample volume (prior to dilution with reagent water) to the nearest 10 ml in the laboratory record book.

6.2. If the pH of the sample was not adjusted to pH 7 during sample collection, add 50 ml of phosphate buffer (Section 3.3.7.) to the sample.

6.2. Add 100 grams of sodium chloride to the sample, seal the separatory funnel, and shake to dissolve the salt.

6.3. Measure 300 ml of methylene chloride in the 1000-ml graduated cylinder. If the original sample container contained more than one liter of water, transfer all the methylene chloride to the 2-l separatory funnel, seal and refreeze the remaining water in the original sample bottle; proceed to Section 6.4. If the entire sample was transferred to the 2-l separatory funnel, add 50 ml of the methylene chloride to the sample bottle, seal, and shake 30 sec to rinse the inner walls. Transfer the solvent to the separatory funnel. Add another 50-ml aliquot of the methylene chloride to the sample bottle, shake for 30 sec, then add to the separatory funnel. Pour the remaining methylene chloride into the separatory funnel.

6.4. Place the stopper in the separatory funnel and shake for 10 sec, venting periodically. Repeat shaking and venting until pressure release is not observed during venting. Place separatory funnel in the automatic separatory funnel shaker, and shake the sample for 30 min. Complete mixing of the organic and aqueous phases should be observed within two min after starting the shaker.

6.5. Remove the separatory funnel from the shaker and place in a ring-stand. Allow the organic layer to separate from the aqueous layer for a minimum of 10 min. If an emulsion interface between the two layers is more than one-third of the organic solvent layer, the analyst must employ techniques to complete the phase separation. The optimum technique depends on the sample, but may include stirring, filtration through glass wool, centrifugation, or other physical methods. Collect the methylene chloride extract in a 500-ml flask containing approximately 5 g anhydrous sodium sulfate. Swirl the flask to dry the extract; more sodium sulfate may be added if necessary to absorb excess water. Allow the flask to sit for 15 min.

6.6. Assemble a K-D concentrator by attaching a 25-ml concentrator tube to a 500-ml flask. Decant the methylene chloride extract into the K-D concentrator. Rinse the sodium sulfate with two 25-ml portions of methylene chloride; decant the rinses into the K-D concentrator.

6.7. Add 1-2 clean boiling chips to the evaporative flask and attach a macro-Snyder column which has been prewetted with methylene chloride. Place the K-D apparatus in a 65-70 C water bath; the concentrator tube should be partially immersed in the hot water and the flask should be bathed with hot vapor. At the proper rate of distillation, the balls of the column will actively chatter, but the chambers will not flood. When the apparent volume reaches 2 ml, remove the K-D apparatus from the bath and allow to cool for at least 10 min.

6.8. Remove the macro-Snyder column and rinse the flask and its lower joint into the concentrator tube with 1-2 ml of methylene chloride. Add 5 ml of hexane and a fresh boiling chip to the sample. Attach a micro-Snyder column which has been pre-wetted with hexane to the concentrator tube. Place the concentrator tube in a 85-90 C water bath. When the apparent volume reaches 2 ml, remove the apparatus from the water bath and allow to cool for 10 min. Add 5 ml hexane and reconcentrate the sample to approximately 0.5 ml. The sample may be concentrated under a stream of nitrogen, if desired. Remove the sample from the bath and allow it to cool. Remove the micro-Snyder column, and adjust the volume to 1 ml with hexane.

6.9. Florisil cleanup --

6.9.1. Add 1 g of Florisil to a serological pipet containing a plug of glass wool in the tip. Thoroughly wet the Florisil with hexane. Do not allow the Florisil to become exposed to the air during the fractionation procedure. When the liquid level is just above the adsorbent, apply the sample. Rinse the concentrator tube with 2 ml of hexane and add the rinse to the column; repeat. Add an additional 6 ml of hexane to the column. Discard the eluate.

6.9.2. Apply 2 ml, 2 ml, then 6 ml of 50% ethyl ether in hexane to the column. Collect the eluate in a concentrator tube.

6.9.3. Concentrate the eluate to approximately 0.5 ml in a water bath or under a stream of nitrogen; dilute the extract to 1 ml with hexane.

6.10. Silica gel cleanup (alternative to Florisil cleanup) --

6.10.1. Add 1 g of silica gel to a serological pipet containing a plug of glass wool in the tip. Thoroughly wet the silica gel with hexane. Do not allow the silica gel to become exposed to the air during the fractionation procedure. When the liquid level is just above the adsorbent, apply the sample. Rinse the concentrator tube with 2 ml of hexane and add the rinse to the column; repeat. Add an additional 6 ml of hexane to the column. Discard the eluate.

6.10.2. Apply 2 ml, 2 ml, then 6 ml of 50% ethyl ether in hexane to the column. Collect the eluate in a concentrator tube.

6.10.3. Concentrate the eluate to approximately 0.5 ml in a water bath (K-D) or under a stream of nitrogen; dilute the extract to 1 ml with hexane.

6.11. Spike the sample with 5 µl of the internal standard spike solution (Section 3.3.13.) and mix on a vortex mixer. Transfer the sample to a GC vial and store the extract at 4±2 C until analysis by GC-ECD. After analysis by GC-ECD, reseal the sample in the GC vial with a new septa and store at -20±2 C or lower.

7. GAS CHROMATOGRAPHY

7.1. Table 1 summarizes the recommended operating conditions for the gas chromatograph. Included in Table 1 are retention times observed using this method. Other GC columns, chromatographic conditions, or detectors may be used if the requirements of Section 5.2. are met.

7.2. Calibrate the system daily as described in Section 4. The standards and extracts must be in hexane.

7.3. Inject 2 µl of the sample extract. Record the resulting peak size in area units.

7.4. If the response for the peak exceeds the calibrated working range of the system, dilute the extract, adjust the internal standard concentration by adding additional internal standard spike solution (Section 3.3.13.), and reanalyze.

8. CALCULATIONS

8.1. Calculate analyte concentrations in the sample extract (C_e) in ng/ml from the relative response of the analyte to the internal standard (RR_a) using the calibration curve described in Section 4. Calculate analyte concentrations in the original sample (C) in ng/l using the equation:

$$C = (C_e \times CF)/1000$$

where CF is the concentration factor (equal to 1000 when one liter of water is processed to yield 1 ml sample extract).

8.2. For samples processed as part of a set where the laboratory control standard recovery falls outside of the control limits in Section 5, data for the affected analytes must be labelled as suspect.

9. RESULTS

9.1. In pond water samples collected in Georgia, a detection limit of 5 ng/l was demonstrated for α-endosulfan, β-endosulfan, and endosulfan sulfate. Recoveries of α-endosulfan, β-endosulfan, and endosulfan sulfate spiked into the pond water at the 5 ng/L level were 131±15 percent, 66±18 percent, and 70±6.7 percent, respectively. These analyses were performed without cleanup.

9.2. In other pond water samples collected in Georgia, a detection limit of 25 ng/l was demonstrated for α-endosulfan, β-endosulfan, and endosulfan sulfate. Coextracted materials made lower detection limits in that specific matrix impossible. Recoveries of α-endosulfan, β-endosulfan, and endosulfan sulfate spiked into the pond water at the 25 ng/L level were 96±9.1 percent, 83±13 percent, and 92±37 percent, respectively. The Florisil cleanup procedure was applied to these samples.

9.2. Examples generated from 5 and 25 ng/ml calibration solutions are given in Figures 1 and 2; these calibration levels represent approximately 5 and 25 ng/l in a pond or runoff water sample, respectively. Examples of chromatograms generated from analyses of a Georgia pond water, unspiked and spiked with endosulfans at the 5 ng/l level, are shown in Figures 3 and 4, respectively. Examples of chromatograms generated from analyses of a Georgia pond water, unspiked and spiked with endosulfans at the 25 ng/l level, are shown in Figures 5 and 6, respectively.

TABLE 1. SUGGESTED CHROMATOGRAPHIC CONDITIONS

Analyte	Chemical Abstracts Registry No.	Retention Time, min (a)
α-Endosulfan	959-98-8	40.8
β-Endosulfan	33213-65-9	43.6
Endosulfan sulfate	1031-07-8	45.7
2,3,4,5,2'-Pentachlorobiphenyl (internal standard)	--	42.0

(a) Suggested GC conditions:

Column: 30 m long x 0.25 mm I.D. DB-5 bonded fused silica column, 0.25 μm film thickness (J&W)
Injection volume: 2 μl splitless with 45 second delay
Carrier gas: He @30 cm/sec linear velocity
Injector temp: 250 C
Detector temp: 320 C
Oven temp: Program from 60 C to 300 C at 4 C/min
Detector: ECD

