Electrospray Liquid Chromatography Quadrupole Ion Trap Mass Spectrometry Determination of Phenyl Urea Herbicides in Water

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Phenyl urea herbicides were determined in water by electrospray quadrupole ion trap liquid chromatography−mass spectrometry (ES-QIT-LC-MS). Over a wide concentration range $[M - H]^-$ and MH^+ ions were prominent in ES spectra. At high concentrations dimer and trimer ions appeared, and sodium, potassium, and ammonium adducts also were observed. In the case of isopturon, source collision-induced dissociation (CID) fragmentation with low offset voltages increased the ion current associated with MH^+ and diminished dimer and trimer ion abundance. In the mass analyzer CID involved common pathways, for example, daughter ions of $[M - H]^-$ resulted from loss of R_2NH in *N*′,*N*′-dialkyl ureas or loss of $C_3H_5NO_2$ (87 amu) in *N*′-methoxy ureas. A 2 mm (i.d.) × 15 cm C_{18} reversed phase column was used for LC-MS with a linear methanol/water gradient and 0.5 mL/min flow rate. Between 1 and 100 pg/μL the response was highly linear with instrument detection limits ranging from <10 to 50 pg injected. Whereas the positive ES signal intensity was greater for each of the compounds except fluometuron, negative ion monitoring gave the highest signal-to-noise ratio. Analysis of spiked Colorado River water, a source high in total dissolved solids and total organic carbon, demonstrated that ES-QIT-LC-MS was routinely capable of quantitative analysis at low nanogram per liter concentrations in conjunction with a published C_{18} SPE method. Under these conditions experimental method detection limits were between 8.0 and 36 ng/L, and accuracy for measurements in the 20−50 parts per trillion range was from 77 to 96%. Recoveries were slightly lower in surface water (e.g., 39−76%), possibly due to suppression of ionization.

Keywords: *Electrospray; phenyl urea; herbicides; water*

INTRODUCTION

Phenyl urea herbicides are subject to decomposition in gas chromatograph inlets, and typically any peak detected corresponds to the aniline breakdown product. For this reason high-performance liquid chromatography (HPLC) is widely used in phenyl urea analysis often with diode array detectors (*1*−*5*). HPLC coupled to a photolysis cell in series with a fluoresence detector (*6*), postcolumn derivatization systems (*5*), or mass spectrometers (*7*) also have been used for these compounds.

Liquid chromatography−mass spectrometry (LC-MS) is appealing because of the need for specificity in environmental analysis. Phenyl ureas have been determined using various LC-MS techniques including particle beam (PB) MS (*1*, *8*, *9*), thermospray (TSP) MS (*10*−*12*), and flow fast atom bombardment (FAB) (*13*).

Atmospheric pressure ionization (API) mass spectrometry offers improved sensitivity and other practical advantages, such as operational ruggedness when compared with the previously mentioned LC-MS interfaces (*14*, *15*). API instruments are becoming widely available, particularly in pharmaceutical and biotechnology laboratories. Determination of the phenyl ureas using the two most popular API techniques, electrospray (ES) (*2*, *3*, *16*, *17*) and atmospheric pressure chemical ionization (APCI) (*18*−*21*), have been described.

The objective of this research was to evaluate the practical utility of ES-quadrupole ion trap (QIT)-LC-MS in the quantitative analysis of phenyl ureas in water. Basic aspects were investigated including spectroscopic features in positive and negative ion modes and characteristics of collision-induced dissociation (CID), both inside and outside the mass analyzer. Quantitative aspects, such as linear dynamic range and instrument detection limits, also were studied. This information was incorporated in a multiresidue LC-MS procedure that was further validated by method detection limit (MDL) determination and analysis of spiked Colorado River water.

MATERIALS AND METHODS

Chemicals. Phenyl urea standards were obtained from the U.S. Environmental Protection Agency (Research Triangle Park, NC) as neat solids with purity of ≥99% and were used as received. Methanol used in chromatographic mobile phases and in the preparation of standards was of gas chromatography or residue analysis grade and was purchased from EM Science (Merck, Darmstadt, Germany). Water used in mobile phases was obtained from a Nanopure Infinity UV water purifier (Barnstead/Thermolyne, Dubuque, IA) that polished distilled feedwater with ion exchange and charcoal resins and UV light. Primary standards (milligrams per milliliter) were prepared in methanol/water (9:1, v/v) containing 0.1 mM ammonium acetate, and working standards were prepared by serial dilution in methanol.

Instrumentation. The mass spectrometer was a Finnigan LCQ-Deca (Finnigan, San Jose, CA) fitted with an electrospray probe and fused silica-lined ES needle. The MS was interfaced to an HP 1100 HPLC (Agilent, Wilmington, DE) equipped with an autosampler, solvent degasser, binary pump, and thermo-

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Table 1. Positive Ions Detected at High Phenyl Urea Concentrations[a]

	ion intensity (% of base peak)					
compound	MH^+	MNH_4^+	MNa^+	$[2M + H]^+$	$[2M + Na]^+$	$[3M + Na]^+$
N′,*N′*-Dimethyl Ureas						
chlortoluron	8.1	ND[b]	ND	26	100	6.9
diuron	23	ND	13	37	100	5.0
fluometuron	7.6	ND	8.0	39	100	ND
isopturon	4.0	ND	3.8	12	100	3.0
metoxuron	7.1	ND	4.8	19	100	4.8
monuron	7.6	ND	9.2	16	100	3.0
N′,*N′*-Dialkyl Ureas						
neburon	7.0	ND	7.8	16	100	ND
N′-Methoxy-*N′*-methyl Ureas						
chlorbromuron	100	20	ND	21	21	ND
linuron	100	18	7	60	60	ND
metobromuron	100	5	22	30	76	ND
monolinuron	53	7	16	20	100	4.0
N′-Alkyl Ureas						
siduron	7.5	ND	3.0	36	100	17

[a] Samples were loop injected into the 0.5 mL/min methanol/water (4:1, v/v) mobile phase. [b] Not detected.

stated column compartment. The entire system was controlled by Finnigan Xcalibur software (version 1.0, SR 1), which was also used for data acquisition and processing.

Operating Conditions. Compounds were introduced to the ESI needle by syringe pump at microliter per minute flow rates or via the 0.5 mL/min mobile phase. In some cases compounds were introduced postcolumn via a 10 μL loop injector or from a syringe pump connected via a T-fitting. For routine LC-MS operation, the chromatographic eluent was conducted directly to the ESI needle.

