Gas Chromatographic Determination of Flucythrinate Synthetic Pyrethroid Residues in a Range of Crops

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A gas chromatographic method is described for the determination of flucythrinate residues in cabbage, broccoli, and apples. Minor, but critical, modifications are also described which enable analysis of residues in wheat, barley, and hops. Flucythrinate residues have been quantitated down to 0.05 ppm for all crop types, and mean recoveries of flucythrinate at fortification levels of 0.05–1.00 ppm were greater than 80%.

CYBOLT flucythrinate (trademark of the American Cyanamid Co., also sold under the trademark PAY-OFF flucythrinate) [(*RS*)-α-cyano-3-phenoxybenzyl (*S*)-2-[4-(difluoromethoxy)phenyl]-3-methylbutyrate] is a novel pyrethroid insecticide developed by the American Cyanamid Co. It is a highly active, broad-spectrum insecticide, its high efficacy allowing the use of very low dosages in agricultural treatments (Wettstein and Whitney, 1979). Single-residue and multiresidue methods for the determination of pyrethroid insecticides in a range of crops have been reported. These methods often employ gas chromatography with electron capture detection (Chapman and Harris, 1978; Greenberg, 1981; Baker and Bottomley, 1982). No single-residue or multiresidue method has been reported for the analysis of flucythrinate. This paper describes a sensitive gas chromatographic method, using electron capture detection, for the analysis of flucythrinate residues in cabbage (red, white, savoy), broccoli, and apples. Modifications to the basic method are listed to enable analysis of flucythrinate residues in wheat (foliage, straw, ears, grain), barley (straw, ears, grain), and hops (green, dried).

MATERIALS AND METHODS

Apparatus. A Waring blender was used for the extraction of flucythrinate from all plant tissues. A Buchi rotary evaporator was used with a water bath set at 35 °C. The dimensions of the glass columns used for Florisil column chromatography were 15 mm i.d. × 250 mm. GLC analyses were performed with a Hewlett-Packard 5830 chromatograph fitted with a nickel-63 electron capture detector.

Reagents. All solvents were supplied by Fisons Scientific Ltd. High-performance liquid chromatography and standard laboratory reagent-grade solvents were used in all steps up to Florisil column chromatography. In this step, and in the preparation of standard solutions, Distol distilled-in-glass solvents were used. Florisil, 60–100 U.S. mesh (BDH), and Celite 545AW (Koch-Light Ltd.) were used as supplied.

Sample Extraction and Solvent Partitioning. The steps outlined in detail below are suitable for the analysis of fruit and vegetable crops (e.g., cabbage, broccoli, and apples). Modifications are then given to enable analysis of cereals and hops.

A mixture of 40 g of finely chopped plant tissue, 30 g of anhydrous sodium sulfate, and 10 g of Celite 545AW was blended at moderate speed with 250 mL of dichloromethane for 5 min. The extract was filtered under reduced pressure through a Buchner funnel fitted with a glass-fiber filter paper (Whatman GF/A). The blender jar was rinsed with 100 mL of dichloromethane, and this was used to wash the filter cake. The combined filtrate was taken to a volume of 500 mL with dichloromethane.

A 50-mL aliquot was evaporated to dryness. The residue in the flask was shaken with 25 mL of hexane, and this was transferred to a separating funnel. The flask was shaken with a further 25 mL of hexane and 50 mL of acetonitrile. These rinsings were combined in the separating funnel. The funnel was shaken for 1 min, the acetonitrile was drawn off, and the partitioning was repeated with a further 50 mL of acetonitrile. The combined acetonitrile extracts were evaporated to dryness.

Modifications for Cereals. Only 20 g of sample was used, and a 100-mL aliquot from 500 mL was taken through the cleanup procedures.

Modifications for Hops. Petroleum ether (60–80 °C) was used to extract 10 g of chopped sample. For the analysis of green hops a 250-mL aliquot from 500 mL was taken, and for dried hops a 50-mL aliquot was used. The hexane/acetonitrile partitioning was carried out with 100 mL of hexane and two 100-mL portions of acetonitrile.

