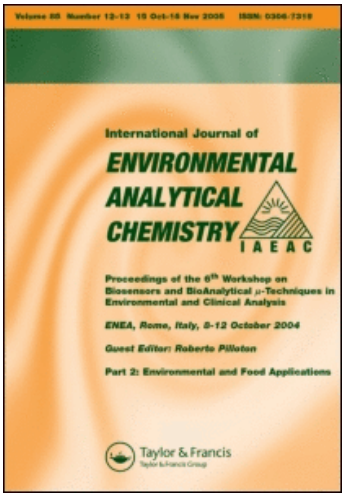


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# The Use of Mobile Phase Additives in the Determination of 55 (Polar) Pesticides by Column Liquid Chromatography-Thermospray-Mass Spectrometry

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# THE USE OF MOBILE PHASE ADDITIVES IN THE DETERMINATION OF 55 (POLAR) PESTICIDES BY COLUMN LIQUID CHROMATOGRAPHY-THERMOSPRAY-MASS SPECTROMETRY

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The effect of various additives (ammonium acetate, ammonium formate, triethyl- and tripropylammonium formate and chloroacetonitrile) on the sensitivity and selectivity in liquid chromatography-thermospray-mass spectrometry (positive and negative ion mode) is studied for a group of 55 pesticides including phenoxy and carboxylic acids, hydroxy-keto-lactones, substituted amides, triazines, (thio)carbamates, phenylureas, substituted phenols, organophosphorus compounds, thiocyanates and anilines.

In the positive ion mode the base peaks correspond to $[M + H]^+$ and $[M + NH_4]^+$ for the majority of the test compounds. Fragmentation is observed in some isolated cases only. In the negative ion mode processes like (dissociative) electron capture and ion-molecule reactions take place. Fragment ions such as $[M - H - CONCH_3]^-$ for the carbamates, $[M - C_3H_6O]^-$ for the hydroxy-keto-lactones and $[M - H]^-$ for the chlorinated phenoxy acids are observed. Depending on the additive used, adduct ions such as $[M + CH_3COO]^-$, $[M + HCOO]^-$ and $[M + Cl]^-$ are formed. Adducts with fragment ions are also observed quite often. The use of triethylammonium formate and tripropylammonium formate instead of ammonium formate increases the sensitivity and selectivity of the method in the NI mode, because their high proton affinity enhances deprotonation.

**KEY WORDS:** Polar pesticides, LC, thermospray, LC-TSP-MS, mass spectrometry.

## INTRODUCTION

Today there is an increasing need for the selective and sensitive detection of priority pollutants such as pesticides in environmental samples. Because of the rather high polarity and/or thermolability of many of these compounds, analysis can not be carried out by means of Gas Chromatography-Mass Spectrometry (GC-MS)[1]. Admittedly, derivatisation, which can provide higher volatility, reduced polarity and/or better thermostability, may be used to overcome this problem[2,3]. However, when dealing with analytes which are not directly

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amenable to GC analysis, column Liquid Chromatography (LC) has to be considered as an alternative separation technique. With medium to highly polar pesticides, Reversed-Phase LC (RPLC) has been shown to be a highly rewarding technique, especially if it is combined with on-line trace enrichment. Recently, several groups of workers have reported interesting applications, one of which involves the fully automated on-line trace enrichment-RPLC-diode array detection of some 40–60 polar pesticides in surface water at the 0.3–5 ppb (μg/l)level[4]. However, although diode array detection can provide structural information[4,5], mass spectrometry is required for unambiguous identification and/or confirmation. LC-MS is therefore becoming increasingly important for the determination of polar, thermolabile and non-volatile compounds, especially since the introduction of the thermospray (TSP)[6] and particle beam (PB)[7] interfaces. These interfaces allow LC flow-rates of 0.5–2.0 ml/min to be introduced into the mass spectrometer. Their main differences concern optimal flow-rates, types of ionisation techniques and application range (analyte polarity)[8]. An important factor in LC-MS is the analyte detectability which is strongly influenced by the nature of the LC eluent. Therefore, the composition of the eluent has to be fine-tuned and it may well differ from the one which is preferably used for separation.

Most of the LC-MS studies reported in the literature have been performed using a TSP interface[9–24]; although analyte quantitation at trace levels still is a major problem[25], interesting applications involving the PB interface have also been published[26–28]. The spectra obtained with LC-TSP-MS resemble chemical ionisation (CI) spectra, i.e. only (de)protonated molecular ions, adduct ions with reactive species in the plasma in the source and some fragments are observed. Solvent adduct ions are often used to confirm the molecular weight of the analytes[10–15,20,21,29–31].

A variety of additives (ammonium acetate, formate, carbonate or bicarbonate, methylammonium acetate and triethylammonium formate) has been used for the determination of certain classes of pesticides (phenylureas, chlorotriazines, organophosphorus compounds, phenoxy acids) in the positive or negative ionisation (PI and NI, respectively) mode. Analyte detectability in the PI and NI mode depends on the proton affinity (PA) and gas-phase acidity ($\Delta H°_{acid}$) of the analyte and additive used. For example, the use of triethylammonium formate enhances deprotonation, because triethylamine has a higher basicity than ammonia[29], and formate enhances the formation of adducts, because of its higher $\Delta H°_{acid}$ than acetate.

Chloride attachment, which can be effected by using a chloroacetonitrile-containing LC eluent, provides complementary molecular weight information[32–34], helps to reduce fragmentation and has been shown to give satisfactory results for chlorine-containing compounds and compounds with electron-withdrawing groups[13,14,20].

The use of a repeller electrode promotes the formation of specific fragments next to that of adducts; collision induced dissociation (CID)-like spectra are obtained in this way[35,36]. Specific fragmentation can enhance the confirmatory power of the technique. Quite often analyte detectability is better in the NI than in the PI mode.

In recent years LC-TSP-MS has developed to the stage where it can be applied to real-life problems in e.g. the field of environmental chemistry[37]. Hammond *et al.*[23] used LC-TSP-MS for the determination of ureas at low levels, reporting detection levels of 10 μg/l with an off-line pre-concentration step for a spiked water sample. Most papers dealing with the use of LC-TSP-MS in environmental analysis report applications for one or two groups or classes

of compounds[10,13–15,17,19–22,24,29,38–40]. Only Bellar and Budde[9] applied LC-TSP-MS to the determination of a large group of pesticides with different polarities, in a spiked water sample. Liquid-liquid and liquid-solid extraction were used for off-line trace enrichment. Using the PI mode with ammonium acetate and formate as additives, $[M+H]^+$ and $[M+NH_4]^+$ ions were mainly observed. Analyte recovery (> 70%) and detectability (μg/l levels) were good, which allowed identification of 26 out of 29 pesticides tested. According to the authors, the sensitivity of the method is sufficient to detect the compounds in real-life samples. Recently, Bagheri *et al.* used on-line trace enrichment, for phenylureas[41] as well as three other classes of compounds[42] to determine sub-μg/l levels of 10–40 analytes by means of LC-TSP-MS. The system showed, besides simplicity, an excellent ruggednes.

In this paper, the use of various additives to flow injection analysis (FIA) carrier streams or LC eluents will be studied in order to optimise conditions for the sensitive detection of a group of 55 (polar) pesticides with widely different structures in LC-TSP-MS (PI or NI mode), the final aim being application in environmental analysis. Additives like ammonium acetate, ammonium formate, triethylammonium formate and tripropylammonium formate will be used. The role of chloroacetonitrile as an additive will also be discussed.



## EXPERIMENTAL

### *Chemicals*

Methanol and acetonitrile were obtained from Baker (Deventer, the Netherlands). Chloroacetonitrile, concentrated formic acid and ammonium acetate were purchased from Merck (Schuchardt, Hohenbrunn bei München, Germany). Ammonium formate was obtained from Aldrich (Beerse, Belgium) and triethylammonium formate (TEAmFo) and tripropylamine were purchased from Fluka Chemie (Buchs, Switzerland). All of these chemicals were of analytical reagent grade; they were used without further purification. Tripropylammonium formate (TPAmFo) was prepared by mixing equimolar amounts of tripropylamine (98%) and formic acid (concentrated). Doubly distilled demineralized water was used. The test compounds were purchased from Riedel de Haan (Seeze, Germany), Promochem (Wesel, Germany), Dr. S. Ehrenstorfer (Augsburg, Germany) and Hoechst (Frankfurt, Germany); they were all at least 95% pure. 10–20μg/ml stock solutions were prepared in methanol.

### *Liquid chromatography*

A Gilson (Villiers-le-Bel, France) Model 302 LC pump delivered the carrier stream or eluent at a flow of 1.0 ml/min in the case of FIA and LC, respectively. A home-made membrane pulse damper was used to deliver a non-pulsating flow of eluent to the TSP interface. A Model 7125 injection valve with a 20-μl loop from Rheodyne (Cotati, CA, USA) was used to introduce the sample. The analytical column was a 150 mm × 4.6 mm I.D. stainless-steel

column home-packed with Rosil C18-bonded silica (5 µm, 80 Å) from Alltech (Zwijndrecht, the Netherlands). Water (with or without 50 mM ionising additive)-acetonitrile (50:50, v/v) and water (with or without 50 mM ionising additive)-acetonitrile-chloroacetonitrile (49:49:2, v/v) mixtures were used as carrier streams during FIA or as the eluent during LC.

### *Mass spectrometry*

A Finnigan (Finnigan MAT, San Jose, CA, U.S.A.) Model 4500 quadrupole mass spectrometer equipped with a Finnigan TSP interface was used. The source temperature was 200 °C. The vaporizer temperature was set between 90 and 120 °C. The discharge voltage, when used, was set at 1000 V. The repeller voltage generally was in the range of +80 to +120 V when operated in the PI mode, and -50 to -80 V in the NI mode. Full-scan spectra (m/z 100–500/sec) were recorded in the PI and NI mode and all three modes of operation (filament-off, filament-on and discharge ionisation) were used.



## RESULTS AND DISCUSSION

The test compounds listed in Table 1 were earlier selected as target compounds in the development of an LC-DAD screening method for surface water monitoring[4]. They represent widely divergent chemical classes as well as several miscellaneous compounds.

The ionisation modes and FIA carrier streams examined in this study are listed in Table 2. Not all combinations were studied. For instance, with a carrier stream of water-acetonitrile-chloroacetonitrile (49:49:2, v/v) hardly any signal will be produced at all in the filament-off mode. In the paper more attention is given to the NI than the PI mode, because the diversity of adducts is larger in the NI (acetate, formate, chloride) than in the PI (ammonium) mode. Hardly any triethylammonium or tripropylammonium adducts were observed during preliminary runs.

### *Reagent gas*

It is well known that analyte detectability in LC-MS strongly depends on the composition of the eluent, which generates the reagent gas responsible for the ionisation of the analytes. Careful examination of the background signal represented by the spectrum of the reagent gas is therefore imperative.

*Positive ion mode.* For relevant carrier streams (cf. Table 2), the main ions in the reagent gas spectrum in the PI mode using discharge ionisation are shown in Table 3. Rather different plasmas are obtained in the absence and presence of ammonium acetate or formate. Without an additive, protonated acetonitrile ions make up the reagent gas; solvation of these ions is also observed. The ion at m/z 56 is probably produced by an intermolecular methyl shift[43].

