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Microbore liquid chromatographic–mass spectrometric determination of atrazine and its major hydroxylated degradate in water at parts-per-trillion levels using electrospray

Zongwei Cai[a,*], Ronald L. Cerny[a], Roy F. Spalding[b]

[a]*Mass Spectrometry Center, Department of Chemistry, University of Nebraska-Lincoln, Lincoln, NE 68588-0304, USA*
[b]*Water Science Laboratory, Water Center, University of Nebraska-Lincoln, Nebraska-Lincoln, NE 68588, USA*

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Abstract

A liquid chromatography–electrospray ionization mass spectrometry (LC–ESI–MS) method was developed to determine atrazine (ATR) and hydroxyatrazine (HA) in water at parts-per-trillion (ppt) levels. In addition to protonated molecular ions, sodiated molecular and dimer ions were observed in full-scan electrospray mass spectra of the analytes, providing additional information for the identification of ATR and HA. The quantification is based on the use of isotopically labeled internal standards that were added to water samples prior to the sample extraction using a carbon black cartridge. The relative response factor for each of the native compounds relative to its corresponding internal standard was measured and used to minimize the analytical error. The precision and accuracy data were obtained from the analysis of standard water samples containing 50 ppt ATR and 100 ppt HA. Relative standard deviation and relative error are less than 15% ($n=6$) for both ATR and HA determinations. The method has been successfully used to analyze agricultural run-off samples for ATR and HA levels. Analytical results compared well with those obtained by using GC–high-resolution and fast atom bombardment high-resolution MS methods developed in this laboratory. Preliminary results for analysis of a standard containing a mixture of ATR, HA, deethylatrazine and deisopropylatrazine suggest that the LC–ESI–MS method can be applied to simultaneous trace-level determination of the major products of atrazine degradation in the environment.

Keywords: Water analysis; Environmental analysis; Atrazine; Pesticides

1. Introduction

Atrazine (2-chloro-4-ethylamino-6-isopropylamino-*s*-triazine), a widely used broad leaf herbicide, is transported to environmental waters during and after its application onto farming land. The trace determination of atrazine in water samples involves enrichment procedures such as liquid–liquid extraction [1,2] and liquid–solid extraction [3–8] and instrumental analysis using gas chromatography (GC) [2,3], gas chromatography–mass spectrometry (GC–MS) [2,5,7,9–12] and high-performance liquid chromatography (HPLC) [8,13–17]. Hydroxyatrazine (2-ethylamino-4-isopropylamino-6-hydroxy-*s*-triazine), generally considered the major degradate of atrazine under a wide range of environmental conditions [18–21], has been detected in soil [20,21], agricultural run-off [8,17], surface water [8,22] and ground water [7,8,17]. Hydroxyatrazine (HA) and other hydroxylated degradation products of atrazine

*Corresponding author.

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can not be directly determined by GC and GC–MS techniques due to their high polarity [10,18,23,24]. A number of liquid chromatographic methods with UV detection have been proposed for the determination of HA [8,9,17,18,23,25,26]. However, these LC methods lack the required sensitivity for detecting HA at parts-per-trillion (ppt) levels. Moreover, a LC method with UV detection is not sufficiently selective for the determination of atrazine (ATR) and HA in complex matrices. The identification of the hydroxylated degradation products of ATR by HPLC using retention time measurement and fixed wavelength data is often difficult because many environmental samples contain complex mixtures of organic molecules so that complete chromatographic resolution of the analytes and other absorbing species is often impossible [17,18]. Mass spectrometry (MS) and tandem mass spectrometry (MS–MS) analyses, which provide information regarding molecular mass and characteristic fragmentation pathways, have been proved to be highly sensitive and highly selective for the trace determination of atrazine and its degradation products [6,7,12,27,28]. Recently, a very sensitive and selective MS method was developed to determine ATR and HA in water at low parts-per-trillion levels [7]. The method involves solid-phase extraction using a graphitized carbon black cartridge for extracting the chemicals and fast atom bombardment high-resolution mass spectrometry (FAB–HRMS) analysis of the sample extracts.

