

# Determination of Corn Herbicides by GC–MS and GC–NPD in Environmental Samples

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Several herbicides widely used to control weeds in corn, atrazine, alachlor, metolachlor, and pendimethalin, have been determined in soil and water. Analysis of herbicides was performed by GC–NPD and GC–MS. Soil was extracted with ethyl acetate on a mechanical shaker, and water was extracted with dichloromethane in a separatory funnel. The average recoveries through the method were higher than 90% in soil and 85% in water. The detection limit was lower than 0.01 ppm in soil and 0.1 ppb in water for GC–MS and GC–NPD, respectively. Soil samples from corn fields, after harvest, were taken from the surface (0–10 cm) of several fields located in Albacete and Guadalajara, Spain, and water was taken from wells located in the fields. These samples were analyzed according to the proposed GC methods, and statistically similar values between the two methods were obtained. The identities of the detected herbicides were confirmed by GC–MS.

**Keywords:** Atrazine; alachlor; metolachlor; pendimethalin; soil; water

## INTRODUCTION

Weed control in maize is widely accomplished by using atrazine, a chlorotriazine herbicide, in binary mixture with another herbicide, such as the chloroacetamide alachlor or metolachlor or the nitroaniline pendimethalin. The potential for environmental contamination by these herbicides, owing to their physicochemical properties and the cultural practices used, is high and most of them have been detected in surface and ground water in different countries (Economic Commission for Europe, 1992).

The persistence of some of these herbicides, such as atrazine and pendimethalin, together with the variability in environmental conditions among different areas, results in residual amounts of herbicide in the soil that can be phytotoxic to sensitive crops cultivated in rotation (Tadeo et al., 1993; Caverly, 1987). In addition, some herbicide metabolites, such as deethylatrazine, are also phytotoxic and persist over corn harvest (Sironi et al., 1973). Therefore, the determination of these compounds in water and soil is of interest to learn about environmental contamination and possible phytotoxicity problems in crop rotation.

Analysis of chlorotriazines, chloroacetamides, and nitroanilines has been usually carried out by gas chromatography (GC) with nitrogen–phosphorus detector (NPD) (Khan and Purkayastha, 1975; Ambrus et al., 1981; Nash, 1990) or electron capture detection (ECD) (Young and Chu, 1973; Deleu and Copin, 1984; Zimdahl et al., 1984) and also by reversed-phase liquid chromatography (Popl et al., 1983; Battista et al., 1988; Miles and Zhou, 1990). The confirmation of residues has been performed in some cases by GC with mass spectrometry (MS) (Mangani and Bruner, 1983; Dietrich et al., 1988; Durand et al., 1992).

The objective of this work is to study the analysis and confirmation of corn herbicides in soil and water samples by GC–MS and to compare these results with those obtained by the standard GC–NPD determination. The herbicides atrazine, alachlor, metolachlor, and pendimethalin were selected on the basis of their application in corn fields in the area. The phytotoxic atrazine metabolite, deethylatrazine, was also included in the

**Table 1. Main Ions Found in the Mass Spectra of Herbicides**

herbicide	<i>m/z</i>	relative abundance (%)	structural assignment
deethylatrazine	172	100	[M – CH <sub>3</sub> ] <sup>+</sup>
	187	24.5	[M] <sup>++</sup>
atrazine	200	100	[M – CH <sub>3</sub> ] <sup>+</sup>
	215	63.9	[M] <sup>++</sup>
alachlor	173	25.9	[M – C <sub>3</sub> H <sub>7</sub> NH] <sup>+</sup>
	188	100	[M – CH <sub>2</sub> OCH <sub>3</sub> – HCl] <sup>+</sup>
	160	99.1	[M – CH <sub>3</sub> OH – OCCH <sub>2</sub> Cl] <sup>+</sup>
	238	18.9	[M – CH <sub>3</sub> OH] <sup>+</sup>
metolachlor	162	100	[M – CH <sub>2</sub> OCH <sub>3</sub> – OCCHCl] <sup>+</sup>
	238	49.1	[M – CH <sub>2</sub> OCH <sub>3</sub> ] <sup>+</sup>
pendimethalin	252	100	[M – CH <sub>3</sub> – CH <sub>3</sub> ] <sup>+</sup>
I.S.	140	100	[M – OCCH <sub>3</sub> – CH <sub>3</sub> ] <sup>+</sup>

