ELSEVIER

Journal of Chromatography A, 761 (1997) 322–326

JOURNAL OF CHROMATOGRAPHY A

Short communication

Direct determination of bromacil and diuron residues in environmental water samples by coupled-column liquid chromatography and large-volume injection

J.V. Sancho*, C. Hidalgo, F. Hernández

Analytical Chemistry, Experimental Sciences Department, University Jaume I, P.O. Box 224, 12080 Castelló, Spain

Received 28 May 1996; revised 25 September 1996; accepted 27 September 1996

Abstract

A rapid procedure for the determination of two herbicides in environmental water samples is described. The method makes use of the potential of coupled-column liquid chromatography for the trace determination of polar compounds by direct large-volume injection of aqueous samples. With direct injection of 2000-μl samples, detection limits down to 0.1 μg/l and an analysis time of about 10 min can be achieved, rendering an overall procedure with a throughput of 50 samples per day. Drinking and surface water samples spiked at levels between 0.2 and 1 μg/l yielded average recoveries between 83–107% ($n=5$) with relative standard deviations between 3–8%. The calibration graphs are linear over at least three orders of magnitude.

Keywords: Water analysis; Pesticides; Environmental analysis; Large-volume injections; Bromacil; Diuron; Pesticides

1. Introduction

Reversed-phase column liquid chromatography (RPLC) can be used to determine small amounts of polar organic compounds in environmental water samples. An important advantage of RPLC in conjuction with aqueous samples is that the low eluotropic strength of water samples allows the injection of large sample volumes [1].

In recent papers, RPLC column-switching with ultraviolet or fluorescence detection using two separation columns was used for the sensitive and selective determination of polar analytes in environmental samples [2–5] in less than 15 min, which makes this technique very attractive for screening purposes. Relevant aspects of column-switching optimization for the determination of moderately polar pesticides have been reviewed [6,7].

Bromacil and diuron, the active ingredients in the commercial herbicide Krovar, marketed by DuPont, are widely used in the Castellón area for weed control in citrus orchards. Residues of the former have been found recently in surface water samples from a wet area nearby which is within a pesticide monitoring program [8]. Both compounds can be analysed either by gas chromatography (GC) or liquid chromatography (LC), but the later approach easily allows the simultaneous determination of the analytes without additional pre-derivatization steps. Therefore the development of an LC procedure will

*Corresponding author.

PII S0021-9673(96)00827-8

be very useful for fast screening in monitoring programs.

RPLC has been used for the determination of diuron, among other phenylureas [9–17] as well as for the multiresidual analysis of polar herbicides, including bromacil and diuron [18–22] in environmental water samples, but herbicides usually applied in weed control have not been focused on. Bromacil and diuron have been analysed by RPLC following a column-switching procedure [23] by performing an off-line liquid–liquid extraction of a 1 l water sample and subsequent concentration by a Kuderna–Danish evaporator, which makes the overall procedure long and tedious. Therefore, the availability of a large volume injection LC–LC procedure makes screening of these analytes more sensitive, more selective and faster.

In this paper, the suitability of large-volume injection coupled-column LC technique was investigated for the rapid determination of residues of bromacil and diuron in environmental water samples.

2. Experimental

2.1. Chemicals

Bromacil and diuron (content >99%) were obtained from Riedel-de Haën (Seelze, Germany). Methanol and acetonitrile, both HPLC-grade, were purchased from Scharlau Science (Barcelona, Spain). HPLC-grade water was obtained by purifying demineralized water in a Nanopure II system (Barnstead, Newton, MA, USA).

Stock standard solutions (ca. 400 μg/ml) of bromacil and diuron were prepared in methanol. Meanwhile, mixed diluted standards were prepared with HPLC-grade water. MeOH–water (30:70) was used as the first (M-1) mobile phase and MeOH–water (65:35) as the second (M-2) mobile phase.

2.2. Equipment

The modular LC system consisted of a Model 1050 sampler (Hewlett-Packard, Waldbronn, Germany), a manual injector which, equipped with a 2.0-ml loop, was used to perform large-volume injections (LVI), a Model 1050 gradient LC pump, (Hewlett-Packard), a Model C6W six-port switching valve driven by a WE-II actuator from Valco (VIGI, Schenkon, Switzerland) and time controlled by the sampler, a Model 2150 isocratic LC pump from LKB (Bromma, Sweden), a Model 1050 ultraviolet detector (Hewlett-Packard) set at 277 nm and 250 nm (time programmed), a 30×4.6 mm I.D. first separation column (C-1) packed with 5-μm Spherisorb ODS-2 from Scharlau Science and a 100×4.6 mm I.D. second separation column (C-2) packed with 3-μm Microsphere C_{18} from Chrompack (Middleburg, Netherlands). C-2 was kept at 30°C in the column heater of the Model 1050 pump.