LC coupled to MS offers an excellent combination of the advantages of both techniques; on-line HPLC separation with the possibility of easy automation and the MS capacity for molecular mass and structure information on separated compounds. Coupling LC with MS is becoming a viable alternative for the analysis of small biological and environmental molecules with chemical properties that are not suitable for GC–MS. ATR and other pesticides have been analyzed by on-line LC–MS using interfaces such as particle beam [29], thermospray [30–35] and atmospheric pressure chemical ionization [36]. However, no data has been shown for the determination of the hydroxylated products of atrazine using LC–MS.

Electrospray ionization (ESI) has rapidly become a very promising interface for LC and MS. In addition to being used to obtain molecular mass information of very large molecules, LC–ESI–MS has recently been applied for the determination of relatively small molecules such as environmental polar pollutants [37,38], pesticides [39–41] and phospholipids [42]. Molina et al. [41] recently reported a LC–high flow pneumatically assisted ESI–MS method for the determination of several herbicides in estuarine waters with instrumental detection limits of 450 pg for ATR and its dealkylated degradates [41]. However, the method was not applied for the hydroxylated degradation products of ATR.

The purpose of this paper is to describe an LC–ESI–MS method for the determination of ATR and HA in environmental waters. The relatively simple LC–MS using a benchtop mass spectrometer can be an effective substitute for the FAB–HRMS method which uses expensive and complicated sector instrument.

2. Experimental

2.1. Chemicals and materials

Methanol and dichloromethane (HPLC-grade) were purchased from Fisher Scientific (Pittsburg, PA, USA). Other chemicals were of reagent grade and were used as received. Purified water was obtained by passing distilled water through a Fisher Barnstead 4-Module Nanopure cartridge system. The graphitized carbon black cartridge (3 ml/250 mg) was purchased from Supelco (Bellefonte, PA, USA).

Native atrazine standard was purchased from AccuStandard (New Haven, CT, USA). The ring-labeled [$^{13}C_3$]-atrazine was synthesized by Merck Frost (Canada). Synthesized native HA (>99% purity) was obtained from Dr. James Carr of the Department of Chemistry, University of Nebraska-Lincoln. Ring-labeled [$^{13}C_3$]-HA was synthesized via hydroxylation of [$^{13}C_3$]-ATR [7]. Both [$^{13}C_3$]-ATR and [$^{13}C_3$]-HA were used as internal standards and were added to the water samples prior to sample preparation.

Stock standard solutions were prepared by dissolving 2 mg of chemical in 10 ml of methanol. The

stock solutions were further diluted with purified water to obtain calibration and spiking standard solutions.

2.2. Water samples

Standard water samples were prepared by fortifying purified water with native standard solutions of ATR and HA to obtain levels of 50 and 100 ppt. Environmental water samples were collected from the Beaver Creek near York (NE, USA) after the first run-off event on June 24, 1994. Details of sample collection were described previously [17]. Some water samples collected after the run-off event have been analyzed using a HPLC method with UV detection [17]. However, the samples analyzed for the LC–MS method development were collected at times different from those water samples analyzed previously.

After addition of the [$^{13}C_3$]-labeled internal standards, 100 ml of the water sample were extracted by the liquid–solid extraction procedure using the carbon black cartridge [7]. The analytes were adsorbed to the graphitized carbon black cartridge and eluted with dichloromethane–methanol (80:20, v/v). The sample extract was concentrated to 5 μl under a nitrogen stream. Typical sample volume was 5 μl for the LC–ESI–MS and 1 μl each for GC–HRMS and FAB–HRMS analyses.

2.3. Liquid chromatography–electrospray mass spectrometry

LC analysis was carried out with a Gilson HPLC System (Middleton, WI, USA). A C_8 reversed-phase microbore column (Nova-Pak 4 μm, 50×0.1 mm, Microtech. Sci., Sunnyvale, CA, USA) and eluents of a mixture of methanol and water were used for the LC separation. The initial composition was 45% methanol, programmed linearly to 65% after 2 min. The flow-rate of the mobile phase was 50 μl/min. The entire column eluent was introduced to the electrospray source through a 50-cm length of a fused-silica capillary tube (75 μm I.D.). A Fisons VG Platform benchtop mass spectrometer (Fisons, Manchester, UK) equipped with an electrospray LC–MS interface, a single quadrupole mass analyzer and VG Masslynx data software was used for the determination of ATR and HA. The mass spectrometer was operated in positive mode by applying a voltage of 3.5 kV to the capillary. The skimmer cone voltage was set at 50 V and the source temperature was maintained at 65°C. Nitrogen gas was used as the drying and nebulizing gas. Typical flow-rates of the drying and nebulizing gas were set at 250 and 15 l/h, respectively. Under the above LC–MS conditions, the retention times of HA and ATR were 1.5 and 4.3 min, respectively.