**Table 1** Compounds used in this study.

| *Compound* | *M.W.* | *Chemical class* | *Compound* | *M.W.* | *Chemical class* |
|---|---|---|---|---|---|
| benzenesulfonamide | 157 | (N-substituted) amide | 2,6-dichlorophenol | 162 | chlorophenol |
| propachlor | 211 | N-substituted amide | pentachlorophenol | 264 | chlorophenol |
| metazachlor | 277 | N-substituted amide | 2-nitrophenol | 139 | nitrophenol |
| alachlor | 269 | N-substituted amide | 2,4-dinitro orthocresol | 198 | nitrophenol |
| metachlor | 283 | N-substituted amide | dinoterb | 240 | nitrophenol |
| aniline | 93 | aniline | dinoseb | 240 | nitrophenol |
| 2,6-dimethylaniline | 121 | aniline | dicamba | 220 | carboxylic acid |
| 3,3-dichlorbenzidine | 252 | aniline | triclorpyr | 255 | phenoxy acid |
| 2-chloroaniline | 127 | chloroaniline | dichlorprop | 234 | phenoxy acid |
| 2,4,5-trichloroaniline | 195 | chloroaniline | mecoprop | 214 | phenoxy acid |
| captan | 299 | captan-like (misc.) | permethrin | 300 | pyrethroid (misc.) |
| aldicarbsulfone | 222 | carbamate | maleic hydrazide | 112 | pyridine-like |
| asulam | 112 | carbamate | metamitron | 202 | pyridine-like |
| oxamyl | 219 | carbamate | chloridazon | 221 | pyridine-like |
| carbendazim | 191 | carbamate | bromacil | 260 | pyridine-like |
| aldicarb | 190 | carbamate | bentazone | 240 | pyridine-like |
| barban | 257 | carbamate | diquat | 184 | quaternary ammonium |
| metham sodium | 129 | thiocarbamate | paraquat | 186 | quaternary ammonium |
| thiram | 240 | thiocarbamate | methyl isothiocyanate | 73 | thiocyanate |
| fenaminosulf | 251 | diazosulphonate (misc.) | benzothiazole | 135 | thiocyanate |
| warfarin | 308 | hydroxy-keto-lactone | simazine | 201 | triazine |
| coumafuril | 298 | hydroxy-keto-lactone | atrazine | 215 | triazine |
| sethoxydim | 281 | miscellaneous | diuron | 232 | phenylurea |
| alloxidim | 345 | miscellaneous | monolinuron | 214 | phenylurea |
| dimethoate | 229 | organophosphorus | linuron | 248 | phenylurea |
| phoxim | 298 | organophosphorus | ethylene thiourea | 102 | thiourea |
| bromoxynil | 275 | bromophenol | propylene thiourea | 116 | thiourea |
| | | | isocarbamid | 185 | (thio) urea |



**Table 2** Ionisation modes and carrier-stream compositions examined in this study.

| *Solvent* | *Filament-off* | *Filament-on* | *Discharge* |
|---|---|---|---|
| Water-acetonitrile (50:50, v/v) + | - | PI/NI | PI/NI |
| $NH_4Ac$ | PI/NI | PI/NI | PI/NI |
| $NH_4Fo$ | PI/NI | PI/NI | PI/NI |
| TEAmFo | - | - | NI |
| TPAmFo | - | - | NI |
| chloroacetonitrile (2%) | - | NI | NI |
| chloroacetonitrile (2%) + $NH_4Ac$ | NI | NI | NI |
| chloroacetonitrile (2%) + $NH_4Fo$ | NI | NI | NI |

-, not examined

PI: Positive ion, NI: Negative ion, $NH_4Ac$: 50 mM ammonium acetate, $NH_4Fo$: 50 mM ammonium formate, TEAmFo: 25 mM triethylammonium formate, TPAmFo: 25 mM tripropylammonium formate.

**Table 3** Reagent gas ions and their abundances for various solvent mixtures in the PI mode with discharge ionisation.
Carrier stream: water-acetonitrile (50:50, v/v) without or with ionising additive (50 mM). $NH_4Ac$ = ammonium acetate, $NH_4FO$ = ammonium formate. Flow-rate, 1 ml/min. Discharge voltage, 1 kV.

| | | *Relative abundance (%) in* | | |
|---|---|---|---|---|
| *m/z* | *Ion* | *$H_2O/CH_3CN$* | *$H_2O/CH_3CN$ +$NH_4Ac$* | *$H_2O/CH_3CN$ +$NH_4FO$* |
| 18 | $[NH_4]^+$ | | 5 | 5 |
| 19 | $[H_3O]^+$ | 5 | | |
| 36 | $[H_2O.NH_4]^+$ | | 2 | 3 |
| 42 | $[CH_3CN.H]^+$ | 100 | 1 | 1 |
| 56 | $[CH_3CN.CH_3]^+$ | 35 | 2 | |
| 59 | $[CH_3CN.NH_4]^+$ | | 100 | 100 |
| 60 | $[CH_3CN.H_2O.H]^+$ | 3 | | |
| 77 | $[CH_3CN.H_2O.NH_4]^+$ | | 2 | 5 |
| 83 | $[(CH_3CN)_2.H]^+$ | 55 | 5 | 15 |
| 100 | $[(CH_3CN)_2.NH_4]^+$ | | 60 | 75 |
| 124 | $[(CH_3CN)_3.H]^+$ | 3 | 1 | 2 |



The use of filament-on instead of discharge ionisation enhanced the ion at m/z 83, which then became the base peak. When ammonium acetate or formate was added, ammonium ions were formed which produced clusters with acetonitrile and water molecules. This resulted in a base peak at m/z 59, due to the $[CH_3CN.NH_4]^+$ ion. Protonated acetonitrile molecules (m/z 42 and 83) were still present, but to a much lower extent, i.e. max. 5% relative abundance. Filament-off and filament-on ionisation – i.e. softer ionisation than discharge ionisation – enhanced the intensity of the $[(CH_3CN)_2.NH_4]^+$ ion at m/z 100 to 100 % and 90 %, respectively.

*Negative ion mode.* Without an additive, the mass spectrum of the reagent gas showed deprotonated acetonitrile and water molecules as well as adducts of these ions with water or acetonitrile. When ammonium acetate or formate was added the main ions in the plasma were acetate or formate and clusters of these ions with water, as can be read from Table 4. No significant change compared to ammonium formate addition was observed when TEAmFo or TPAmFo was used as ionising additive. The use of filament-off and filament-on instead of discharge ionisation resulted in a higher relative intensity of the multiple adduct ions, but the total intensity dropped by a factor of 8 upon going from discharge ionisation via filament-on to filament-off ionisation.

Carrier streams containing chloroacetonitrile were already examined in some detail before[11,13,14]; they were also used in this work. Briefly, the main ions which appeared in the mass spectra of the background were adducts of chloride ions with water, acetonitrile, chloroacetonitrile, acetic acid and formic acid.

**Table 4** Reagent gas ions and their abundances for various solvent mixtures in the NI mode with discharge ionisation.

Carrier stream: (A) water-acetonitrile (50:50, v/v) with (B) 50 mM ammonium acetate, (C) 50 mM ammonium formate, (D) 25 mM triethylammonium formate, (E) 25 mM tripropylammonium formate, (F) 2% chloroacetonitrile, (G) chloroacetonitrile + ammonium acetate and (H) chloroacetonitrile + ammonium formate. Flow-rate, 1 ml/min. Discharge voltage, 1kV.

| | | *Relative abundance (%) in* | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| *m/z* | *Ion* | *A* | *B* | *C* | *D* | *E* | *F* | *G* | *H* |
| 17 | $[OH]^-$ | 1 | | | | | | | |
| 26 | $[CN]^-$ | 3 | | | | | | | |
| 35 | $[H_2O.OH]^-$ | 10 | | | | | | | |
| 40 | $[CH_2CN]^-$ | 100 | | | | | | | |
| 53 | $[(H_2O)_2.OH]^-$ | 40 | | | | | | | |
| 58 | $[CH_3CN.H_2O\text{-}H]^-$ | 35 | | | | | | | |
| 71 | $[(H_2O)_3.OH]^-$ | 5 | | | | | | | |
| 76 | $[CH_3CN.(H_2O)_2\text{-}H]^-$ | 5 | | | | | | | |
| 81 | $[CH_3CN.CH_2CN]^-$ | 15 | | | | | | | |
| 59 | $[CH_3COO]^-$ | | 60 | | | | | 1 | |
| 77 | $[CH_3COOH.H_2O\text{-}H]^-$ | | 2 | | | | | | |
| 95 | $[CH_3COOH.(H_2O)_2\text{-}H]^-$ | | 1 | | | | | | |
| 119 | $[CH_3COOH.CH_3COO]^-$ | | 100 | | | | | 2 | |
| 45 | $[HCOO]^-$ | | | 70 | 10 | 10 | | | 1 |
| 63 | $[HCOOH.H_2O\text{-}H]^-$ | | | 1 | 2 | 2 | | | |
| 91 | $[HCOOH.HCOO]^-$ | | | 100 | 100 | 100 | | | 2 |
| 137 | $[HCOOH)_2.HCOO]^-$ | | | 8 | 10 | 30 | | | |
| 35 | $[Cl]^-$ | | | | | | 40 | 15 | 25 |
| 53 | $[Cl.H_2O]^-$ | | | | | | 40 | 2 | 3 |
| 76 | $[ClCH_2CN]^-$ | | | | | | 30 | 2 | 2 |
| 81 | $[Cl.HCOOH]^-$ | | | | | | | | 100 |
| 95 | $[Cl.CH_3COOH]^-$ | | | | | | | 100 | |
| 110 | $[Cl.ClCH_2CN]^-$ | | | | | | 100 | 5 | 1 |
| 128 | $[Cl.ClCH_2CN.H_2O]^-$ | | | | | | 10 | | |



## *Mass spectra*

*Positive ions.* Table 5 presents the results obtained when using discharge ionisation in the PI mode and a carrier stream of water-acetonitrile (50:50, v/v) with or without ammonium acetate or ammonium formate, for one or two representatives of each group of pesticides. If, in a few cases, other members of a group showed a different behaviour, this is mentioned in the text. The thiocarbamates, hydroxy-keto-lactones, captan, permethrin, substituted phenols and the phenoxy and carboxylic acids are not included in this table because they did not show a noticeable signal even when 200 ng of analyte were injected.

*Without additive.* When no additive was present in the carrier stream, $[M+H]^+$ ions were the base peak for most of the pesticides. However, for the carbamates severe fragmentation was observed (see below). The fragments have a rather high intensity. A relatively hard ionisation plasma, i.e. a plasma which transfers more energy than is required for ionisation, is present in the source in this case. This can be explained as follows. Especially when

**Table 5** Main ions and their relative abundances for representative polar pesticides in FIA-PI-TSP-MS with discharge ionisation.
Carrier stream; (A) water-acetonitrile (50:50, v/v) with (B) 50 mM ammonium acetate and (C) 50 mM ammonium formate; flow-rate, 1 ml/min. Discharge voltage, 1 kV.

| | | *Relative abundance (%) in* | | |
|---|---|---|---|---|
| *m/z* | *Proposed identification* | *A* | *B* | *C* |
| Propachlor | | | | |
| 212 | $[M+H]^+$ | 100 | 100 | 100 |
| 229 | $[M+NH_4]^+$ | | 15 | 20 |
| 2,6-Dimethylaniline | | | | |
| 122 | $[M+H]^+$ | 100 | 100 | 100 |
| 163 | $[M+H+CH_3CN]^+$ | 15 | 15 | 15 |
| Oxamyl | | | | |
| 147 | $[M-CON(Me)_2]^+$ | | 5 | 70 |
| 163 | $[M+H-CONCH_3]^+$ | 100 | 50 | 35 |
| 220 | $[M+H]^+$ | 2 | 15 | 20 |
| 237 | $[M+NH_4]^+$ | | 100 | 100 |
| 251 | | | 2 | 10 |
| Alloxidim | | | | |
| 266 | $[M+H-C_3H_6O]^+$ | 100 | 100 | 100 |
| 324 | $[M+H]^+$ | | 60 | 60 |
| Dimethoate | | | | |
| 230 | $[M+H]^+$ | 100 | 100 | 100 |
| 247 | $[M+NH_4]^+$ | | 10 | 10 |
| Chloridazon | | | | |
| 222 | $[M+H]^+$ | 100 | 100 | 100 |
| Diquat | | | | |
| 172 | $[M-14]^+$ | n.d. | 100 | 80 |
| 187 | $[M+H]^+$ | n.d. | 90 | 100 |
| Benzothiazole | | | | |
| 136 | $[M+H]^+$ | 100 | 100 | 100 |
| 177 | $[M+H+CH_3CN]^+$ | | 20 | 20 |
| Simazine | | | | |
| 202 | $[M+H]^+$ | 100 | 100 | 100 |
| 243 | $[M+H+CH_3CN]^+$ | | 10 | 8 |
| Monolinuron | | | | |
| 215 | $[M+H]^+$ | 100 | 100 | 100 |
| 232 | $[M+NH_4]^+$ | | 10 | 10 |
| Propylenethiourea | | | | |
| 117 | $[M+H]^+$ | 100 | 100 | 100 |
| 158 | $[M+H+CH_3CN]^+$ | | 15 | 15 |