Recordings of chromatograms and quantitative measurements of peak areas were performed with a Hewlett-Packard HPLC Chem Station (software version G1034A). A MicropH 2001 pH meter and Pipetmans (200-, 1000- and 5000-μl) were obtained from Crison Instruments (Barcelona, Spain) and Gilson, respectively.

Recordings of spectra were performed with a spectrophotometer Model UV-240 from Shimadzu.

2.3. Procedure

The mobile phases were set at a flow-rate of 1 ml/min. 2 ml of the water sample, prefiltered for surface waters, was injected onto C-1. After clean-up with a certain volume of M-1 (injection volume included), C-1 was switched on-line with C-2 for a short time to transfer the fraction containing bromacil and diuron to C-2. After transferring, C-1 was rinsed and conditioned with M-1 while the analytes were separated on C-2 with M-2. Quantification of analytes was done by external calibration with standard solutions in water.

The clean-up and transfer times were matrix-dependent for bromacil, so the clean-up and transfer times were slightly different, as shown in Table 1.

3. Results and discussion

The UV spectra of both analytes were recorded in order to evaluate their maxima wavelength and UV response. In the case of diuron, $\lambda_{max}=250$ nm and $\epsilon^0=22144$ l mol^{-1} cm^{-1}, and for bromacil, $\lambda_{max}=277$ nm and $\epsilon^0=7885$ l mol^{-1} cm^{-1}.

Table 1
LC–LC coupling conditions for bromacil and diuron in different water matrices

	Clean-up volume M-1 (ml)	Transfer volume M-2 (ml)
HPLC water	5.64	1.26
Drinking water	3.74	2.86
Surface water	5.00	0.90

As discussed in earlier work [4], the attainable sensitivity and selectivity of a coupled-column procedure will depend on how much sample can be injected on the first column and transferred to the second column without excessive band broadening of the analytes. Two processes are important, the elution of the analytes during injection and the peak compression prior to transfer. Applying large-volume injections (LVI), elution on C-1 must be considered as a step gradient elution in which the same volume acts as the first mobile phase. An important advantage of working with aqueous samples is that the low eluotropic strength of water samples allows the injection of large sample volumes.

Earlier studies [1–3,7] have shown that successful coupled-column LC analysis of highly polar organic compounds in aqueous samples can be carried out, even with low retention on C_{18} bonded silica, non-selective UV detection wavelength or low molar extinction coefficient. Better results are expected for analytes for which the stated parameters are more favourable, as for bromacil and diuron (Table 2), showing selective detection wavelengths, high molar extinction coefficients and high k' values in pure aqueous solutions, allowing large volume injection without additional band broadening.

Therefore, the volume injection was studied injecting 400 and 200 ng for bromacil and diuron respectively, on C-1 (30 mm long, packed with 5-μm C_{18}) with a mobile phase MeOH–water (40:60, v/v). The volumes studied range from 100 to 4000 μl, showing the possibility of performing LVI with good peak shape (width and asymmetry) even up to 4.0-ml injection.

Selecting a sample volume of 2.0 ml, as a compromise between required sensitivity and speed of analysis, the mobile phase compositions were optimized for both M-1 and M-2, in order to obtain the best performance in sensitivity and selectivity. Mobile phase compositions (M-1) ranging between 30–40% MeOH and M-2 between 60–65% MeOH were assayed. Finally, 30% MeOH and 65% MeOH were chosen as M-1 and M-2, respectively.