ESI–MS spectra collected in full-scan mode were obtained by direct flow injection and by scanning over the mass range of m/z 100 to 500 at 6 s/scan. Selected-ion monitoring mode with a scan rate of 0.1 s/scan was used for the quantification. Protonated molecular ions of the analytes and the $^{13}C_3$-labeled internal standards were selected as quantification ions. The mass chromatograms of the ions at m/z 216 for ATR, m/z 219 for [$^{13}C_3$]-ATR, m/z 198 for HA and m/z 201 for [$^{13}C_3$]-HA were acquired for the water sample extracts.

Because the LC–MS instrument is intensively used for the analysis of proteins and peptides, the interference background is often intensive. It was necessary to clean the counter-electrode and the sampling cone before the herbicide analysis. Typically, a formic acid–water mixture (1:1, v/v), clean water, acetone and acetonitrile were used in a cleaning sequence.

2.4. GC–HRMS and FAB–HRMS analyses

The GC–HRMS analysis of ATR was performed on a Carlo-Erba GC/Kratos MS-50 double-focusing mass spectrometer system. The details of the GC–HRMS method were described elsewhere [6].

FAB–HRMS was used to determine HA at trace levels [7]. The quantitative determination was conducted on a Fison/VG Autospec sector mass spectrometer with mass resolution of 10 000 (10% valley definition). Acceleration voltage scanning over a small mass range of m/z 197.5 to 201.5 was conducted. The peak intensities of the protonated molec-

ular ions of HA and [$^{13}C_3$]-HA were used for the quantitative analysis.

3. Results and discussion

3.1. Full-scan electrospray mass spectra

The positive electrospray mass spectra demonstrated in Fig. 1a–d were obtained by directly injecting 10 ng each of ATR and HA as well as of the corresponding $^{13}C_3$-labeled internal standards into the ESI source. The spectra show no significant cross interference for the selected quantification ions, the protonated molecular ions. In addition to the protonated molecular ions, sodiated molecular and dimer ions were also observed with the MS conditions used. This additional peak information was used to confirm the identification of ATR and HA, especially when a complicated water sample extract was analyzed. Several impurity peaks were observed in the spectra of [$^{13}C_3$]-ATR and [$^{13}C_3$]-HA (Fig. 1b and Fig. 1d).

3.2. LC–ESI–MS quantification

Quantitative determination was based on the use of an isotopically labeled internal standard method on LC–ESI–MS. The quantification was performed by comparing peak areas of the native analytes and the spiked $^{13}C_3$-labeled standards. Data were acquired in the selected-ion monitoring (SIM) mode for each selected ion. Typical SIM chromatograms for the HPLC separation of ATR and HA are demonstrated in Fig. 2. The injection amounts were 200 pg of ATR and 500 pg of [$^{13}C_3$]-ATR as well as 500 pg of HA and 200 pg of [$^{13}C_3$]-HA. The results indicate that the LC–MS method provides for a high throughput of samples as less than 6 min is needed per LC–MS analysis.