n.d. = not detected

filament-on or discharge ionisation is used, the analyte ionisation resembles that occurring under CI conditions. Water-acetonitrile generates a plasma consisting of protonated acetonitrile and some adducts, whereas the carrier streams containing an ammonium-salt generate an ammonia plasma (cf. above). The PAs of ammonia and acetonitrile are 858 KJ/mole and 795 KJ/mole, respectively[46]. That is, ionising a compound with ammonia is less exothermic

than ionising it with acetonitrile, and more fragmentation will be observed in the latter case. This is demonstrated in Figure 1, where the mass spectra of metazachlor, a N-substituted amide, without and with ammonium acetate show more abundant fragmentation – i.e. ions at m/z 142, 210 and 244 – in the absence of ammonium acetate. This is especially true for the fragment at m/z 142. The fragments at m/z 210 and 142 are produced by the loss of the pyrazole moiety and the two substituents on the secondary nitrogen, respectively. The ion at 244, also observed when ammonium acetate is present in the carrier stream, is formed by an exchange of a chlorine atom by a proton in the protonated molecule. In the case of metachlor and alachlor an ion at $[M-31]^+$, corresponding to the loss of methanol from the protonated molecular ion, is observed. In the latter case, this ion even is the base peak. As another example, the three N-methylcarbamates, oxamyl, aldicarbsulfone and aldicarb, all show intense $[M+H-CH_3NCO]^+$ ions. The intensity of these ions is much higher than in the presence of an additive, e.g. 100% instead of 45%. These ions were earlier reported in LC-MS with a TSP [9,38,40,47] as well as a DLI[48] interface. With some of the miscellaneous compounds, fragmentation was also observed. For instance, alloxidim shows a base peak at m/z 266, corresponding to $[M+H-58]^+$. This ion is tentatively identified as $[M+H-C_3H_6O]^+$. One of the pyridine-like compounds, bentazone, showed a very intense fragment at $[M-61]^+$, which was the base peak without and with additive. This ion is probably formed by the addition of an ammonium ion with subsequent loss of the $NH-SO_2$ moiety; however, further investigation seems necessary.

*With additive.* When ammonium acetate or formate is used as additive, next to $[M+H]^+$ which still is the base peak for most pesticides, $[M+NH_4]^+$ is present for 10–30%. For the carbamates the latter ion is the base peak. The carbamates also produce an ion at $[M+32]^+$. Saar and Salomon[47] also reported this ion with low abundance (<1%) and proposed the formation of $[M + CH_3NH_3]^+$, from the methanol and ammonia used in the eluent, via a multistep reaction. However, our use of acetonitrile rules out this suggestion. For the rest, an ion at $[M+CH_3NH_2CO]^+$ was present at a low level (1–2%). Adducts of the carbamates with acetonitrile reported by others[15,39,40], were not observed in this study. The above mentioned fragmentations are illustrated for oxamyl in Figure 2, where the mass spectra in the absence and presence of ammonium formate clearly show the adducts at m/z 237 and 251, corresponding to $[M+NH_4]^+$ and $[M+32]^+$, respectively.

The anilines (except alachlor), organophosphorus and pyridine-like compounds, triazines and ureas mainly show protonated or ammoniated molecular ions and in some cases an adduct with protonated acetonitrile. The $[M+H+CH_3CN]^+$ ion, which is reported to be very prominent in the case of the triazines by Durand *et al.*[15] has a relative abundance of only 10% in our study. The use of different TSP interfaces may well explain this discrepancy[13]. With the anilines, the pyridine-like compounds, thiocyanates and (thio)ureas the same ion is also present for 5–20%. This indicates that the PA of these compounds is comparable to or slightly higher than that of ammonia[46].

In the case of diquat a protonated molecular ion and a not yet understood fragment at $[M-14]^+$ were observed. It is interesting to add that in pure water, intense signals at $M^{+\cdot}$ and $[M-H]^+$ with hardly any fragmentation[44] were observed for paraquat and diquat, respectively. The latter ion can probably be explained by an unusual fragmentation of the intact dication as suggested by Volmer *et al.*[45]. The formation of the $M^{+\cdot}$ ion, observed for paraquat, is not yet fully understood. Obviously, a reduction takes place in the interface, spray or ion source.

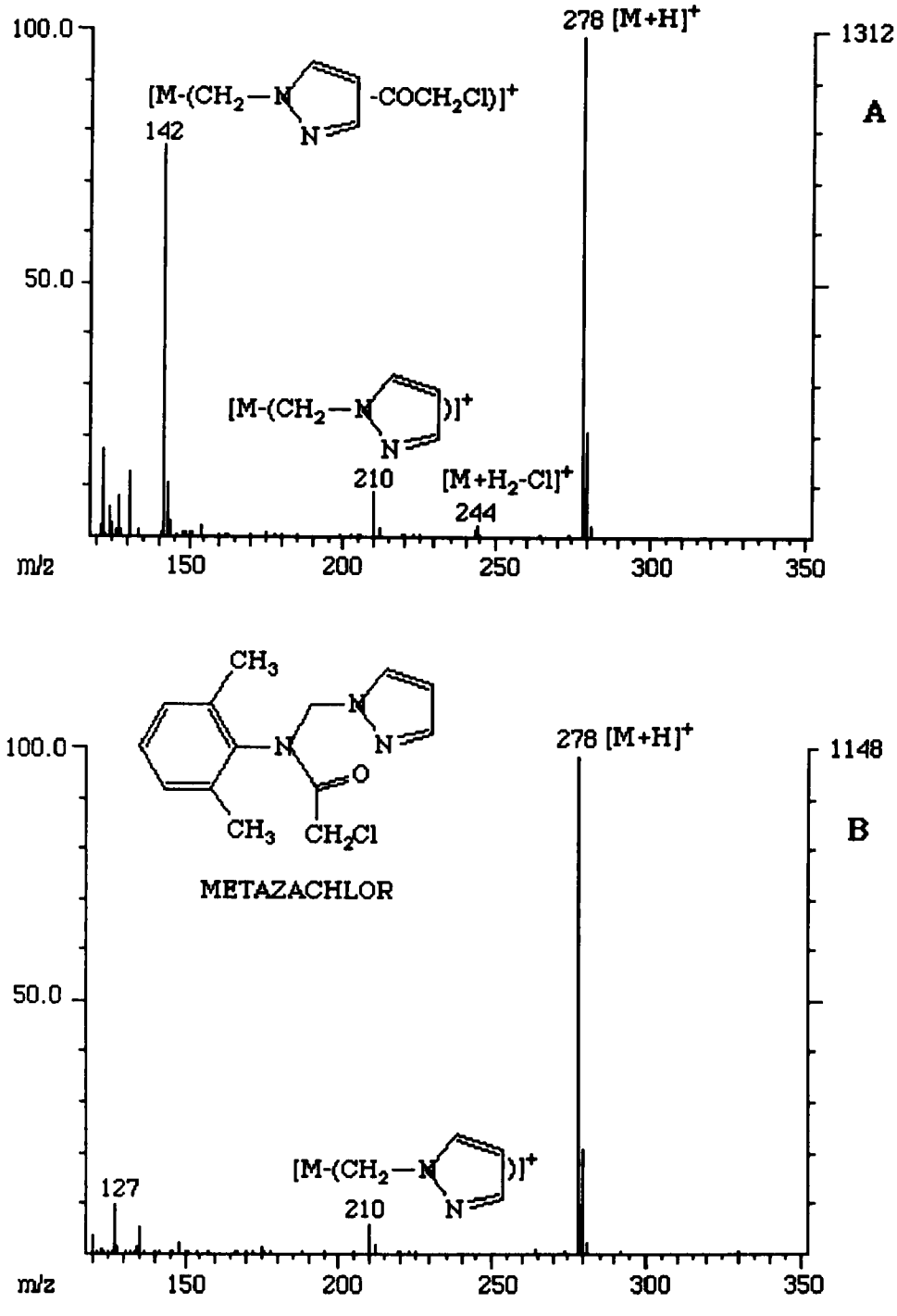



**Figure 1** Mass spectra of metazachlor in the PI mode using [A] acetonitrile-water (50:50, v/v), containing [B] 50 mM ammonium acetate as carrier stream. Flow, 1 ml/min. Discharge ionisation, 1 kV. Amount injected, ca. 200 ng.

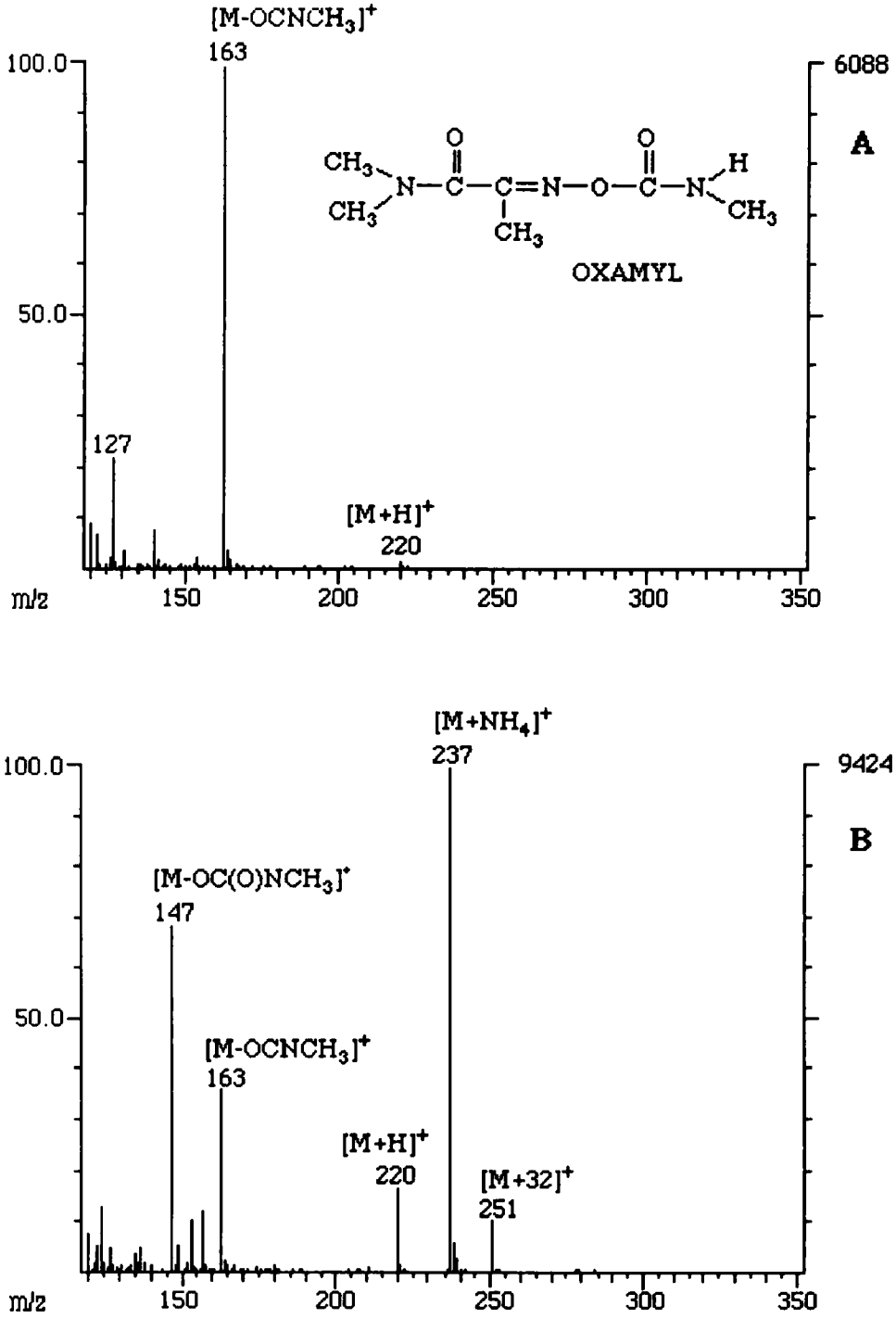



**Figure 2** Mass spectra of oxamyl in the PI mode using [A] acetonitrile-water (50:50, v/v), containing [B] 50 mM ammonium formate as carrier stream. Flow, 1 ml/min. Discharge ionisation, 1 kV. Amount injected, ca. 200 ng.