**Table 2. Recovery of Herbicides Added to Soil Samples**

herbicide	added (ppm)	recovery <sup>a</sup> (%)	
		GC-ITD	GC-NPD
deethylatrazine	1	99.0 ± 2.1	100.0 ± 1.1
	0.2	96.0 ± 1.7	99.4 ± 2.3
atrazine	1	95.3 ± 10.4	100.0 ± 0.6
	0.2	99.2 ± 3.4	91.0 ± 5.9
alachlor	1	91.1 ± 7.6	99.0 ± 6.6
	0.2	96.1 ± 3.0	98.5 ± 1.7
metolachlor	1	90.4 ± 7.6	101.0 ± 4.1
	0.2	97.5 ± 6.7	98.0 ± 2.3
pendimethalin	1	90.0 ± 4.9	98.3 ± 2.3
	0.2	95.3 ± 4.1	99.5 ± 1.3

<sup>a</sup> Values are the mean of five replicates ± standard deviation.

study. The concentration of these compounds in well water and soil from several corn fields, after harvest, was determined.

## EXPERIMENTAL PROCEDURES

**Materials.** Herbicide standards were obtained from commercial sources: atrazine [6-chloro-*N*-ethyl-*N'*-(1-methylethyl)-1,3,5-triazine-2,4-diamine], deethylatrazine [6-chloro-*N'*-(1-methylethyl)-1,3,5-triazine-2,4-diamine], and metolachlor [2-chloro-*N*-(2-ethyl-6-methylphenyl)-*N'*-(2-methoxy-1-methylethyl)acetamide] from Ciba Geigy Co., Buchs, Switzerland; alachlor [2-chloro-*N*-(2,6-diethylphenyl)-*N'*-(methoxymethyl)acetamide] from Monsanto, St. Louis, MO; and pendimethalin

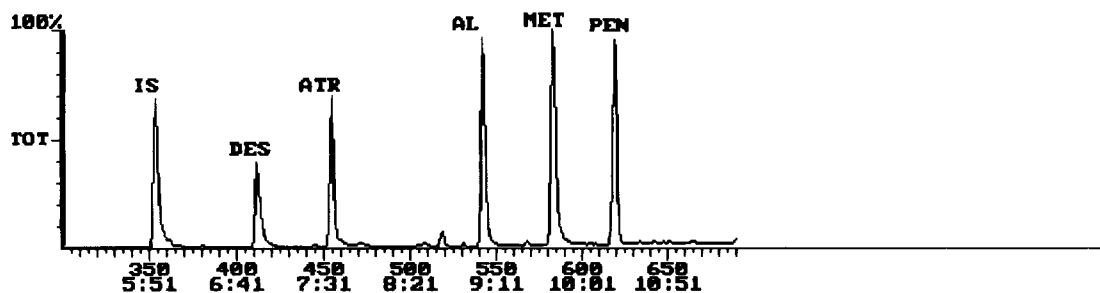


Figure 1. GC-ITD total ion chromatogram of a mixture of the herbicides and the internal standard (1  $\mu\text{g/mL}$ ).

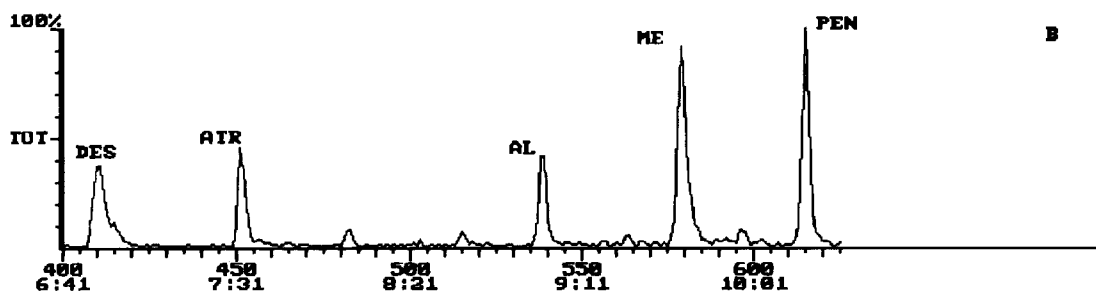
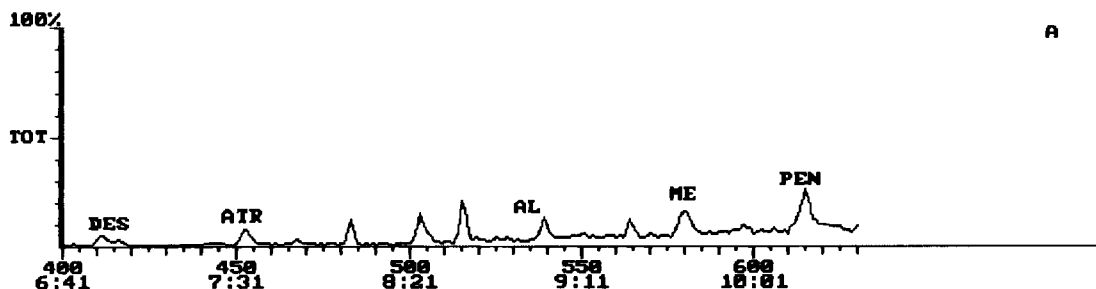