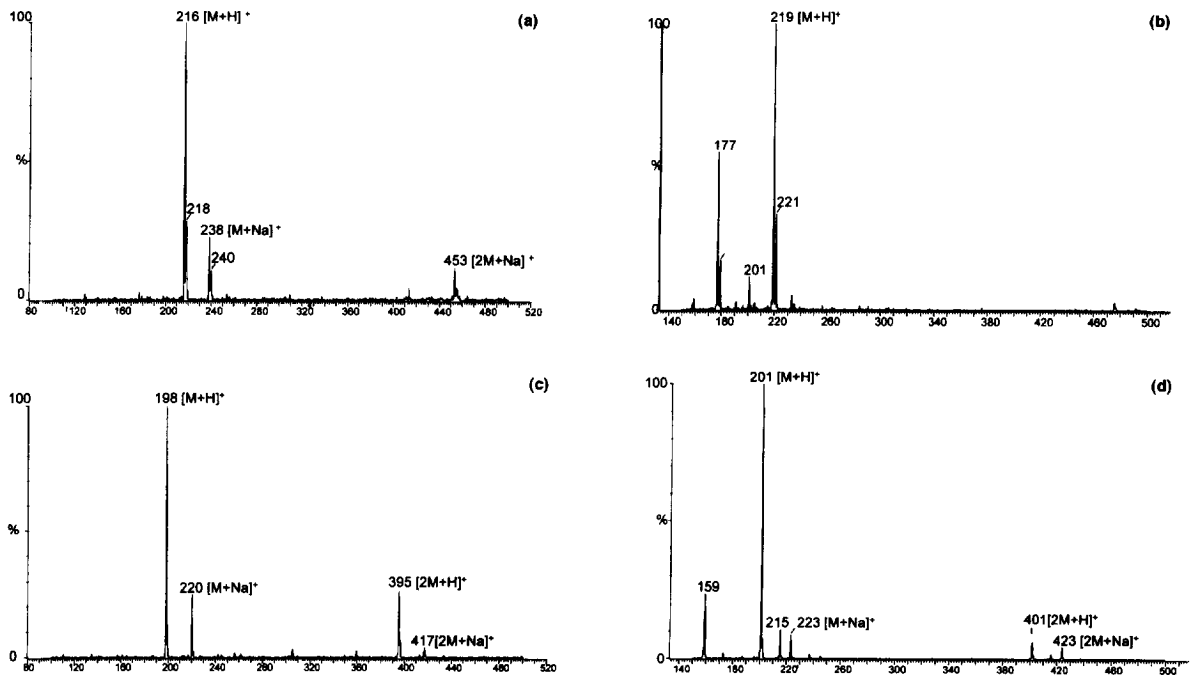

Fig. 1. Full-scan ESI–MS spectra of ATR (a), [$^{13}C_3$]-ATR (b), HA (c), and [$^{13}C_3$]-HA (d). The spectra were obtained by analyzing 10 ng of each standard using direct flow injection.

