was removed from the extract by washing with several portions of distilled water. All extracts were dried by filtration through anhydrous sodium sulfate.

Cleanup of Plant Extracts. Interfering materials from surface strippings of apples, pears, and their leaves were removed by shaking an aliquot of the extract for 2 minutes with Hyflo Super-Cel or Attaclay. The cleaned-up extract was filtered and the flask and filter paper were washed several times with *n*-hexane. After shaking, the extract may be centrifuged rather than filtered.

To remove interfering materials from extracts of chopped or ground crops, 100 ml. (or an aliquot diluted to 100 ml.) of the *n*-hexane solution was shaken for 1 minute with 10 ml. of 85% sulfuric acid. Acid was allowed to separate from *n*-hexane and was discarded. The sulfuric acid wash was repeated until all color was removed (usually two washes were sufficient). The *n*-hexane solution was then washed repeatedly (3 to 5 times) with 50 ml. of distilled water until the washings were neutral to methyl orange, then dried by shaking for 30 seconds with 20 ml. of saturated sodium chloride. The solution was dried further by filtration through a sodium sulfate pad, and flask and pad were washed with *n*-hexane. Excess solvent was evaporated off and volume adjusted to that of the original aliquot.

Analysis of Plant Extracts. Plant extracts were analyzed in the same manner as for the preparation of the standard curve using an aliquot of the cleanedup plant extract. Control or blank samples of the same food crop, not treated with Kelthane, were analyzed and found to contribute interference equivalent to from 0.0 p.p.m. of Kelthane (surface residues on apples) to 0.8 p.p.m. in plant extracts of asparagus.

Known amounts of Kelthane, in *n*-hexane, were added to the samples just prior to addition of the solvent for surface stripping or extraction and percentage recovery of the added Kelthane was determined. Values obtained are presented in Tables I and II.

Interferences. Kelthane, an acaricide, is generally used in combination with other pesticides. To check possible interferences, a number of insecticides were analyzed by this method. No interferences were found from 100 μg. of Chlorobenzilate (ethyl 4,4′-dichlorobenzilate), DDT, dieldrin, Dimite (4,4′-dichloro-α-methylbenzhydrol), endrin, Guthion [*O,O*-dimethyl *S*-(4-oxo-1,2,3-benzotriazin-3-(4*H*)-ylmethyl) phosphorodithioate], heptachlor epoxide, lindane, methoxychlor, Sevin (1-naphthyl methylcarbamate), and Tedion (2,4,4′,-5-tetrachlorodiphenyl sulfone). One hundred micrograms of chlordan gave an interference equivalent to 7.5 μg. of Kelthane. A similar amount of heptachlor gave an interference equivalent of 15 μg. of Kelthane and when these samples were heated for 5 minutes, instead of the suggested 2-minute period, the interference increased to 22.5 μg. Giang, Barthel, and Hall (*3*) reported interference for some insecticides that gave no interference when analyzed by this method. Giang and others mentioned that impurities in insecticides tested may have been responsible.

Recovery of Chloroform. Known amounts of chloroform, in a pyridine solution, added directly to the test tubes were analyzed by this method and results obtained were similar to those reported by Rosenthal and coworkers (*6*).

Discussion

Reaction products of Kelthane and chloroform gave identical transmittance wave-length curves between 350 and 600 mμ. Maxima on each curve occurred at 370 and 525 mμ. The curves were similar to those reported by Rosenthal and coworkers (*6*).

The simplicity of the method and the absence of special equipment or reagents make the method adaptable to the rapid routine examination of a large number of samples on a variety of crops.

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HERBICIDE DETERMINATION

A New Basic Procedure for Determining Phenoxy Acid Herbicides in Agricultural Products

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THE NEW and increased uses of phenoxy acid herbicides in agricultural products made it desirable to seek a better analytical method than that of Marquardt and Luce (*3*), which is limited to the determination of phenoxyacetic acids and occasionally gives high blanks when interfering materials are difficult to remove. Besides being applicable to phenoxy acids in general and giving low blanks, a procedure that shortens analysis time and improves recoveries was sought. As a result of our investigations, a new analytical scheme was developed that involves cleavage with pyridine hydrochloride of the ether linkage common to phenoxy acids.

