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Note

High-performance liquid chromatographic separation and determination of quinalphos in technical and commercial formulations[a]

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O,O-Diethyl O-2-quinoxalinyl phosphorothioate (quinalphos) is a pesticide used effectively against caterpillars on cotton, groundnuts, vegetables and rice and its widespread use necessitates a simple, specific and rapid method for its determination in formulations. Many methods based on gas chromatography (GC)[1–4] and thin-layer chromatography (TLC)[5–8] have been reported for the determination of organophosphorus pesticides. GC methods for the determination of quinalphos lack not only precision but also accuracy as the compound is highly sensitive to heat and often decomposes in the column before it is detected and quantified. The separation of quinalphos by paper chromatography followed by its combustion for spectrophotometric determination is an accurate procedure[9], but it is tedious and time consuming. Chemical methods[10,11] in which the quinalphos content is measured by phosphorus or nitrogen determination suffer from interference from O,O-diethyl phosphorochloridothioate, ethyl phosphorodithioate and 2-ethoxyquinoxaline. Rastogi *et al.*[12] described a gravimetric method for the determination of quinalphos using copper(I) chloride as a precipitating agent. However, it has the drawback that 2-hydroxyquinoxaline and the emulsifier interfere during precipitation. Therefore, a rapid and reliable method for the determination of quinalphos in formulations is still needed.

High-performance liquid chromatographic (HPLC)[13–16] methods for the separation of several organophosphorus pesticides have been reported, but these are neither specific nor selective for quinalphos in formulations. We report here a simple and selective HPLC method for the separation and determination of quinalphos in technical and commercial formulations. The proposed method is also applicable to the analysis of food and agricultural products for residues of quinalphos.

EXPERIMENTAL

Materials and reagents

All reagents were of analytical-reagent grade unless stated otherwise. HPLC-

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grade 2,2,4-trimethylpentane and 2-propanol were obtained from Spectrochem (Bombay, India) and 1,3-dinitrobenzene from BDH (Poole, U.K.).

Quinalphos was prepared by condensing 2-hydroxyquinoxaline with O,O-diethyl phosphorochloridothioate in the presence of anhydrous potassium carbonate and acetonitrile according to Schmidt and Hammann[17]. The product was stabilized by adding epichlorohydrin and diluted with *o*-xylene to obtain a sample of technical-grade quinalphos. Commercially formulated samples, Ekalux and Suquin (25% emulsifiable concentrate) and dust powder (1.5% quinalphos), manufactured by Sandoz (Bombay, India) and Sudarshan Chemicals (Poona, India), respectively were used.

Apparatus

A high-performance liquid chromatograph (Shimadzu, Kyoto, Japan) with a 20-μl loop injector having an high-pressure six-way valve was used. A Model SPD-6AV variable-wavelength UV–VIS spectrophotometric detector (Shimadzu, Kyoto, Japan) was connected after the column. A Zorbax SIL (DuPont, Wilmington, DE, U.S.A.) column (250 mm × 4.6 mm I.D.; 5 μm particle size) was used for separation. The chromatograms and the integrated data were recorded by a Chromatopac C-R3A processing system.

Chromatographic conditions

The mobile phase was 2,2,4-trimethylpentane–2-propanol (9:1, v/v). Samples were dissolved in the mobile phase. The analysis was carried out under isocratic conditions at a flow-rate of 1 ml/min and a chart speed of 5 mm/min at room temperature (27°C). Chromatograms were recorded at a wavelength of 245 nm.

Sample preparation

Samples of quinalphos (50 mg) and 1,3-dinitrobenzene (10 mg) used as the internal standard (I.S.) were dissolved in the mobile phase (25 ml).

Analytical procedure

Standard mixtures containing 2–10 mg of internal standard, 5–20 mg of quinalphos, 15–50 mg of *o*-xylene and 1–2 mg of epichlorohydrin were prepared by dissolving known amounts of the compounds in 10 ml of the mobile phase. A 5-μl volume of each standard mixture was injected and chromatographed under the above conditions. From the areas of the peaks the response factor of quinalphos with respect to the internal standard was calculated.

Synthetic mixtures and commercial formulations were analysed under identical conditions. Quinalphos of technical grade or a commercial formulation (50 mg) together with the internal standard (10 mg) was dissolved in 25 ml of mobile phase and chromatographed. The percentage of quinalphos was calculated from the peak areas.