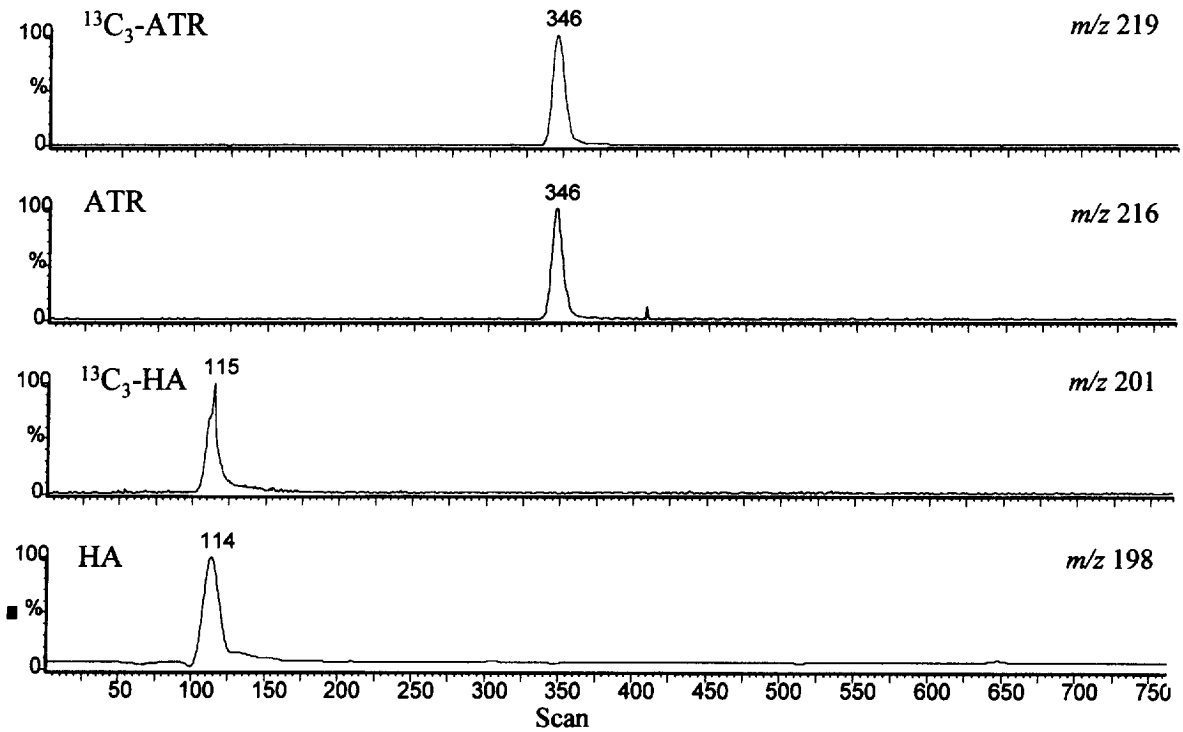

Fig. 2. Selected-ion monitoring chromatograms obtained from the LC–ESI–MS analysis of 500 pg of [$^{13}C_3$]-ATR; 200 pg of ATR; 200 pg of [$^{13}C_3$]-HA and 500 pg of HA. A C_8 reversed-phase microbore column (50×0.1 mm, 4 μm) was used for the separation. The initial composition of the mobile phase was 45% methanol. The methanol was programmed to 65% after 2 min. The flow-rate of the mobile phase was 50 μl/min.

3.3. Response curves and detection limits

Calibration standards were analyzed to evaluate linearity of the determination range. For the calibration data points, the injected amounts of [$^{13}C_3$]-ATR and [$^{13}C_3$]-HA were 500 pg while the amounts of ATR and HA varied from 50 to 3000 pg and from 75 to 5000 pg, respectively. A response curve was obtained by plotting the injected amount of analyte versus the intensity ratio of the analyte signal to that of the corresponding internal standard. The response curves show that the method gives a linear dynamic range over two orders of magnitude for the LC–MS determination of both ATR and HA. The correlation coefficient for the ATR curve is 0.9987 (n=6), with a slope of 0.0021±0.00012 and an intercept of 0.071±0.004. The correlation coefficient for the HA curve is 0.9975 (n=6), with a slope of 0.0024±0.00017 and an intercept of 0.23±0.016.

The typical instrument detection limits at a signal-to-noise (S/N) ratio of 3 are 10 and 30 pg for ATR and HA, respectively, with a sample injection volume of 5 μl. To analyze a water sample extract, a large volume of sample may be injected to achieve lower procedural detection limits. Injection of more than 30 μl, however, resulted in tailing of the HPLC peak of HA under the LC conditions.

Detection limits for the entire analytical procedure depend upon the quality of sample extraction. Procedural detection limits at low ppt levels were achieved for ATR and HA in the standard water samples. A previous study on the solid-phase extraction using the carbon black cartridge has shown that ATR and HA were extracted quantitatively from

water [7]. Moreover, because the isotopically labeled internal standards were added to the water samples prior to sample preparation, recovery of the analytes in the sample extraction procedure should not affect method accuracy. The detection limits can, however, be improved when good recovery is achieved.

3.4. Relative response factors of ATR–[$^{13}C_3$]-ATR and HA–[$^{13}C_3$]-HA

The relative response factor (RRF), i.e. the ratio of the intensity of the signal of the native analyte to that of the corresponding internal standard, was determined for ATR and HA by analyzing the calibration standards. The RRFs obtained from the analysis of each standard are presented in Table 1. The average RRFs of 1.07±0.06 and 1.29±0.09 were used for the determinations of ATR and HA in water samples to minimize the analytical error. The deviation of RRFs from one may be due to the different purities of native and $^{13}C_3$-labeled standards and to errors introduced in the preparation of the calibration standard solutions and in the LC–MS analysis. Impurities existing in [$^{13}C_3$]-ATR and [$^{13}C_3$]-HA standards were observed by full-scan ESI–MS analysis (see Section 3.1) and other mass spectrometric techniques [6,7]. The RRFs were checked periodically throughout the method development and sample analysis stages; the RRFs did not vary by more than 15% for either ATR or HA.

Table 2
Accuracy and precision data from analysis of standard water samples

ATR		HA	
Spiked (ppt)	Found (ppt)	Spiked (ppt)	Found (ppt)
50	46	100	112
50	54	100	104
50	51	100	97
50	48	100	115
50	46	100	102
50	44	100	118
Av.±SD[a]	48±3.7	–	108±8.2
R.S.D.[b] (%) (n=6)[c]	7.4	–	8.2

[a] Average result±standard deviation.
[b] Relative standard deviation.
[c] Number of determinations.