Figure 2. Detection limit of these herbicides in soil samples: (A) total ion current; (B) selected ion monitoring.

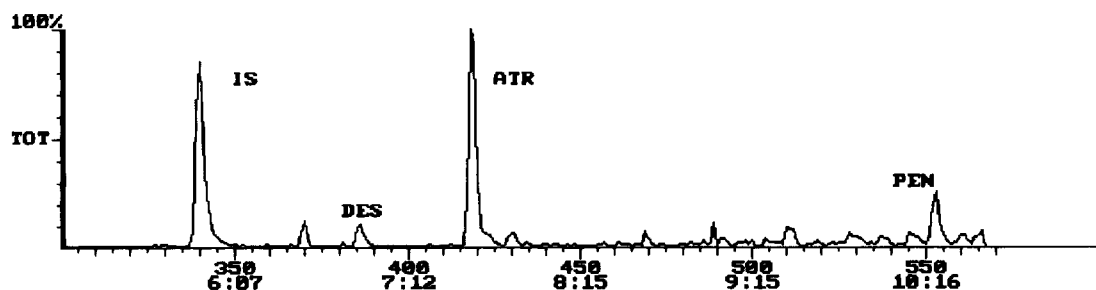


Figure 3. GC-ITD chromatogram, in the selected ion mode, of a soil sample from a maize field.

[*N*-(1-ethylpropyl)-3,4-dimethyl-2,6-dinitrobenzenamine] from American Cyanamid, Princeton, NJ. The solvents, dichloromethane, toluene, ethyl acetate, and hexane AR (Panreac, Barcelona, Spain), were used without further purification.

The internal standard, *N*-ethyl-*p*-chloroacetanilide, was prepared in the laboratory. Sodium hydride (432 mg) and ethyl iodide (0.6 mL) were added to *p*-chloroacetanilide (200 mg) dissolved in dimethyl sulfoxide (20 mL) and left overnight at room temperature. The mixture was diluted with hexane and hydrolyzed with water. The organic phase was decanted, dried over anhydrous sodium sulfate, and diluted to an adequate volume.

**Apparatus.** A Perkin-Elmer Model 8500 gas chromatograph equipped with an ion trap detector Finnigan (ITD) was used for the analysis of herbicides. A fused silica capillary column, BP-1 (12 m  $\times$  0.22 mm) was employed. Helium was used as carrier gas at 10 psig. The injection port and detector temperatures were 250 and 300  $^{\circ}\text{C}$ , respectively. Samples

were injected splitless, with the split valve closed for 1 min. The oven temperature was held at 85  $^{\circ}\text{C}$  for 1 min and then programmed at 20  $^{\circ}\text{C}/\text{min}$  to 180  $^{\circ}\text{C}$ , held for 0.5 min, and programmed at 10  $^{\circ}\text{C}/\text{min}$  to 250  $^{\circ}\text{C}$ , 5 min.

Mass spectrometric acquisition parameters were the following: transfer line temperature, 250  $^{\circ}\text{C}$ ; mass range, 50–350 Da; scan rate, 1 s/scan, 5- $\mu\text{s}$ scan; radio frequency voltage, 1.1 MHz and 0–7.5 kV; automatic gain control from 78  $\mu\text{s}$  to 25 ms; solvent delay, 5 min.

A Varian Model 3700 chromatograph was used for the GC-NPD analysis; it was equipped with a fused silica capillary column, BP-5 (12 m  $\times$  0.53 mm). The column temperature was maintained at 120  $^{\circ}\text{C}$  for 1 min and then programmed at 15  $^{\circ}\text{C}/\text{min}$  to 230  $^{\circ}\text{C}$ , 1 min. Injection port and detector temperatures were 270 and 300  $^{\circ}\text{C}$ , respectively. Nitrogen was used as carrier gas at 8 psig.