Florisil Column Chromatography. Final cleanup is achieved by Florisil column chromatography. The procedures given below are suitable for the analysis of fruit and vegetables. Modifications are required for other crop types, and these are given separately.

Florisil (2.3 g) was added to 50 mL of hexane in a column, and the hexane was drained to within 0.5 cm of the surface of the Florisil. The residue was vigorously shaken with 10 mL of hexane. This solution was applied to the column and drained to within 0.5 cm of the surface of the Florisil at a rate of approximately 8 mL/min. A further 10 mL of hexane was similarly processed, the eluate from both additions being discarded. The residue was shaken with 75 mL of toluene, and this solution was added to the column and collected at a rate of approximately 5 mL/min. The eluate was evaporated to dryness and reconstituted in 2 mL of toluene for gas chromatographic analysis.

Modifications for Wheat and Barley, Grain and Ears. The column was rinsed with 50 mL of 50% hexane in toluene at a rate of approximately 8 mL/min prior to the elution with 75 mL of toluene.

Modifications for Wheat Foliage and Barley Straw. A column of 4.6 g of Florisil was used with a rinse of 150 mL of 50% hexane in toluene prior to elution with 150 mL of toluene.

Modifications for Wheat Straw. A column of 13.8 g of Florisil was used with a rinse of 300 mL of 50% hexane in toluene prior to elution with 200 mL of toluene.

Modifications for Hops. Two columns were used. The first used 9.2 g of Florisil, 200 mL of 50% hexane in toluene for rinsing, and 200 mL of toluene for the elution. The eluant was evaporated to dryness, and the residue was

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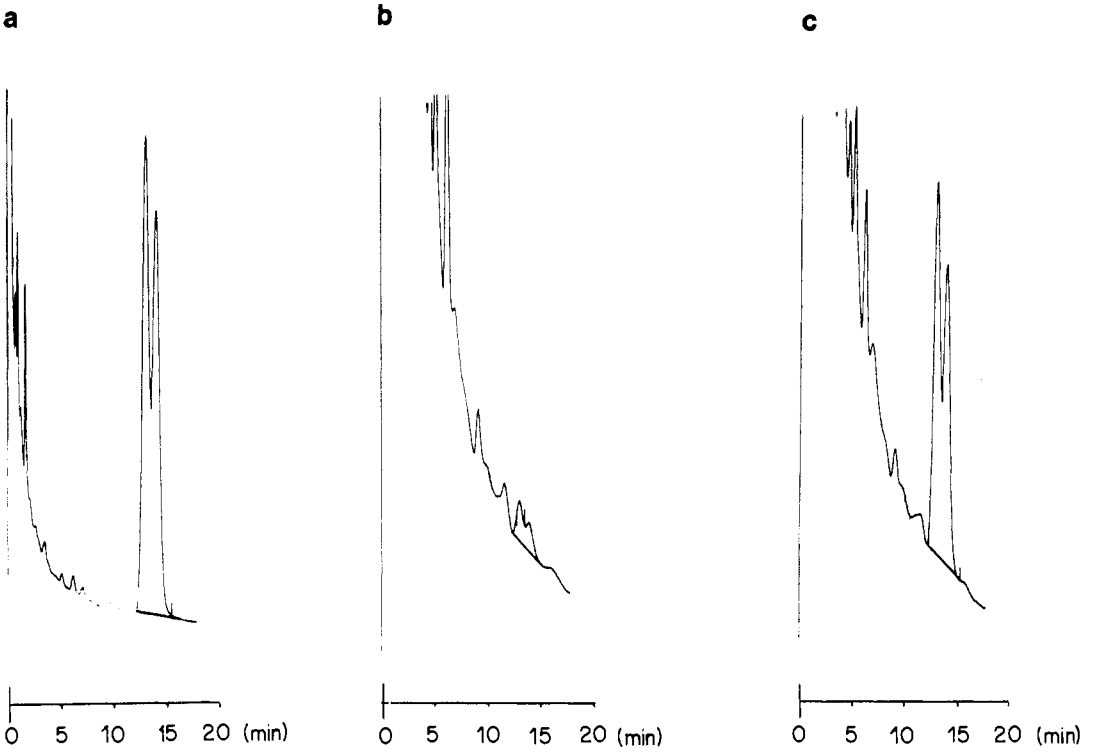

Figure 1. Chromatograms of flucythrinate in toluene at attenuation of 1 × 32: (a) 0.625-ng standard; (b) extract from untreated apple; (c) extract from untreated apple fortified with 0.05 ppm of flucythrinate.