Pyridine hydrochloride is a very acidic onium salt, especially in the fused state (*1*). Prey (*4*) showed that the molten compound cleaves phenyl ethers. The authors found that under the same conditions, pyridine hydrochloride cleaves phenoxy acids, liberating the corresponding phenol derivatives.

In the following procedures illustrating a general scheme for determination of phenoxy acids in agricultural products, the phenol derivatives involved are determined photometrically by aminoantipyrine (*2*) methods as measures of the original compounds.

Determination of 2,4-Dichlorophenoxyacetic Acid (2,4-D Acid) in Sugar Cane Juice

This method without modification is applicable to the determination of 0.05 to 2.0 p.p.m. of 2,4-dichlorophenoxyacetic acid in sugar cane juice. After removal and suitable cleanup, cleavage of the ether linkage in 2,4-dichlorophenoxyacetic acid with pyridine hydro-

This work was undertaken because existing analytical methods for determining phenoxy-acid herbicides in agricultural products were not always satisfactory. Phenoxy acids are cleaved with pyridine hydrochloride, releasing the respective phenol derivatives which are determined colorimetrically as measures of the original herbicides. Cleanups are shortened and concentrations as low as 0.05 p.p.m. are easily determined.

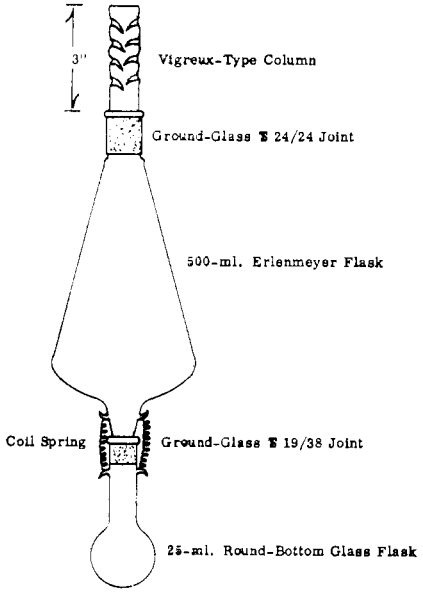

Figure 1. Kuderna-Danish evaporator concentrator

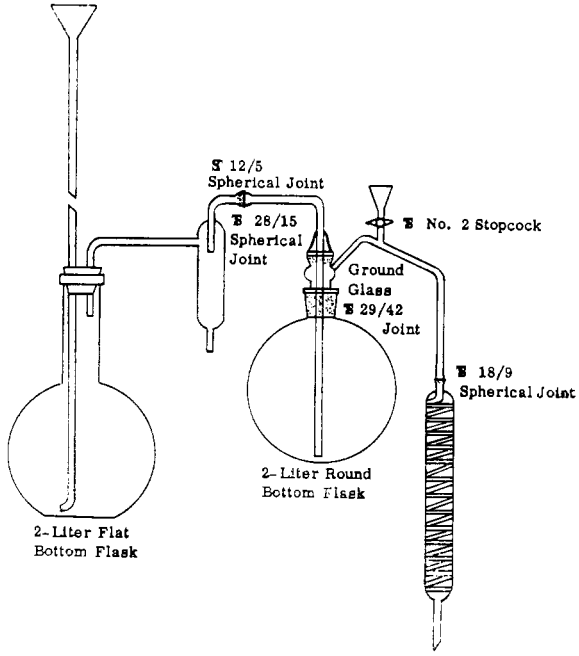

Figure 2. Steam distillation apparatus

chloride produces 2,4-dichlorophenol, which is determined photometrically as a measure of the original compound.

Reagents. Hyflo Super-Cel. A Celite product, diatomaceous silica (Johns Manville Co.).

Phosphotungstic acid solution. Dissolve 200 grams of phosphotungstic acid (approximately $P_2O_5.24WO_3.xH_2O$) in water and dilute the solution to 500 ml. Disregard any insoluble matter in the solution.