3.5. Method accuracy and precision

Six standard samples containing 50 ppt ATR and 100 ppt HA were analyzed using LC–ESI–MS after the solid-phase extraction. Accuracy and precision data are presented in Table 2. The average error for both ATR and HA determinations is less than 15%, indicating that the method achieved an accuracy of

Table 1
Relative response factor of ATR–[$^{13}C_3$]-ATR and HA–[$^{13}C_3$]-HA

ATR		HA	
Injected amount[a] (pg)	RRF[b]	Injected amount (pg)	RRF
50	1.14	75	1.20
250	1.04	250	1.35
500	1.12	500	1.38
1000	1.10	1000	1.31
2000	0.99	2500	1.34
3000	1.03	5000	1.17
Av.±SD[c]	1.07±0.06	–	1.29±0.09
R.S.D.[d] (%) (n=6)[e]	5.5	–	7.0

[a] Injected amount of [$^{13}C_3$]-ATR and [$^{13}C_3$]-HA was kept as 500 pg for each calibration point.
[b] Relative response factor.
[c] Average RRF±standard deviation.
[d] Relative standard deviation.
[e] Number of determinations.

greater than 85%. The relative standard deviations (R.S.D.) are 7.3 and 8.2 ($n=6$) for the determination of ATR and HA, respectively.

3.6. Analysis of agricultural run-off water

Five surface water samples collected after the run-off event on June 24, 1994 were analyzed. After the sample extraction, water sample extracts were analyzed for the levels of ATR and HA using LC–ESI–MS, GC–HRMS [6] and FAB–HRMS [7]. The spiked levels of the internal standards were 5.0 ppb [$^{13}C_3$]-ATR and 1.0 ppb [$^{13}C_3$]-HA for all unknown samples as well as for the control samples. The analytical results are presented in Table 3. The data indicate that the concentration levels of ATR and HA in the water samples vary slightly from sample to sample. This is because the water samples were collected at different times during the run-off event and thus, under different environmental conditions. The data show that the levels of HA are approximately five times lower than those of ATR in the surface water samples. A comparison of LC–ESI–MS with GC–HRMS and FAB–HRMS analyses, showed that the results varied by less than 15%. For quality assurance and quality control, samples of blank and duplicate were analyzed. The results of the blank and duplicate determinations show that no precontamination occurred and that there was good precision.

The LC–ESI–MS method was also applied to the determination of major dealkylated degradation products of atrazine such as deethylatrazine (2-amino-4-chloro-6-isopropylamino-*s*-triazine, DEA) and deisopropylatrazine (2-amino-4-chloro-6-ethylamino-*s*-triazine, DIA). A typical HPLC separation of ATR and its major dealkylated and hydroxylated degradation products under current LC and MS conditions is illustrated in Fig. 3. The total ion current chromatogram for selected ions was obtained by analyzing 100 pg each of HA, DIA, DEA and ATR. The results demonstrate the feasibility of simultaneous determination of ATR and its degradation products by using LC–ESI–MS. The application of the method for simultaneous analysis of the chemicals in environmental waters and soils, however, will depend on the cleanliness of the sample extract.

4. Conclusion

The method using solid-phase extraction with a carbon black cartridge and LC–ESI–MS is satisfactory for the trace analysis of atrazine and hydroxyatrazine in water. Quantification by using corresponding $^{13}C_3$-labeled compounds as internal standards was accurate and precise at parts-per-trillion to parts-per-billion levels. The method was successfully applied to analyzed surface water samples for atrazine and its major hydroxylated degradation product. The analytical procedure can be expanded to other degradates of atrazine as well as to other related herbicides present in environmental waters.

Table 3
Analytical results for ATR and HA in the agricultural run-off water samples

Sample ID[a]	ATR (ppb)		HA (ppb)	
	LC–ESI–MS	GC–HRMS	LC–ESI–MS	FAB–HRMS
94-2667	13.2	14.4	3.0	2.8
94-2667 dup.[b]	12.4	12.1	2.7	2.6
94-2671	12.7	13.5	2.5	2.7
94-2673	11.8	13.1	1.9	2.1
94-2674	12.6	11.7	2.3	2.5
94-2679	10.9	11.3	2.3	2.2
blank	ND[c]	ND	ND	ND

[a] Sample identification name.
[b] Duplicated sample.
[c] Not detected.