When attempting to apply this procedure to drinking and surface water samples spiked with bromacil and diuron, the procedure failed in the determination of the more polar herbicide, bromacil. After disregarding degradation or wavelength maxima shift, we concluded that the problem was a wrong 'heart-cutting' due to matrix interferences. Bromacil was affected by the water matrix, changing slightly its retention on C-1, but enough for completely loosing

Table 2
Properties of polar compounds analyzed by LVI–LC–LC with UV detection

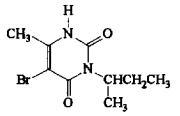
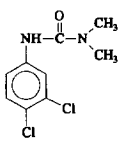
Compound	Formula	S_{H_2O} (g/l)	k'	λ (nm)	ε (l mol^{-1}cm^{-1})	Sample Vol. (μl)	LOD (μg/l)
Bromacil		0.815	88	277	8000	2000	0.1
Diuron		0.042	>100	250	22 000	2000	0.1

Table 3
Recoveries and relative standard deviations (%) for bromacil and diuron in different water samples spiked at two levels ($n=5$)

	Bromacil		Diuron	
	1 μg/l	0.4 μg/l	0.5 μg/l	0.2 μg/l
Drinking water	107 (8)	88 (8)	86 (3)	96 (8)
Surface water	102 (6)	103 (8)	107 (3)	99 (7)

it with the proposed procedure developed with standard solutions. Therefore, there are several approaches to solve this problem, by avoiding sample handling in order to speed up analysis, different coupling conditions were established depending on water sample source, yielding a robust procedure, as after three months of daily use the clean-up and transfer volumes remained constant.

The response of bromacil and diuron was linear for standard solutions as well as for spiked drinking and surface water samples, with concentrations between 0.5 and 400 μg/l ($r>0.9996$, $n=6$). The described procedure (see Section 2) was validated by analysing various types of water samples spiked with bromacil and diuron. The recoveries at several levels are given in Table 3. The performance of the procedure is illustrated in Fig. 1 and Fig. 2, which

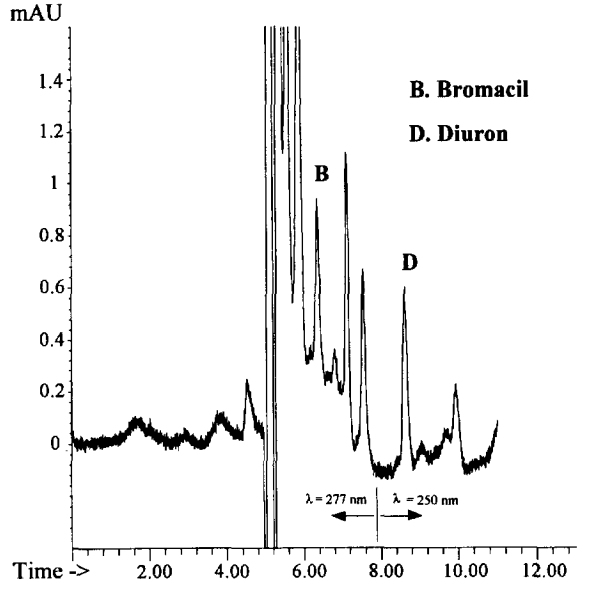


Fig. 1. LC–LC chromatogram of a drinking water sample spiked with bromacil (0.4 μg/l) and diuron (0.2 μg/l). Clean-up time 3.74 min, transfer time 2.86 min.

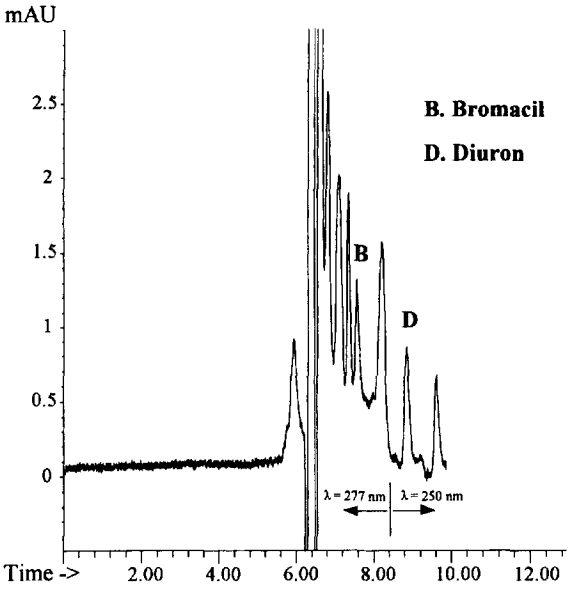


Fig. 2. LC–LC chromatogram of a surface water sample spiked with bromacil (0.4 μg/l) and diuron (0.2 μg/l). Clean-up time 5.00 min, transfer time 0.90 min.

show the LC–LC analysis of drinking and surface water samples spiked at 0.2–0.4 μg/l, respectively, obtaining limits of detection ($S/N=3$) for both bromacil and diuron below 0.1 μg/l, which meet the requirements of the EU Drinking Water Directive [24].