**Soil Samples Analysis.** Soil (20 g) was extracted with ethyl acetate (2  $\times$  100 mL) by shaking in a flask for 30 min

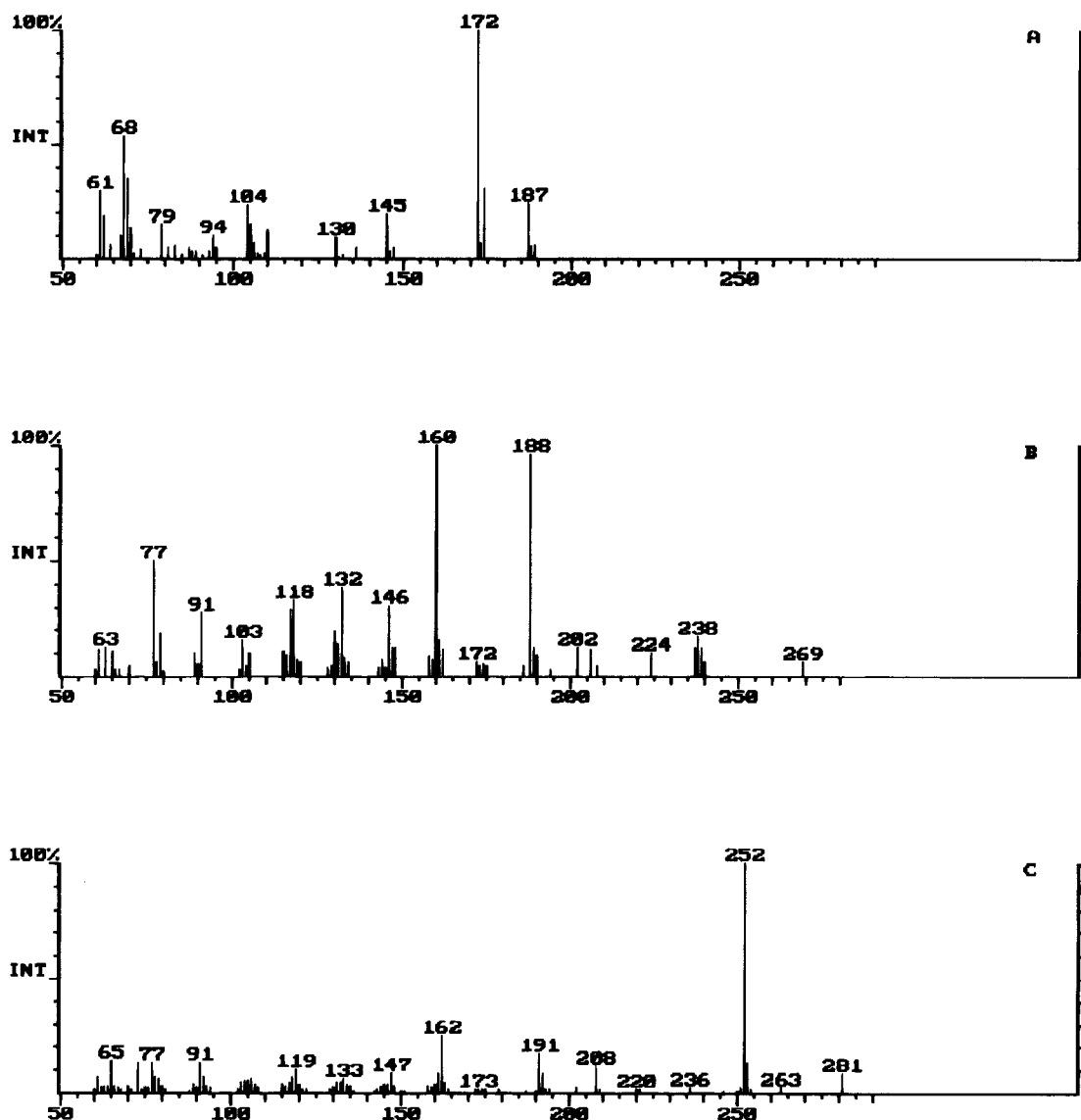


Figure 4. Mass spectra of some compounds identified in the samples: (A) deethylatrazine; (B) alachlor; (C) pendimethalin.

on an orbit shaker. The organic phase was filtered under suction through Whatman No. 1 filter paper and Celite, and the filter cake was washed with 30 mL of ethyl acetate. Solvent was removed under vacuum and the residue transferred to a tube with ethyl acetate and concentrated to a suitable volume. An aliquot was determined by gas chromatography under the conditions described above.

