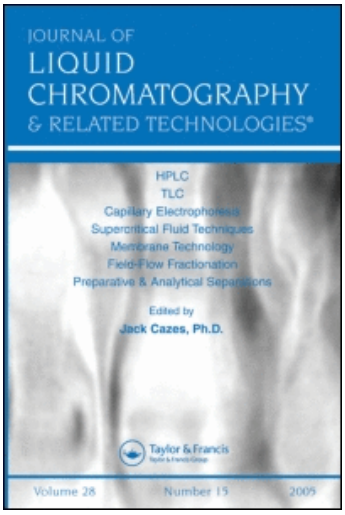

Journal of Liquid Chromatography & Related Technologies
Publication details, including instructions for authors and subscription information:
http://www.informaworld.com/smpp/title~content=t713597273

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N. Delhom[a]; Y. Balanant[a]; J. C. Ader[b]; A. Lattes[b]
[a] Société Sétric International Chemin Des Monges, Deyme, France [b] Université P. Sabatier Lab, Toulouse, Cedex, France

To cite this Article Delhom, N. , Balanant, Y. , Ader, J. C. and Lattes, A.(1996) 'High Performance Liquid Chromatographic Determination of Diazinon in Polymeric Matrix', Journal of Liquid Chromatography & Related Technologies, 19: 11, 1735 — 1743
To link to this Article: DOI: 10.1080/10826079608014001
URL: http://dx.doi.org/10.1080/10826079608014001

J. LIQ. CHROM. & REL. TECHNOL., 19(11), 1735–1743 (1996)

HIGH PERFORMANCE LIQUID CHROMATOGRAPHIC DETERMINATION OF DIAZINON IN POLYMERIC MATRIX

N. Delhom, Y. Balanant

Société Sétric International
Chemin Des Monges
31450 Deyme, France

J. C. Ader, A. Lattes

Université P. Sabatier
Lab. IMRCP, UA CNRS No. 470
118 Route de Narbonne
31062 Toulouse Cedex, France

ABSTRACT

As a part of a study of pesticide migration through a polymer matrix, an HPLC method for analysis of O,O-diethyl-O-(6-methyl-2-(1-methylethyl)-4-pyrimidinyl)-phosphorothioate, commonly called diazinon) was developed.

INTRODUCTION

In human or animal therapy, some medicinal forms use a principle where the active compound is incorporated into a polymer matrix and gradually diffused following controllable kinetics. Control of the active ingredient at

different times during treatment is of great importance.[1-3] Also, realisation of this kind of presentation requires accurate and reliable analytical tools.

This paper describes an HPLC method for determination of an organophosphorus substance, O,O-diethyl-O-(6-methyl-2-(1-methylethyl)-4-pyrimidinyl)-phosphorothioate, commonly called diazinon. This compound is dispersed in a polyvinyl chloride polymer matrix. This kind of formulation, used in the pesticide field, should be considered as an interesting model for long-term treatment by the slow migration of an active substance.

Also, it is necessary, after extraction, to elaborate a chromatographic technique which allows a qualitative and quantitative evaluation. This was achieved by an internal standardization method.[4-8]

Currently, the most common analysis methods for determination of this compound use gas phase chromatography.[9-13] Gas chromatography presents some limitations in the case of a thermolabile substance (the degradation of diazinon begins at 120°C) and liquid chromatography appears to be more suitable. Most of the previous HPLC work on diazinon has been devoted to the environment, especially in water and vegetables.[14-16]

EXPERIMENTAL

Materials and Equipment

The HPLC system consisted of a Millipore Waters unit with a UV/Visible diode array detector (Waters 991), with detection at 250 nm. The analysis was carried out with an ODS C_{18} (250 mm x 4.6 mm I. D., 5 microns) reversed phase column. The mobile phase was a mixture of acetonitrile/water, 85/15 (v/v). The flow rate was 1 mL/min.

Preparation of Solutions

Internal standard solution.

Accurately weigh about 0.12 g of benzophenone into a 20 mL volumetric flask and dilute with dioxane.

Reagent: Benzophénone , purity > 99%, Fluka.

Diazinon standard solution.