Hydrochloric acid, dilute. Dilute 60 ml. of concentrated hydrochloric acid to 1 liter with water.

Chloroform, redistilled. Use center 90% cut of chloroform (technical grade).

Pyridine hydrochloride. Practical grade.

Ammonium hydroxide, 1*N*. Dilute 70 ml. of concentrated ammonium hydroxide (28%) to 1 liter with water.

Skellysolve F. A 35° to 60° C. petroleum fraction (Skelly Oil Co.).

Ammonium hydroxide, 0.05*N*. Dilute 50 ml. of 1*N* ammonium hydroxide to 1 liter with water.

Buffer solution. Dissolve 300 grams of dibasic potassium phosphate in water and dilute the solution to 1 liter. This solution should have a pH of 9.1 ± 0.1 (check with pH meter). If necessary, adjust the pH by adding a small amount of monobasic potassium phosphate or tribasic potassium phosphate.

4-Aminoantipyrine, 1.0% solution. Dissolve 1.000 gram of 4-aminoantipyrine in water and dilute the solution to 100 ml. Store the reagent in an amber-colored bottle in a dark place. Make a fresh solution weekly.

Potassium ferricyanide, 2.0% solution. Dissolve 2.000 grams of potassium ferricyanide in water and dilute the solution to 100 ml. Store the reagent in an amber-colored bottle in a dark place. Make a fresh solution weekly.

2,4-Dichlorophenoxyacetic acid (2,4-D acid), commercial product assaying 99 + %.

Methanol. ACS grade.

Apparatus. Evaporative concentrator. Kuderna-Danish, small, with 500-ml. upper flask and 25-ml. lower flask (Figure 1).

Peanut oil bath. Regulated to a temperature range of 207° to 210° C.

Wire holder. For suspending the 25-ml. flask of the Kuderna-Danish evaporative concentrator in the peanut oil bath.

Steam distillation apparatus (Figure 2).

Coleman spectrophotometer. Model 14, equipped with 4-cm. absorption cells. Any photometer measuring light transmittance at 515 mμ should be suitable.

Procedure. Weigh 150 grams of sugar cane juice in a 250-ml. beaker. Add 5.0 grams of Hyflo Super-Cel, 10 ml. of phosphotungstic acid, and 10 ml. of concentrated hydrochloric acid. Stir the mixture in the beaker occasionally for 15 minutes.

Filter the solids on a 7-cm. Büchner funnel and wash with three 25-ml. portions of dilute hydrochloric acid. Discard the solids. Pour the combined solution of filtrate and washings into a 500-ml. separatory funnel.

Place a few boiling chips in the small Kuderna-Danish evaporative concentrator. Extract the 2,4-D acid from the solution with three 50-ml. portions of chloroform. Combine the extract solutions in the concentrator and attach the Vigreux column.

Using a steam bath, evaporate the chloroform. When the liquid level is in the 25-ml. flask of the concentrator, remove the Vigreux column and continue the evaporation. After all the chloroform has been evaporated, blow a gentle stream of air in the concentrator for 1 minute.

Disconnect the 25-ml. flask from the concentrator and add 10 grams of pyridine hydrochloride to the residue. Using the wire holder, suspend the flask with contents in a peanut oil bath kept at a temperature of 207° to 210° C. After 10 minutes, swirl the contents of the flask to make a homogeneous solution and then keep the flask suspended in the oil bath for an additional 50 minutes. Cool the flask and contents to room temperature.

Dissolve the pyridine hydrochloride by filling the flask with water from a 150-ml. portion of water. Add the

aqueous solution to 150 ml. of water and 10 ml. of concentrated hydrochloric acid in the 2-liter round-bottomed flask of the steam distillation apparatus. Wash the 25-ml. flask with the remainder of the 150-ml. portion of water, adding the washings to the solution in the 2-liter flask. Steam-distill 500 ml. of distillate into a 1-liter Erlenmeyer flask containing 25 ml. of 1*N* ammonium hydroxide.