For sample infusion via syringe pump, the mass spectrometer was tuned with a three-component tuning mixture containing caffeine, a tripeptide, and a synthetic polymer mixture. For operation with the mobile phase, a milligram per milliliter metobromuron tuning solution was used—the herbicide was dissolved in methanol/water (9:1, v/v) with 0.1 mM ammonium acetate. Metobromuron was introduced at 1 μL/min via a T-fitting into 0.45 mL/min 80% methanol/20% water. The capillary temperature was 350 °C, and the nitrogen sheath and auxiliary gas flows were 80 and 20 units, respectively, typical for this mobile phase flow rate. The instrument was tuned on *m/z* 257 ($[M - H]^-$) or *m/z* 259 (MH^+) ions for negative and positive ion modes, respectively. All voltages and offsets were determined by autotune software.

LC-MS Operation. A 2.1 mm × 15 cm Supelcosil LC-18 column (Supelco, Bellefonte, PA) with 3 μm packing was used exclusively. The HPLC separation was a 14 min methanol/water gradient from 30 to 80% methanol with a consistent 0.5 mL/min flow rate. The spray voltage was 5 kV. The mass spectrometer acquired for 18 min in centroid mass scanning mode from 175 to 300 amu in both positive and negative ion modes after a 0.5 min start delay. The mass analyzer was operated with automatic gain control with three total microscans and a 50 ms maximum injection time. The electron multiplier voltage was 880 V.

RESULTS AND DISCUSSION

ES MS of Phenyl Ureas. *Positive Ions.* Phenyl urea ES spectra were concentration-dependent, particularly positive ion spectra. At high concentrations, where the response was beginning to saturate, intense dimer species were observed, for example, $[2M + H]^+$ and $[2M + Na]^+$. Trimeric species such as $[3M + H]^+$ were also present. At these high concentrations, especially for the *N′*,*N′*-dimethyl ureas, dimer ions were more intense than the MH^+ and MNa^+ pseudomolecular ions. Spectral data are summarized in Table 1. The strong tendency for *N′*,*N′*-dimethyl ureas to form dimers relative to the *N′*-methoxy-*N′*-methyl analogues suggests that polarity (e.g., octanol−water partition coefficient) may influence aggregation in electrospray ionization.

Table 2. Negative Ions Detected at High Phenyl Urea Concentrations

	ion intensity (% of base peak)		
compound	$[M - H]^-$	$[M + acetate]^-$	$[2M - H]^-$
N′,*N′*-Dimethyl Ureas			
chlortoluron	100	56	5.0
diuron	100	15	6.2
fluometuron	100	28	13
isopturon	100	15	9.0
metoxuron	100	17	ND
monuron	61	22	100
N′,*N′*-Dialkyl Ureas			
neburon	100	7.0	ND
N′-Methoxy-*N′*-methyl Ureas			
chlorbromuron	100	ND	ND
linuron	100	ND	ND
metobromuron	100	10	ND
monolinuron	100	16	ND
N′-Alkyl Ureas			
siduron	53	100	56

Over a wide concentration range where the instrument response was dynamic and pertinent to trace analysis, MH^+ ions predominated. MNa^+ ions were less prevalent and occurred at lower intensity. In 0.1 mM ammonium acetate solution, ammonium adduct ions were formed, but only by the *N′*-methoxy-*N′*-methyl ureas.

Negative Ions. In negative ion mode, even at high concentrations, the $[M - H]^-$ ions always were prominent. In the presence of 0.1 mM acetate $[M + CH_3COO]^-$ adducts were observed and minor $[2M - H]^-$ dimers were present in several cases (Table 2). The *N′*-methoxy-substituted ureas again were less inclined to form dimer ions (and acetate adducts) than the *N′*,*N′*-dimethyl ureas. In both negative and positive ion modes, fragment ions were largely absent. However, the low end of the scan window was 150 amu. The lack of fragmentation is typical of electrospray, which is among the "softest" mass spectrometry ionization methods. In summary, the negative ion spectra were less complex than the corresponding positive ion ES spectra.

Source Fragmentation. Source fragmentation was investigated as a means to reduce the intensity of the dimer and trimer ions and to increase the ion current carried by MH^+. In source fragmentation an offset voltage is applied in the region external to the mass analyzer. In this region there is sufficient pressure for accelerated ions to collide with neutrals causing fragmentation. Two phenyl ureas were selected for study, isopturon, an *N′*,*N′*-dimethyl urea with a strong tendency to form dimers and trimers, and chlorbromuron, an *N′*-methoxy urea that did not.

For chlorbromuron very small offset voltages (e.g., 5−10 V) induced fragmentation of $[2M + H]^+$ and $[2M + Na]^+$ dimers, but the intensity of the MH^+ ion (*m/z* 293) did not increase at any applied potential. At offset voltages of >10 V smaller fragments (e.g., *m/z* 182 and 204) appeared at the expense of MH^+ (Figure 1).

Higher voltages (e.g., >10 V) were needed to fragment the *m/z* 435 isopturon dimer ($[2M + Na]^+$). This ion was diminished by 20% at 5 V, by 60% at 10 V, and by 90% at 15 V offsets. Application of 20 V produced a spectrum with MNa^+ (*m/z* 229) as the base peak. Thus, source fragmentation achieved the objective of enhancing two