Accurately weigh about 0.12 g of diazinon into a 25 mL volumetric flask and dilute with ethanol.

Reagent: Diazinon standard, purity > 99.1% ,Cluzeau, 33 Ste. Foy la Grande.

Calibration solutions.

Successively, accurately, measure 4.5 mL; 3.5 mL; 2.0 mL; 1.0 mL; 0.5 ml of the diazinon standard solution and introduce into separate 10 mL volumetric flasks. Next, add 0.4 mL of the internal standard solution and dilute to volume with a mixture of ethanol/THF 4/1 (v/v).

Method of Sample Preparation.

Polyvinyl chloride is used as the polymer matrix. PVC and stabilizer are mixed, at 80°C. Diazinon and plasticizer (dibutyl phtalate) are incorporated. The resulting mixture is then heated to 110°C. After cooling, this dry blend is extruded into a continuous strap into a cold water bath and dried in an air current.

Sample solution.

To 1 g of sample, add 10 mL of tetrahydrofuran. After total dissolution, add 35 ml of ethanol; the polyvinyl chloride precipitates. This preparation is filtered into a 100 mL volumetric flask. This operation is repeated a second time. Then add 4 mL of internal standard solution and dilute with ethanol to 100 mL.

Analysis was performed after preparation of a polymeric sample. This sample contained approximately 15% of diazinon. It corresponded to the veterinary licence quantity. This diazinon quantity should be contained between the limit values 13.5% and 16.5% in mass percentage.

The diazinon quantity incorporated was equal to 16%, an amount of active compound practiced to compensate for eventual loss of diazinon during the extrusion phase.

Determination of Diazinon Content.

P1 = diazinon standard purity,
Ws = diazinon weight in standard solution,
Wt = sample weight in sample solution,
We = benzophenone weight in diazinon standard solution,
W'e = benzophenone weight in sample solution,
Xs = benzophenone peak area obtained with diazinon standard solution,
Xt = benzophenone peak area obtained with a sample solution,
Ys = diazinon peak area obtained with diazinon standard solution,
Yt = diazinon peak area obtained with sample solution,
Rs = Ys/Xs ratio,
Rt = Yt/Xt ratio

The content of diazinon in the sample was calculated from the following equation:

$$\% \text{ Diazinon} = Rt/Rs \times ((Ws/We \times P1) / (Wt/W'e)) \times 100$$

RESULTS AND DISCUSSION

Presently, diazinon is analyzed by a gas chromatographic method. Diazinon is a thermolabile substance, its degradation temperature is 120°C. Hence, the gas chromatographic method is likely a destructive method.

The temperature conditions during GC analysis were 190°C for the column, 250°C for the injector and 280°C for the detector (FID). With this method, a good separation was obtained. The retention time for the benzophenone used as an internal standard was 2.63 minutes, for the diazinon, it was 3.30 minutes. A quantitative result was obtained; however, the diazinon was determined with a 4.3% variation coefficient (CV%). CV% was evaluated as an average after 10 injections. We obtained a low recovery.

In the literature, the referee CG method[10] gave the retention time for internal standard (Aldrin) at 10 to 12 minutes, for the diazinon, it was 5 to 6 minutes. Repeatability was equal to 1.9% and reproductibility to 2.0%.

The determination of diazinon by high performance liquid chromatography during preliminary analysis gives the retention time of benzophenone (t_R = 4.23 min) and Diazinon (t_R = 5.77 min). Using a diode array detector permits a tri-dimensional analysis in wavelength spectrum from

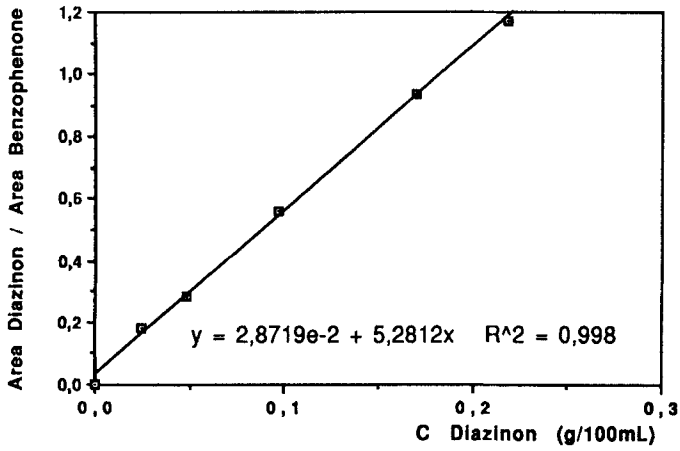

Figure 1. Calibration curve of diazinon. Graph representing peak area ratio of diazinon to benzophenone versus diazinon concentration.