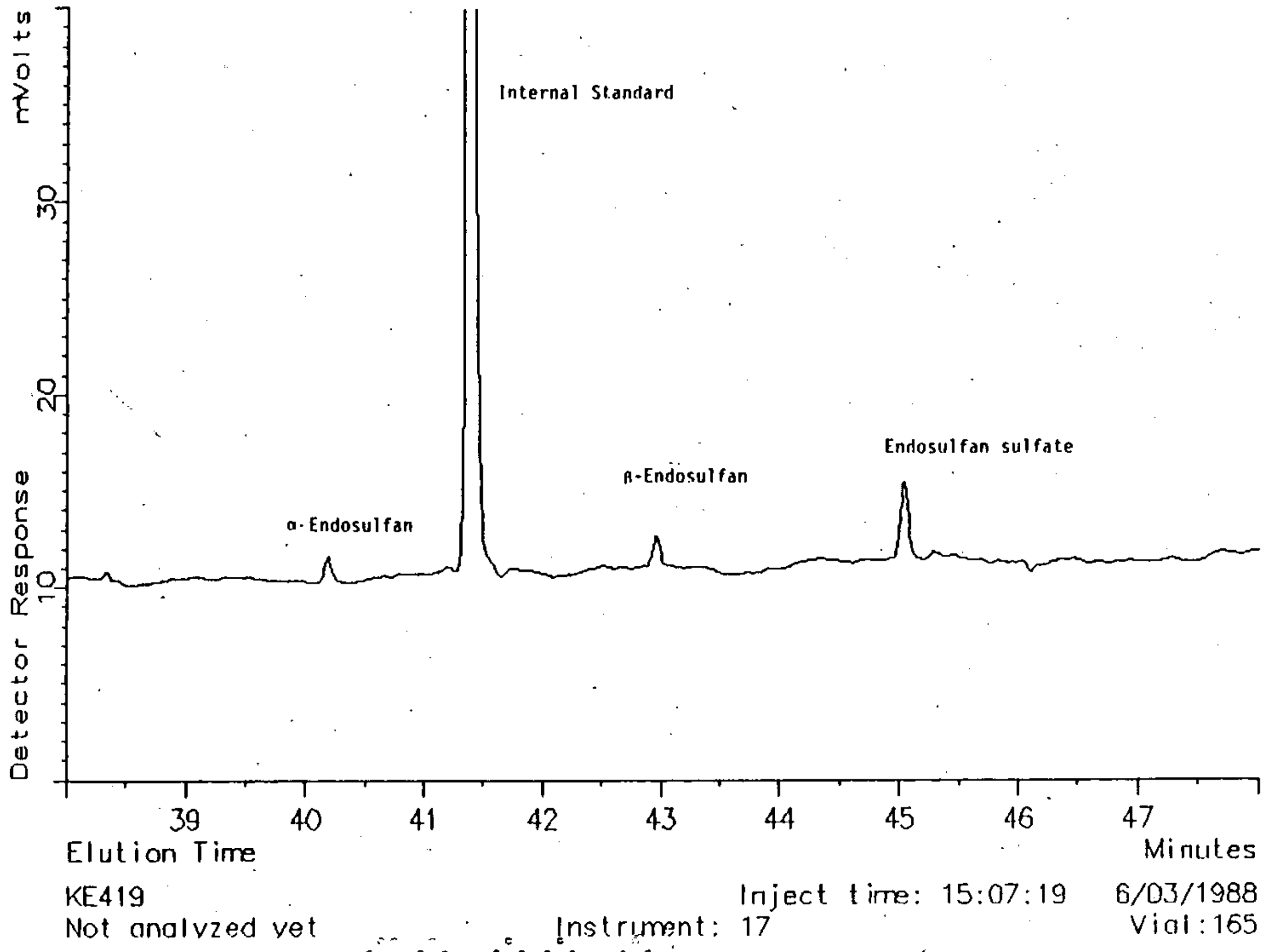


FIGURE 1. GC-ECD CHROMATOGRAM OF A 5 NG/ML CALIBRATION SOLUTION

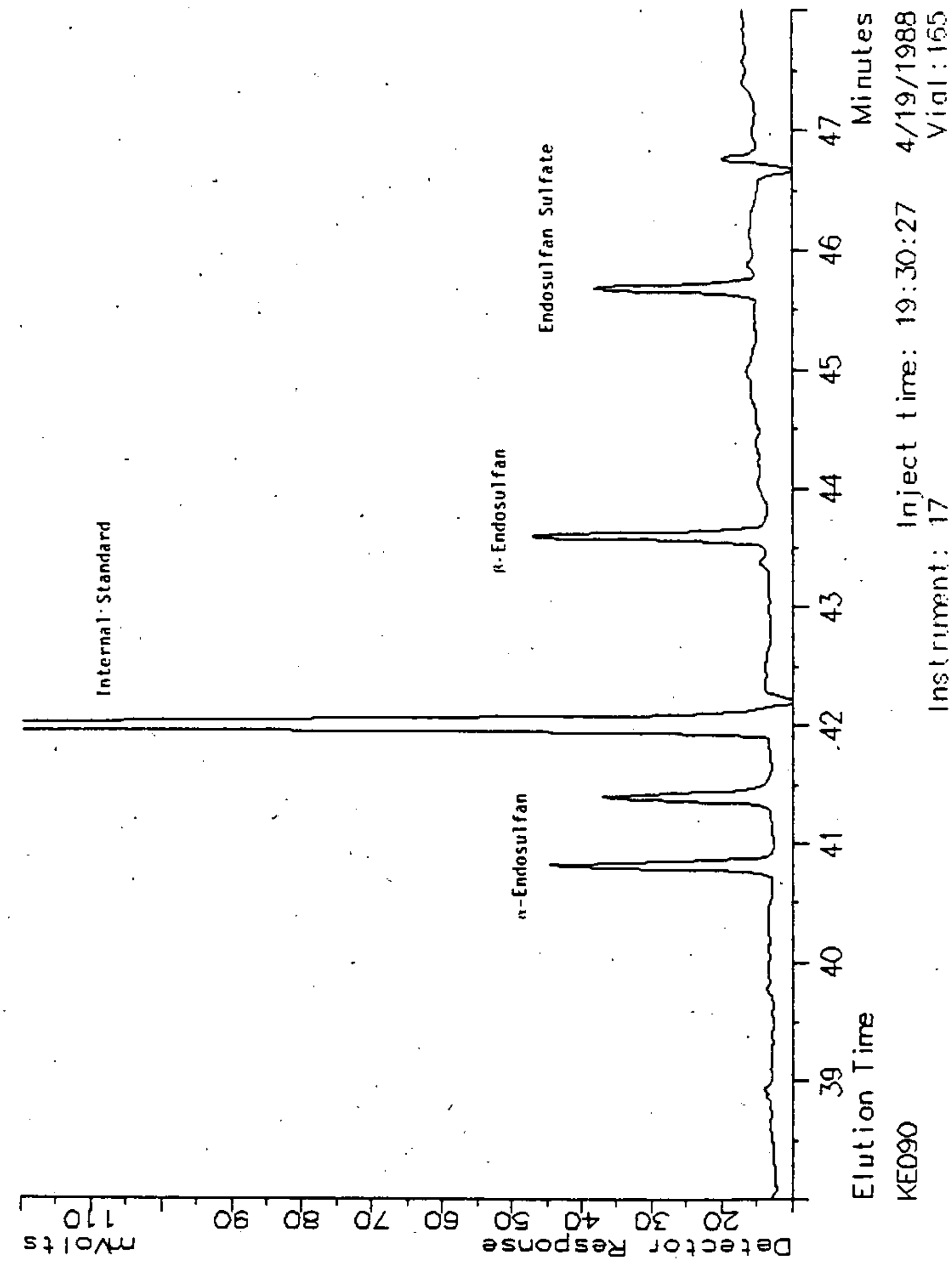


FIGURE 2. GC-ECD CHROMATOGRAM OF A 25 NG/ML CALIBRATION SOLUTION

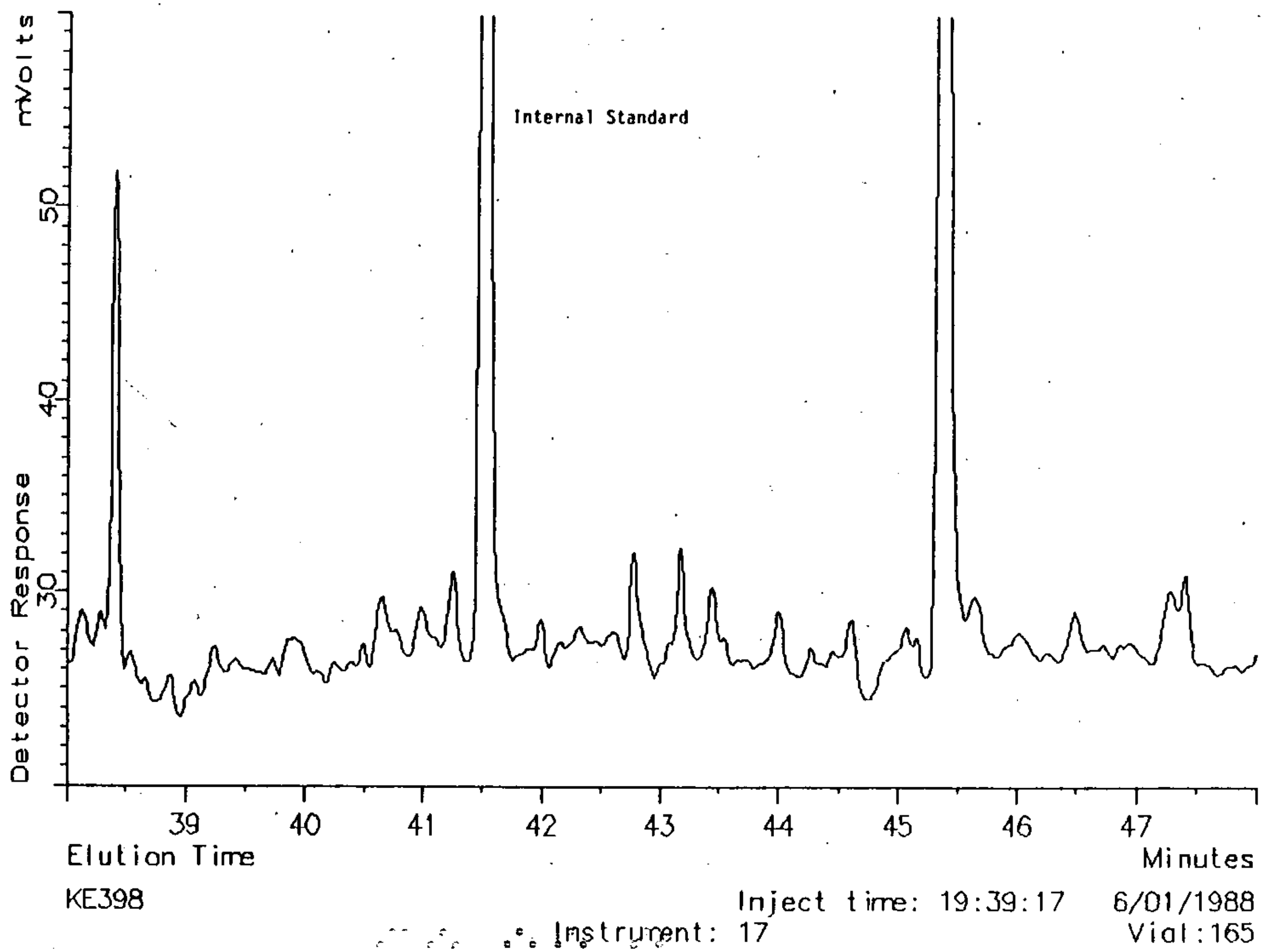


FIGURE 3. GC-ECD CHROMATOGRAM OF AN EXTRACT FROM AN UNSPIKED POND WATER SAMPLE

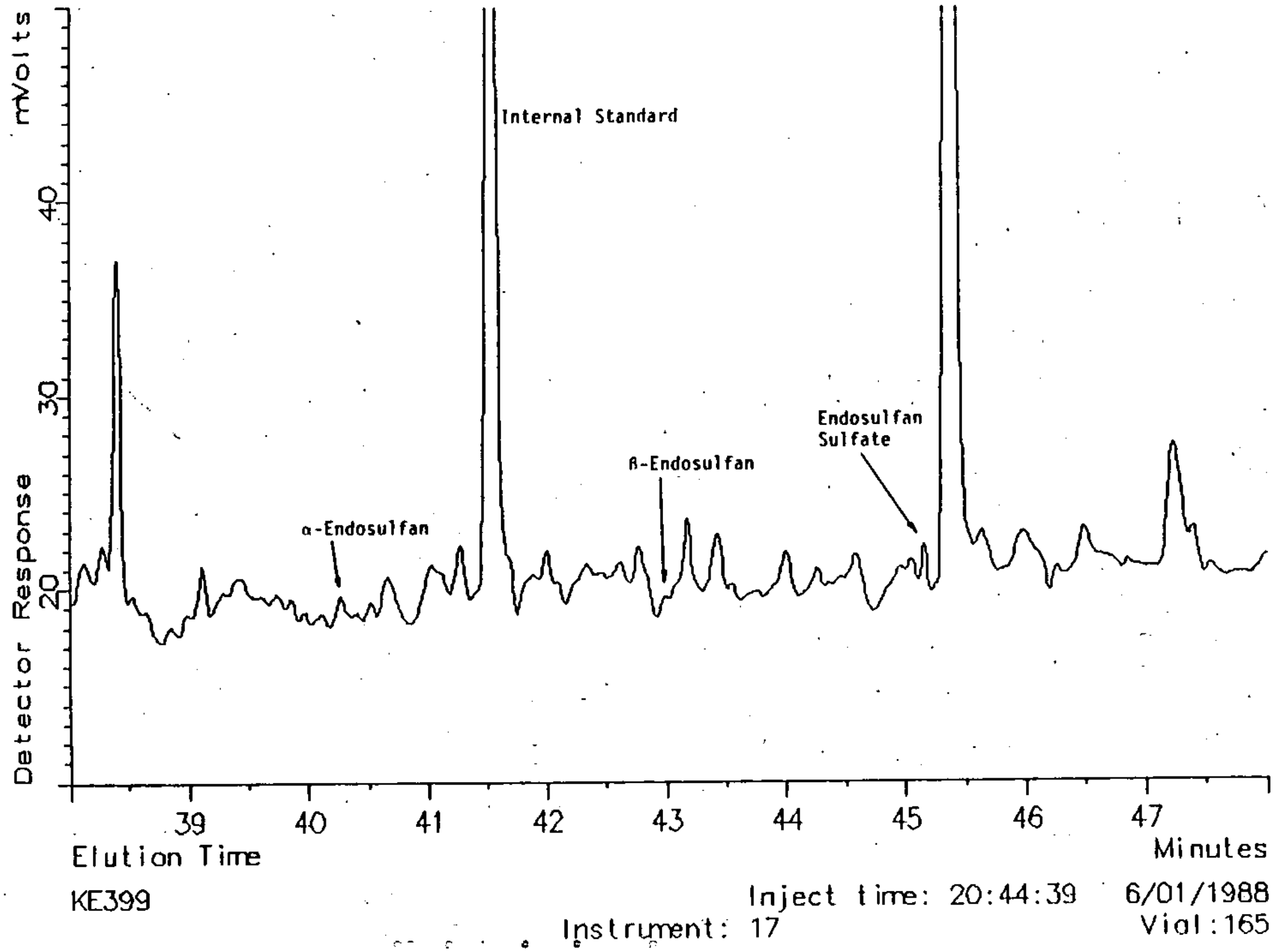


FIGURE 4. GC-ECD CHROMATOGRAM OF ~~AN~~ EXTRACT FROM A POND WATER SAMPLE SPIKED WITH ENDOSULFANS AT THE 5 NG/L LEVEL

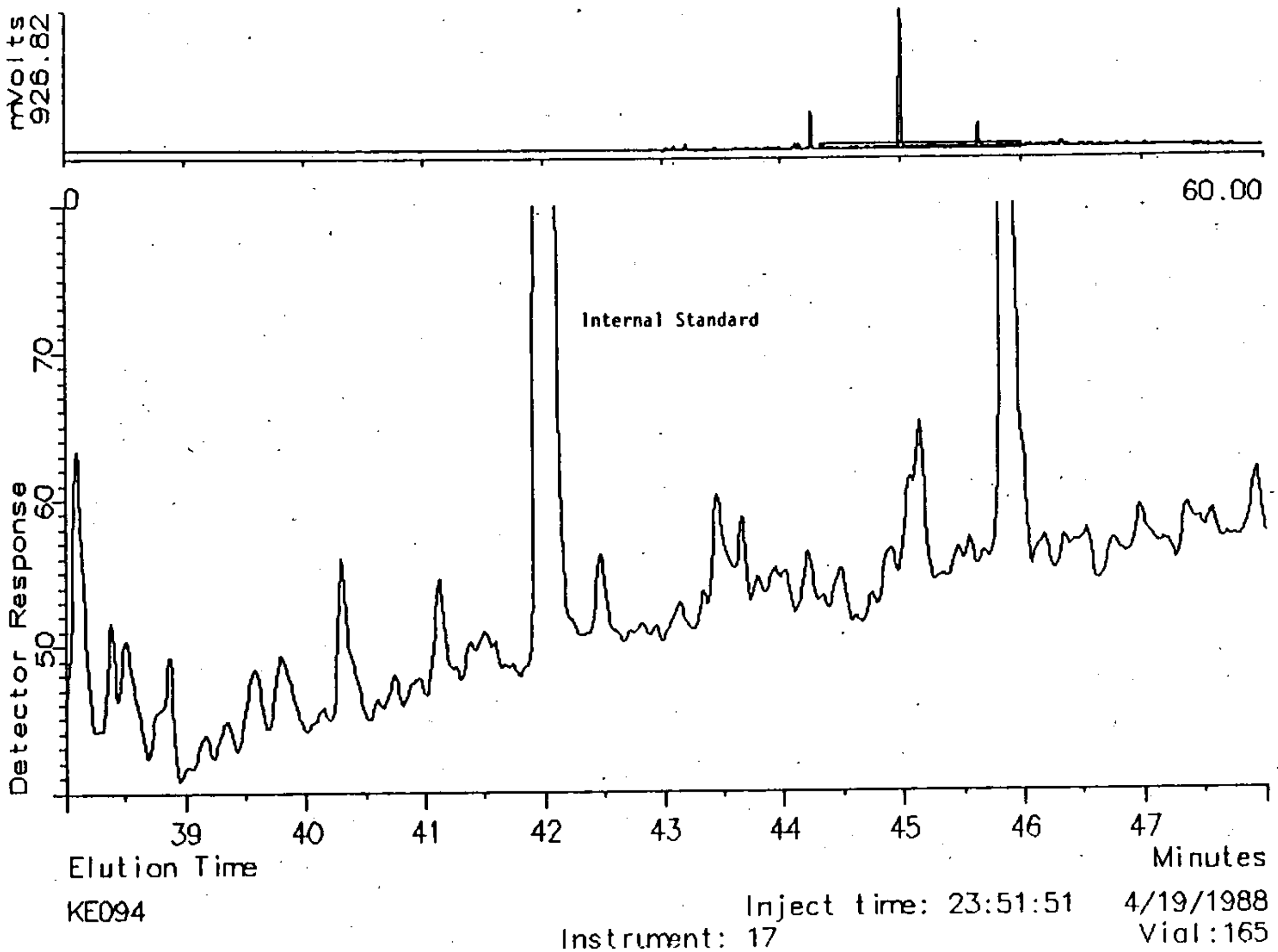


FIGURE 5. GC-ECD CHROMATOGRAM OF AN EXTRACT FROM AN UNSPIKED POND WATER SAMPLE

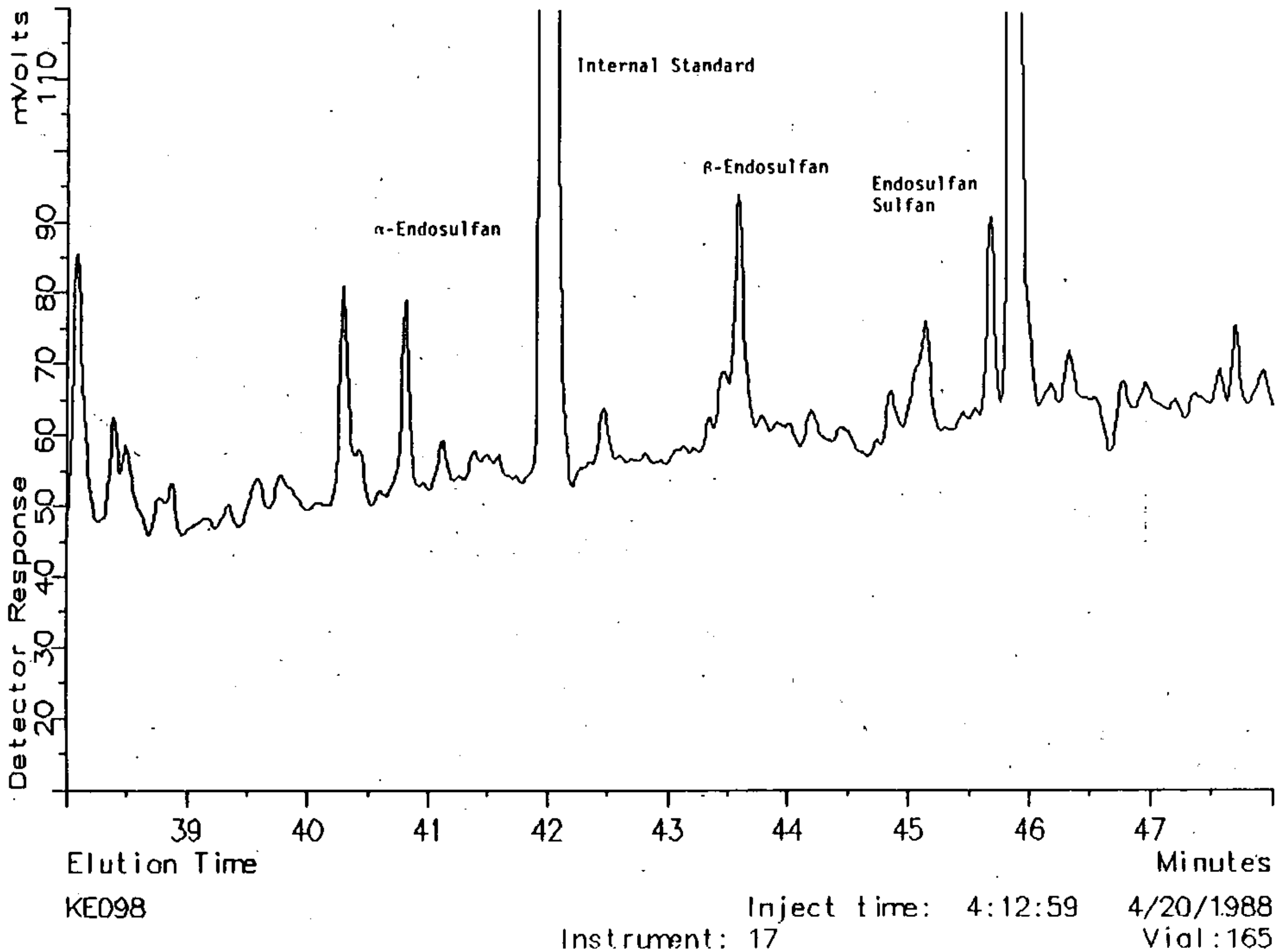


FIGURE 6. GC-ECD CHROMATOGRAM OF AN EXTRACT FROM A POND WATER SAMPLE SPIKED WITH ENDOSULFANS AT THE 25 NG/L LEVEL

Pond and Runoff Water

Chromatograms KM223 through KM229 are calibration chromatograms for alpha- and beta-endosulfan and endosulfan sulfate. The endosulfans are calibrated at 2.5, 5.0, 10, 25, 50, 100, and 