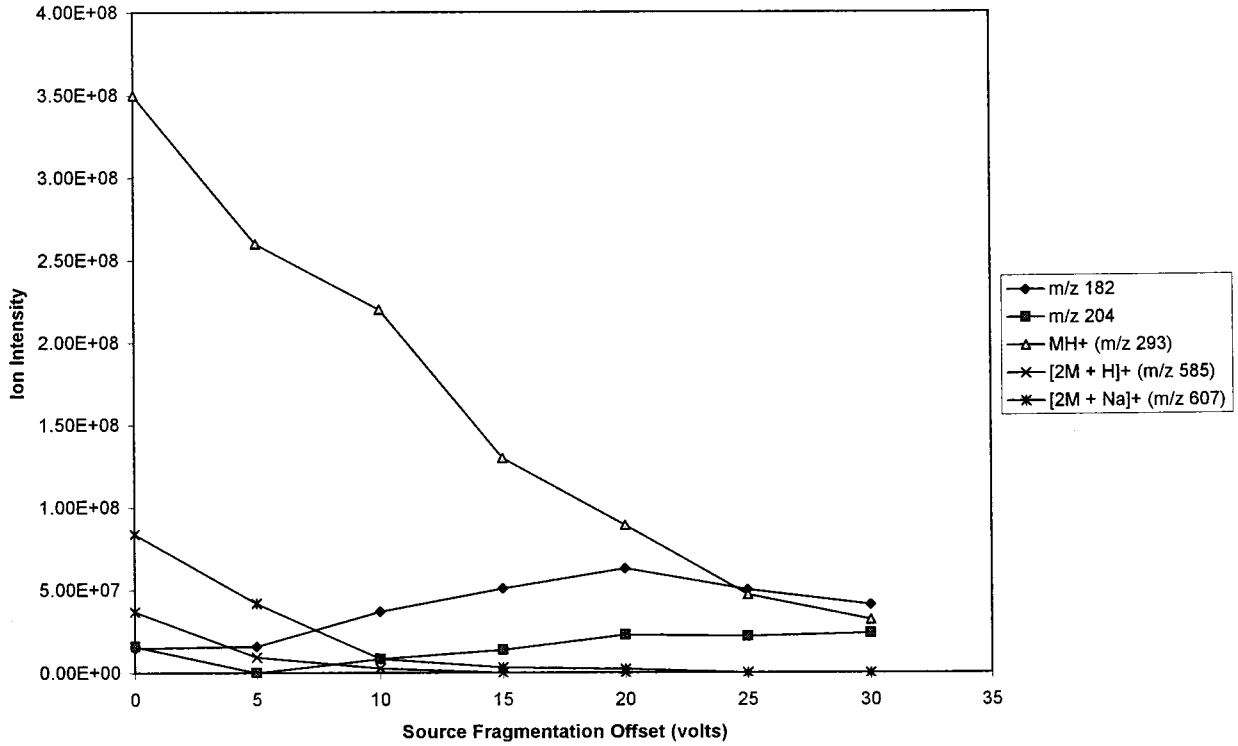

Figure 1. Source CID of chlorbromuron protonated molecule ion.

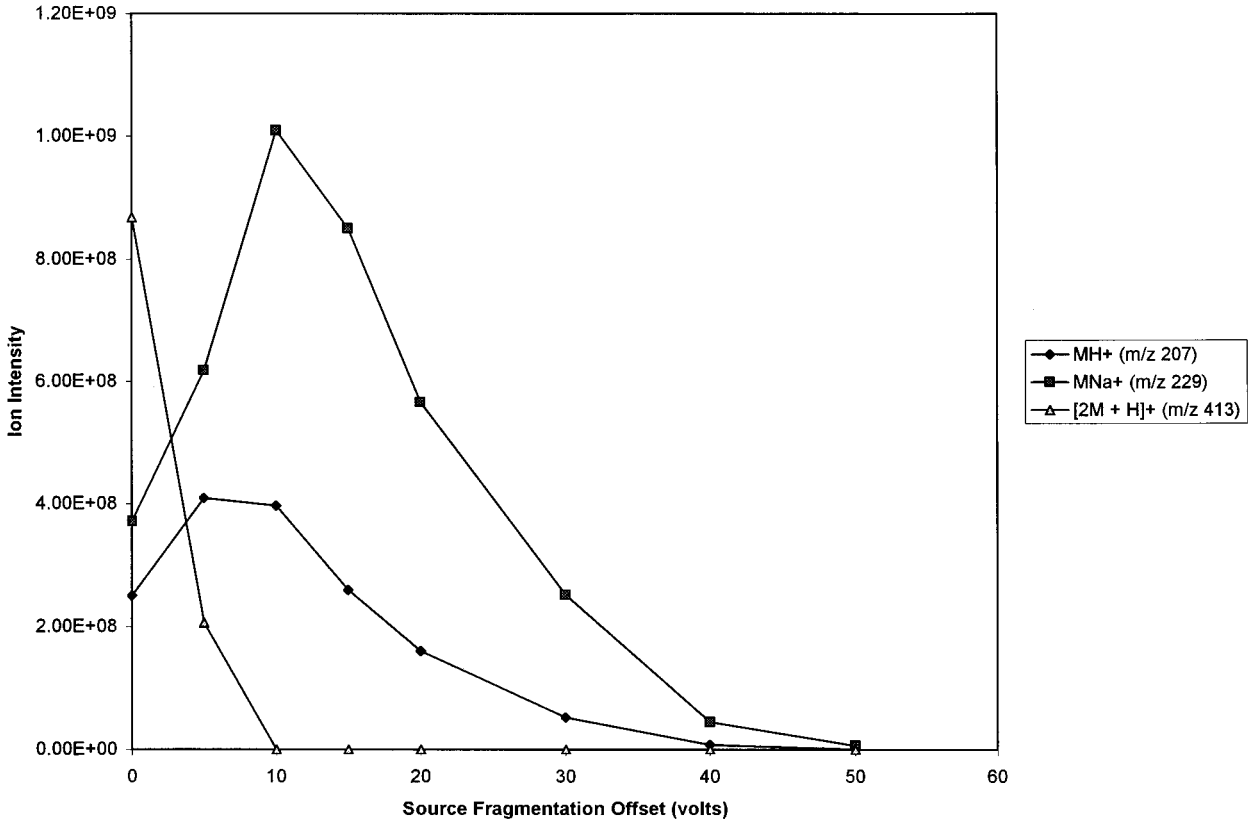

Figure 2. In-source CID of isopturon MH^+.

molecular adduct ions, MH^+ and MNa^+, by as much as 150% but only in the case of isopturon. Figure 2 plots the intensity of isopturon molecular adduct ions and a dimer, $[2M + H]^+$, at various source CID offset voltages.

CID in the Mass Analyzer. *Negative Ions.* MS/MS or multiple stages of fragmentation (e.g., $MS^{n \leq 10}$) can be accomplished in various commercial ion trap instruments. The precursor ion—the monoisotopic MH^+ and $[M - H]^-$ ions in this study—is accumulated in the ion trap and subsequently subjected to successive stages of CID. By applying a sequence of waveforms the quadrupole ion trap accomplishes multiple stages of fragmentation in time, whereas quadrupole instruments carry out CID in different stages, typically only MS^2.

Table 3. Collision-Induced Dissociation of Phenyl Urea $[M - H]^-$ Ions

compound	parent ion ($[M - H]^-$)	collision energy (V)	loss of R_2NH (−45 amu/ −87 amu*)	$HNPh^-$ (−87 amu)
N′,N′-Dimethyl Ureas				
chlortoluron	211	35	*m/z* 166 (100%)	*m/z* 140 (13%)
diuron	231	35	*m/z* 186 (100%)	*m/z* 160 (25%)
flumeturon	231	35	*m/z* 186 (100%)	*m/z* 160 (7%)
isopturon	205	35	*m/z* 160 (100%)	*m/z* 134 (11%)
metoxuron[a]	227	25	ND[b]	*m/z* 156 (<1%)
monuron	197	35	*m/z* 152 (100%)	*m/z* 126 (4%)
N′,N′-Dialkyl Ureas				
neburon	273	35	*m/z* 186 (21%)[c]	*m/z* 160 (100%)
N′-Methoxy-*N′*-methyl Ureas				
chlorbromuron	291	25	ND	*m/z* 204 (100%)
linuron	247	25	ND	*m/z* 160 (100%)
metobromuron	257	20	ND	*m/z* 170 (100%)
monolinuron	213	20	*m/z* 152 (<1%)	*m/z* 126 (100%)

[a] Metoxuron with a unique methoxy substitutent on the aromatic ring fragmented by loss of methyl radical to give a prominent *m/z* 212 daughter ion as base peak. [b] Not determined. [c] Loss of *N′*-methyl-*N′*-butyl amine from neburon; a prominent *m/z* 217 daughter ion (37%) due to loss of butene also was found.