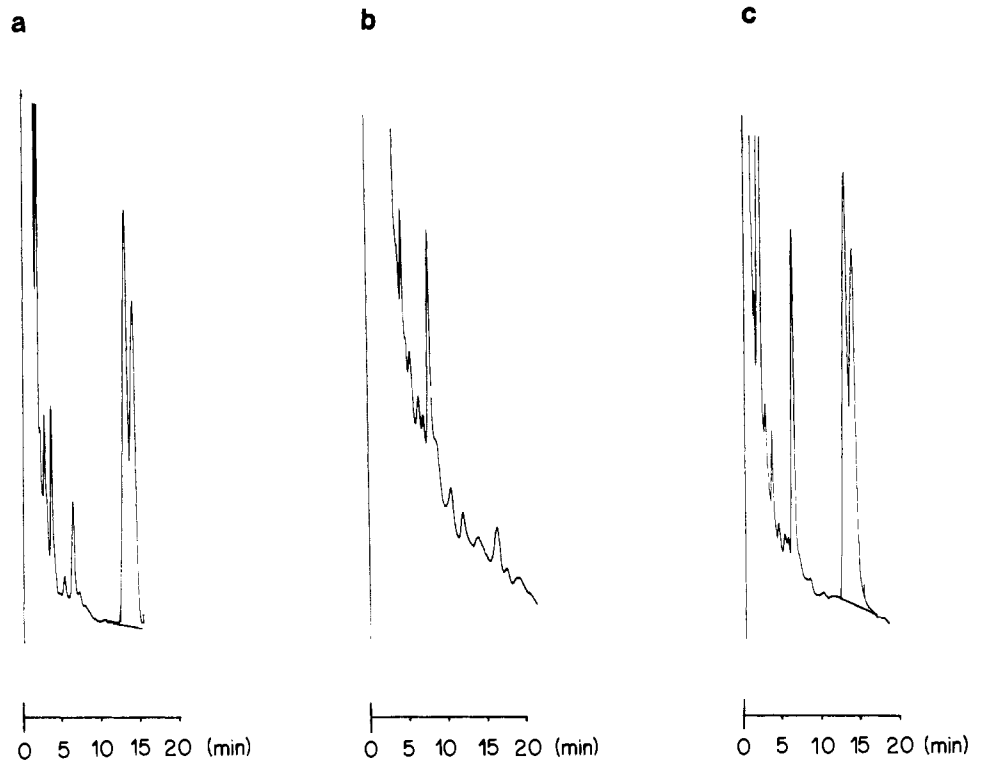

Figure 2. Chromatograms of flucythrinate in toluene at attenuation of 1 × 32: (a) 0.625-ng standard; (b) extract from untreated wheat straw; (c) extract from untreated wheat straw fortified with 0.05 ppm of flucythrinate.

stored overnight in a freezer at −6 °C. The second column used 4.6 g of Florisil. The sample was applied to the column in two 10-mL portions of hexane. No rinsing was required, elution being carried out with 100 mL of 10% diethyl ether in petroleum ether (60–80 °C) at a rate of approximately 5 mL/min, discarding the first 25 mL of effluent.

Fortification. Recoveries of flucythrinate were determined by the extraction of untreated chopped tissue fortified at 0.05, 0.10, 0.25, 0.50, and 1.00 ppm with standard solutions in acetone. Single recovery experiments were carried out at each level. The levels of flucythrinate residues were quantitated by comparison with standard solutions injected under identical gas chromatographic conditions.