Pour the distillate into a 1-liter separatory funnel and wash it three times with 50-ml. portions of Skellysolve. Discard the washings.

Acidify the washed distillate with 5 ml. of concentrated hydrochloric acid. Extract the 2,4-dichlorophenol with three 50-ml. portions of Skellysolve and combine the extract solutions in a 250-ml. separatory funnel. Wash the solution with three 25-ml. portions of water and discard the washings.

Extract the 2,4-dichlorophenol from the washed Skellysolve solution with one 10.0-ml. and two 5.0-ml. portions of 0.05*N* ammonium hydroxide and combine the extract solutions in a 50-ml. volumetric flask. Add 20.0 ml. of buffer solution (pH 9.1) and 1.0 ml. of a 1.0% solution of 4-aminoantipyrine and mix well. Add 1.0 ml. of a 2.0% solution of potassium ferricyanide and again mix well. After 1 minute, dilute to the mark with water and mix well. Fill a 4-cm. absorption cell with the solution. Three minutes after the addition of potassium ferricyanide, determine the absorbance with a spectrophotometer at 515 mμ, using water as a reference liquid.

Determine the micrograms of 2,4-D acid represented by the absorbance reading by referring to a standard calibration curve. Subtract any apparent 2,4-D acid found in the control sugar cane juice and correct for per cent recovery of 2,4-D acid obtained from the juice.

Calculate, parts per million, as based on the weight of the juice sample:

$$\frac{\text{Micrograms in sample}}{150} = \text{p.p.m. 2,4-D acid}$$

Preparation of Standard Calibration Curve. Prepare standard solution I as follows: Dissolve 0.100 gram of 2,4-D acid in about 50 ml. of methanol and dilute the solution with methanol to 1 liter. Each milliliter of this solution contains 100 μg. of 2,4-D acid.

Prepare standard solution II by diluting 15.0 ml. of standard solution I to 200 ml. with methanol. Each milliliter of this solution contains 7.5 μg. of 2,4-D acid.

Prepare standard solution III by diluting 30.0 ml. of standard solution I to 200 ml. with methanol. Each milliliter of this solution contains 15.0 μg. of 2,4-D acid.

Pipet 0, 1.0, 2.0, 5.0, 10.0, 15.0, and 20.0 ml. of standard solution II and 15.0 and 20.0 ml. of standard solution III into 25-ml. flasks of the small Kuderna-Danish evaporative concentrator. Proceed with the known amount of 2,4-D acid in each flask as follows:

Add a few boiling chips and assemble the small evaporative concentrator. Using a steam bath, evaporate the methanol. After all of the methanol has evaporated, blow a gentle stream of air in the concentrator for 1 minute.

Disconnect the 25-ml. flask from the concentrator and add 10 grams of pyridine hydrochloride. Continue the determination as described in the above procedure.

Prepare a standard calibration curve by plotting the data on graph paper. Beer's law is followed over the range from 0 to 300 μg. of 2,4-D acid.

Recovery of 2,4-D Acid Added to Sugar Cane Juice. Prepare standard solution A as follows: Dissolve 0.100 gram of 2,4-D acid in 25 ml. of 1*N* ammonium hydroxide and dilute the solution with water to 1 liter. Each milliliter of this solution contains 100 μg. of 2,4-D acid.

Prepare standard solution B by diluting 15.0 ml. of standard solution A with water to 200 ml. Each milliliter of this solution contains 7.5 μg. of 2,4-D acid.

Prepare standard solution C by diluting 30.0 ml. of standard solution A with water to 200 ml. Each milliliter of this solution contains 15.0 μg. of 2,4-D acid.

Weigh 150 grams of nontreated sugar cane juice in a 250-ml. beaker. Using standard solution B or C, add a known amount of 2,4-D acid in the same range as for the standard calibration curve.

Continue with the determination as directed in the procedure.

Determine the micrograms of 2,4-D acid represented by the absorbance reading by referring to the standard calibration curve and subtract the apparent 2,4-D acid content. Calculate the per cent recovery obtained.