Phenyl urea MH^+ or $[M - H]^-$ parent ions were subjected to successively higher collision energies in 5 V increments. The compounds were introduced to the source in mobile phase via loop injection of milligram per milliliter standards.

CID fragmentation pathways involved a few common cleavage steps consistent with the type of substitution. For example, $[M - H]^-$ ions of the *N′,N′*-dimethyl ureas lost 45 amu corresponding to loss of dimethyl amine (Table 3). The only exception was metoxuron, a molecule with a unique aromatic methoxy substituent. Another prominent daughter ion was $HNPh^-$ (Table 3).

The *N′*-methoxy-*N′*-methyl ureas formed $HNPh^-$ ions almost exclusively as daughter ions. Loss of *N*-methoxy-*N*-methyl amine (analogous to dimethyl amine) was not observed. The methoxy compounds fragmented at about half the collision energy required for the *N′,N′*-dialkyl ureas. Each of the methoxy compounds also lost formaldehyde and methanol, with the corresponding daughter ions detected as follows: chlorbromuron, $-CH_2O$ (13%), $-CH_3OH$ (3%); linuron, $-CH_2O$ (17%), $-CH_3OH$ (5%); metobromuron, $-CH_2O$ (23%), $-CH_3OH$ (11%); and monolinuron, $-CH_2O$ (16%), $-CH_3OH$ (9%) (for collision energies see Table 3).

Figure 3 depicts CID of the diuron $[M - H]^-$ ion. Loss of dimethyl amine produces the *m/z* 186 ion, probably the indicated bicyclic carbanion. Fragmentation of the *m/z* 186 daughter ion in MS^3 occurs with loss of the $^{\bullet}CN$ radical (*m/z* 160) or HCl (*m/z* 150). HCl is also lost from the *m/z* 160 anions in MS^4. Thus, two fragmentation sequences, $231^- > 186^- > 160^- > 124^-$ and $231^- > 186^- > 150^- > 122^-$, are available for selected reaction monitoring (SRM) of diuron. Analogous sequences are expected for the other phenyl ureas.

Positive Ions. CID of MH^+ was investigated in both diuron and linuron. In the case of diuron, loss of dimethyl amine was important and yielded the *m/z* 188 cation, possibly a bicyclic oxonium ion or isocyanate cation (Figure 4). An anilinium ion, *m/z* 160, is the proposed product ion from $233^+ > 188^+ > 160^+$. Dimers fragment by loss of M or dimethyl amine. For example, diuron $[2M + Na]^+$ ion (*m/z* 487) dissociated to both MNa^+ (*m/z* 255) and $[2M + Na - (CH_3)_2NH]^+$ (*m/z* 442).

Linuron MH^+ ion (*m/z* 249) yielded four daughter ions: *m/z* 182, *m/z* 160 (anilinium), *m/z* 188 (loss of

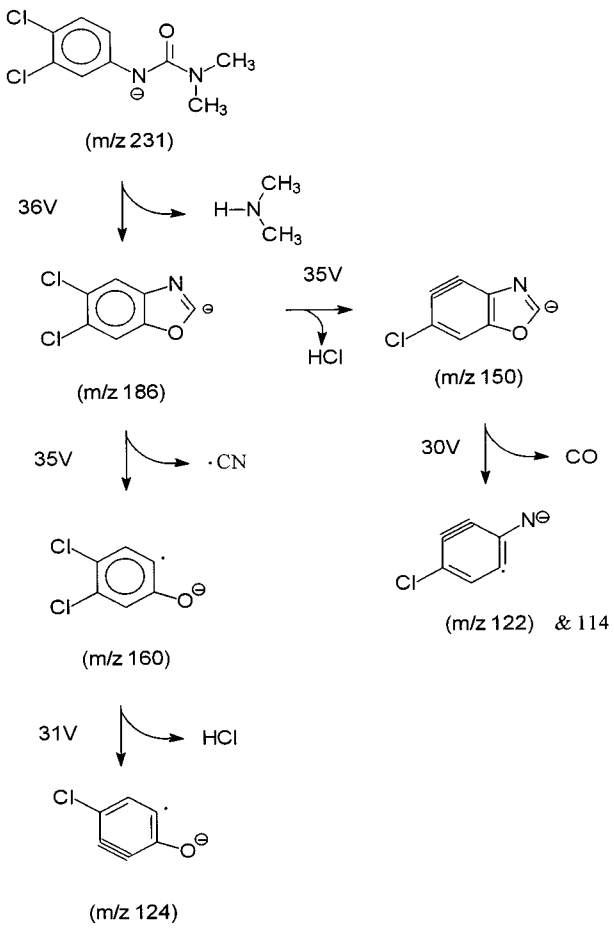

Figure 3. Proposed daughter ions from CID of diuron $[M - H]^-$ anion.

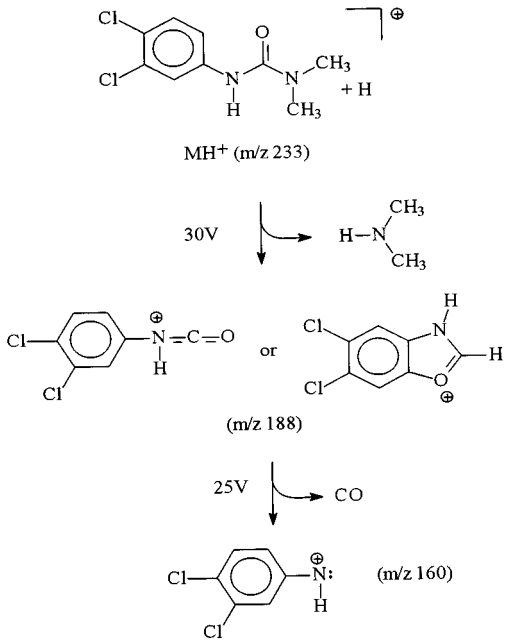

Figure 4. Proposed CID of diuron protonated molecule ion.

N-methoxy-*N*-methyl amine), and *m/z* 218 (loss of methoxy radical). An MS^3 experiment of the major daughter ion resulted in sequential loss of fragments weighing 67 and 29 amu (e.g., $249^+ > 182^+ > 153^+$).