180 nm to 380 nm. The wavelength selected was 250 nm in accordance with the absorption spectra of benzophenone and diazinon.

A calibration curve was established with a concentration range from 0.02 g/100 mL to 0.2 g/100 mL (three injections, 5 microliters each). For each concentration, the peak area ratio of diazinon to benzophenone Rs = $\Sigma n(Ys/Xs)/n$ were calculated. The results are reported in Table 1.

We have taken the detector response at a wavelength of 250 nm, for a diazinon range quantity from 0.2 µg to 200 µg. We have obtained a good linearity with a correlation coefficient of 0.998.

The precision of the analytical method was evaluated from the degree of repeatability of the peak area ratio of diazinon to an internal standard in a series of 10 injections. The precision of this method is usually expressed as the coefficient of variation (CV%).

Data reported in Table 2 are relative to a sample solution obtained after diazinon extraction from the polymeric matrix, as indicated in the experimental part.

Table 1

Calculation of Peak Area Ratio of Diazinon to Benzophenone

Std. Solution	Concentration (g/100mL)	Rs*
1	0.2187	1.167
2	0.1701	0.936
3	0.0972	0.560
4	0.0486	0.283
5	0.0243	0.178

* $Rs = \Sigma n(Ys/Xs)/n$

Table 2

Repeatability of Retention Times and Peak Area Ratios of Diazinon to Benzophenone*

	Benzophéone		Diazinon		R
Inj. No.	tR (min)	Xt	tR (min)	Yt	Yt/Xt
1	4.26	0.11831	5.84	0.07065	0.597
2	4.26	0.11899	5.83	0.07008	0.588
3	4.24	0.12353	5.81	0.07257	0.587
4	4.25	0.14002	5.79	0.08265	0.590
5	4.24	0.11543	5.78	0.06875	0.595
6	4.24	0.13314	5.79	0.07862	0.590
7	4.23	0.13197	5.75	0.07794	0.590
8	4.21	0.12485	5.73	0.07332	0.587
9	4.22	0.1283	5.74	0.07535	0.587
10	4.22	0.13364	5.74	0.0786	0.588
Average	4.237	0.12682	5.78	0.07485	0.590
CV %	0.46%	5.93%	0.69%	5.73%	0.56%

* Calculated from 10 analyses of a sample prepared as indicated in the Experimental section.

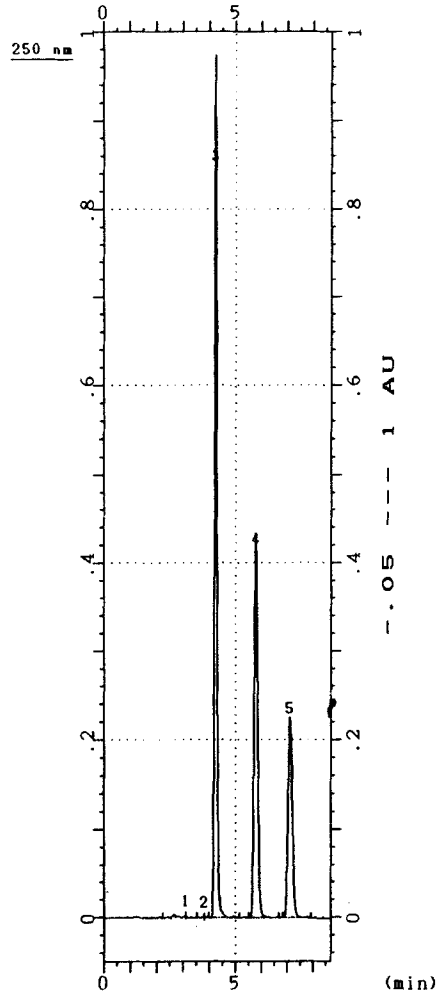

Figure 2. Chromatogram of benzophenone (3), diazinon (4) and plasticizer: dibutyl phtalate (5).