Gas Chromatography. A glass column (1.2 m × 4 mm i.d.) packed with 5% OV-1 on 100/120 mesh Gas Chrom Q was used. Methane (5%) in argon was the carrier gas

Table I. Percent Recovery of Flucythrinate from Fortified Control Samples

sample	% recovery[a] 0.05 ppm	0.10 ppm	0.25 ppm	0.50 ppm	1.00 ppm	mean
red cabbage	75	93	90	90	89	87
white cabbage	90	85	89	83	88	87
savoy cabbage	80	98	92	85	94	90
broccoli	92	88	83	82	92	87
apples	85	85	90	107	90	91
wheat grain	85	75	93	74	77	81
wheat ears	85	100	96	99	98	96
wheat foliage	70	83	83	94	78	82
wheat straw	105	100	96	89	86	95
barley grain	95	98	90	101	93	95
barley ears	90	73	80	83	95	84
barley straw	110	73	79	83	95	88
green hops	73	83	104	76	85	84
dried hops	94	74	118	90	97	95

[a] Each value is the result of a single experiment.

at 30 mL/min. The operating parameters were as follows: detector temperature, 300 °C; injection port temperature 250 °C; column temperature, 240 °C. Under these conditions, flucythrinate gives two peaks at retention times of approximately 13 and 14 min, due to the two diastereoisomeric pairs (*RR*, *SS*, *RS*, *SR*). A linear response (the summed height for each peak) was observed over the range 0.06–1.25 ng of flucythrinate, using 5-μL injections of standard solutions. Quantitiation of apparent residues found was achieved by comparing the total peak height from the sample injection (5 μL) with the total peak height of a 0.50- or 0.625-ng standard injection. [Samples were reconstituted in a volume of toluene such that when injected (5 μL) the response observed was within the linear range tested.]

RESULTS AND DISCUSSION

The percent recoveries of flucythrinate from crops, fortified with standard material in the range 0.05–1.00 ppm prior to extraction, are given in Table I. The recoveries given are the results of single experiments. Typical chromatograms of standards and untreated and fortified samples are shown in Figures 1 and 2. Different sample weights were used for different crops in order to aid solvent extraction. Petroleum ether (60–80 °C) was used in the extraction of hops in order to reduce the amount of crop-related coextractives. Changes in the Florisil column chromatography were found to be necessary with different crop types in order to achieve adequate sample cleanup. For example, fruit and vegetable crops need no hexane/toluene column wash prior to elution with toluene whereas hops required two Florisil columns. For hops, overnight storage of the sample, at −6 °C before the second column, was found to be essential to remove interference.

The methods have been used successfully for the analysis of flucythrinate residues in treated crops. The basic method (cabbage, broccoli, apples) allows a throughput of approximately six samples per analyst per day whereas the modified methods allow analysis of four samples per analyst per day. The methods listed should be readily applicable to other crops.

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Validation of an Analytical Residue Method for Analysis of Glyphosate and Metabolite: An Interlaboratory Study

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A new residue method for the analysis of glyphosate and (aminomethyl)phosphonic acid has been validated with an interlaboratory study. Five different analysts from Monsanto Co. and other laboratories participated in testing of five different matrixes: alfalfa forage, cabbage, grapes, soybean grain, environmental water. These were chosen to represent the wide variety of matrixes analyzed for glyphosate-related residues. The cornerstone of the method is concentration and isolation via chelation ion exchange, with subsequent quantitation by HPLC with postcolumn reaction detection. The method was validated over the concentration range from 0.05 to 5.00 ppm with overall analytical recoveries of 80.9 ± 13.8% for glyphosate and 79.2 ± 13.8% for (aminomethyl)phosphonic acid. The coefficient of variation for both analytes was 17%, which fits well with that predicted for the analysis of compounds in this concentration range.

INTRODUCTION

There has been significant interest in recent years in analytical residue methodology for glyphosate [*N*-(phosphonomethyl)glycine] and its metabolite [(aminomethyl)phosphonic acid, AMPA] as a result of the increasing use of the herbicides Roundup, Rodeo, and Bronco. Numerous methods have been developed that determine the two compounds from specific matrices [Moye and St. John (1980); Guinivan et al., (1982); Roseboom and Berkoff (1982); Friestad and Bronstad (1982); Moye et al., (1983)], but many of these methods have been applied primarily to matrices that contain a high percentage of water and, by experience, have proven to be

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