Closer inspection of the original diuron spectra revealed *m/z* 160 and 188 as positive fragment ions and *m/z* 186, 150, and 160 as fragment ions in negative ES

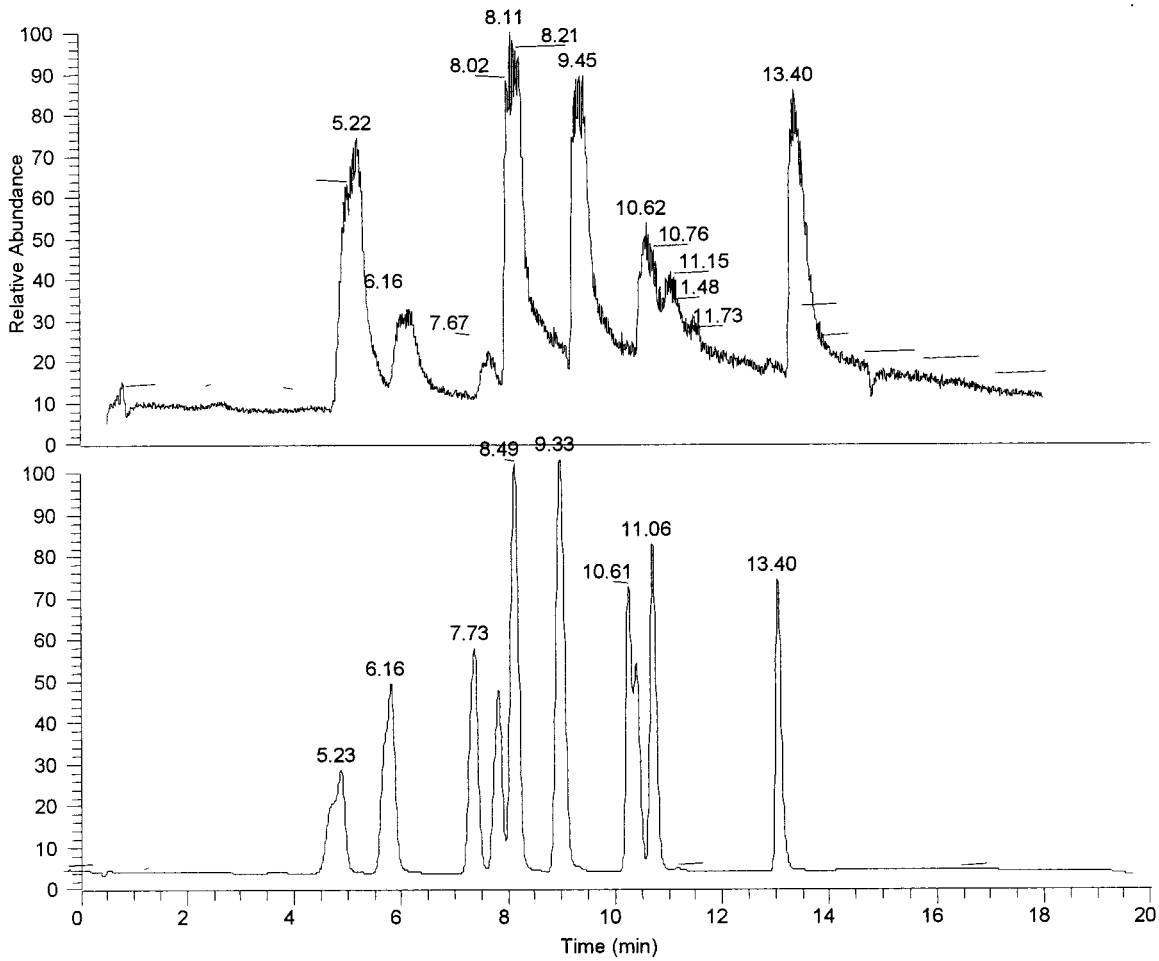

Figure 5. Chromatographic separation of 12-component phenyl urea mixture on C_{18} reversed phase column: ES mass spectrometer total negative ion current (upper trace) and UV absorbance chromatogram from inline diode array detector (lower trace).

spectra. However, only the *m*/*z* 160 positive ion was very abundant. In each case the chlorine isotope was present. Similarly, linuron's primary spectrum exhibited fragments at *m*/*z* 160 and 182.

LC-MS. *ES-Compatible HPLC Separation.* The phenyl urea herbicides have minor variations in substitution with slight differences in polarity. Despite the slight differences, most of the analogues can be separated using a moderately efficient C_{18} reversed phase column using acetonitrile/water or acetonitrile/dilute phosphoric acid gradients (*5*). In ES-LC-MS a few restrictions must be observed including minimizing mobile phase flow rate and avoiding high concentrations of involatile buffers.

In this study a 2.1 mm (i.d.) × 15 cm C_{18} reversed phase column was used with various 0.5 mL/min methanol/water gradients. A 14 min gradient from 30 to 80% methanol (Figure 5) with the 12-component mixture eluting between ~5 and ~13 min provided adequate separation. For these conditions the heated capillary was held at 350 °C, and both sheath and auxiliary gas were needed. Typical retention times (t_R), quantitation ions, and other details appear in Table 4.

Negative and Positive Ion ES Sensitivity. To determine detector sensitivity (the slope of the calibration response curve), the instrument was calibrated with mixed standards ranging in concentration from 1 pg/μL to 100 ng/μL. A 10 μL injection volume was used. Regression lines based on peak areas in reconstructed ion chromatograms were determined using Excel software (Microsoft, Redmond, WA) to determine the slope of the best fit line. Monolinuron was present in each of the mixtures at 2.0 ng/μL as an internal standard (IS). The instrument was operated in mass scanning mode, and calibration was repeated on different days, each time after retuning with metobromuron. The sensitivities reported represent the average of two separate experiments.

Sensitivity to the phenyl ureas varied considerably with chemical structure. In positive ion mode the greatest sensitivity was found for siduron, whereas chlorbromuron was least sensitive, ~15% that of sidu-

Table 4. Chromatographic Retention Times and $[M - H]^-$ Electrospray MS Ions Used for LC-MS Determination of Phenyl Ureas

compound	t_R (min)	halogen(s)	negative ions
metoxuron	5.13	Cl	*m*/*z* 227, *m*/*z* 229
monuron	6.08	Cl	*m*/*z* 197, *m*/*z* 199
monolinuron	7.60	Cl	*m*/*z* 213, *m*/*z* 215
fluometuron	8.07	F (3)	*m*/*z* 231
metobromuron	8.36	Br	*m*/*z* 257, *m*/*z* 259
chlortoluron	8.40	Cl	*m*/*z* 211, *m*/*z* 213[a]
isopturon	9.12		*m*/*z* 205
diuron	9.28	Cl (2)	*m*/*z* 231, *m*/*z* 233
linuron	10.48	Cl (2)	*m*/*z* 247, *m*/*z* 249
siduron	10.57, 10.86		*m*/*z* 231
chlorbromuron	10.93	Cl, Br	*m*/*z* 291, *m*/*z* 293[b]
neburon	13.22	Cl (2)	*m*/*z* 273, *m*/*z* 275

[a] To readily distinguish chlortoluron, only the *m*/*z* 211 ion is monitored. [b] The *m*/*z* 293 ion is avoided because of an interference noted in the text.