250 ng/mL. The peak heights for each endosulfan was divided by the peak height of the internal standard. Calibration plots were generated by the computer and follow the standard chromatograms. The equation of the best line through the data is also given.

Chromatograms KM211 through KM215 are water samples. The computer uses peak height ratios from these samples and calculates the final sample concentration in ng/mL from the calibration plot. This value is adjusted using the Standard and Sample Weights and Factors 3 and 4 shown on each report. The concentration is multiplied by Standard Weight and Factor 3 and divided by Sample Weight and Factor 4. The values used for these factors are as follows:

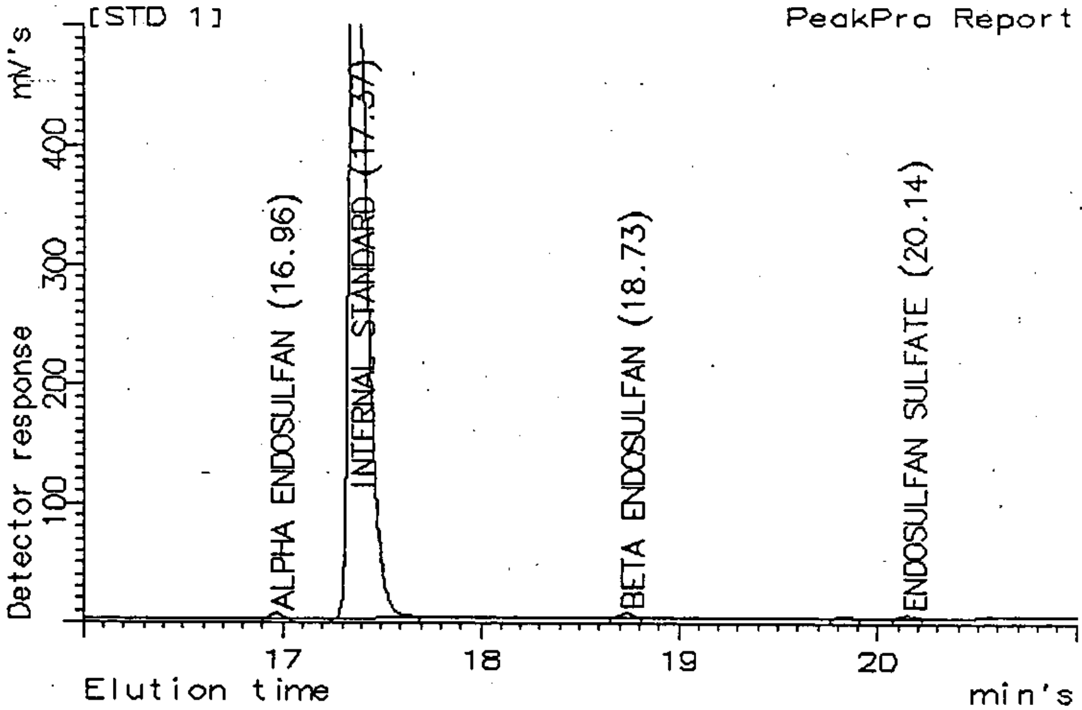
Standard Weight =	final sample volume in mL before further dilution.
Sample Weight =	100. This is a requirement of the software for Internal Standard calculations.
Factor 3 =	Sample dilution factor.
Factor 4 =	Volume of water sample in L.