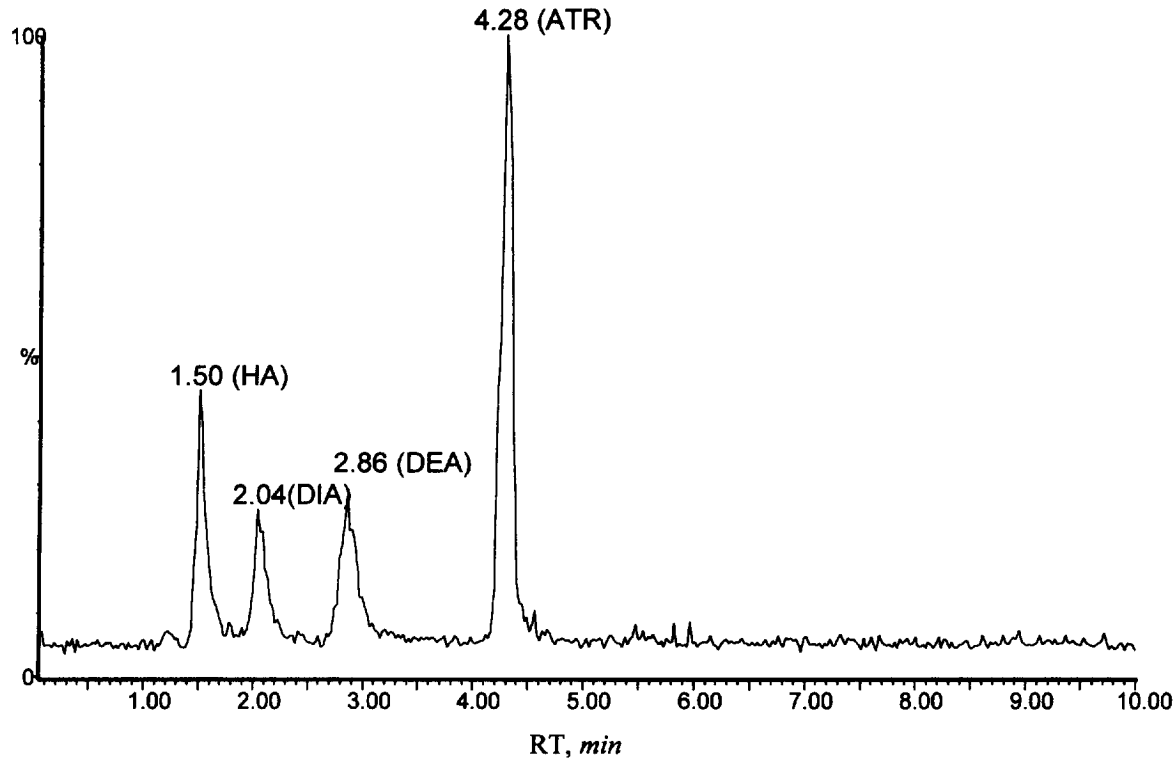

Fig. 3. LC–ESI–MS–SIM chromatogram of 100 pg of HA, DIA, DEA and ATR in purified water. The LC conditions were the same as those stated in Fig. 2.