*Negative ions.* For the majority of the polar pesticides in this group more fragmentation is observed in the NI than in the PI mode because of the better stabilisation of the negative charge in the fragment than in the deprotonated molecular ion. Unfortunately, fragment ions are often produced by rearrangements which are not well understood.

Table 6 shows the ions observed using discharge ionisation in the NI mode for one or two representatives of the various groups of pesticides. However, not all examples discussed in the text are included in the table, and vice versa. The N-substituted amides, anilines, quaternary ammonium compounds, thiocyanates and thioureas are not represented in the table because they exhibit at best very weak signals for injected amounts of 200 ng. Only discharge ionisation is discussed because in this case the highest degree of fragmentation is observed while the analyte detectability was 3–5 fold better than with filament-on ionisation. The use of filament-off ionisation resulted in ion intensities similar to or up to 8-fold lower than in filament-on ionisation; the relative abundance of adduct ions was however higher in the former case.

*Without additive.* When no additive was present, most of the test compounds showed deprotonated molecular ions, even the N-substituted amides and anilines, which produced very weak signals. Some groups however, e.g. the carbamates, organophosphorus compounds, phenoxy acids and fenaminosulf, captan, permethrin and alloxidim, displayed extensive fragmentation. For example, aldicarbsulfone showed only the $[M-76]^-$ ion, which can be identified as $[M-H-HOOCNHCH_3]^-$, and dimethoate showed intense $[M-CH_{2CONHCH;3}]^-$ and $[CH_3OP(S)SCH_3]^-$ ions (see Figure 3). The latter ion can be formed from the molecular ion after the loss of methylisocyanate and a methoxy group. The relatively high fragmentation rate can be explained by the $\Delta H°_{acid}$ of the reagent ions. As the acidities of water and formic acid are 1635 KJ/mole and 1445 KJ/mole, respectively, the exothermicity of analyte deprotonation will obviously be higher when no additive is used, and therefore more fragmentation will be observed.

*With ammonium acetate or formate.* The use of additives in LC-TSP-MS in the NI mode can yield additional information through adduct formation. Generally speaking, the use of formate results in more adduct formation than that of acetate, because of its higher $\Delta H°_{acid}$, viz. 1445 KJ/mole versus 1457 KJ/mole[46].

For most of the compounds tested, deprotonated molecular ions and/or adducts with formate or acetate ions were observed as base peaks. The amount of adducts present in the mass spectra also depended on the mode of ionisation used. Generally speaking, the filament-off mode of ionisation produced the highest abundance of adduct ions, e.g. up to 100% for dimethoate, while with discharge ionisation the abundance of these ions decreased to only 10–40%. With some analytes however, e.g. sethoxidim and aldicarbsulfone, the formate or acetate adduct was still present in the mass spectrum for 100%.

Even when additives were used, severe fragmentation was observed with fenaminosulf, captan, alloxidim, permethrin and the N-methylcarbamates. As in carrier streams containing no ammonium acetate or formate, captan produced the $[M-(SCCl_3)]^-$ ion as base peak. Aldicarbsulfone produced fragments at m/z $[M-97]^-$ and $[M-12]^-$, which can tentatively be identified as $[CH_3SO_2H + HCOO]^-$ and $[M+HCOO-OCNCH_3]^-$, respectively. The acetate analogue of the latter ion was also observed under filament-off conditions with ammonium acetate as additive: the ion at m/z 224 is the fragment produced by the loss of the isothiocyanate group from the acetate adduct of the molecule. Some of these ions were also

**Table 6** Main ions and their relative abundances for representative polar pesticides in FIA-NI-TSP-MS with discharge ionisation.

Carrier stream; (A) water-acetonitrile (50:50, v/v) with (B) 50 mM ammonium acetate, (C) 50 mM ammonium formate, (D) 25 mM triethylammonium formate, (E) 25 mM tripropylammonium formate, (F) water-acetonitrile-chloroacetonitrile (49:49:2), (G) 50 mM ammonium acetate, (H) 50 mM ammonium formate; flow-rate, 1 ml/min. Discharge voltage, 1 kV.

| | | *Relative abundance (%) in* | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| *m/z* | *Proposed identification* | *A* | *B* | *C* | *D* | *E* | *F* | *G* | *H* |
| Aldicarbsulfone | | | | | | | | | |
| 125 | $[CH_3SO_2H+HCOO]^-$ | | | 30 | 30 | 90 | | | |
| 146 | $[M-H-HOOCNHCH_3]^-$ | 100 | | | | | | | |
| 210 | $[M+HCOO-OCNCH_3]^-$ | | | 60 | | | | | |
| 257 | $[M+Cl]^-$ | | | | | | 100 | 100 | 100 |
| 267 | $[M+HCOO]^-$ | | | 100 | 100 | 100 | | | |
| 281 | $[M+CH_3COO]^-$ | | 100 | | | | | 40 | |
| Carbendazim | | | | | | | | | |
| 132 | $[M-H-OCOCH_2]^-$ | 15 | | 100 | | | | | |
| 158 | | 25 | 95 | 90 | | | | | |
| 168 | $[M+Cl-OCOCH_2]^-$ | | | | | | | | 10 |
| 178 | $[M+HCOO-OCOCH_2]^-$ | | | 50 | 100 | 100 | | | |
| 190 | $[M-H]^-$ | 100 | 100 | | 50 | 25 | | 10 | |
| 226 | $[M+Cl]^-$ | | | | | | 100 | 100 | 100 |
| 236 | $[M+HCOO]^-$ | | | | 15 | 5 | | | |
| 250 | $[M+CH_3COO]^-$ | | 5 | | | | | | |
| Warfarin | | | | | | | | | |
| 250 | $[M-C_3H_6O]^-$ | | | 20 | 40 | 30 | 50 | | |
| 307 | $[M-H]^-$ | 100 | 100 | 100 | 100 | 100 | 100 | 100 | 100 |
| Sethoxydim | | | | | | | | | |
| 238 | $[M-C_2H_3O]^-$ | | | 30 | 15 | | 50 | | |
| 280 | $[M-H]^-$ | 100 | | | | | | | |
| 282 | $[M+H]^-$ | | 90 | 5 | 30 | 30 | | | |
| 297 | $[M+CH_3COO-C_2H_3O]^-$ | | 15 | | | | | | |
| 326 | $[M+HCOO]^-$ | | 100 | 100 | 100 | 100 | | | |
| 340 | $[M+CH_3COO]^-$ | | 20 | | | | | | |
| Dimethoate | | | | | | | | | |
| 141 | $[CH_3OPSSCH_3]^-$ | 60 | | | | | | | |
| 157 | $[M-CH_2CONHCH_3]^-$ | 100 | | 100 | 100 | 100 | | | |
| 214 | $[M-15]^{-*}$ | 5 | 85 | 15 | 5 | 10 | 100 | 20 | 40 |
| 228 | $[M-H]^-$ | 80 | 5 | | | | | | |
| 264 | $[M+Cl]^-$ | | | | | | 75 | 100 | 100 |
| 274 | $[M+HCOO]^-$ | | | 10 | 65 | 65 | | | |
| 288 | $[M+CH_3COO]^-$ | | 100 | | | | | | |
| Bromoxynil | | | | | | | | | |
| 196 | $[M-Br]^-$ | | 10 | 15 | 10 | 20 | | | |
| 274 | $[M-H]^-$ | 100 | 100 | 100 | 100 | 100 | 100 | 100 | 100 |
| Dinitro orthocresol | | | | | | | | | |
| 181 | $[M-OH]^-$ | 20 | 25 | 60 | | | | | |
| 197 | $[M-H]^-$ | | | | 100 | 100 | 100 | 100 | 100 |
| 198 | $M^{\cdot-}$ | 100 | 100 | 100 | | | | | |
| 243 | $[M+HCOO]^-$ | | | | 5 | | | | 8 |

**Table 6** Continued.

| | | *Relative abundance (%) in* | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| *m/z* | *Proposed identification* | *A* | *B* | *C* | *D* | *E* | *F* | *G* | *H* |
| Mecoprop | | | | | | | | | |
| 213 | $[M-H]^-$ | 100 | 100 | 100 | 100 | 100 | 100 | 25 | 40 |
| 249 | $[M+Cl]^-$ | | 10 | 10 | 10 | 10 | 55 | 100 | 100 |
| 259 | $[M+HCOO]^-$ | | | 15 | 65 | 80 | | | |
| 273 | $[M+CH_3COO]^-$ | | 5 | | | | | | |
| Chloridazon | | | | | | | | | |
| 220 | $[M-H]^-$ | 100 | 100 | 100 | 85 | 90 | 15 | 20 | |
| 256 | $[M+Cl]^-$ | | | 15 | 10 | 7 | 100 | 100 | 100 |
| 266 | $[M+HCOO]^-$ | | | 12 | 100 | 100 | | | |
| 280 | $[M+CH_3COO]^-$ | | 5 | | | | | | |
| Simazine | | | | | | | | | |
| 182 | $[M+HCOO-Cl-C_2H_5]^-$ | | | 30 | | | | | |
| 200 | $[M-H]^-$ | 100 | | | | | 100 | | |
| 203 | $[M+HCOO-C_2H_5N]^-$ | | | 100 | 100 | 100 | | | |
| 242 | $[M+CH_3COO-H_2O]^-$ | | 100 | | | | | | |
| 283 | | | | 10 | | | | | |
| Diuron | | | | | | | | | |
| 160 | $[M-CON(Me)_2]^-$ | 35 | | | | | | | |
| 203 | | | | 100 | | | | | |
| 217 | | | 10 | | | 20 | 20 | | |
| 231 | $[M-H]^-$ | 100 | 25 | 25 | 100 | 70 | | | |
| 239 | | | 50 | 30 | | | | | 100 |
| 249 | $[M+HCOO-CO]^-$ | | | 95 | | 15 | | | |
| 262 | | | 100 | | | | | | |
| 267 | $[M+Cl]^-$ | | | | | 5 | 100 | 100 | |
| 277 | $[M+HCOO]^-$ | | | 25 | 70 | 100 | | | |

* = $M+OH^-$,$Cl^-$,$HCOO^-$ and subsequent loss of $CH_3OH$, $CH_3Cl$,$HCOOCH_3$ or $CH_3OOCCH_3$.



reported by Voyksner *et al.*[21]. Asulam showed a very intense $[M-NHC(O)OCH_3]^-$ ion in the presence of formate. The thiocarbamates metham sodium and thiram showed unidentified base peaks at $[M+45]^-$ and $[M+63]^-$, respectively.

The hydroxy-keto-lactones produced a group-characteristic ion at $[M-58]^-$, corresponding to the loss of $C_3H_6O$ via ß-cleavage[50].

Sethoxydim produced a fragment at m/z $[M-43]^-$ corresponding to the loss of a $C_2H_3O$ group. The adduct of this ion with acetate was also observed. Alloxidim showed an intense specific fragment at $[M-57]^-$, corresponding to $[M-C_3H_5O]^-$, in all eluents tested.