Units for the final calculated result are:

$$\frac{ng}{mL} * \frac{ml}{L} = \frac{ng}{L}$$

Sample KM211 is a field spike and is treated slightly differently. Factor 4 is 1.0 (resulting in an assumed sample volume of 1 L) in order to calculate total nanograms of spike added.

A calibration check sample is analyzed with every 5 to 10 samples to make sure that the instrument performance does not change with time. Samples KM210 and KM216 are such checks at 50 ng/mL. The Factors do not include any dilution or sample volume data to yield results in ng/mL.



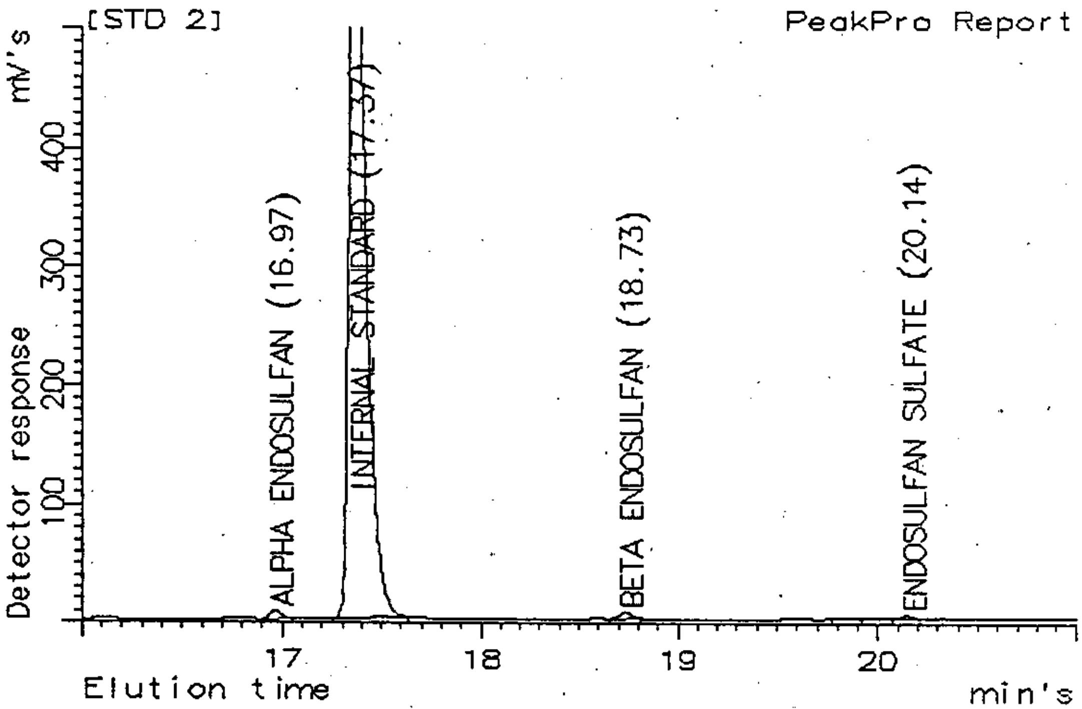
INTERNAL STANDARD HEIGHT CALIBRATION

Acquisition Information:
Chromatogram: KM223 43481-60-04 2.5 NG/ML 20:50:38 12/09/1988
AC Method: 1510 Inst 19 Vial # 0

Analysis Information:
AN Method: GC1520 [STD 1] 20:31:29 12/11/1988
Analysis Method revision number: 72
Analyst: KIM ANDREWS Channel 0
RRF update code: Replace
RRT update code: Check

Testing Conditions
COLUMN: SPB-5 30 METER
OVEN: 60(1) - 220/20; 220 - 280/4

Component Name	RRT	RRF	Pk	NG/ML	Area	Height
ALPHA ENDOSULFAN	16.96	n/a	BCB	2.500	16.876	4.586
INTERNAL STANDARD	17.37I	1.000	BCB	1.000	4523.564	858.618
BETA ENDOSULFAN	18.73	n/a	BCB	2.500	17.934	4.417
ENDOSULFAN SULFATE	20.14	n/a	BCB	2.500	9.020	2.242



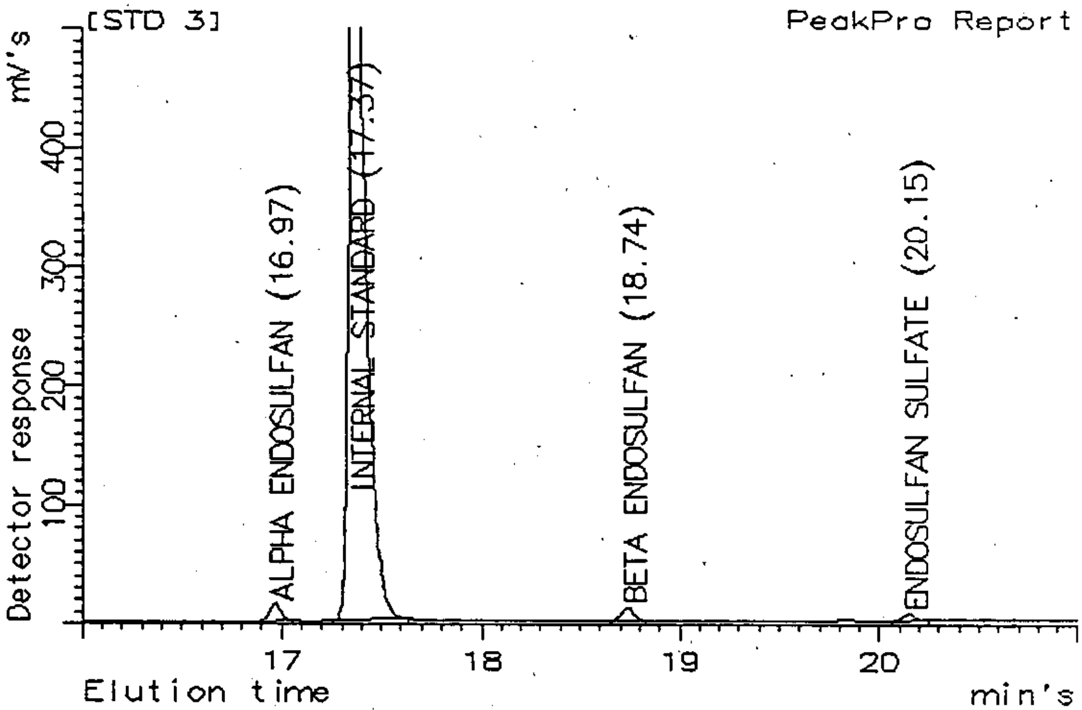
INTERNAL STANDARD HEIGHT CALIBRATION

Acquisition Information:
Chromatogram: KM224 43481-60-05 5 NG/ML 21:19:55 12/09/1988
AC Method: 1510 Inst 19 Vial # 0

Analysis Information:
AN Method: GC1520 [STD 2] 20:31:35 12/11/1988
Analysis Method revision number: 72
Analyst: KIM ANDREWS Channel 0
RRF update code: Replace
RRT update code: Check

Testing Conditions
COLUMN: SPB-5 30 METER
OVEN: 60(1) - 220/20; 220 - 280/4

Component Name	RRT	RRF	Pk	NG/ML	Area	Height
ALPHA ENDOSULFAN	16.97	n/a	BCB	5.000	27.384	7.273
INTERNAL STANDARD	17.37I	1.000	BCB	1.000	4041.297	776.427
BETA ENDOSULFAN	18.73	n/a	BCB	5.000	22.925	5.709
ENDOSULFAN SULFATE	20.14	n/a	BCB	5.000	11.149	2.888



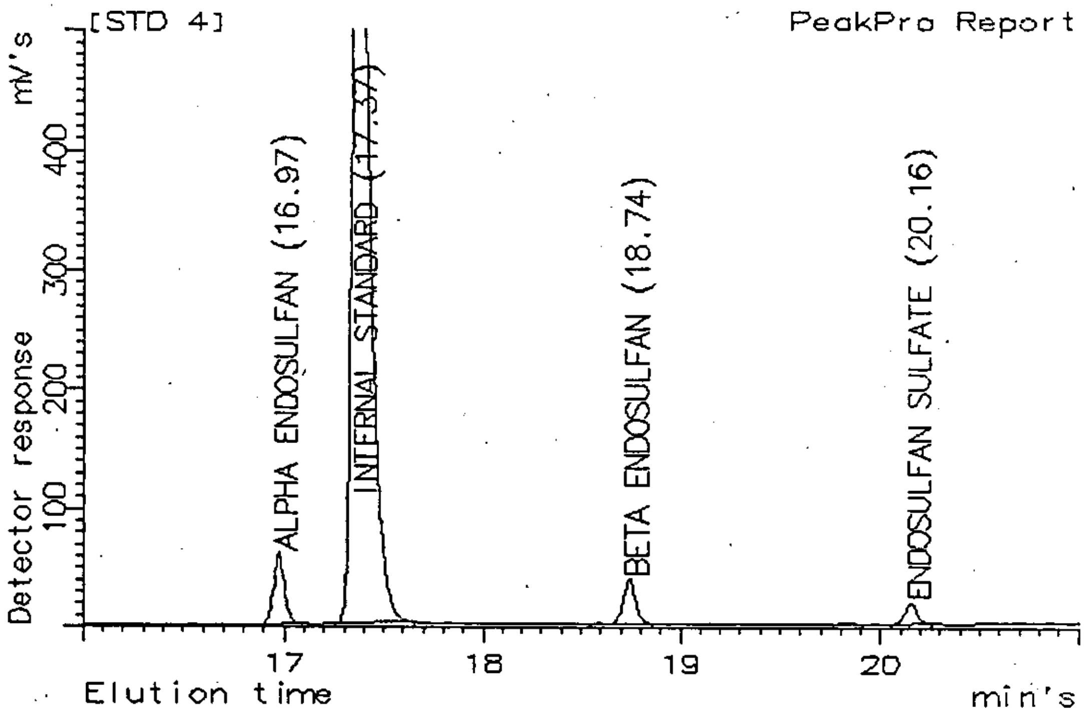
INTERNAL STANDARD HEIGHT CALIBRATION

Acquisition Information:
Chromatogram: KM225 43481-60-06 10 NG/ML 21:49:11 12/09/1988
AC Method: 1510 Inst 19 Vial # 0

Analysis Information:
AN Method: GC1520 [STD 3] 20:31:41 12/11/1988
Analysis Method revision number: 72
Analyst: KIM ANDREWS Channel 0
RRF update code: Replace
RRT update code: Check -