**Water Samples Analysis.** Water (1000 mL) was placed in a 2 L separatory funnel and extracted with  $3 \times 50$  mL of dichloromethane. The organic solvent was filtered through anhydrous sodium sulfate and evaporated to dryness under vacuum; small volumes of hexane were added to the residue and evaporated again to remove dichloromethane completely. The residue was transferred to a tube with ethyl acetate and concentrated to 1 mL, and an aliquot was analyzed by gas chromatography under the conditions described above.

The concentration of each compound was calculated by comparing the ratio of the peak areas in the sample with the ratio found for mixtures of herbicides and the internal standard of known concentration. *N*-Ethyl-*p*-chloroacetanilide was used as internal standard.

## RESULTS AND DISCUSSION

**GC-MS Characterization.** Figure 1 shows the total ion chromatogram obtained with a mixture of these herbicides and the internal standard. The major ions

found in the mass spectra of the herbicides obtained by gas chromatography with ion trap detection are shown in Table 1.

The mass spectrum of deethylatrazine showed the base peak at  $m/z$  172. Atrazine showed two characteristic ions at  $m/z$  215 and 200. The main ions of the mass spectrum of alachlor were at  $m/z$  188 and 160. Metolachlor presented a fragmentation pattern similar to that of alachlor, with main ions at  $m/z$  238 and 162 (Durand et al., 1992). Pendimethalin showed the most abundant ion at  $m/z$  252. The internal standard, *N*-ethyl-*p*-chloroacetanilide, presented the most important ion at  $m/z$  140. The different ions shown in Table 1 were used as diagnostic ions for screening purposes, as they correspond to most abundant ions at high  $m/z$  values. The base peak of each spectrum was monitored for quantitation of the studied compounds.

**Herbicide Determination in Soil.** Soil samples were fortified with 0.2 and 1 ppm of deethylatrazine, atrazine, alachlor, metolachlor, and pendimethalin and analyzed following the procedure described above. The average recoveries obtained were higher than 90% (Table 2), with good agreement between GC-ITD and GC-NPD determination in most cases.