RESULTS AND DISCUSSION

The HPLC separation of quinalphos, epichlorohydrin, *o*-xylene and 1,3-dinitrobenzene is shown in Fig. 1. The peaks were identified by injecting the individual compounds. It can be seen that quinalphos is not only resolved from its formulation

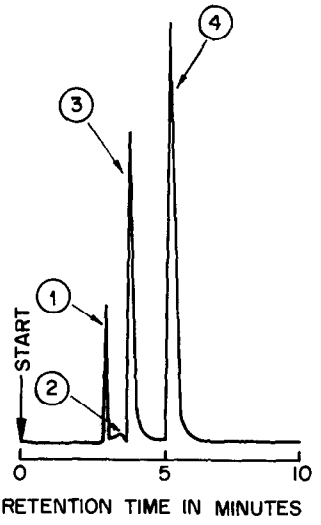

Fig. 1. Typical chromatogram of technical-grade quinalphos (30 μg). For conditions, see text. Peaks: 1 = *o*-xylene; 2 = epichlorohydrin; 3 = quinalphos; 4 = 1,3-dinitrobenzene (10 μg).

additives but also from 1,3-dinitrobenzene. The λ_{max} and retention time data for all the compounds are given in Table I. A wavelength of 245 nm, at which all the compounds under investigation absorb UV light, was selected for analysis not only because detection is ensured but also a good linearity between mass and integral response is obtained. When the UV detector is set at 0.001 a.u.f.s. the limit of detection for quinalphos is $3 \cdot 10^{-9}$ g with a signal-to-noise ratio of 4.0. Response factors for quinalphos were determined in the range 25–75% of quinalphos and are recorded in Table II. They were found to remain constant throughout this range.

Standard mixtures containing different amounts of quinalphos were prepared and analysed by HPLC. The results are given in Table III. The accuracy of the method was determined by the standard addition technique. Subsequent additions of quinalphos were accurately reflected in the peak heights. The measured amounts of quinalphos agreed well with the actual values to within 1.88%. Linear regression analysis of the data yielded the line $y = 0.9978x + 0.1999$ with a correlation coefficient of 0.9998.

TABLE I

RETENTION DATA

Compound	*Retention time (min)*	*Relative retention time*	λ_{max} *(nm)*
1,3-Dinitrobenzene	5.61	1.00	233
Quinalphos	4.12	0.73	237
Epichlorohydrin	3.57	0.64	210
o-Xylene	3.25	0.58	263

TABLE II

DETERMINATION OF RESPONSE FACTORS FOR QUINALPHOS

Sample No.	*Weight of quinalphos (mg)*	*Weight of I.S. (mg)*	*Area of quinalphos peak*	*Area of I.S. peak*	*Response factor*[a]
1	16.53	8.10	15 557	13 902	1.8236
2	16.53	8.10	15 814	14 068	1.8154
3	16.53	5.40	15 505	9271	1.8303
4	16.53	5.40	15 721	9627	1.8745
5	12.40	5.40	11 813	8907	1.7314
6	12.40	5.40	11 915	9639	1.8577
7	8.27	5.40	7783	8957	1.7625
8	8.27	5.40	7515	8770	1.7872
9	8.27	2.70	7768	4514	1.7799
10	8.27	2.70	7724	4469	1.7722
Mean					1.8035
S.D.					0.0423
R.S.D.[b] (%)					2.4

[a] Response factor of quinalphos = [A (I.S.)/W (I.S.)] · [W (quinalphos)/A (quinalphos)], where A = peak area and W = weight.

[b] Relative standard deviation.

Technical and commercial formulations of quinalphos were analysed by the proposed method and also by the Indian Standards Institution procedure[9]. The results (Table IV) are in good agreement, with an error of 1.56%.

Vegetables spiked with quinalphos at the level of 1 mg kg^{-1} were been crushed and dried, extracted with acetone and analysed using the developed method. The chromatograms of a cabbage blank and the same sample spiked with quinalphos are shown in Fig. 2. Even at the lowest detector attenuation setting, the blank showed a fairly flat baseline in the region where quinalphos and 1,3-dinitrobenzene elute and the limit of detection for quinalphos in spiked vegetables was found to be 0.05 mg kg^{-1}. The results are given in Table V.

It is concluded that the proposed method is not only suitable for the routine determination of the quinalphos in technical and commercial formulations but may

TABLE III

DETERMINATION OF QUINALPHOS IN STANDARD MIXTURES

Sample No.	*Quinalphos (%)*		*Error (%)*
	Taken	*Found*[a]	
1	20.36	20.72	+1.77
2	25.07	25.54	+1.88
3	29.14	28.69	−1.54
4	65.20	65.09	−0.17
5	70.05	70.32	+0.39

[a] Averages of triplicate analyses.