Testing Conditions
COLUMN: SPB-5 30 METER
OVEN: 60(1) - 220/20; 220 - 280/4

Component Name	RRT	RRF	Pk	NG/ML	Area	Height
ALPHA ENDOSULFAN	16.97	n/a	BCB	10.000	56.801	14.747
INTERNAL STANDARD	17.37I	1.000	BCB	1.000	3859.575	742.325
BETA ENDOSULFAN	18.74	n/a	BCB	10.000	46.390	11.146
ENDOSULFAN SULFATE	20.15	n/a	BCB	10.000	23.415	5.462
Total				31.000	3986.181	773.680



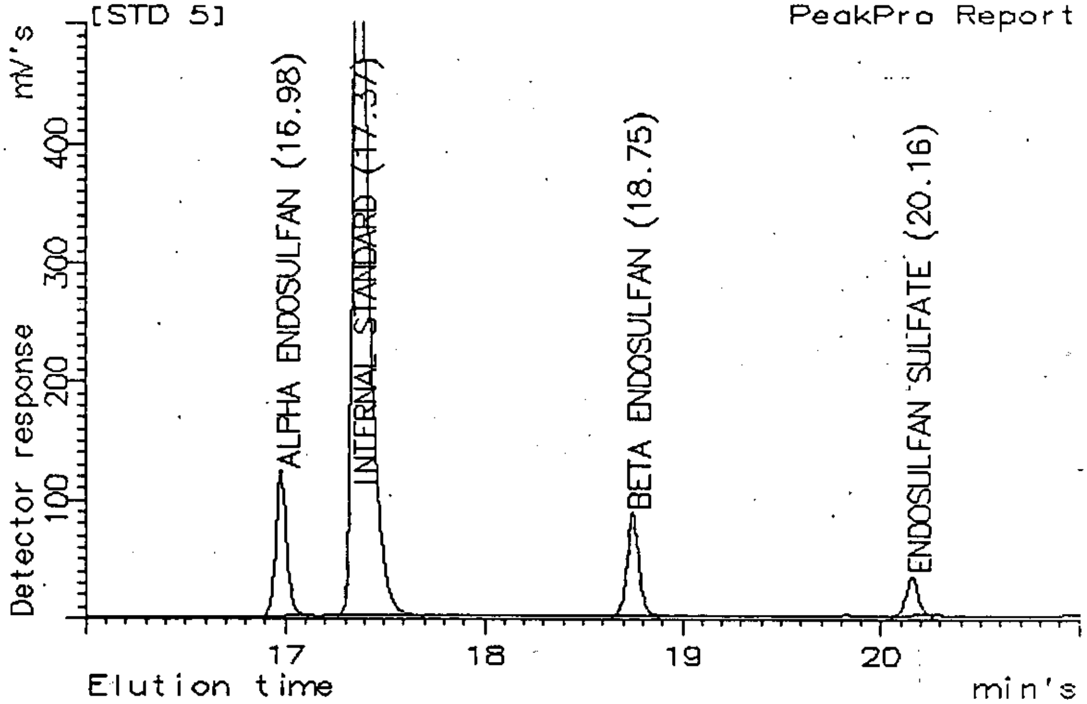
INTERNAL STANDARD HEIGHT CALIBRATION

Acquisition Information:
Chromatogram: KM226 43481-60-07 25 NG/ML 22:18:31 12/09/1988
AC Method: 1510 Inst 19 Vial # 0

Analysis Information:
AN Method: GC1520 [STD 4] 20:31:47 12/11/1988
Analysis Method revision number: 72
Analyst: KIM ANDREWS Channel 0
RRF update code: Replace
RRT update code: Check

Testing Conditions
COLUMN: SPB-5 30 METER
OVEN: 60(I) - 220/20; 220 - 280/4

Component Name	RRT	RRF	Pk	NG/ML	Area	Height
ALPHA ENDOSULFAN	16.97	n/a	BCB	25.000	225.786	59.644
INTERNAL STANDARD	17.37I	1.000	BCB	1.000	5033.636	954.893
BETA ENDOSULFAN	18.74	n/a	VCB	25.000	158.295	37.893
ENDOSULFAN SULFATE	20.16	n/a	BCB	25.000	73.534	17.180



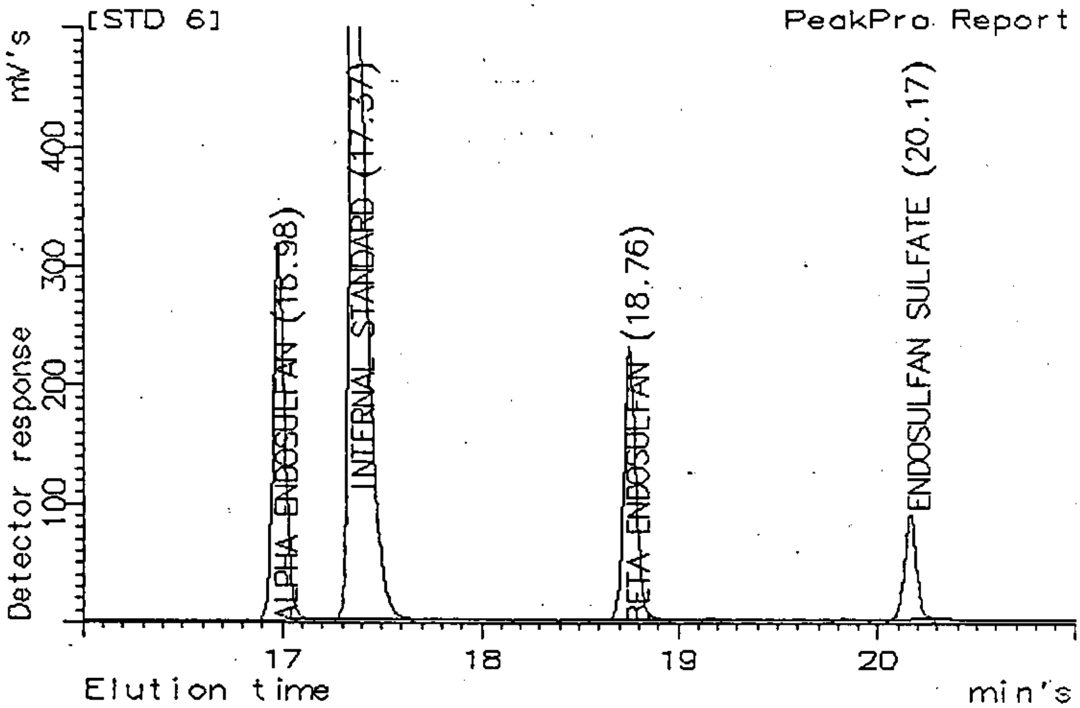
INTERNAL STANDARD HEIGHT CALIBRATION

Acquisition Information:
Chromatogram: KM227 43481-60-08 50 NG/ML 22:47:52 12/09/1988
AC Method: 1510 Inst 19 Vial # 0

Analysis Information:
AN Method: GC1520 [STD 5] 20:31:53 12/11/1988
Analysis Method revision number: 72
Analyst: KIM ANDREWS Channel 0
RRF update code: Replace
RRT update code: Check

Testing Conditions
COLUMN: SPB-5 30 METER
OVEN: 60(1) - 220/20; 220 - 280/4

Component Name	RRT	RRF	Pk	NG/ML	Area	Height
ALPHA ENDOSULFAN	16.98	n/a	BCB	50.000	464.575	121.535
INTERNAL STANDARD	17.37I	1.000	BCB	1.000	3959.545	761.178
BETA ENDOSULFAN	18.75	n/a	BCB	50.000	355.933	86.023
ENDOSULFAN SULFATE	20.16	n/a	BCB	50.000	134.070	31.850
Total				151.000	4914.125	1000.586



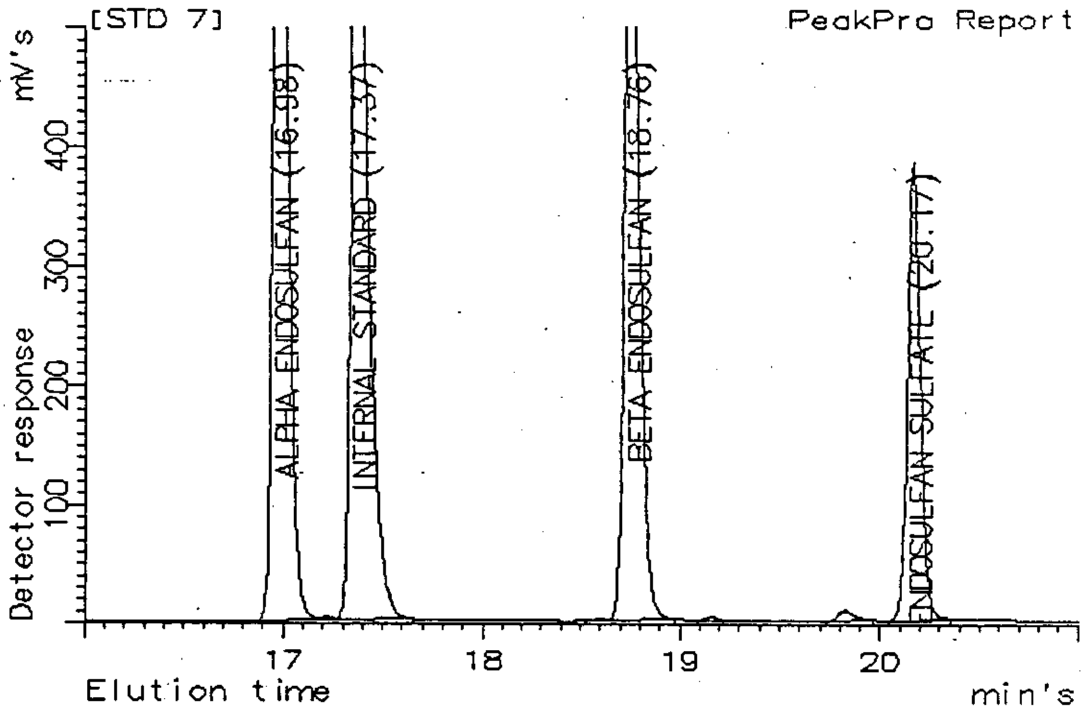
INTERNAL STANDARD HEIGHT CALIBRATION

Acquisition Information:
Chromatogram: KM228 43481-60-09 100 NG/ML 23:17:10 12/09/1988
AC Method: 1510 Inst 19 Vial # 0

Analysis Information:
AN Method: GC1520 [STD 6] 20:31:59 12/11/1988
Analysis Method revision number: 72
Analyst: KIM ANDREWS Channel 0
RRF update code: Replace
RRT update code: Check

Testing Conditions
COLUMN: SPB-5 30 METER
OVEN: 60(1) - 220/20; 220 - 280/4

Component Name	RRT	RRF	Pk	NG/ML	Area	Height
ALPHA ENDOSULFAN	16.98	n/a	BCB	100.000	1281.364	314.292
INTERNAL STANDARD	17.37I	1.000	BCB	1.000	4051.294	767.669
BETA ENDOSULFAN	18.76	n/a	VCB	100.000	983.176	227.398
ENDOSULFAN SULFATE	20.17	n/a	BCB	100.000	365.320	87.152