Recovery data obtained by the authors from known amounts of 2,4-D acid added to nontreated sugar cane juice are shown in Table I. Analysis of the nontreated juice itself showed no apparent 2,4-D acid contact.

Table I. Recovery of 2,4-D Acid from Sugar Cane Juice

Added, P.P.M.	Found, P.P.M.	Recovery, %
0.050	0.0450	90
0.050	0.0425	85
0.10	0.100	100
0.10	0.100	100
0.25	0.220	88
0.25	0.222	89
0.50	0.460	92
0.50	0.460	92
0.75	0.692	92
0.75	0.702	94
1.00	0.919	92
1.00	0.914	91
1.00	0.943	94
1.50	1.485	99
1.50	1.354	90
2.0	1.984	99
2.0	1.907	95

Determination of Propylene Glycol Butyl Ether Ester of 2-(2,4,5-Trichlorophenoxy)propionic Acid in Sugar Cane Juice

This method without modification is applicable to the determination of 0.05 to 2.0 p.p.m. of the propylene glycol butyl ether ester of 2-(2,4,5-trichlorophenoxy)propionic acid in sugar cane juice. The commercial product is the active ingredient in Kuron (The Dow Chemical Co.) formulations. After removal and suitable cleanup, cleavage of the ether linkage in the ester with pyridine hydrochloride produces 2,4,5-trichlorophenol, which is determined photometrically as a measure of the original commercial product.

Reagents. The following reagents are in addition to or supplant reagents already listed.

Methanol, dilute. Mix equal volumes of methanol and water.

Carbon tetrachloride, redistilled. Use center 90% cut of carbon tetrachloride (technical grade).

Buffer solution. Dissolve 200.0 grams of dibasic potassium phosphate and 20.0 grams of monobasic potassium phosphate in water and dilute the solution to 1 liter. This solution should have a pH of 7.8 ± 0.1 (check with pH meter). If necessary, adjust the pH by adding a small amount of dibasic potassium phosphate or monobasic potassium phosphate.

4-Aminoantipyrine, 0.3% solution. Dissolve 0.300 gram of 4-aminoantipyrine in water and dilute the solution to 100 ml. Store the reagent in an amber-colored bottle in a dark place. Make a fresh solution weekly.

Potassium ferricyanide, 1.0% solution. Dissolve 1.000 gram of potassium ferricyanide in water and dilute the solution to 100 ml. Store in an amber-colored bottle in a dark place. Make a fresh solution weekly.

Propylene glycol butyl ether ester of 2-(2,4,5-trichlorophenoxy)-propionic acid, the active ingredient of Kuron. Commercial product used in Kuron formulations.

Apparatus. Same as already listed.

Procedure. Weigh 150 grams of sugar cane juice in a 400-ml. beaker. Add 150 ml. of methanol, 10 grams of Hyflo Super-Cel, and 10 ml. of phosphotungstic acid solution. Stir the mixture in the beaker occasionally for 30 minutes.

Table II. Recovery of Propylene Glycol Butyl Ether Ester of 2-(2,4,5-Trichlorophenoxy)propionic Acid from Sugar Cane Juice

Added, P.P.M.	Found, P.P.M.	Recovery, %
0.050	0.033	66
0.050	0.047	94
0.10	0.087	87
0.10	0.082	82
0.25	0.224	90
0.25	0.247	99
0.50	0.461	92
0.50	0.492	98
0.75	0.651	87
0.75	0.618	82
1.00	0.839	84
1.00	0.879	88
1.50	1.320	88
1.50	1.356	90
2.00	1.834	92
2.00	1.834	92

Filter the solids on a 7-cm. Büchner funnel and wash with three 25-ml. portions of dilute methanol. Discard the solids. Pour the combined solution of filtrate and washings into a 1-liter separatory funnel. Add 150 ml. of water and 10 ml. of concentrated hydrochloric acid to the solution.

Place a few boiling chips in the small Kuderna-Danish evaporative concentrator. Extract the propylene glycol butyl ether ester of 2-(2,4,5-trichlorophenoxy) propionic acid from the solution in the 1-liter separatory funnel with three 50-ml. portions of carbon tetrachloride. Combine the extract solutions in the concentrator and attach the Vigreux column.