The developed coupled-column procedure with direct injection of the sample is capable of assaying bromacil and diuron down to a level of 0.1 μg/l in environmental water samples with a total analysis time of less than 10 min, yielding a high sample throughput of about 50 samples per day. Therefore, LVI–LC–LC is a useful technique for the fast trace analysis of polar herbicides in aqueous environmental samples. The procedure will be applied to real water samples; for such samples, additional confirmation of positive samples will be sought. The results will be published.

4. Conclusions

A sensitive and selective, coupled-column RPLC method was developed for the trace-level determination of bromacil and diuron in drinking and

surface water samples. The high molar extinction coefficients combined with the relatively high injection volume and C_{18} retention, provides sufficient sensitivity, while the application of small transfer volumes yields the required selectivity. The method has a high sample throughput, which makes it useful for screening purposes in environmental water samples at a level of 0.1 μg/l.

Acknowledgments

The scientific collaboration of Dr. E.A. Hogendoorn is greatly acknowledged.

References

[1] E.A. Hogendoorn, C. Verschraagen, P. van Zoonen and U.A.Th. Brinkman, Anal. Chim. Acta, 268 (1992) 205.
[2] E.A. Hogendoorn, A.P.J.M de Jong, P. van Zoonen and U.A.Th. Brinkman, J. Chromatogr., 511 (1990) 243.
[3] E.A. Hogendoorn, P. van Zoonen and U.A.Th. Brinkman, Chromatographia, 31 (1991) 285.
[4] J.V. Sancho, F.J. López, F. Hernández, E.A. Hogendoorn and P. van Zoonen, J. Chromatogr. A, 678 (1994) 59.
[5] J.V. Sancho, C. Hidalgo, F. Hernández, F.J. López, E.A. Hogendoorn and E. Dijkman, Int. J. Environ. Anal. Chem., 62 (1996) 53.
[6] E.A. Hogendoorn, C.E. Goewie and P. van Zoonen, Fresenius J. Anal. Chem., 339 (1991) 348.
[7] E.A. Hogendoorn and P. van Zoonen, J. Chromatogr. A, 703 (1995) 149.
[8] F. Hernández, R. Serrano, M.C. Miralles and N. Font, Chromatographia, 42 (1996) 151.
[9] T.D. Behymer, T.A. Bellar and W.L. Budde, Anal. Chem., 62 (1990) 1686.
[10] A. Di Corcia and M. Marchetti, J. Chromatogr., 541 (1991) 365.
[11] M.J. Incorvia Mattina, J. Chromatogr., 549 (1991) 237.
[12] H. Bagheri, E.R. Brouwer, R.T. Ghijsen and U.A.Th. Brinkman, Analusis, 20 (1992) 475.
[13] H. Bagheri, E.R. Brouwer, R.T. Ghijsen and U.A.Th. Brinkman, J. Chromatogr., 647 (1993) 121.
[14] A. Balinova, J. Chromatogr., 643 (1993) 203.
[15] A. Lagana, A. Marino, G. Fago and B.P. Martínez, Analusis, 22 (1994) 63.
[16] P. Parrilla, J.M. Martínez-Vidal, M. Martínez-Galera and A.G. Frenich, Fresenius J. Anal. Chem., 350 (1994) 633.
[17] R.M. Marcé, H. Prosen, C. Crespo, M. Calull, F. Borrull and U.A.Th. Brinkman, J. Chromatogr. A, 696 (1995) 63.
[18] C. Schlett, Fresenius J. Anal. Chem., 339 (1991) 344.
[19] J.M. Huen, R. Gillard, A.G. Mayer, B. Baltensperger and H. Kern, Fresenius J. Anal. Chem., 348 (1994) 606.
[20] J. Slobodnik, M.G.M. Groenewegen, E.R. Brouwer, H. Lingeman and U.A.Th. Brinkman, J. Chromatogr., 642 (1993) 359.
[21] W.A. Minnard, J. Slobodnik, J.J. Vreuls, K.P. Hupe and U.A.Th. Brinkman, J. Chromatogr. A, 696 (1995) 333.
[22] S. Sennert, D. Volmer, K. Levsen and Wünsch, Fresenius J. Anal. Chem., 351 (1995) 642.
[23] C.E. Goewie and E.A. Hogendoorn, J. Chromatogr., 410 (1987) 211.
[24] 80/778/EEC, Council directive of 15th July 1980 relating to the quality of water for human consumption, European Comission, Official Journal L229/11-29, August 30th 1980.