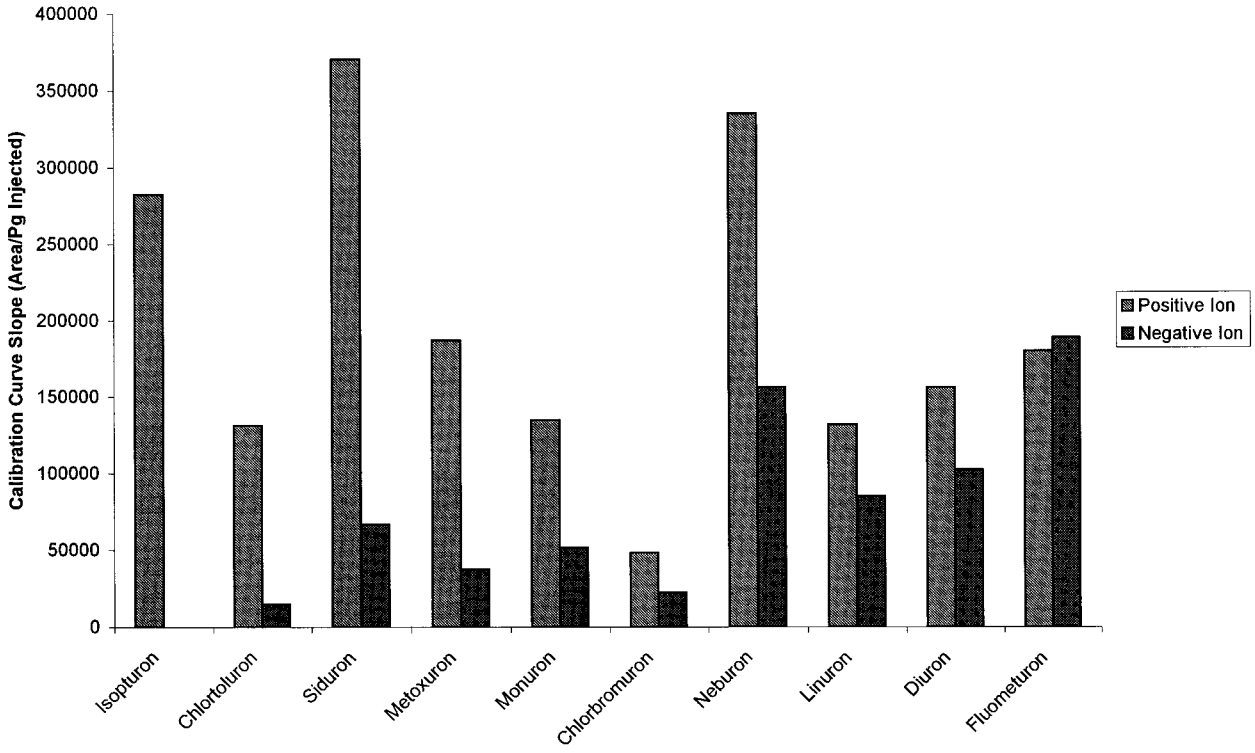

Figure 6. Electrospray sensitivity (calibration curve slope) for phenyl ureas monitored in positive and negative ion modes.

ron. Electron-rich compounds, that is, siduron, neburon, and isopturon, had the steepest response curves. For all phenyl ureas studied except fluometuron, positive ion sensitivity was greater than negative ion sensitivity (Figure 6).

Each of the phenyl ureas, with the exception of isopturon, gave a satisfactory signal in the negative ion mode. Negative ion sensitivity was greatest for compounds with *para*- or *para*- and *meta*- halogen substitution (e.g., diuron, linuron, neburon, chlorbromuron, and monuron) or *meta* electron-withdrawing groups (e.g., fluometuron).

Linearity and Instrument Detection Limits. The instrument response was very linear at the low end of the dynamic range (Figure 7). At higher concentrations the response was quadratic (Figure 8), indicating the onset of detector saturation, which is typical of ES ionization (*22*).

For calibration over a wide concentration range, added standards that bracket sample concentrations are required for accuracy. The software used has provisions for linear, quadratic, and other calibration curves. The maximum "smoothing" was used when the ion chromatograms were processed, and internal standard quantitation was employed. Monolinuron was used routinely as the IS with a concentration of 2 or 5 ng/μL in the final extract. Because the instrument was tuned with metobromuron there was some system contamination, which made low-level metobromuron determinations impossible. Accordingly, no quantitative data are presented for monolinuron or metobromuron, although simlar accuracy and precision are expected for these analogues.

Quantitative Analysis. Although the phenyl urea compounds gave a stronger signal in positive ion mode, negative ion monitoring was preferred for several reasons. Negative ion spectra were simpler and tended less to exhibit intense dimer and trimer species. Positive ion spectra appeared to be more concentration-dependent, suggesting a greater potential for matrix effects. Although source fragmentation enhanced the intensity of molecular adducts in some cases, the effect was compound specific. Overall, negative ion detection had substantially reduced background signal and greater S/N.

Negative quantitation ions are summarized in Table 4. For most of the halogenated analytes the signals for the two isotopes were summed, except as noted below. The two compounds with no halogens were monitored exclusively by single ion monitoring (SIM). In each case except isopturon, instrument detection limits (Table 5) are lower in the negative ion mode due to reduced noise levels and enhanced S/N.

MDLs and Surface Water Analyses. *MDLs.* MDLs were determined by analysis of laboratory reagent water spiked at 20 or 50 ng/L. Samples were subjected to automated solid phase extraction (SPE) using C_{18} disk cartridges as described elsewhere (*5*). SPE achieved a 1000-fold analyte enrichment with absolute recoveries for the phenyl ureas ranging from 48 to 70%.

MDLs were determined using negative ion ES-LC-MS by analysis of seven replicates. The extracts were dissolved in 1 mL of ethyl acetate with 5 ng/μL of monolinuron as IS. Without reiterating the MDL determination—there was ample signal in most cases for lower spike concentrations—the experimental MDLs ranged from ~10 to 40 ng/L (ppt) (Table 5). Method accuracy ranged from 77 to 96%, high accuracy considering the very low fortification levels studied.

Similar phenyl urea MDLs were reported by Ferrer and Barcelo (*3*) using an SPE API mass spectrometry technique. ES-LC-MS detection limits are comparable or lower than those achievable using diode array detection in agreement with previous reports (*2*, *4*).