In the case of dimethoate an ion at $[M-15]^-$ was observed which probably corresponds to the addition of $OH^-$, $Cl^-$, acetate or formate, depending on the additive, with a subsequent loss of methanol, methylchloride, methyl acetate or methyl formate. These kinds of reactions—addition of reagent ion and subsequent loss of a small molecule—are quite common in TSP-MS, as reported by Maeder[30]. The straightforward attribution of this ion to a $[M-CH_3]^-$ ion, as suggested by others [17,33,51,52], is somewhat strange and the sudden occurrence of the negative charge is not readily explained. Phoxim, the other organophosphorus compound

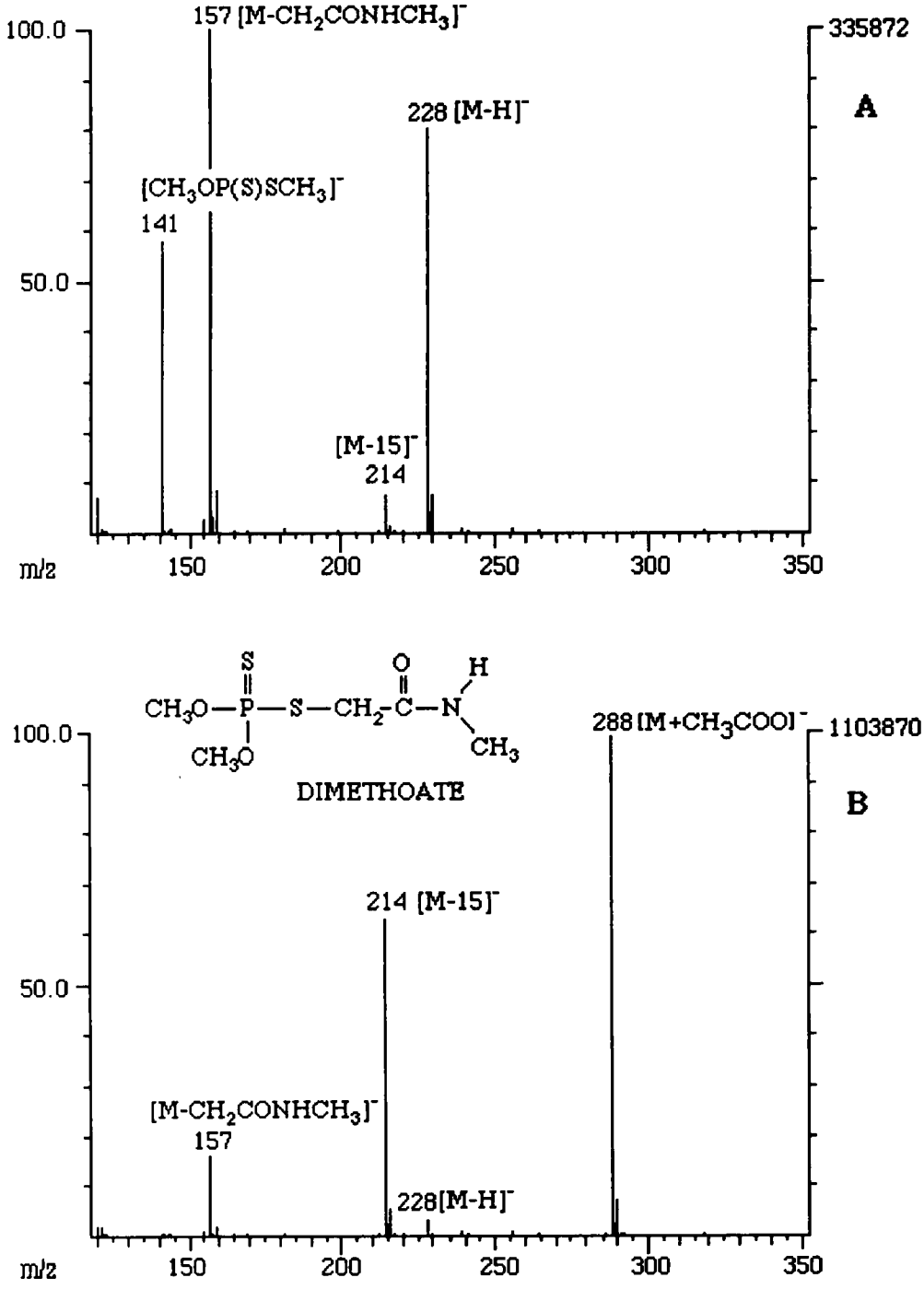



**Figure 3** Mass spectra of dimethoate in the NI mode using [A] acetonitrile-water (50:50, v/v), containing [B] 50 mM ammonium acetate as carrier stream. Flow, 1 ml/min. Discharge ionisation, 1 kV. Amount injected, ca. 200 ng.

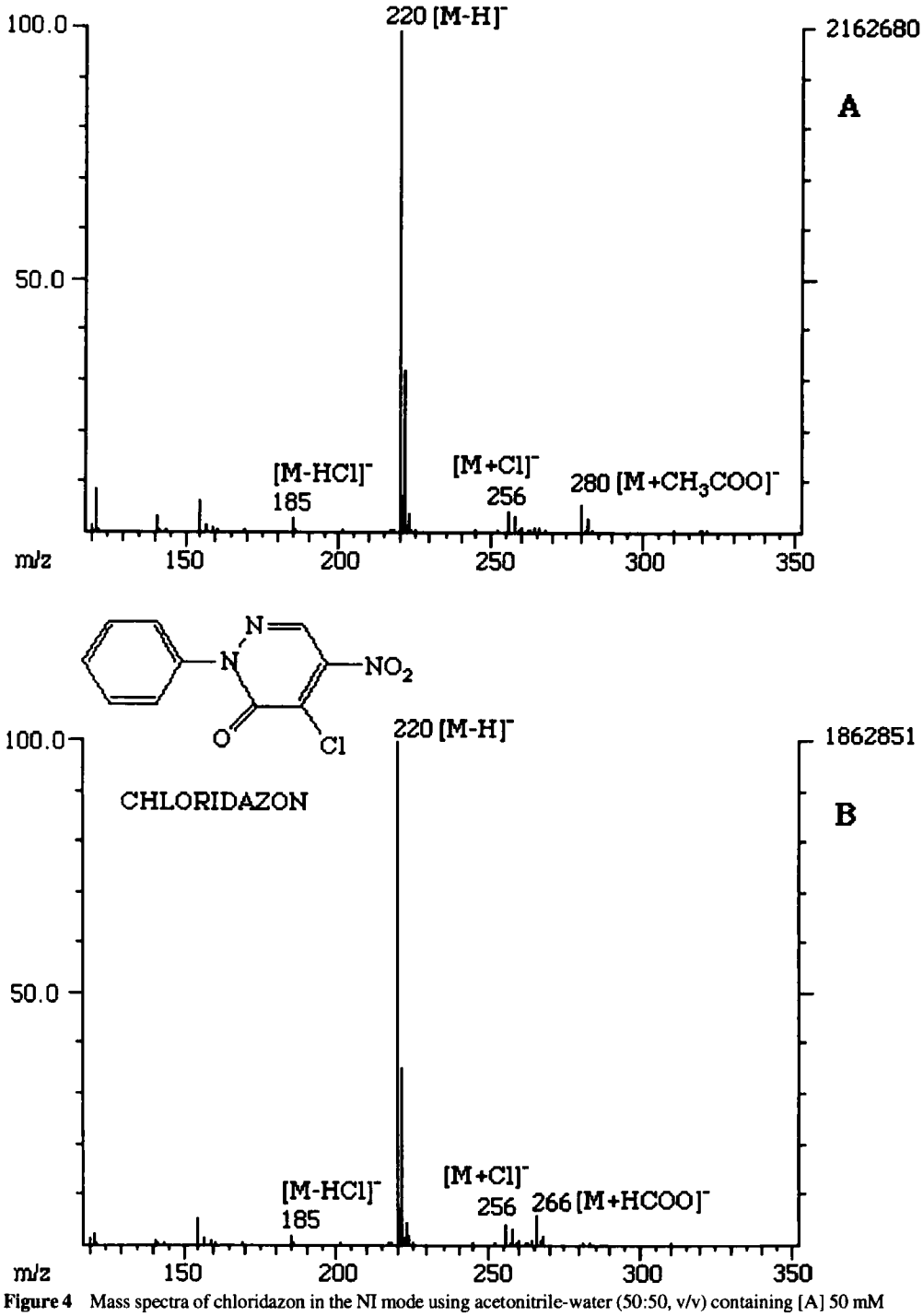
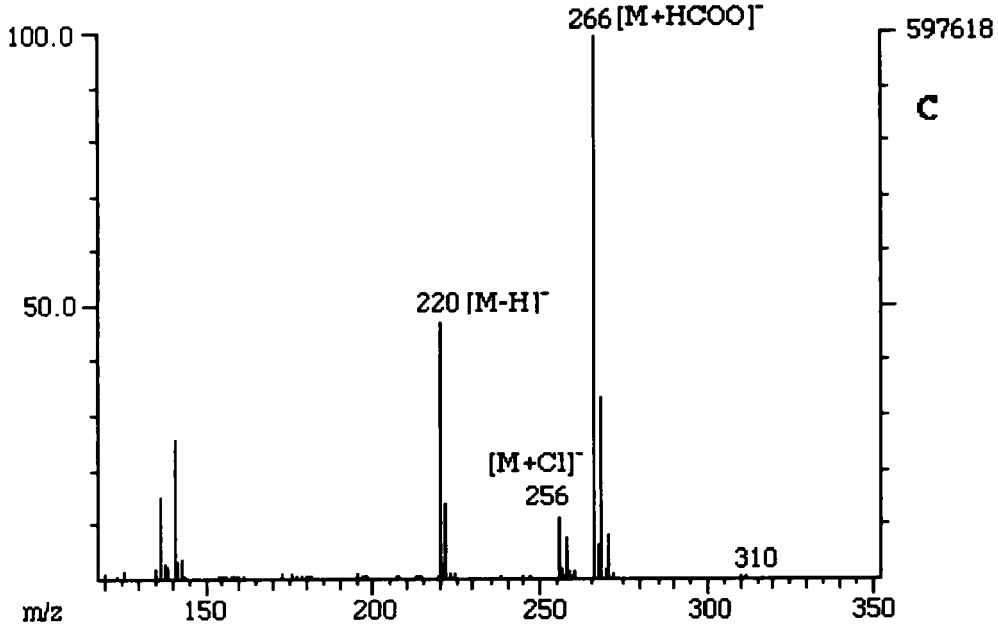
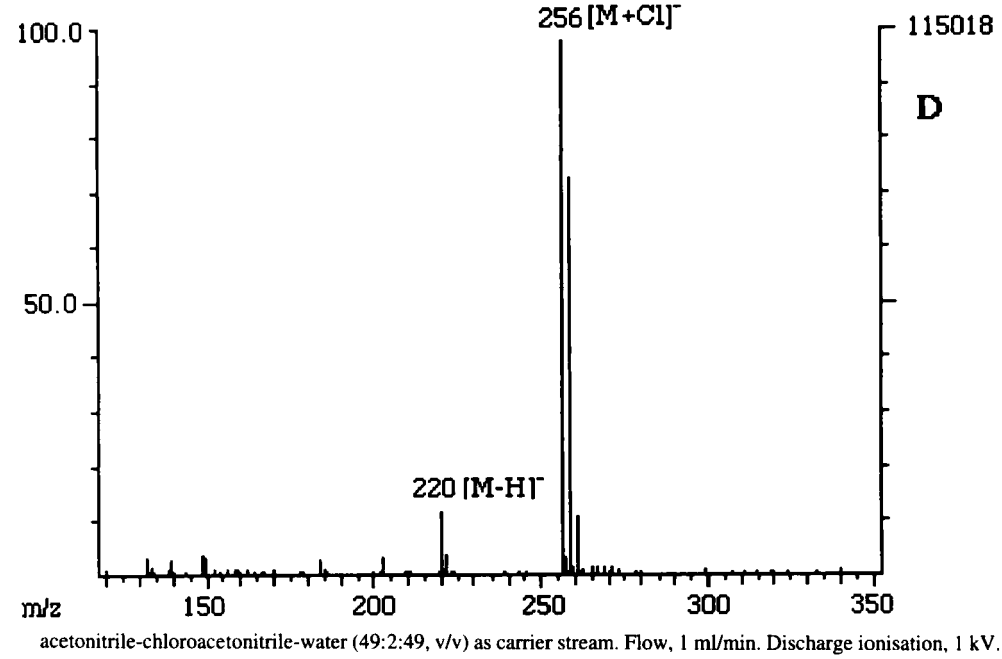



**Figure 4** Mass spectra of chloridazon in the NI mode using acetonitrile-water (50:50, v/v) containing [A] 50 mM ammonium acetate,[B] 50 mM ammonium formate or [C] 25 mM triethylammonium formate, and [D]




acetonitrile-chloroacetonitrile-water (49:2:49, v/v) as carrier stream. Flow, 1 ml/min. Discharge ionisation, 1 kV. Amount injected, ca. 200 ng.

tested, showed, as in all other cases in the NI mode, a base peak at m/z 169, corresponding to the $[(C_2H_5O)_2P(S)O]^-$ ion. Hardly any fragments were observed.