Continue the determination as in the procedure for 2,4-D acid until ready to add the buffer solution.

Add 20.0 ml. of buffer solution (pH 7.8) and 1.0 ml. of 0.3% 4-aminoantipyrine solution into the 50-ml. volumetric flask and mix the solution well. Add 1.0 ml. of 1.0% potassium ferricyanide solution and again mix the solution well. After 1 minute, dilute the solution to the mark with water and mix it well. Fill a 4-cm. absorption cell with the solution. Three minutes after the addition of the potassium ferricyanide, determine the absorbance with a spectrophotometer at 505 mμ, using water as a reference liquid.

Determine the micrograms of propylene glycol butyl ether ester of 2-(2,4,5-trichlorophenoxy)propionic acid represented by the absorbance reading by referring to a standard calibration curve. Subtract any apparent ester found in the control sugar cane juice and correct for per cent recovery of the ester obtained from the juice.

Calculate parts per million, as based on the weight of the juice sample.

Table III. Recovery of Propylene Glycol Butyl Ether Esters of 2-(2,4,5-Trichlorophenoxy)propionic Acid from Undelinted Cottonseed

Added, P.P.M.	Found, P.P.M.	Recovery, %
0.10	0.106	106
0.10	0.082	82
0.20	0.169	85
0.20	0.188	94
0.30	0.281	94
0.30	0.300	100
0.50	0.455	91
0.50	0.435	87
0.70	0.591	84
0.70	0.578	83
1.00	0.804	80
1.00	0.830	83
1.50	1.245	83
1.50	1.277	85
2.00	1.766	88
2.00	1.633	82
2.50	2.188	88
2.50	2.281	91
3.00	2.438	81
3.00	2.450	82
3.50	3.005	86
3.50	3.037	87
4.00	3.307	83
4.00	3.351	84

Preparation of Standard Calibration Curve. Prepare standard solutions of propylene glycol butyl ether ester of 2-(2,4,5 - trichlorophenoxy)propionic acid with methanol in the same way as described for 2,4-D acid.

Obtain absorption data on known amounts of the ester mixture in the same manner as described for 2,4-D acid, but using the modified procedure.

Prepare a standard calibration curve by plotting the data on graph paper. Beer's law is followed in the range from 0 to 300 μg. of propylene glycol butyl ether ester of 2-(2,4,5-trichlorophenoxy) propionic acid.

Recovery of Propylene Glycol Butyl Ether Ester of 2-(2,4,5-Trichlorophenoxy)propionic Acid Added to Sugar Cane Juice

Weigh 150 grams of nontreated sugar cane juice in a 400-ml. beaker. Using standard methanol solutions already prepared, add a known amount of propylene glycol butyl ether ester of 2-(2,4,5-trichlorophenoxy)propionic acid in the same range as for the standard calibration curve.

Continue with the determination as directed in the procedure.

Calculate per cent recovery obtained, in the same manner as with 2,4-D acid in sugar cane juice.

Recovery data obtained by the authors are shown in Table II. Analysis of nontreated juice showed an apparent ester content of 0.028 p.p.m.

Determination of Propylene Glycol Butyl Ether Ester of 2-(2,4,5-Trichlorophenoxy)propionic Acid in Undelinted Cottonseed

Most of the cleanup requirements for the determination of phenoxy acids in agricultural products by this basic analytical procedure should be relatively easy. However, the large amount of oil in cottonseed made the separation of trace amounts of propylene glycol butyl ether ester of 2-(2,4,5-trichlorophenoxy)propionic acid more difficult than usual; therefore, this determination is presented to illustrate a more complicated cleanup procedure. The method without modification is applicable to the determination of 0.1 to 4.0 p.p.m. of herbicide.

Reagents. The following reagents are in addition to or supplant reagents already listed.

Sodium hydroxide, 4*N*. Dissolve 165 grams of 97% sodium hydroxide pellets in water and dilute the solution to 1 liter. Keep the solution in a polyethylene bottle.