The detection limit of these herbicides in soil samples

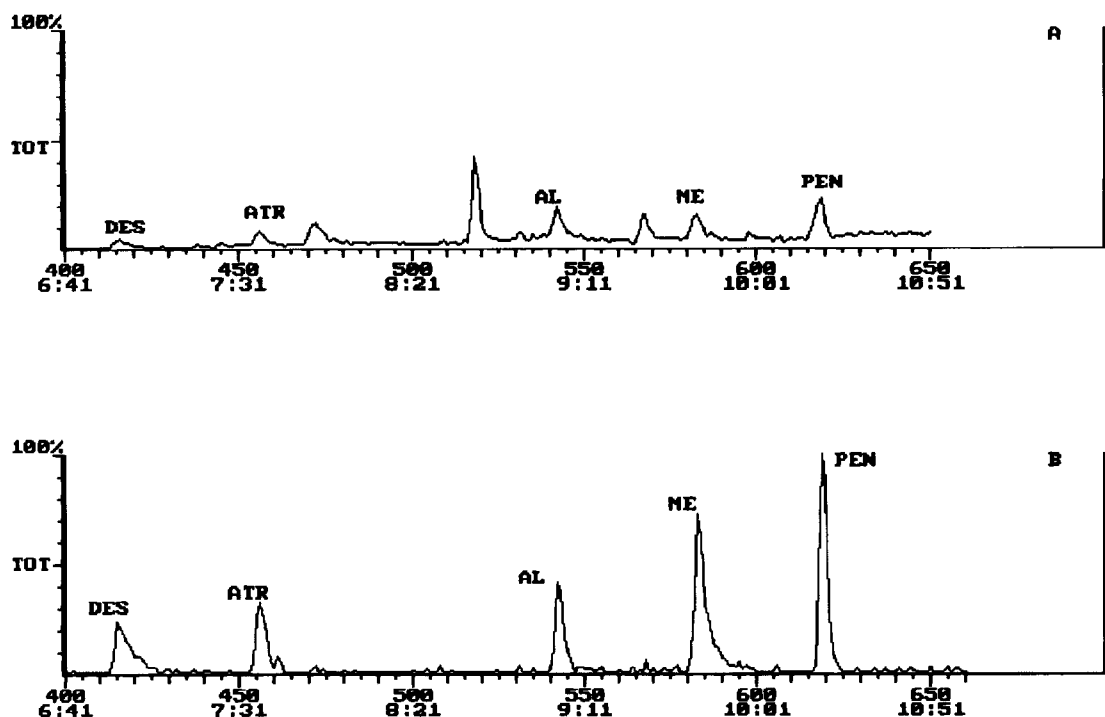


Figure 5. Detection limit of these herbicides in water samples: (A) total ion current; (B) selected ion monitoring.

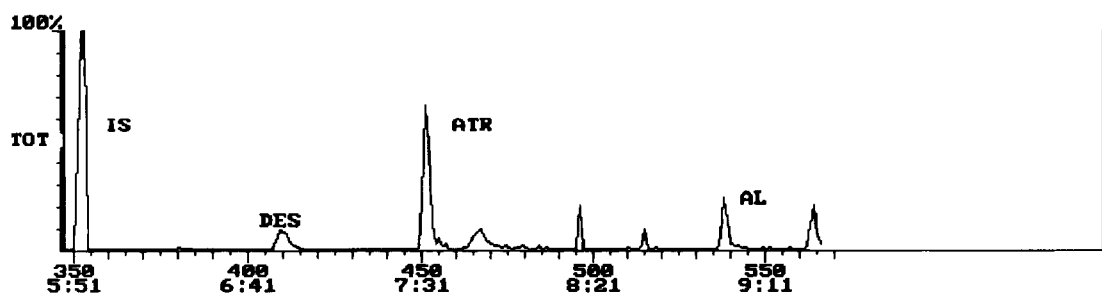


Figure 6. GC-ITD chromatogram, in the selected ion mode, of a water sample from a farm well.

was lower than 0.01 ppm with total ion current. This limit can be lowered about 5 times using selected ion monitoring (Figure 2). The detection limit with NPD was lower than 0.01 ppm.

Several corn fields from Albacete, Spain, were treated with herbicide mixtures of atrazine and alachlor or pendimethalin and sampled after crop harvest. Soil samples were analyzed using the described procedure and herbicides determined by GC-ITD (Figure 3) and confirmed by their mass spectra (Figure 4). Herbicide residues are shown in Table 3. These residues varied according to the herbicide persistence in the different fields and were in some cases higher than 0.1  $\mu\text{g/g}$ , a value that may be phytotoxic to sensitive crops cultivated in rotation with corn (Tadeo et al., 1993).

**Herbicide Determination in Water.** Water samples fortified with 0.2 and 1.0 ppb of these herbicides were analyzed by GC-ITD and GC-NPD in the conditions described above. Herbicides were extracted with dichloromethane (EPA, 1990), and recoveries higher than 85% were obtained, with a good agreement between both of the chromatographic techniques (Table 4).

The detection limit, for these herbicides, obtained using total ion current, was lower than 0.1 ppb. This limit can be improved with selected ion monitoring (Figure 5). The detection limit with NPD was lower than 0.1 ppb.

Water from two wells in Albacete located in the studied corn fields were analyzed following the de-

Table 3. Herbicide Residues Found in Soil of Maize Fields

field <sup>a</sup>	herbicide <sup>b</sup>	concentration <sup>c</sup> ( $\mu\text{g/g}$ )	
		GC-ITD	GC-NPD
1	deethylatrazine	0.02 $\pm$ 0.01	0.02 $\pm$ 0.01
	atrazine	0.14 $\pm$ 0.09	0.12 $\pm$ 0.08
	pendimethalin	0.04 $\pm$ 0.02	0.05 $\pm$ 0.02
2	deethylatrazine	0.01 $\pm$ 0.01	0.01 $\pm$ 0.01
	atrazine	0.05 $\pm$ 0.03	0.06 $\pm$ 0.03
	pendimethalin	0.09 $\pm$ 0.05	0.09 $\pm$ 0.04
3	deethylatrazine	0.03 $\pm$ 0.01	0.03 $\pm$ 0.01
	atrazine	0.11 $\pm$ 0.02	0.12 $\pm$ 0.01
	alachlor	0.04 $\pm$ 0.01	0.03 $\pm$ 0.01
4	deethylatrazine	0.01 $\pm$ 0.01	0.01 $\pm$ 0.01
	atrazine	0.05 $\pm$ 0.02	0.05 $\pm$ 0.02
	alachlor	0.03 $\pm$ 0.01	0.04 $\pm$ 0.01