Surface Water Analyses. Colorado River water was analyzed using the SPE LC-MS method described. The

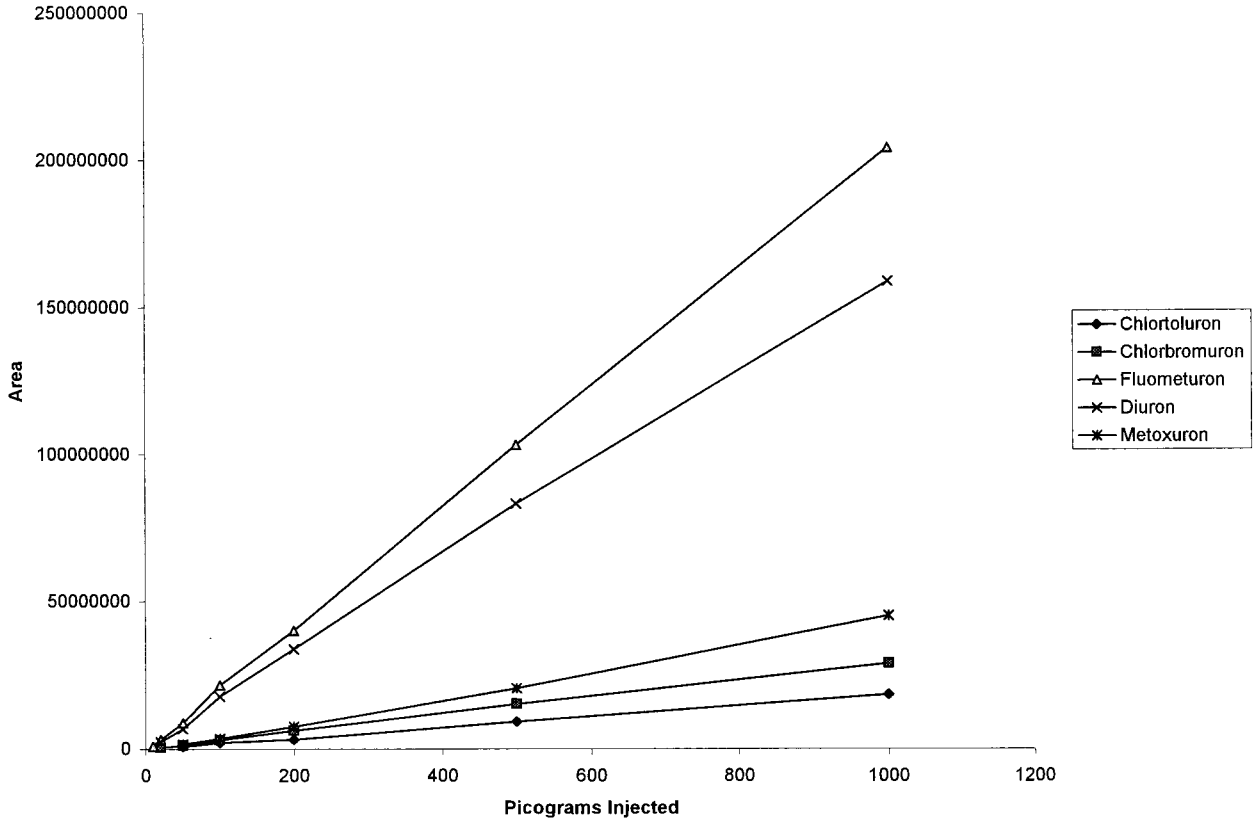

Figure 7. Electrospray linearity at the low end of the dynamic response range between 10 and 1000 pg.

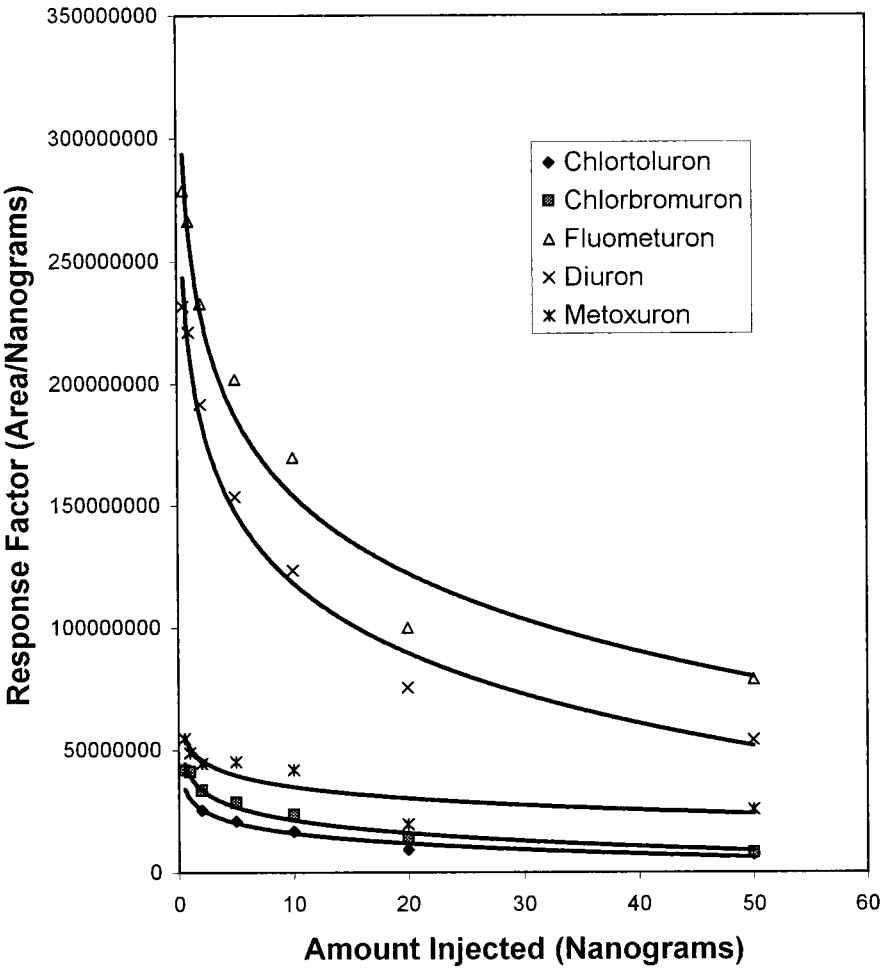

Figure 8. Quadratic multipoint calibration in the low nanogram range.

Table 5. Summary of Instrument Detection Limits, Method Detection Limit Study, and Accuracy for Determinations in Surface Water

analyte	instrument detection limit (pg)		method detection limit study egative Ions			mean recovery from Colorado River water
	negative ions	positive ions	spike level (ng/L)	MDL (ng/L)	accuracy (%)	negative ions (%)
metoxuron	50	50	20	ND[a]	NA[b]	44
monuron	10	10	20	10	77	76
fluometuron	10	20	20	15	96	64
chlortoluron	50	50	20	8	91	39
isopturon	>200	10	50	ND	NA	NA
diuron	20	50	50	30	81	70
linuron	20	50	50	25	94	54
siduron	20	20	50	27	84	75
chlorbromuron	20	50	50	NA	NA	52
neburon	10	20	50	36	87	72

[a] Not detected. [b] Not available.