INTERNAL STANDARD HEIGHT CALIBRATION

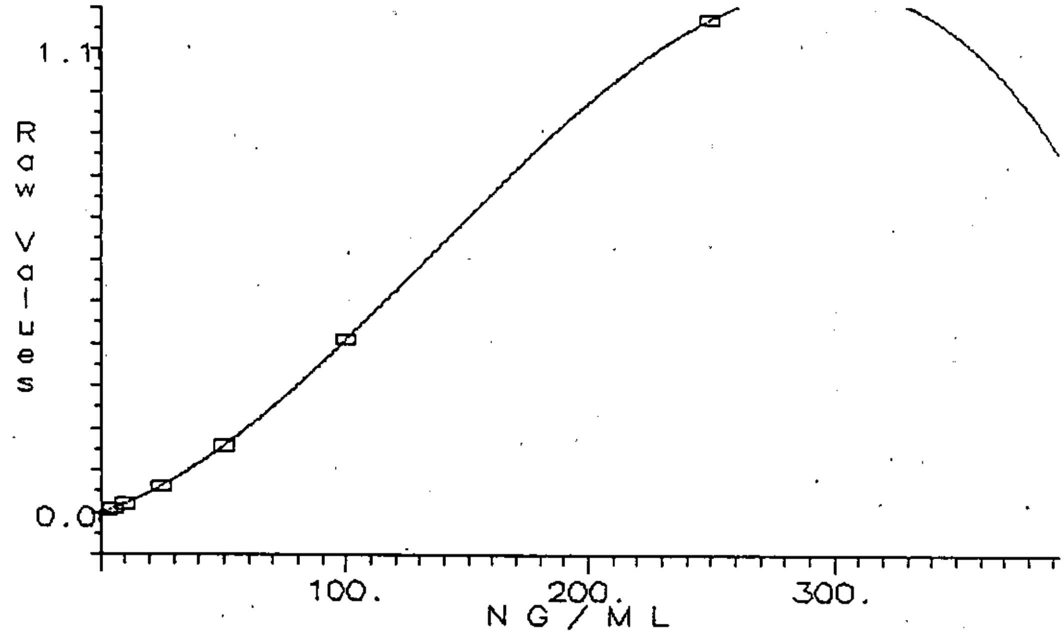
Acquisition Information:
Chromatogram: KM229 43481-60-10 250 NG/ML 23:46:28 12/09/1988
AC Method: 1510 Inst 19 Vial # 0

Analysis Information:
AN Method: GC1520 [STD 7] 20:32:05 12/11/1988
Analysis Method revision number: 72
Analyst: KIM ANDREWS Channel 0
RRF update code: Replace
RRT update code: Check

Testing Conditions
COLUMN: SPB-5 30 METER
OVEN: 60(1) - 220/20; 220 - 280/4

Component Name	RRT	RRF	Pk	NG/ML	Area	Height
ALPHA ENDOSULFAN	16.98	n/a	BCV	250.000	4487.625	975.437
INTERNAL STANDARD	17.37I	1.000	VCB	1.000	4445.806	838.263
BETA ENDOSULFAN	18.76	n/a	VCB	250.000	3604.752	754.378
ENDOSULFAN SULFATE	20.17	n/a	BCB	250.000	1716.200	378.611

PeakPro CALIBRATION PLOT/REPORT 9:34:02 6/21/1989

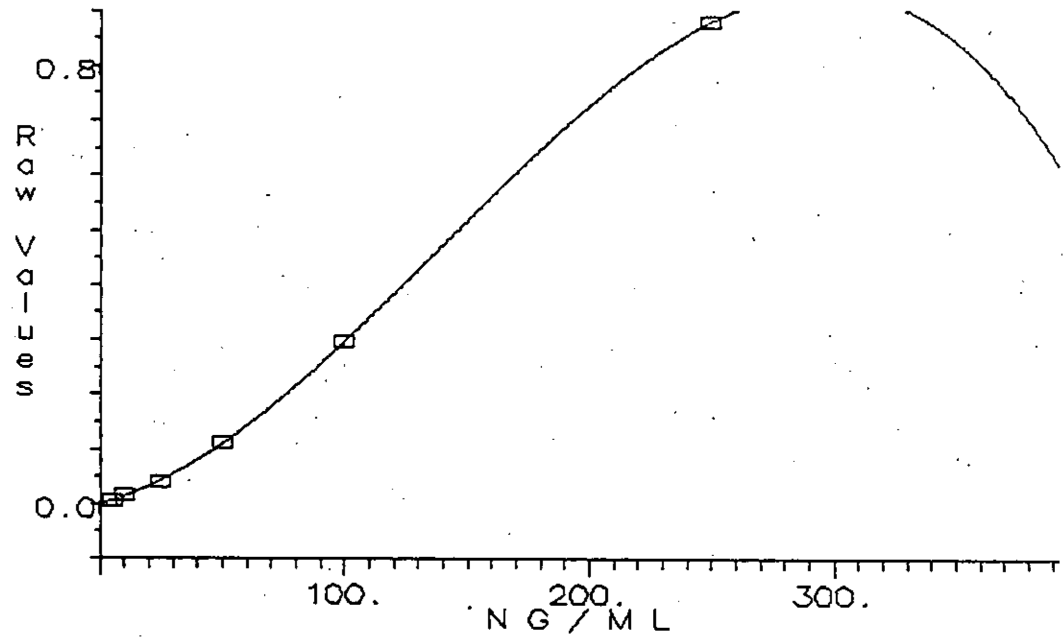


Analysis method: 1192
Method Title: ENDOSULFAN COMPOUNDS - INSTRUMENT 19
Component Name: ALPHA ENDOSULFAN

Third degree polynomial fit
Polynomial Y = -0.0009 + 0.0019x + 0.0000x^2 - 0.0000x^3
RMS Deviation: 0.0010

Calibration Level		NG/ML	Raw Values
1	STD 1	2.500	0.0053
2	STD 2	5.000	0.0094
3	STD 3	10.000	0.0199
4	STD 4	25.000	0.0625
5	STD 5	50.000	0.1597
6	STD 6	100.000	0.4094
7	STD 7	250.000	1.1696

PeakPro CALIBRATION PLOT/REPORT 9:34:53 6/21/1989

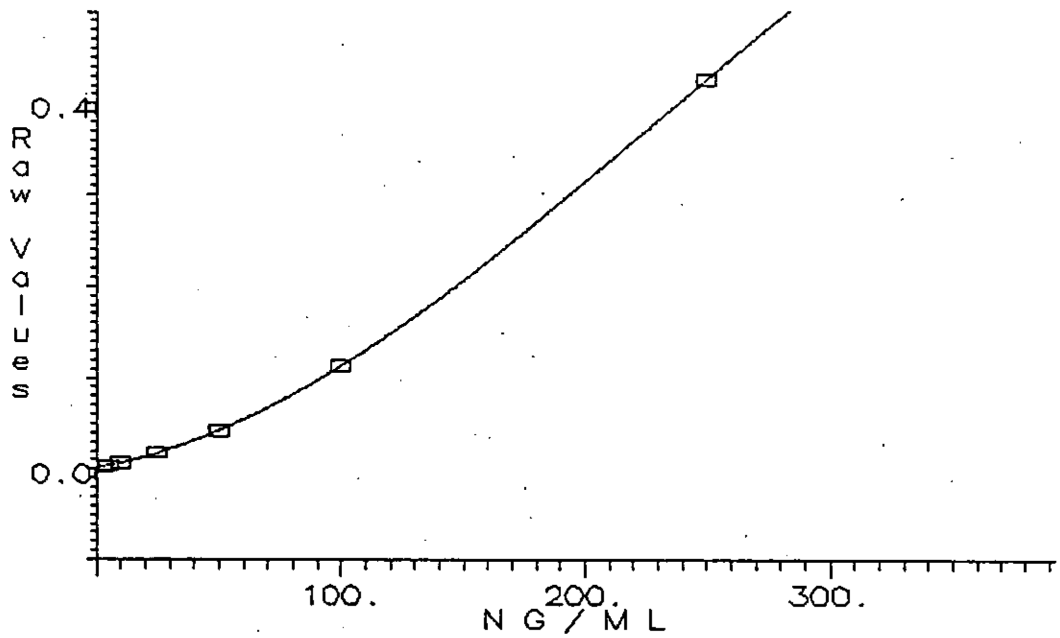


Analysis method: 1192
Method Title: ENDOSULFAN COMPOUNDS - INSTRUMENT 19
Component Name: BETA ENDOSULFAN

Third degree polynomial fit
Polynomial Y = 0.0012 + 0.0011x + 0.0000x^2 - 0.0000x^3
RMS Deviation: 0.0019

Calibration Level		NG/ML	Raw Values
1	STD 1	2.500	0.0051
2	STD 2	5.000	0.0074
3	STD 3	10.000	0.0150
4	STD 4	25.000	0.0397
5	STD 5	50.000	0.1130
6	STD 6	100.000	0.2962
7	STD 7	250.000	0.8788

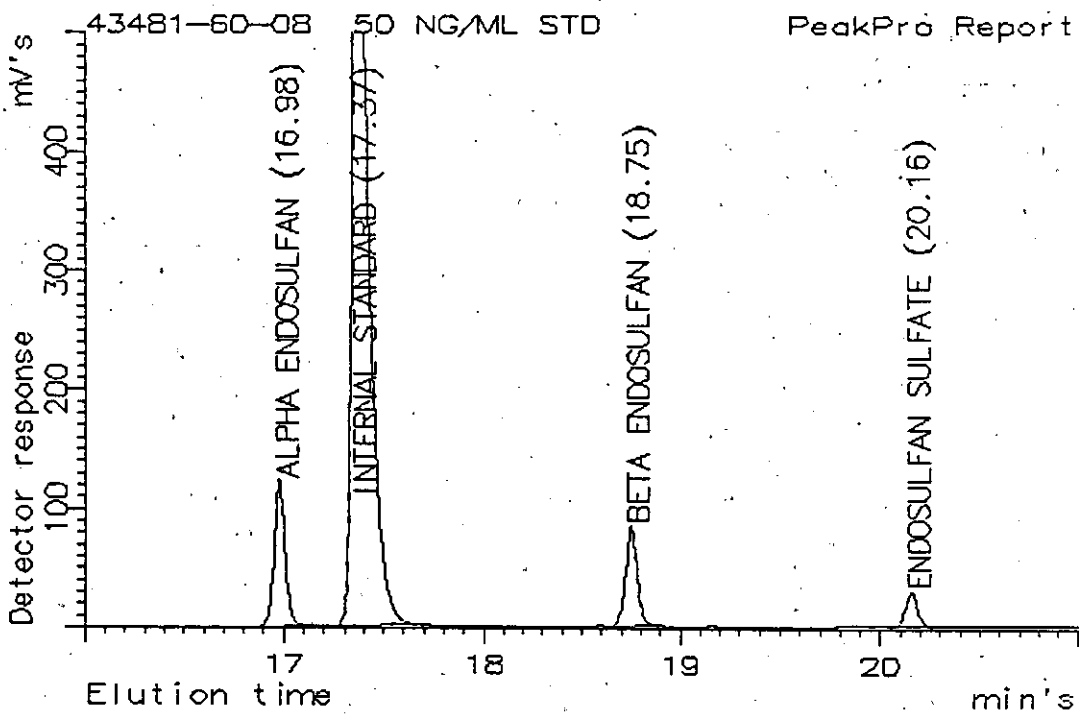
PeakPro CALIBRATION PLOT/REPORT 9:35:49 6/21/1989



Analysis method: 1192
Method Title: ENDOSULFAN COMPOUNDS - INSTRUMENT 19
Component Name: ENDOSULFAN SULFATE

Third degree polynomial fit
Polynomial Y = 0.0016 + 0.0005x + 0.0000x^2 - 0.0000x^3
RMS Deviation: 0.0003

Calibration Level		NG/ML	Raw Values
1	STD 1	2.500	0.0026
2	STD 2	5.000	0.0037
3	STD 3	10.000	0.0074
4	STD 4	25.000	0.0180
5	STD 5	50.000	0.0418
6	STD 6	100.000	0.1135
7	STD 7	250.000	0.4256