For the nitrophenols, next to ion-molecule reactions like deprotonation, electron-capture behaviour was observed. Resonance electron capture—quite common for compounds that contain nitro groups—as well as dissociative electron capture (ions at m/z $[M-17]^-$, i.e. $[M-OH]^-$ ions) was observed.

Some phenoxy acids gave ions produced by the loss of the carboxylic moiety, with or without the alkyl chain. These ions were also observed by Jones *et al.*[20] using a TSP interface, and admittedly, methanol-water mixtures containing ammonium acetate as mobile phase.

The triazines showed ions at m/z $[M+2]^-$ and $[M-19]^-$, especially when ammonium formate was used in the carrier stream. The ions can be tentatively explained by the loss of a side chain, i.e. the $NCH_2CH_3$group, and the loss of the ethyl moiety and the chlorine atom (confirmed by the absence of the chlorine isotope), respectively, with subsequent adduct formation with formate. This is the first time these ions are reported; until now only PI data were published for triazines.

The phenylureas also showed fragmentation when additives were used. They produced fragments at $[M-14]^-$, $[M-28]^-$ or $[M-29]^-$, depending on the groups attached to the nitrogen atom, and $[M-CONR_2]^-$, with abundances of up to 100%. Some of these ions were also observed by Barcelo[14] and Voyksner and Haney[10]. Next to adducts of formate or acetate with the molecular ions, adducts with fragments also showed up in the spectra. For example, ions at m/z $[M+15]^-$ and $[M+17]^-$ can be attributed to $[M+HCOO-CH_2O]^-$ and $[M+HCOO-CO]^-$, respectively. The ions at m/z $[M+7]^-$ and $[M+30]^-$ have not been identified yet. A study on the ions observed and the dependence of their nature on the structure of the phenylureas will be published elsewhere[49].

As is to be expected from the $\Delta H°_{acid}$, the adducts with formate are more abundant than with acetate. This was especially observed for sethoxydim, some phenols, the phenoxy acids, the pyridine-like compounds and the phenylureas. As an illustration of this effect, the mass spectra of chloridazon in Figure 4 with discharge ionisation, using various carrier streams, clearly demonstrate the usefulness of the adducts with acetate and—distinctly more so—with formate or chlorine for molecular weight confirmation. The chloride adducts observed without added chloroacetonitrile, are produced by self-chlorination—a process originating from pyrolysis of part of the analyte molecules and subsequent addition to intact molecules[13–15,20]. Self-chlorination or self-bromination was observed for nearly all phenols, pyridine-like compounds, phenoxy acids and the carbamate, barban. The $[M+Cl]^-$ or $[M+Br]^-$ ion was present in the spectra with a relative abundance of 30–100%.

*With TEAmFo or TPAmFo.* When using discharge ionisation in the presence of ammonium formate, most of the mass spectra contained a $[M+HCOO]^-$ ion. Its relative abundance for the carboxylic and phenoxy acids was typically 5–10%. When TPAmFo was used as ionising additive instead of ammonium formate for, e.g., triclorpyr, next to an enhancement of the relative abundance of this ion from 3% to 45% (see Figure 5) also the signal intensity was enhanced. This is in accordance with the results obtained by Steffenrud *et al.*[29] who reported—next to an increase of signal intensity—also an enhanced relative abundance of the $[M+HCOO]^-$ ions for some acidic and amphoteric compounds with TEAmFo instead of ammonium acetate. The enhanced adduct formation can be explained by the PA of the bases and the $\Delta H°_{acid}$ of the acids involved. In the case of TEAmFo, triethylamine (PA=967

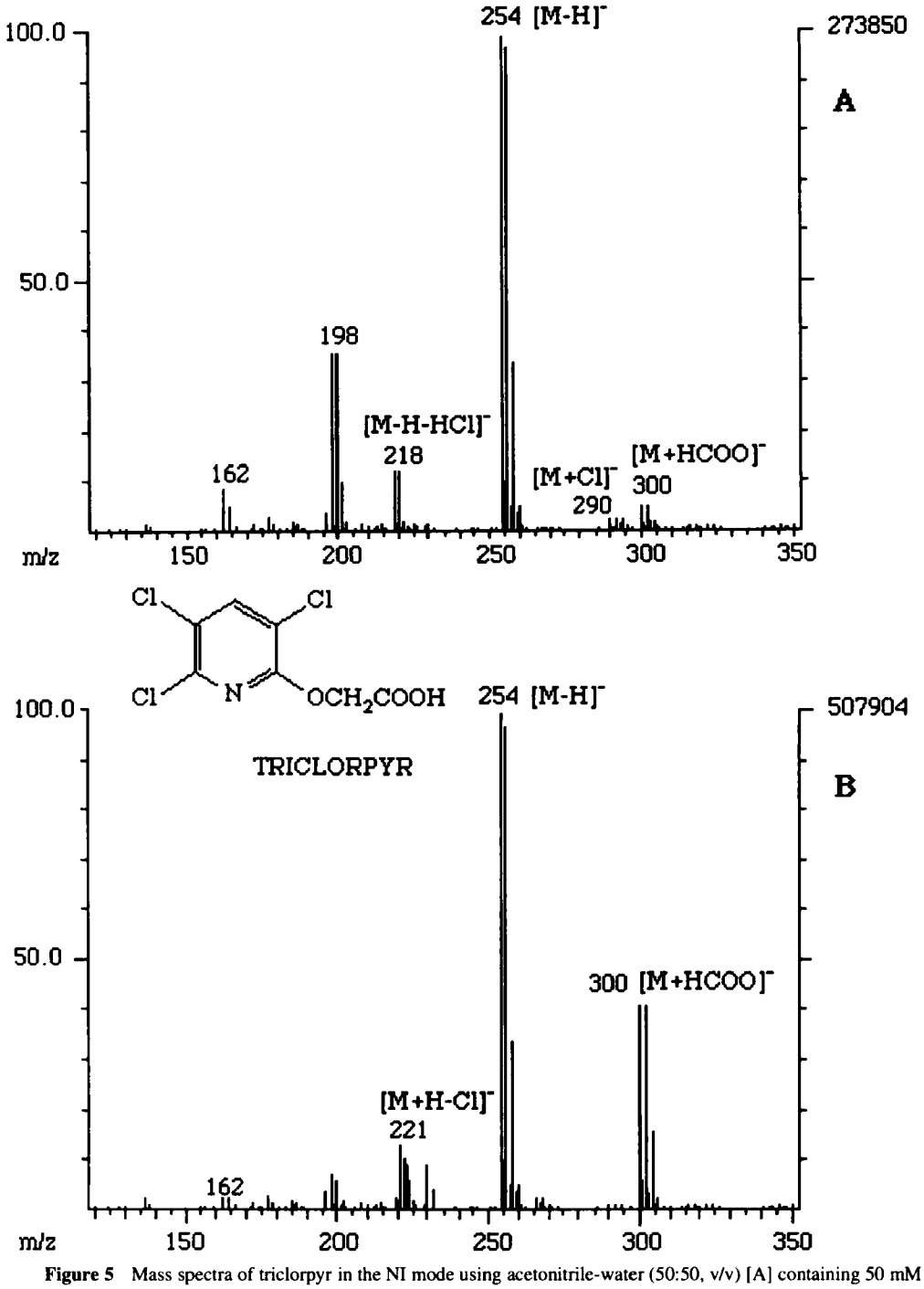



**Figure 5** Mass spectra of triclorpyr in the NI mode using acetonitrile-water (50:50, v/v) [A] containing 50 mM ammonium formate and [B] containing 25 mM tripropylammonium formate, as carrier stream. Flow, 1 ml/min. Discharge ionisation, 1 kV. Amount injected, ca. 200 ng.

KJ/mole), which is a stronger base than ammonia (PA=858 KJ/mole), is present in the ion source. This enhances deprotonation of the analyte. In other words, the use of a stronger base, e.g. tripropylamine (PA=976 KJ/mole), should result in a still further increased deprotonation. Since the $\Delta H°_{acid}$ of formate is higher than that of acetate, i.e. 1445 KJ/mole vs. 1457 KJ/mole, formation of the $[M+HCOO]^-$ ion may be expected to be stronger than that of the $[M+CH_3COO]^-$ ion[11,22].

In Table 7 the relative abundances of the formate adducts and the intensity of the base peak, i.e. $[M-H]^-$, are presented for the carboxylic and phenoxy acids studied, using a carrier stream containing TEAmFo or TPAmFo. Because the difference in PA of triethylamine and tripropylamine is only 9 KJ/mole, only a relatively small, i.e. 2-fold, increase of the signal intensity and a somewhat larger relative abundance of the $[M+HCOO]^-$ ion was observed when TEAmFo was replaced by TPAmFo. These results confirm the idea that the use of a strong base enhances the deprotonation and therefore the signal intensity in the NI mode.

However, when the data obtained with TPAmFo were compared with those obtained with ammonium acetate (difference in PA, 118 KJ/mole) a mere 5-fold instead of the expected 20-fold (29) increase in signal intensity was found. Varying the concentration of TPAmFo or TEAmFo in the carrier stream between 5 and 50 mM did not affect the signal intensity or the relative abundance of the formate adduct ion. Below 5 mM the signal decreased rapidly. Quite clearly other factors known to influence sensitivity in LC-TSP-MS like e.g. type of analyte, vaporizer temperature and kind of interface[13,49] play an important role in this case.

The use of TEAmFo and TPAmFo also promoted the formation of $[M+HCOO]^-$ ions—in some instances up to 100% of the base peak—in the case of dimethoate, several carbamates, the lower chlorinated phenols, the pyridine-like compounds (see Figure 4C) and the phenylureas (see Figure 6B below). In the case of fenaminosulf, captan and permethrin the base peaks in carrier streams containing ammonium salts, found at m/z $[M-95]^-$, $[M-149]^-$ and $[M-183]^-$, respectively, shifted 46 mass units upwards to the corresponding formic acid adducts.

*With chloroacetonitrile.* The use of chloride attachment to obtain complementary structural information is well known. For a distinct majority (ca.70%) of the test compounds in this survey, mass spectra could be generated using a carrier stream containing 2% of chloroacetonitrile and discharge ionisation in the NI mode. From this group benzenesulfonamide, the carbamates, dimethoate, the phenoxy acids, the pyridine-like compounds except chloridazon and the phenylureas showed a chloride adduct in the mass spectrum. In most cases this ion was the base peak (see Figure 4D). However,a distinct relationship between the chemical structure of the analyte and the appearance of a chloride adduct in the mass spectrum was not found. Nevertheless, the $[M+Cl]^-$ ion can be used for screening purposes for several classes of compounds. Unfortunately, however, as was to be expected from previous work of our group concerning chlorophenols and phenoxy acids[13], no gain, and occasionally even a decrease, in signal intensity was observed when this additive was used.