References

[1] H.B. Lee and Y.D. Stokker, J. Assoc. Off. Anal. Chem., 69 (1986) 568.
[2] V. Lopez-Avila, P. Hirata, S. Kraska, M. Flanagan, J.H. Taylor and S.C. Hern, Anal. Chem., 57 (1985) 2797.
[3] G.A. Junk and J.J. Richard, Anal. Chem., 60 (1988) 451.
[4] J. Sherma, J. Liq. Chromatogr., 9 (1986) 3433.
[5] A. Di Corcia, M. Marchetti and R. Samperi, J. Chromatogr., 405 (1987) 357.
[6] Z. Cai, R.V.M. Sadagopa, D.E. Giblin and M.L. Gross, Anal. Chem., 65 (1993) 21.
[7] Z. Cai, R.V.M. Sadagopa, M.L. Gross, S.J. Monson, D.A. Cassada and R.F. Spalding, Anal. Chem., 66 (1994) 4202.
[8] M. Berg, S.R. Mueller and R.P. Schwarzenbach, Anal. Chem., 67 (1995) 1860.
[9] G. Durand and D. Barcelo, J. Chromatogr., 502 (1990) 275.
[10] E.M. Thurman, M. Meyer, M. Pomes, C.A. Perry and A.P. Schwab, Anal. Chem., 62 (1990) 2043.
[11] A. Kraut-Vass and J. Thoma, J. Chromatogr., 538 (1991) 233.
[12] D.A. Cassada, R.F. Spalding, Z. Cai and M.L. Gross, Anal. Chim. Acta, 287 (1994) 7.
[13] D.S. Owens and P.E. Sturrock, Anal. Chim. Acta, 188 (1986) 269.
[14] R. Reupert and E. Ploger, Fresenius' J. Anal. Chem., 331 (1988) 503.
[15] A. Di Corcia and M. Marchetti, Anal. Chem., 63 (1991) 580.
[16] M. Batista, A. Di Corcia and M. Marcetti, Anal. Chem.. 61 (1989) 935.
[17] Z. Cai, S.J. Monson and R.F. Spalding, J. Assoc. Off. Anal. Chem., 19 (1996) 929.
[18] R.N. Lerch and W.W. Donald, J. Agric. Food Chem., 42 (1994) 922.
[19] H.D. Skipper, G.M. Gilmour and G.R. Furtick, Soil Sci. Am. Proc., 31 (1967) 653.
[20] T.H. Dao, T.L. Lavy and R.C. Sorensen, Soil Sci. Soc. Am. J., 43 (1979) 1129.
[21] A.M. Rustum, S. Ash, A. Saxena and K. Balu, J. Chromatogr., 514 (1990) 209.
[22] C.D. Adams and S.T. Randtke, Environ. Sci. Technol.. 26 (1992) 2218.
[23] D.C.G. Muir and Baker, J. Agric. Food Chem., 26 (1978) 420.
[24] J.E. Bakke, J.D. Larson and C.E. Price, J. Agric. Food Chem., 20 (1972) 602.
[25] N.M.J. Vermeulen, Z. Apostolides and D.J.J Potgieter, J. Chromatogr., 240 (1982) 247.
[26] T.R. Steinheimer and M.G. Ondrus, USGS/Water Resources Investigation Report 89-4193, 1990, p. 27.

[27] K.A. Caldwell, R.V.M. Sadagopa, Z. Cai and M.L. Gross, Anal. Chem., 65 (1993) 2372.
[28] Z. Cai, R.F. Spalding and M.L. Gross, Anal. Chim. Acta, 304 (1995) 67.
[29] A. Cappiello, G. Famiglini and F. Bruner, Anal. Chem., 66 (1994) 1416.
[30] R.J. Vreeken, W.D. van Dongen, R.T. Ghijsen and U.A.Th. Brinkman, Int. J. Environ. Anal. Chem., 54 (1994) 119.
[31] T.A. Beller and W.L. Budde, Anal. Chem., 60 (1988) 2076.
[32] D. Volmer and K. Levsen, J. Am. Soc. Mass Spectrom., 5 (1994) 655.
[33] S. Chiron, S. Dupas, P. Scribe and D. Barcelo, J. Chromatogr. A, 665 (1994) 295.
[34] H. Bagheri, E.R. Brouwer, R.T. Ghijsen and U.A.Th. Brinkman, J. Chromatogr., 647 (1993) 121.
[35] D. Volmer, K. Levsen and G. Wunsch, J. Chromatogr. A, 660 (1994) 231.
[36] D.R. Doerge and S. Bajic, Rapid Commun. Mass Spectrom., 6 (1992) 663.
[37] H. Lin and R.D. Voyksner, Anal. Chem., 65 (1993) 451.
[38] C. Crescenzi, A. Di Corcia and R. Samperi, Anal. Chem., 67 (1995) 1797.
[39] G. Hopfgartner, T. Wachs, K. Bean and J. Henion, Anal. Chem., 65 (1993) 439.
[40] C. Crescenzi, A. Di Corcia, S. Marchese and R. Samperi, Anal. Chem., 67 (1995) 1986.
[41] C. Molina, G. Durand and D. Rarcelo, J. Chromatogr. A, 712 (1995) 113.
[42] H.Y. Kim, T.L. Wang and Y.C. Ma, Anal. Chem., 66 (1994) 3977.