<sup>a</sup> Soil properties: (1) silty clay, OM (organic matter) = 3.94%, pH 7.6; (2) clay, OM = 0.99%, pH 7.77; (3) loam, clay-sandy, OM = 1.22%, pH 8.6; (4) loam-sandy, OM = 1.23%, pH 8.5. <sup>b</sup> Doses: (fields 1 and 2) Zeastomp 30/20 (1.5 kg/ha of pendimethalin plus 1.0 kg/ha of atrazine); (fields 3 and 4) Lasso GD (0.8 kg/ha of atrazine). <sup>c</sup> Values are the mean of three replicates  $\pm$  standard deviation.

scribed procedure. Herbicides were not detectable in water samples from one well, but in the other well alachlor and atrazine, together with deethylatrazine, were detected at levels higher than 0.1 ppb, the maximum allowable concentration for each pesticide in drinking water (EC, 1980). Table 5 shows herbicide

**Table 4. Recovery of Herbicides Added to Water**

herbicide	added (ppb)	recovery <sup>a</sup> (%)	
		GC-ITD	GC-NPD
deethylatrazine	1	97.7 ± 1.2	103.0 ± 3.1
	0.2	94.5 ± 4.0	100.0 ± 2.2
atrazine	1	98.6 ± 5.8	88.8 ± 6.1
	0.2	100.4 ± 4.2	98.5 ± 5.5
alachlor	1	89.0 ± 7.1	94.2 ± 5.8
	0.2	100.2 ± 4.3	98.7 ± 5.4
metolachlor	1	90.4 ± 6.0	94.5 ± 4.7
	0.2	103.1 ± 4.7	108.6 ± 8.3
pendimethalin	1	95.1 ± 4.4	98.5 ± 4.4
	0.2	108.4 ± 2.1	104.5 ± 5.8

<sup>a</sup> Values are the mean of five replicates ± standard deviation.

**Table 5. Determination of Herbicides in Albacete Well Water**

herbicide	sampling date	concentration <sup>a</sup> (μg/L)	
		GC-ITD	GC-NPD
deethylatrazine	Dec 15, 1992	0.15 ± 0.02	0.13 ± 0.02
	Jan 21, 1993	0.11 ± 0.02	0.12 ± 0.02
atrazine	Dec 15, 1992	0.89 ± 0.06	0.74 ± 0.05
	Jan 21, 1993	0.65 ± 0.03	0.50 ± 0.04
alachlor	Dec 15, 1992	0.23 ± 0.03	0.24 ± 0.02
	Jan 21, 1993	0.22 ± 0.02	0.23 ± 0.02

<sup>a</sup> Mean of two replicates ± standard error.

levels determined in that well, and Figure 6 displays a GC-ITD chromatogram of a water sample from the same well.

**Comparison of Results.** Both analytical methods, GC-ITD and GC-NPD, were compared including all of the samples (Simal et al., 1993). The equation for calculating  $t$  is

$$t = X_d(n)^{(1/2)} / S_d \quad (1)$$

where  $n$  is the number of samples ( $n = 22$  for soil and  $n = 16$  for water samples),  $X_d$  is the mean of the difference between both methods in the value obtained for each sample ( $-1.5823$  and  $-0.7256$  for soil and water samples, respectively), and  $S_d$  is the standard deviation ( $\pm 3.8676$  and  $\pm 4.1047$  for soil and water samples, respectively). The calculated values of  $|t|$  (1.9189 for soil and 0.7071 for water samples) are lower than the critical value (3.182 at  $p = 0.05$ ); therefore, the methods do not give statistically different values.

**Conclusions.** The proposed methods are reproducible and sensitive enough to determine these chlorotriazine, chloroacetamide, and dinitroaniline herbicides in soil and water by gas chromatography with ITD and NPD detection. The GC-ITD results are in agreement with those determined by GC-NPD, with the advantage that the compounds can be confirmed at residue level by their mass spectra.

Several herbicides were detected, and their identities confirmed by GC-ITD, in farm well water and in soil after corn harvest.

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