**Table 7** Relative abundance of the formate adducts and the intensity of the base peak of dicamba and three phenoxy acids using negative ion discharge ionisation.
Carrier stream: water-acetonitrile (50:50, v/v) with 25 mM triethylammonium formate (pH=4.5) or tripropylammonium formate (pH=7.5); flow, 1 ml/min. Discharge voltage, 1 kV.

| *Compound* | *Signal intensity of [M-H]⁻ in TIC * 1000* | | *Relative abundance of [M+HCOO]⁻ (%)* | |
|---|---|---|---|---|
| | *TEAmFo* | *TPAmFo* | *TEAmFo* | *TPAmFo* |
| Dicamba | 150 | 275 | 10 | 35 |
| Triclorpyr | 130 | 145 | 30 | 45 |
| Dichlorprop | 175 | 290 | 60 | 50 |
| Mecoprop | 315 | 375 | 65 | 80 |

*Detection limits and LC-TSP-MS*

The intensity of the ion chromatogram of each pesticide after FIA was extrapolated to a signal of about 500 counts—which can be considered as the level of three times the mean noise—and the corresponding amount of analyte was calculated. Since changing from



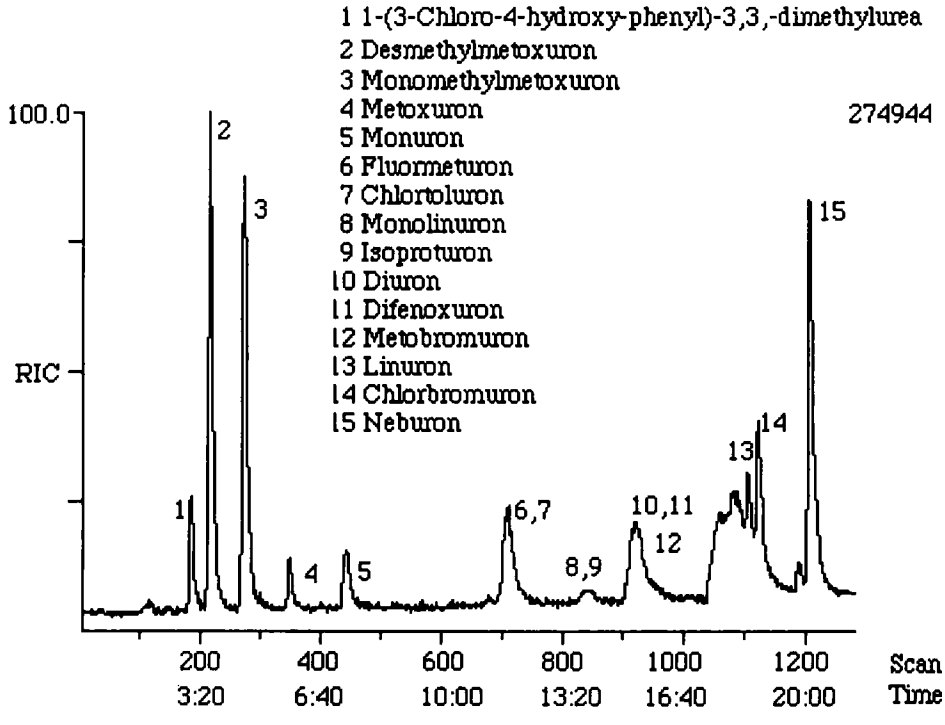



**Figure 6** Reconstructed total ion chromatogram in the NI mode with discharge ionisation obtained after a loop injection (5 µl) of a standard mixture of 13.2 µg/ml of 15 phenylureas. LC column: 150×4.6 mm i.d.; stationary phase: 5 µm, 80 Å Rosil C18-bonded silica. Eluent: water (25 mM TPAmFo, pH=7.5) – acetonitrile (65:35, v/v and at t=15 min, 30:70, v/v). Discharge ionisation, 1 kV. For further information, see text.

**Table 8** Calculated limits of detection (LODs)* for various groups of pesticides in LC-TSP-(FULL-SCAN)-MS.
Carrier stream: water-acetonitrile (50:50, v/v) containing 50 mM ammonium acetate (PI) or 25 mM TPAmFo (pH =7.5) (NI). Flow-rate, 1 ml/min. Discharge voltage, 1 kV.

| *Class of pesticides* | *LOD (ng) in* | | | | | |
|---|---|---|---|---|---|---|
| | *PI* | | | *NI* | | |
| | *≥200* | *20–200* | *≤20* | *≥200* | *20–200* | *≤20* |
| N-Substituted amides | | + | | + | | |
| Anilines | + | | | + | | |
| Carbamates | | + | | | + | |
| Thiocarbamates | + | | | | | + |
| Hydroxy-keto-lactones | | + | | | | + |
| Miscellaneous compounds** | + | | | | + | |
| Organophosphorus compounds | | + | | | | + |
| Phenols | + | | | | | + |
| Phenoxy and carboxylic acids | + | | | | | + |
| Pyridine-like compounds | | + | | | | + |
| Quaternary ammonium compounds | + | | | + | | |
| Thiocyanates | | + | | + | | |
| Triazines | | + | | | + | |
| Phenylureas | | + | | | | + |
| Thioureas | | | + | + | | |

*LOD = amount corresponding to an ion trace of 500 counts * 10 (see text); for details of compounds included, cf. Table I.
**Captan, fenaminosulf, sethoxydim, alloxidim and permethrin.



FIA-TSP-MS to LC-TSP-MS will introduce additional peak broadening, these values were multiplied by a factor of 10 to obtain limits of detection (LODs). Three groups of analytes were discerned, viz. with LODs for a full-scan spectrum of ≤ 20 ng, 20 – 200ng, or ≥ 200 ng. In Table 8, the LODs are presented for the various groups of pesticides using discharge ionisation (PI and NI mode).

*Positive ions.* Only small differences were observed when the signal intensities of the analytes in the various carrier streams (see Table 8) were compared. The addition of ammonium acetate or ammonium formate generally caused an up to 2-fold higher signal intensity. The signal intensity also changed with the mode of ionisation applied. Filament-off ionisation gave the worst sensitivity. Changing to filament-on ionisation enhanced the sensitivity by a factor of 3–4. A similar further gain in sensitivity was observed when switching to discharge ionisation. However, background signals increased by a factor of 3 upon going form filament-off via filament-on to discharge ionisation. Summarizing, analyte detectability improved 3–5-fold when using discharge instead of filament-off ionisation. At the same time the selectivity of the method improved because of the enhanced production of fragment ions.

As can be seen from Table 8, only the signals of the thioureas and isocarbamid were strong enough to meet the criterion of the first group, i.e. LOD ≤ 20 ng. The majority of the compounds tested could be classified in the second group, while the quaternary ammonium

compounds, thiocarbamates, phenols, phenoxy acids, anilines and miscellaneous compounds needed 200 ng or more for a full-scan mass spectrum.

*Negative ions.* The signal intensities of the pesticides under NI conditions varied with the additive used but were highest when TPAmFo was used. As is evident from Table 8, several classes of compounds—i.e., the phenoxy acids, hydroxy-keto-lactones, phenylureas, phenols, thiocarbamates and the organophosphorus and pyridine-like compounds can be sensitively detected by LC-TSP-MS under NI conditions (LOD ≤ 20 ng). With the triazines, carbamates, fenaminosulf, sethoxydim, captan and alloxidim 20–200 ng were required to obtain a full-scan spectrum. Fragmentation was often observed in the NI mode of operation. Although not all the fragments were identified, they were sufficiently specific to allow confirmation. Again discharge ionisation was the most sensitive mode tested. Filament-off and filament-on ionisation were 4–5- and 2–3- fold less sensitive, respectively.

*LC-TSP-MS.* In order to verify the above conclusions under real-life LC conditions, 15 phenylureas were separated on a C18-bonded silica column with water (25 mM of TPAmFo, pH 7.5)-acetonitrile (65:35, v/v and at t=15 min, 30:70, v/v) as eluent. A typical TIC chromatogram of a solution containing 13.2 μg/ml of each phenylurea—corresponding with ca. 65 ng injected—is shown in Figure 6. Full-scan negative ion data were acquired using discharge ionisation. The intense signal at 17 min originates from the applied step gradient. As is clear not all of the phenylureas are separated. Nevertheless, using the ion chromatograms, they could be separately quantified. The figure illustrates also that the response is not the same for all the test compounds. The signal intensities for the primary and secondary amine, i.e. desmethylmetoxuron and monomethylmetoxuron, respectively, and neburon are about one order higher than for the other compounds tested. Nevertheless, much lower levels can be analysed. Figure 7A (lower trace) shows the TIC chromatogram obtained for a solution containing 0.67 μg/ml, i.e. 3.5 ng injected, of the phenylureas. As is often the case in LC-TSP-MS at low levels, most of the analytes are not readily visible in the TIC chromatogram; actually, only desmethylmetoxuron, monomethylmetoxuron and neburon—scan Nos. 220, 278 and 1212, respectively—can be unambiguously stated to be present. Nonetheless, the ion chromatograms of the $[M-H]^-$ and $[M+HCOO]^-$ ions of the various phenylureas were clearly discernible from the background. As an example, this is demonstrated for monuron in Figure 7A (upper two traces). Here the ion traces for m/z 197 and 243 are shown, which correspond with the $[M-H]^-$ and $[M+HCOO]^-$ ions, respectively. Figure 7B shows the mass spectrum of monuron that can be obtained from the TIC chromatogram (see arrow). Next to ions at m/z 197 and 243, i.e. $[M-H]^-$ and $[M+HCOO]^-$ ions, also an ion at m/z [M+101]$^-$ was observed, as for all the phenylureas tested[49]. Its origin is still not clear. Obviously, even a few nanograms suffice to obtain a good full-scan spectrum. Actually, rather similar results were obtained for all other phenylureas: their LODs ranged from 1 to 10 ng. Comparison of these values with the calculated LODs for a full-scan spectrum, discussed above, shows that there is a nice mutual agreement.

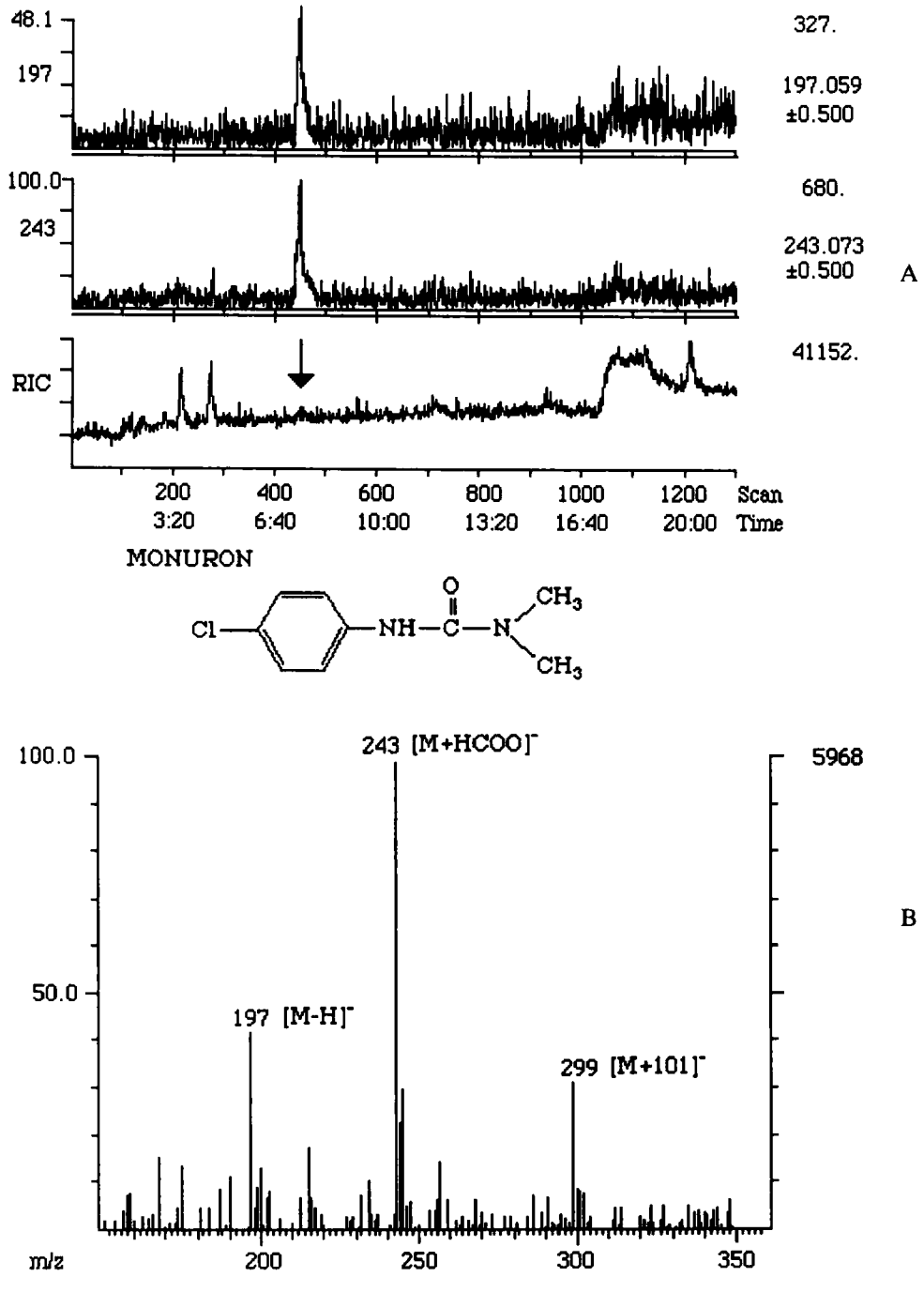



**Figure 7** [A] Reconstructed ion chromatograms of the $[M-H]^-$ and the $[M+HCOO]^-$ ion of monuron (upper two traces) and the total ion chromatogram (lower trace) in the NI mode with discharge ionisation obtained after a loop injection (5 µl) of a standard mixture of 0.67 µg/ml of 15 phenylureas.[B] Mass spectrum of monuron, eluting around 7.3 min (see arrow). Conditions see Figure 6.