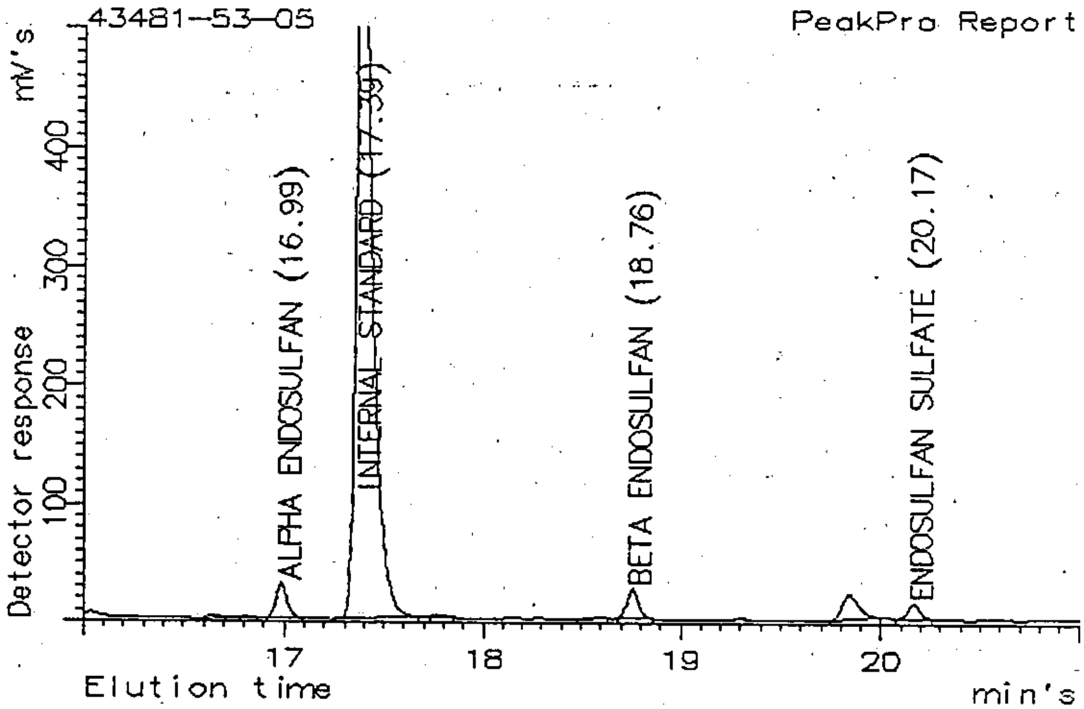
INTERNAL STANDARD HEIGHT ANALYSIS

Acquisition Information:
Chromatogram: KM210 43481-60-08 50 NG/ML STD 14:16:27 12/09/1988
AC Method: 1510 Inst 19 Vial # 0

Analysis Information:
AN Method: GC1520 43481-60-08 50 NG/ML STD 15:10:41 12/09/1988
Analysis Method revision number: 72
Analyst: KIM ANDREWS Channel 0
Standard Weight: 1.0000 Factor 3: 1.0000
Sample Weight: 100.0000 Factor 4: 1.0000
[STANDARDxFACTOR3]/[SAMPLExFACTOR4] weight %: 1.0000

Testing Conditions
COLUMN: SPB-5 30 METER
OVEN: 60(1) - 220/20; 220 - 280/4

Component Name	RRT	RRF	PkNG/ML	Area	Height
ALPHA ENDOSULFAN	16.98	n/a BCB	51.157	464.407	121.867
INTERNAL STANDARD	17.37I	1.000 BCB		4109.174	761.451
BETA ENDOSULFAN	18.75	n/a BCB	51.528	349.567	83.928
ENDOSULFAN SULFATE	20.16	n/a BCB	50.013	120.077	28.290
Total			152.698	934.051	234.085



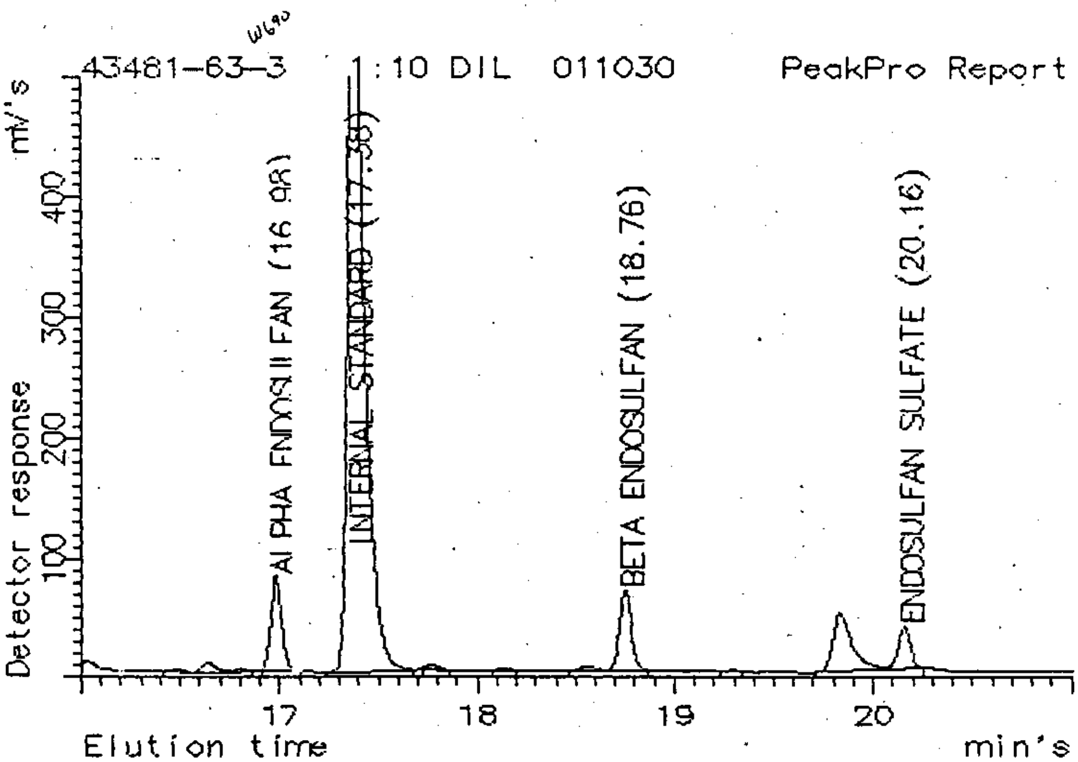
INTERNAL STANDARD HEIGHT ANALYSIS

Acquisition Information:
Chromatogram: KM211 43481-53-05 1:10 DIL 007693C 14:58:49 12/09/1988
AC Method: 1510 Inst 19 Vial # 0

Analysis Information:
AN Method: GC1190 43481-53-05 16:19:10 12/12/1988
Analysis Method has not been modified
Analyst: KIM ANDREWS Channel 0
Standard Weight: 1.0000 Factor 3: 10.0000
Sample Weight: 100.0000 Factor 4: 1.0000
[STANDARDxFACTOR3]/[SAMPLExFACTOR4] weight %: 10.0000

Testing Conditions
COLUMN: SPB-5 30 METER
OVEN: 60(1) - 220/20; 220 - 280/4

Component Name	RRT	RRF	PkNG/ML	Area	Height
ALPHA ENDOSULFAN	16.99	n/a	BCB 187.097	110.700	29.071
INTERNAL STANDARD	17.39I	1.000	BCB	3482.662	654.886
BETA ENDOSULFAN	18.76	n/a	BCB 224.273	101.456	24.731
ENDOSULFAN SULFATE	20.17	n/a	BCB 254.676	48.927	11.828



INTERNAL STANDARD HEIGHT ANALYSIS

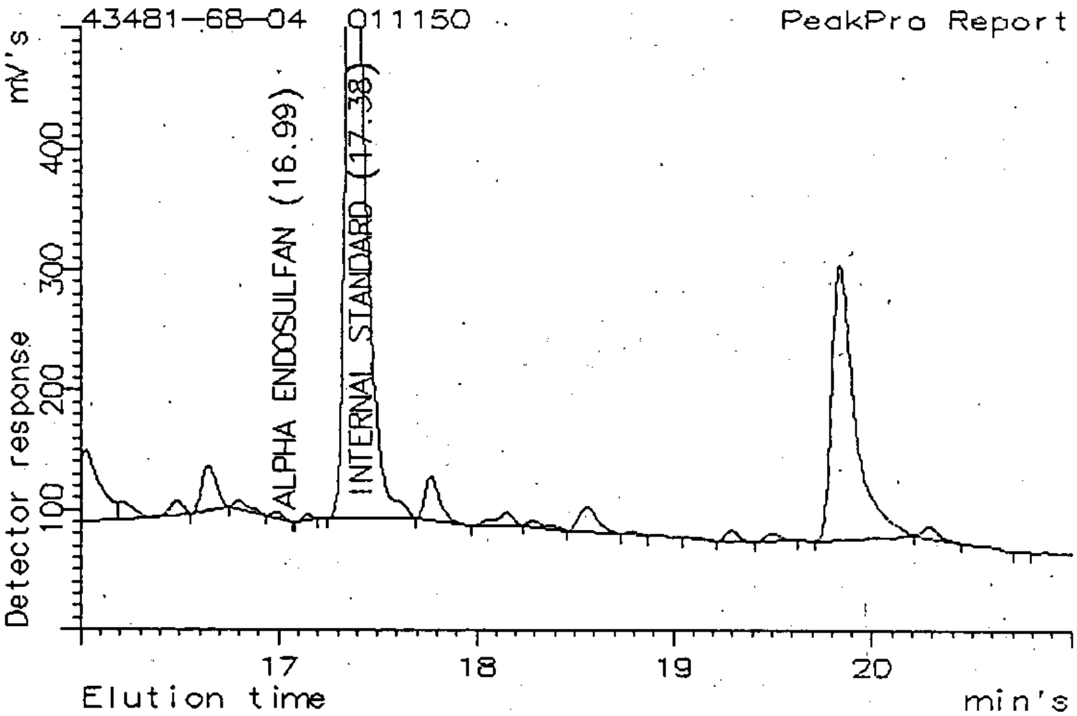
Acquisition Information:
Chromatogram: KM212 43481-63-31 1:10 DIL 011030 15:28:09 12/09/1988
AC Method: 1510 Inst 19 Vial # 0

Analysis Information:
AN Method: GC1520 43481-63-31 1:10 DIL 011030 20:41:04 12/11/1988
Analysis Method revision number: 72
Analyst: KIM ANDREWS Channel 0
Standard Weight: 1.0000 Factor 3: 10.0000
Sample Weight: 100.0000 Factor 4: 1.0000
[STANDARDxFACTOR3]/[SAMPLExFACTOR4] weight %: 10.0000

Testing Conditions
COLUMN: SPB-5 30 METER
OVEN: 60(1) - 220/20; 220 - 280/4

Component Name	RRT	RRF	PkNG/ML	Area	Height
ALPHA ENDOSULFAN	16.98	n/a	BCB 388.809	298.585	78.667
INTERNAL STANDARD	17.38I	1.000	BCB	3768.798	696.290
BETA ENDOSULFAN	18.76	n/a	VCB 453.687	280.206	66.989
ENDOSULFAN SULFATE	20.16	n/a	VCB 562.787	146.141	34.531