Barium chloride (analytical reagent grade), saturated aqueous solution.

Sulfuric acid, 87%. Slowly add 500 ml. of concentrated sulfuric acid (95.5%) to 100 ml. of water, while cooling the solution in a cold water bath.

Potassium phosphate, tribasic, 7.5% solution. Dissolve 75.0 grams of tribasic potassium phosphate in water and dilute the solution to 1 liter in a volumetric flask. Fill a wash bottle with some of the solution.

Apparatus. The following apparatus are in addition to those already listed.

Wiley mill. Fitted with 6-mm. screen.

French-square bottle. Wide-mouthed, 1-liter, cap lined with polyethylene film.

Jar mill. Roller-type.

Evaporative concentrator. Kuderna-Danish, large, with 1500-ml. upper flask and 200-ml. lower flask (Figure 3).

Shaker. Burrell Wrist Action, Model BB, or equivalent.

Procedure. Grind a sufficient quantity of undelinted cottonseed in the Wiley mill.

Weigh 75 grams of ground cottonseed in a 1-liter French-square bottle. Add 20 grams of Hyflo Super-Cel and 750 ml. of methanol. Roll the bottle and contents for 30 minutes.

Filter the solids on a 9-cm. Büchner funnel. Then return any cottonseed in the funnel to the bottle, leaving most of the Hyflo Super-Cel in the funnel. Add 600 ml. of methanol and roll the bottle and contents on the jar mill for another 30 minutes. Filter and discard the solids, using the same Büchner funnel and combining filtrates.

Transfer the solution to a 2-liter

beaker. Add 20 grams of Hyflo Super-Cel and 50 ml. of 4*N* sodium hydroxide. Stir the mixture well.

Filter the solids on a 9-cm. Büchner funnel. Wash the beaker and solids with 50 ml. of methanol and add the filtered wash solution to the filtrate. Discard solids.

Transfer the filtrate to a 2-liter round-bottomed flask. Add some boiling chips, 20 grams of Hyflo Super-Cel, and 25 ml. of saturated barium chloride solution. Reflux the solution for 1 hour, using an electric heating mantle. Cool the flask and contents in a cold water bath.

Filter the solids, using a 9-cm. Büchner funnel. Wash the solids with 50 ml. of methanol and combine the filtered wash solution and filtrate. Discard solids.

Transfer the filtrate to the large Kuderna-Danish evaporative concentrator. Add some boiling chips and attach the Vigreux column to the concentrator. Using a steam bath, evaporate methanol until 150 to 175 ml. of liquid remain. Cool the concentrator and contents to room temperature. Remove the Vigreux column.

Add 500 ml. of water to the contents of the concentrator and mix the solution thoroughly.

Filter the solids, using a 10-cm. funnel and folded filter paper. Wash the concentrator and solids with 100 ml. of water, filter the solids from the wash solution, and combine filtrates. Discard solids and pour the filtrate into a 1-liter separatory funnel.

Wash the filtrate with three 50-ml. portions of carbon tetrachloride. Discard the washings. Acidify the washed solution with 25 ml. of concentrated hydrochloric acid and extract the 2-(2,4,5-trichlorophenoxy)propionic acid with three 50-ml. portions of carbon tetrachloride. Combine the extract solutions in a 250-ml. cylindrical separatory funnel.

Wash the combined extract solution with three 5.0-ml. portions of 87% sulfuric acid. For each wash, use a shaker to mix the contents of the funnel vigorously for 5 minutes. Discard the wash solutions.

Extract the 2-(2,4,5-trichlorophenoxy) propionic acid with three 10-ml. portions of concentrated sulfuric acid. For each extraction use a shaker to mix the contents of the funnel vigorously for 2 minutes. Add the extract solutions to 500 ml. of water in a 1-liter separatory funnel.

Place a few boiling chips in the small Kuderna-Danish evaporative concentrator. Extract 2-(2,4,5-trichlorophenoxy)propionic acid from aqueous solution with three 50-ml. portions of carbon tetrachloride. Combine the extract solutions in the evaporative concentrator and attach the Vigreux column.