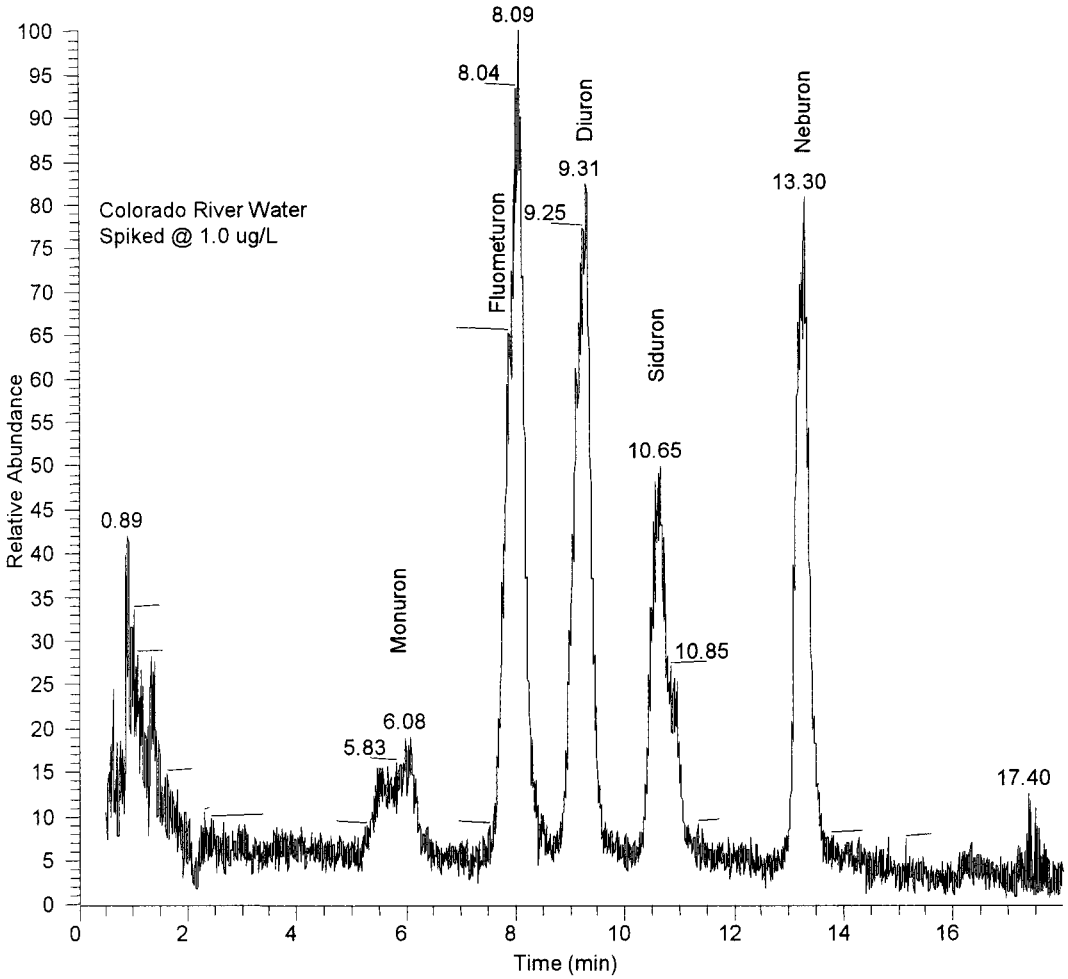

Figure 9. Colorado River water spiked with 1 μg/L concentrations of the indicated phenyl urea herbicides. The unsmoothed reconstructed ion chromatogram summing negative ions for five of the target compounds is shown.

river water was high in total dissolved solids (TDS) (590 mg/L) and total organic carbon (TOC) (3.2 mg/L) on the basis of standard water quality tests. No phenyl ureas were detected in river water samples prior to spiking. At the 1 μg/L spike level, all of the phenyl ureas were detected ($n = 3$). Accuracy in surface water was in the range of 39−76%, lower than that found in laboratory reagent water. The apparent low bias encountered in the analysis of high TDS/high TOC surface water cannot be attributed to lowered SPE recoveries, which actually are improved by dissolved salts (5). A better possibility is signal suppression. A surface water chromatogram is presented in Figure 9.

MS-MS and SRM Confirmation. An interference corresponding in retention time to chlorbromuron was present in SPE extracts of laboratory reagent water. This unidentified material may have originated from the extraction disks, extraction solvent, septum caps, or other source. The interfering compound was not chlorinated and was detected only in the *m/z* 293 channel. Accordingly, chlorbromuron detection was limited to the *m/z* 291 ion. There was no indication of interference with any of the other herbicides in parts per trillion analyses.

Distinguishing chlorbromuron from the interference was possible using MS-MS techniques such as daughter ion scanning or SRM. With a collision energy of 21 V

the daughter ions from *m*/*z* 293, the $[M - H]^- + 2$ isotope peak, included *m*/*z* 206 (base peak) and *m*/*z* 263 (25%). In daughter ion scanning mode, a reconstructed ion chromatogram of *m*/*z* 206 or 263 or their sum allowed specific chlorbromuron detection. In SRM the signal of only one (or several) daughter ions is acquired. By either MS-MS technique, chlorbromuron was detected with high specificity in laboratory reagent water spiked with 20 ng/L chlorbromuron. The present study did not examine the use of MS-MS in quantitative analysis.

Conclusion. API LC-MS is establishing a reputation for reliability, ruggedness, and sensitivity and is now routinely used in pharmaceutical, agrochemical, and biotechnology laboratories. Electrospray MS could also greatly extend the capabilities of environmental testing laboratories. In the current study, phenyl urea herbicides were determined with instrument detection limits ranging from <10 to 50 pg injected. Although sensitivity was typically greater in positive ion ES, negative ion monitoring offered improved S/N ratios and was selected as the basis for a multiresidue LC-MS phenyl urea method. Negative ion ES spectra were simpler, consisting primarily of $[M - H]^-$ over a wide concentration range.

Quantitative performance and linearity were very satisfactory, especially at the low end of the calibration range where trace analyses are performed. In conjunction with a conventional SPE sample workup, experimental MDLs in the low parts per trillion range were determined. The quadrupole ion trap instrument offered various approaches to confirmation (e.g., daughter ion scanning or SRM) as demonstrated in an instance where SIM had insufficient specificity.

Dissolved substances in surface water appeared to affect accuracy to some extent. Minor signal suppression was noted in a high TDS/high TOC surface water. More thorough study is needed to confirm this observation, to elucidate the mechanisms involved, and possibly to develop procedures to mitigate the effect.

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NOTE ADDED AFTER ASAP POSTING

An earlier version of Figure 8 was inadvertently used for initial Web posting on May 18, 2001. The correct version of Figure 8 is shown in this posting.

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