DB 12/16/88



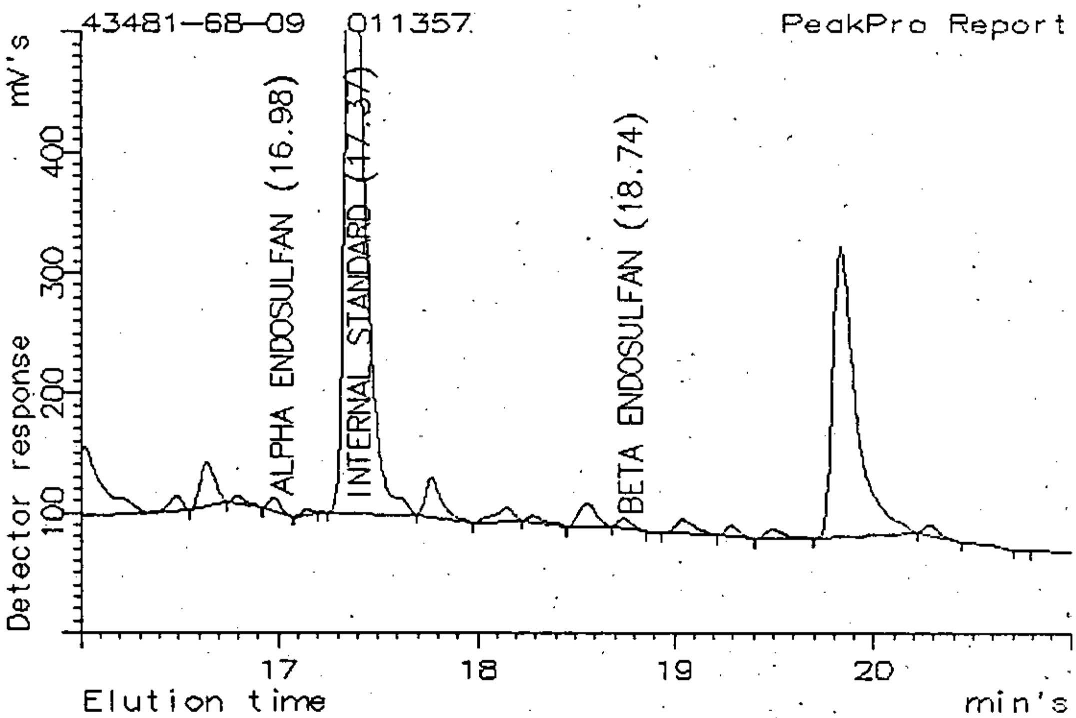
INTERNAL STANDARD HEIGHT ANALYSIS

Acquisition Information:
Chromatogram: KM213 43481-68-04 011150 15:57:29 12/09/1988
AC Method: 1510 Inst 19 Vial # 0

Analysis Information:
AN Method: GC1520 43481-68-04 011150 9:30:17 12/15/1988
Analysis Method revision number: 80
Analyst: KIM ANDREWS Channel 0
Standard Weight: 1.0000 Factor 3: 1.0000
Sample Weight: 100.0000 Factor 4: 1.0000
[STANDARDxFACTOR3]/[SAMPLExFACTOR4] weight %: 1.0000

Testing Conditions
COLUMN: SPB-5 30 METER
OVEN: 60(1) - 220/20; 220 - 280/4

Component Name	RRT	RRF	PkNG/ML		Area	Height
ALPHA ENDOSULFAN	16.99	n/a	BCB	2.629	18.652	5.178
INTERNAL STANDARD	17.38I	1.000	BCB		4427.160	736.698
BETA ENDOSULFAN	18.75					
ENDOSULFAN SULFATE	20.16					



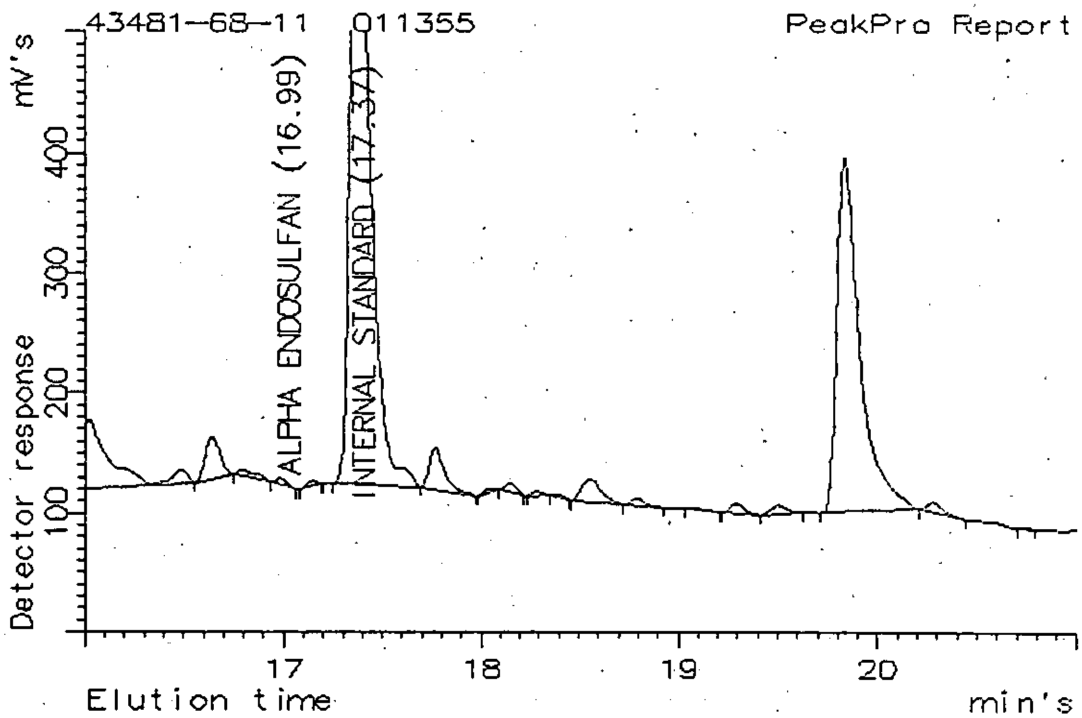
INTERNAL STANDARD HEIGHT ANALYSIS

Acquisition Information:
Chromatogram: KM214 43481-68-09 011357 16:26:48 12/09/1988
AC Method: 1510 Inst 19 Vial # 0

Analysis Information:
AN Method: GC1520 43481-68-09 011357 9:31:27 12/15/1988
Analysis Method revision number: 80
Analyst: KIM ANDREWS Channel 0
Standard Weight: 1.0000 Factor 3: 1.0000
Sample Weight: 100.0000 Factor 4: 1.0000
[STANDARDxFACTOR3]/[SAMPLExFACTOR4] weight %: 1.0000

Testing Conditions
COLUMN: SPB-5 30 METER
OVEN: 60(1) - 220/20; 220 - 280/4

Component Name	RRT	RRF	PkNG/ML		Area	Height
ALPHA ENDOSULFAN	16.98	n/a	BCB	5.101	43.847	11.134
INTERNAL STANDARD	17.37I	1.000	BCB		4214.671	711.249
BETA ENDOSULFAN	18.74	n/a	BCB	4.671	39.737	8.283
ENDOSULFAN SULFATE	20.16					



INTERNAL STANDARD HEIGHT ANALYSIS

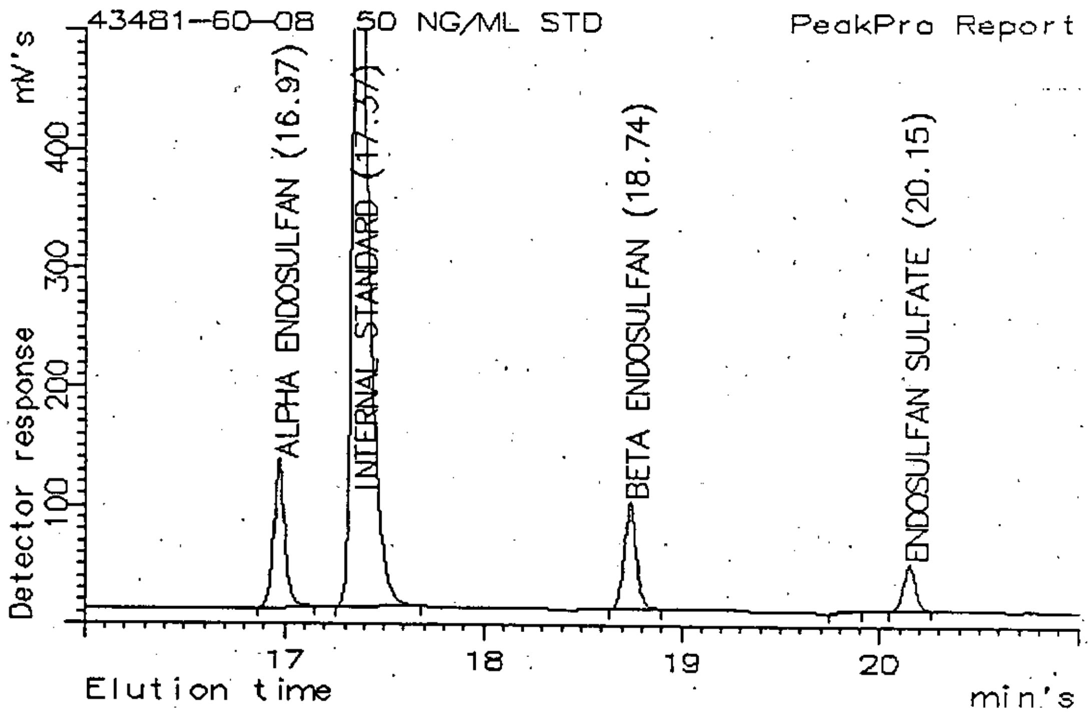
Acquisition Information:
Chromatogram: KM215 43481-68-11 011355 16:56:09 12/09/1988
AC Method: 1510 Inst 19 Vial # 0

Analysis Information:
AN Method: GC1520 43481-68-11 011355 9:32:30 12/15/1988
Analysis Method revision number: 80
Analyst: KIM ANDREWS Channel 0
Standard Weight: 1.0000 Factor 3: 1.0000
Sample Weight: 100.0000 Factor 4: 1.0000
[STANDARDxFACTOR3]/[SAMPLExFACTOR4] weight %: 1.0000

Testing Conditions
COLUMN: SPB-5 30 METER
OVEN: 60(1) - 220/20; 220 - 280/4

Component Name	RRT	RRF	PkNG/ML	Area	Height
ALPHA ENDOSULFAN	16.99	n/a	BCB 2.694	18.091	5.364
INTERNAL STANDARD	17.37I	1.000	BCB	4424.059	739.499
BETA ENDOSULFAN	18.75				
ENDOSULFAN SULFATE	20.16				

[handwritten initials] *[handwritten initials] 12/16/88*



INTERNAL STANDARD HEIGHT ANALYSIS

Acquisition Information:
Chromatogram: KM216 43481-60-08 50 NG/ML STD 17:25:29 12/09/1988
AC Method: 1510 Inst 19 Vial # 0

Analysis Information:
AN Method: GC1520 43481-60-08 50 NG/ML STD 20:34:36 12/11/1988
Analysis Method revision number: .72
Analyst: KIM ANDREWS Channel 0
Standard Weight: 1.0000
Sample Weight: 100.0000 Standard/Sample weight %: 1.0000

Testing Conditions
COLUMN: SPB-5 30 METER
OVEN: 60(1) - 220/20; 220 - 280/4

Component Name	RRT	RRF	Pk	NG/ML	Area	Height
ALPHA ENDOSULFAN	16.97	n/a	BCB	51.058	482.462	122.429
INTERNAL STANDARD	17.37I	1.000	BCB		3992.555	751.735
BETA ENDOSULFAN	18.74	n/a	BCB	52.581	378.844	88.941
ENDOSULFAN SULFATE	20.15	n/a	BCB	57.456	163.026	38.362