TABLE IV

DETERMINATION OF QUINALPHOS IN TECHNICAL AND COMMERCIAL FORMULATIONS

Sample No.	*Formulation*	*Quinalphos present (%)*	*Quinalphos found[a] (%)*		*Mean ± R.S.D. (%)*		*Error (%)*
			By HPLC	*By ISI method[9]*	*By HPLC*	*By ISI method[9]*	
1	Ekalux	25.00	24.73	26.36	25.03 ± 1.33	24.82 ± 4.38	+0.85
			25.49	24.15			
			24.86	23.96			
2	Suquin	25.00	24.51	25.92	24.81 ± 1.23	25.17 ± 2.69	−1.43
			24.70	25.31			
			25.23	24.28			
3	Technical I	65.00	63.79	65.45	64.58 ± 1.36	65.59 ± 1.20	−1.54
			65.80	66.62			
			64.15	64.70			
4	Technical II	70.00	71.36	68.57	70.96 ± 1.03	69.87 ± 1.37	+1.56
			71.59	70.84			
			69.94	70.20			
5	Dust	1.50	1.53	1.43	1.48 ± 6.47	1.46 ± 6.83	+1.37
			1.57	1.35			
			1.35	1.59			

[a] Average values obtained from duplicate analyses.

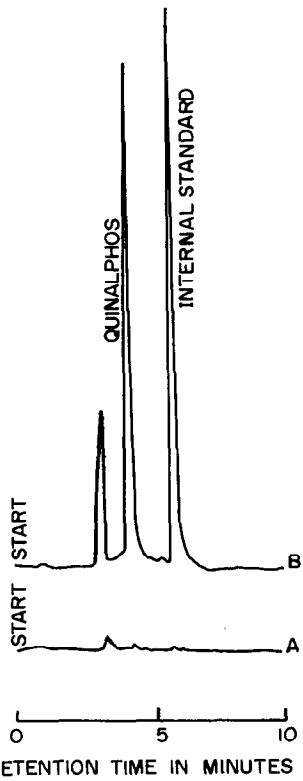

Fig. 2. Chromatograms of (A) a cabbage blank and (B) the same cabbage spiked with 7.3 mg kg^{-1} of quinalphos and 10 mg of internal standard. For conditions, see text.

TABLE V

DETERMINATION OF TRACE AMOUNTS OF QUINALPHOS IN VEGETABLES

Sample	*Amount of sample (g)*	*Content of quinalphos*	
		mg	*ppm*
Cabbage	125.70	0.92	7.32
Cauliflower	104.52	0.50	4.78
Potato	139.86	0.17	1.26

also be useful for the analysis of environmental samples and agricultural products for its residues. It is simple, specific, rapid and inexpensive compared with methods reported earlier[9–12]. Moreover, impurities such as O,O-diethyl phosphorochloridothioate and 2-hydroxyquinoxaline, which are generally present in quinalphos, do not interfere because they elute at 3.43 and 8.95 min, respectively.

REFERENCES

1 M. Horiba, *J. Chromatogr.*, 287 (1984) 189.
2 M. Wisson, C. V. Hoek and H. H. Sauer, in G. Zweig and J. Sherma (Editors), *Analytical Methods for Pesticides and Plant Growth Regulators*, Vol. XI, Academic Press, New York, 1980, pp. 147–163.
3 J. Kjølholt, *J. Chromatogr.*, 325 (1985) 231.
4 Y. V. Subbarao, S. Siddhan and P. N. Sarma, *Proceedings of the National Symposium on Recent Developments in Applied Analytical Chemistry*, Institution of Chemists, India, 1978, pp. 79–83.
5 G. L. Brun and V. Mallet, *J. Chromatogr.*, 80 (1973) 117.
6 G. L. Braequer, *Pflanzenschutz-Nachr.*, 22 (1969) 308; *C.A.*, 74 (1971) 9830lz.
7 I. Vukusic and B. Laskarin, *J. High Resolut. Chromatogr. Chromatogr. Commun.*, 4 (1981) 659.
8 V. Mallet and G. L. Brun, *Bull. Environ. Contam. Toxicol.*, 12 (1974) 739.
9 *Specifications for Quinalphos Technical, IS : 8072*, Indian Standards Institution, New Delhi, 1976, pp. 6–10.
10 *Quantitative Determination of Organophosphorus Pesticides in Technical and Formulated Products*, Sandoz, Basle, 1975, Appendix 10.
11 *Specifications for Quinalphos Technical, IS : 8072*, Indian Standards Institution, New Delhi, 1976, pp. 10–11.
12 R. C. Rastogi, N. C. Dey and P. Baruah, *Analyst (London)*, 109 (1984) 1361.
13 P. Bottomley and P. G. Baker, *Analyst (London)*, 109 (1984) 85.
14 G. Szalontai, *J. Chromatogr.*, 124 (1976) 9.
15 L. G. Rice, *J. Chromatogr.*, 317 (1984) 523.
16 Working Group of the Committee for Analytical Methods for Residues of Pesticides and Veterinary Products in Foodstuffs and Working Party on Pesticide Residue of the Ministry of Agriculture, Fisheries and Food, *Analyst (London)*, 105 (1980) 515.
17 K. J. Schmidt and I. Hammann, *Br. Pat.*, 1 160 493 (1969); *C.A.*, 71 (1969) 113 084q.