A good repeatability of retention times was obtained, $t_R = 4.23 \pm 0.02$ min for benzophenone and $t_R = 5.77 \pm 0.04$ min for diazinon. The coefficients of variation for the retention times are: 0.46% for benzophenone and 0.7% for diazinon.

The addition of an internal standard allowed us to neglect any error in the injected volume. After 10 injections, the precision of this method was evaluated from a ratio Yt/Xt of the variation coefficient and estimated at 0.56%. The active substance quantity in polymeric matrix has a mean value equal to 15.85 ± 0.08% w/w. It corresponds to a concentration range advocated by the veterinary license.

Figure 2 shows the chromatogram of a polymer sample analysis. Analysis conditions:

Column:	Kromasil® 110 ODS C_{18}
	250 mm x 4.6 mm I. D.
Mobil phase:	CH_3CN/H_2O, 85/15 v/v
Flow rate:	1 mL/mn
Detector:	UV/Visible diode array detector
Wavelength:	$\lambda = 250$ nm

CONCLUSION

This study shows the possibility of validating this HPLC method for qualitative and quantitative analysis of O,O-diethyl-O-(6-methyl-2-(1-methyl ethyl)-4- pyrimidinyl)-phosphorothioate (diazinon). The precision is evaluated by the coefficient of variation at 0.56%. The HPLC procedure developed appears to offer an attractive alternative approach to the currently employed gas chromatographic method in the veterinary license and CIPAC (Collaborative International Pesticides Analytical Council) referee method. This method presents a greater recovery and has the advantage of being very rapid. It is a non-destructive method and permits easy control of diazinon distribution kinetics during external therapeutic treatment.

REFERENCES

1. J. P. Varion, L. Yean, J. C Meslard, F. Subira, C. Bunel, L'Actualité Chimique, Sept./Oct., 330-335 (1992).

2. L. Cassiani-Ingoni, F. Subira, C. Bunel, J. P. Vairon, J. L. Halary, Makromol. Chem., Macromol. Symp., **19**, 287-293 (1988).

3. J. Grove, C. Bunuel, J. C. Meslard, J. P. Vairon, B. Plazonnet, **IVth World Conf. on Clin. Pharm. Therap.**, Mannheim (1989).

4. Peter L. Carter, Keith C. Overton, J. Assoc. Off. Anal. Chem., **69**, (5) 908-911 (1986).

5. U. Nemmer, N. Dimov, J. Chromatogr., **288**, 227-229 (1984).

6. Rice, L. Grant, J. Chromatogr., **317**, 523-526 (1984).

7. M. W. Dong, F. L. Vandermark, P. V. Passalacqua, LC Views, May, 1992.

8. R. Rosset, M. Caude, A. Jardy, **Manuel Pratique de Chromatographie en Phase Liquide**, 2nd edition, Masson, Paris, 1982.

9. Sétric International, Dossier Analytique AMM No. 69794100485.

10. CIPAC Method, **CIPAC Handbook**, Vol IA, p. 1199, 1980.

11. R. Turle, B. Levac, Bull. Environ. Contam. Toxicol., **38(5)**, 793 (1987).

12. A. F. Machin, C. R. Morris, Analyst, **97(1153)**, 289-293 (1972).

13. Karr J. James, J. Assoc. Off. Anal. Chem., **65(1)**, 115-119 (1982).

14. S. Farhat, M. M. H. Baig, M. A. Khan, Karachi Univ. J. Sci., **20(1-2)**, 143-149 (1992).

15. M. R. Driss, M. C. Hennion, M. L. Bouguerra, J. Chromatogr., **639(2)**, 352-358 (1993).

16. Mallet N. Victorin, M. Dugay, M. Bernier, N. Trottier, Int. J. Environ. Anal. Chem., **39(3)**, 271-279 (1990).

Received June 28, 1995
Accepted October 6, 1995
Manuscript 3913