## CONCLUSIONS

The addition of ammonium acetate or formate, TEAmFo or TPAmFo, or chloroacetonitrile, to the mobile phase in LC-TSP-MS provides extremely useful additional information. In both the PI and the NI mode, discharge ionisation was preferred above filament-off or filament-on ionisation because more fragmentation, i.e. more selectivity, and lower detection limits were obtained.

In the PI mode protonated and ammoniated molecular ions were mainly observed. For some compounds, e.g. the N-methylcarbamates and the miscellaneous compounds specific fragmentations were observed which provide more valuable confirmation of the analyte identity. Using water-acetonitrile (50:50, v/v) containing 50 mM ammonium acetate or ammonium formate as mobile phase and discharge ionisation, the LODs for a full-scan spectrum in LC-TSP-MS were calculated to be between 20 and 200 ng for most of the test compounds.

In the NI mode the deprotonated molecular ion or an adduct ion with acetate or formate generally was the base peak. Using TEAmFo or TPAmFo instead of ammonium acetate or formate enhanced the abundance of the $[M+HCOO]^-$ ion and improved the analyte detectability. The use of a stronger base than ammonia as additive obviously enhances the sensitivity and selectivity of the ionisation process, especially in the NI mode. Specific fragmentations were observed for the carbamates, the organophosphorus compounds, the hydroxy-keto-lactones, the phenoxy acids and the miscellaneous compounds. Analyte detectability was distinctly better than when working in the PI mode, and the LODs for a full-scan spectrum were less than 20 ng for several of the compound classes tested (see Table 8).

At first sight detection limits of ca. 20–200 ng, do not appear to be satisfactory for real-life environmental applications such as the analysis of surface or ground water samples, because in such matrices detection limits (in concentration units) of around 1 ng/ml are required. However, two aspects should be considered here. Firstly, in the case of target analysis of specific compounds or classes of compounds, selected ion monitoring and/or multiple ion detection will be the detection principle of choice; in other words, analyte detectability then can be easily improved 10–50-fold. Secondly, LC-TSP-MS can be coupled on-line with trace enrichment procedures involving the use of, typically, 50–100 ml water samples. This may be expected to increase analyte detectability by some two orders of magnitude. In this way, it should be possible to set up both group-specific as well as general screening procedures displaying the required sensitivity. Actually, in sequel to the work described in this paper, first results in this area have recently been presented by Bagheri *et al.*[41,42], for various classes of polar pesticides. Using slightly different experimental conditions (mainly because of the use of another mass spectrometer), these authors determined sub-ng/ml levels of phenylureas, and also carbamates, organophosphorus pesticides and triazines in several river water samples. Current research aims at the use of LC-TSP-MS in the NI mode, and with TPAmFo as additive, in order to further extend the application range of the technique for monitoring purposes.

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*References*

1. D. Barcelo, *TrAC*, **10,** 323–329 (1991)
2. J. Drodz in "Chemical Derivatisation in Gas Chromatography" (ed. J. Drodz, Vol. 19, Elsevier, Amsterdam).
3. R.J. Anderegg, *Mass Spectrom. Rev.*, **7,** 395–424 (1988).
4. I. Liska, E.R. Brouwer, A.G.L. Ostheimer, H. Lingeman, U.A.Th. Brinkman, R.B. Geerdink and W.H. Mulder, *Intern. J. Environm. Anal. Chem.*, **47,** 267–291 (1992).
5. E.R. Brouwer, I. Liska, R.B. Geerdink, P.C.M. Frintrop, W.H. Mulder, H. Lingeman and U.A.Th. Brinkman, *Chromatographia*, **32,** 445–452 (1991).
6. C.R. Blakley and M.L. Vestal, *Anal. Chem.*, **55,** 750–754 (1983).
7. R.C. Willoughby and R.F. Browner, *Anal. Chem.*, **55,** 2626–2631 (1984).
8. J. van der Greef, W.M.A. Niessen and U.R. Tjaden, *J. Chromatogr.* **474,** 5–19 (1989).
9. T.A. Bellar and W.L. Budde, *Anal. Chem.*, **80,** 2076–2083 (1988).
10. R.D. Voyksner and C.A. Haney, *Anal. Chem.*, **57,** 991–996 (1985).
11. D. Barcelo, G. Durand, R.J. Vreeken, G.J. de Jong, H. Lingeman and U.A.Th. Brinkman, *J. Chromatogr.*, **553,** 311–328 (1991).
12. H.Fr. Schröder, *J. Chromatogr.*, **554,** 251–266 (1991).
13. R.J. Vreeken, G.J. de Jong, U.A.Th Brinkman and D. Barcelo, *Biomed. Environm. Mass Spectrom.*, **19,** 481–492 (1990).
14. D. Barcelo, *Org. Mass Spectrom.*, **24,** 898–902 (1989).
15. G. Durand, N. de Bertrand and D. Barcelo, *J. Chromatogr.*, **562,** 507–523 (1991).
16. R.C. Dougherty and J.D. Wander, *Biomed. Mass Spectrom.*, **7,** 401–409 (1980).
17. D. Barcelo, G. Durand, R.J. Vreeken, G.J. de Jong and U.A.Th. Brinkman, *Anal. Chem.*, **62,** 1696–1700 (1990).
18. P. Arpino, *Mass Spectrom. Rev.*, **9,** 631–669 (1990).
19. L.M. Shalaby and S.W. George in "Liquid Chromatography/Mass Spectrometry. Applications in Agriculture, Pharmaceutical, and Environmental Chemistry", (ed. M.A. Brown, Amercian Chemical Society, Washington, 1990, ACS Symposium Series 420), pp. 75–91.
20. T.L. Jones, L.D. Betowski and J. Yinon in "Liquid Chromatography/Mass Spectrometry. Applications in Agriculture, Pharmaceutical, and Environmental Chemistry" (ed. M.A. Brown, Amercian Chemical Society, Washington, 1990, ACS Symposium Series 420) pp. 62–74.
21. R.D. Voyksner, T. Pack, C. Smith, H. Swaisgood and D. Chen, in "Liquid Chromatography/Mass Spectrometry. Applications in Agriculture, Pharmaceutical and Environmental Chemistry" (ed. M.A. Brown, Amercian Chemical Society, Washington, 1990, ACS Symposium Series 420) pp. 14–39.
22. D. Barcelo, *Biomed. Environm. Mass Spectrom.*, **17,** 363–369 (1988).
23. I. Hammond, K. Moore, H. James and C. Watts, *J. Chromatogr.*, **474,** 175–180 (1989).
24. T. Cairns and E.G. Siegmund in "Liquid Chromatography/Mass Spectrometry. Applications in Agriculture, Pharmaceutical, and Environmental Chemistry" (ed. M.A. Brown, Amercian Chemical Society, Washington, 1990, ACS Symposium Series 420) pp. 40–47.
25. Bellar, T.D. Behymer and W.L. Budde, *J. Am. Soc. Mass Spectrom.*, **1,** 92–101 (1991).
26. M.A. Brown, R.D. Stephens and I.S. Kim, *TrAC*, **10,** 330–336 (1991).
27. M.A. Brown, I.S. Kim, F.I. Sasinos and R.D. Stephens in "Liquid Chromatography/Mass Spectrometry. Applications in Agriculture, Pharmaceutical, and Environmental Chemistry" (ed. M.A. Brown, Amercian Chemical Society, Washington, 1990, ACS Symposium Series 420) pp. 198–214.
28. I.S. Kim, F.I. Sasinos, R.D. Stephens, J. Wang and M.A. Brown, *Anal. Chem.*, **63,** 819–823 (1991).
29. S. Steffenrud, E. Dewey and G. Maylin, *Rapid Commun. Mass Spectrom.*, **4,** 463–466 (1990).
30. H. Maeder, *Rapid Comm. Mass Spectrom.*, **3,** 183–185 (1989).

31. D. Barcelo, *Org. Mass Spectrom.*, **24,** 219–224 (1989).
32. R.B. Geerdink, F.A. Maris, G.J. de Jong, R.W. Frei and U.A.Th. Brinkman, *J. Chromatogr.*, **394,** 51–64 (1987).
33. D. Barcelo, F.A. Maris, R.B. Geerdink, R.W. Frei, G.J. de Jong and U.A.Th. Brinkman, *J. Chromatogr.*, **394,** 65–76 (1987).
34. C.E. Parker, K. Yamaguchi, D.J. Harvan, R.W. Smith and J.R. Hass, *J. Chromatogr.*, **319,** 273–283 (1985).
35. J. Saar, M. Haimberg and C. Chenier, *Org. Mass Spectrom.*, **26,** 263–266 (1991).
36. C.E.M. Heeremans, R.A.M. van der Hoeven, W.M.A. Niessen, J. van der Greef and N. Nibbering, *Org. Mass Spectrom.*, **26,** 519–527 (1991).
37. J.D. Rosen in "Applications of New Mass Spectrometry Techniques in Pesticide Chemistry" (ed. J.D. Rosen, Wiley, New York, 1987).
38. K.S. Chiu, A. van Langenhove and C. Tanaka, *Biomed. Environm. Mass Spectrom.*, **18,** 200–206 (1989).
39. R.D. Voyksner, J.T. Bursey and E.D. Pellizzari, *Anal. Chem.*, **56,** 1507–1514 (1984).
40. T. Cairns, E.G. Siegmund and J.J. Stamp, *Rapid Comm. Mass Spectrom.*, **1,** 89–94 (1987).
41. H. Bagheri, E.R. Brouwer, R.T. Ghijsen and U.A.Th. Brinkman, *Analusis*, **8,** 475–482 (1992).
42. H. Bagheri, E.R. Brouwer, R.T. Ghijsen and U.A.Th. Brinkman, submitted for publication.
43. H. Wincel, S. Wlodek and D.K. Bohme, *Int. J. Mass Spectrom. Ion Processes*, **84,** 69–87 (1988).
44. R.J. Vreeken, unpublished results.
45. D. Vollmer, A. Preiss, K. Levsen and G. Wunsch, presented at the 9th Montreux Symposium on Liquid Chromatography-Mass Spectrometry (LC-MS, SFC-MS, CZE-MS, MS-MS), 4–6 Nov. 1992, Montreux, Switzerland.
46. A.G. Harrison, *Chemical Ionization Mass Spectrometry* (CRC Press, Boca Raton, FL, U.S.A. 1983).
47. J. Saar and A. Salomon, *Org. Mass Spectrom.*, **25,** 209–213 (1990).
48. R.D. Voyksner, J.T. Bursey and E.D. Pellizzari, *J. Chromatogr.*, **312,** 221–235 (1984).
49. R.J. Vreeken, H. Bagheri, R.T. Ghijsen and U.A.Th. Brinkman, in preparation.
50. H. Budzikiewicz, C. Djerassi and D.H. Williams in "Mass Spectrometry of Organic Compounds" (ed. H. Budzikiewicz, Holden-Day, San Francisco, 1967) pp. 312.
51. H. J. Stan and G. Kellner, *Biomed. Mass Spectrom.*, **9,** 483–492 (1982).
52. C.E. Parker, C.A. Haney and J.R. Hass, *J. Chromatogr.*, **237,** 233–248 (1982).