Continue the determination as in the procedure for the propylene glycol butyl ether ester of 2-(2,4,5-trichlorophenoxy)propionic acid in sugar cane juice, but with this one modification: Dilute the final solution in the 50-ml. volumetric flask to the mark with 7.5% tripotassium phosphate instead of with water.

With the absorbance reading determine the micrograms of the sought-for compound present by referring to a standard calibration curve and correcting for sample blank and per cent recovery in the usual manner. Calculate parts per million, as based on the weight of the cottonseed sample.

Preparation of Standard Calibration Curve. Prepare standard solutions of propylene glycol butyl ether ester of 2-(2,4,5-trichlorophenoxy)propionic acid in methanol as described.

Obtain absorption data in the same way except for the modification of adding 7.5% tripotassium phosphate instead of water to the final solution.

Prepare a standard calibration curve by plotting the data on graph paper.

Recovery of Propylene Glycol Butyl Ether Ester of 2-(2,4,5-Trichlorophenoxy)propionic Acid Added to Cottonseed

Grind a quantity of nontreated, undelinted cottonseed in the Wiley mill. Weigh 75 grams of ground cottonseed in a 1-liter French-square bottle. Add a known amount of propylene glycol butyl ether ester of 2-(2,4,5-trichlorophenoxy)propionic acid in the same range as for the standard curve.

Continue with the determination as directed in the procedure and calculate per cent recovery in the usual manner.

Recovery data obtained by the authors are shown in Table III. Analysis of nontreated, undelinted cottonseed showed an apparent ester content of 0.041 p.p.m.

Discussion

Chloroform is used in the procedure for 2,4-dichlorophenoxyacetic acid because carbon tetrachloride, employed in the same manner, does not extract this acid quantitatively.

Carbon tetrachloride is used in the described procedures for propylene glycol butyl ether ester of 2-(2,4,5-trichlorophenoxy)propionic acid because it quantitatively extracts 2-(2,4,5-trichlorophenoxy)propionic acid, yet extracts less of other material along with it. However, chloroform can be used if on occasion it should be the preferred solvent.

In the procedure for propylene glycol butyl ether ester of 2-(2,4,5-trichlorophenoxy)propionic acid in cottonseed,

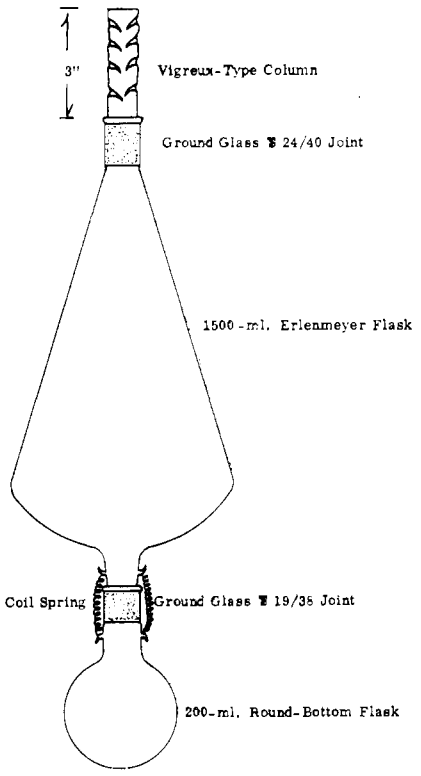

Figure 3. Large Kuderna-Danish evaporative concentrator

7.5% tripotassium phosphate solution is used to increase the pH of the solution for absorbance reading in order to eliminate an opaque condition that may occur with the presence of a trace amount of insoluble organic acids.

Pyridine hydrochloride should not be exposed to air more than necessary, as it is very hygroscopic. However, desiccation of the compound should be avoided, since the small amount of water already present appears to promote proper cleavage of the phenoxy acids.

The analytical scheme illustrated by the described procedures should provide a basic scheme for determination of residues of phenoxy acids and their esters or salts in agricultural products. Cleanup procedures will have to be modified, of course, and colorimetric or other methods adapted for the phenols involved.

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