Negative-Ion Mass Spectrometry of Amphetamine Congeners

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Negative-ion chemical ionization mass spectrometry with a conventional combined gas chromatograph–mass spectrometer has been used for the analysis of amphetamine-like compounds. Ammonia was used as the reagent gas, which gives rise to few but specific sample-fragment ions, such as (M – 1)− as the base peak, m/z 91, and a smaller peak corresponding to the end part of the side chain. Five reference compounds and a urine sample from an overdose case were analyzed. Comparative positive-ion chemical ionization reference spectra were also recorded.

Amines that stimulate the central nervous system have been studied by GC/MS and MS methods, including normal electron impact ionization (EI) and positive ion chemical ionization (PCI) (1–3). In the EI mode the mass spectra of these compounds show low intensity of the molecular ion and the base peak belongs to a fragment of the side chain. In the PCI mode a base peak at (M+1)+ is most frequently shown. The use of negative ion chemical ionization (NCI) mass spectrometry in the field of analytical chemistry has been described in several reports (4–6). The number of parameters is increased when operating the mass spectrometer in the chemical ionization mode as compared to the EI mode. Different reagent gases give often quite specific fragmentation of a compound and it is of importance that the reagent gas is selected for a given class of studied compounds (7, 8). Because NCI mass spectrometry is an unexplored analysis method, it is of interest to study various groups of substances and to compare their NCI mass spectra with those obtained with EI and PCI. In this paper, five reference compounds have been run by NCI mass spectrometry, as well as a urine sample probably containing central nerve stimulating amines. For a comparison, these compounds were also run in the PCI mode.

Materials and Methods

The reference compounds were dissolved in ethanol. The urine was alkalized and extracted with diethyl ether. The samples were received from the Government Laboratory for Forensic Chemistry in Stockholm.

The mass spectra were recorded with the LKB 2091 gas chromatograph–mass spectrometer, modified for chemical ionization as previously described (9). For the work in the NCI mode, the polarities of the magnetic current, accelerating voltage, and repeller voltage were reversed (7). A 1.5 m × 2 mm (i.d.) column containing 5% Carbowax 20 M and 5% KOH on Chromosorb G/AW-DMCS (80–100 mesh) was used, with a helium flow rate of 15 ml/min. The column temperature was 160 °C for the reference samples. The urine sample was run from 120 to 160 °C with a temperature programming of 9 °C/min. The ion source temperature was 220 °C. The electron energy was kept at 250 eV and the accelerating voltage was −3.5 kV. The magnetic current was repetitively scanned at a mass range of m/z 20–500, about 3 s per cycle. Ammonia was used as reagent gas and the pressure in the ion source pumping line was 13.3 mPa (10−4 Torr). The mass spectrometer is interfaced to a PDP 11/34 computer system, equipped with a floating point processor, two RK06 disk storages, a magnetic tape unit, a Versatec electrostatic printer/ploter, a DECwriter terminal, and a Tektronix alphanumeric-graphic terminal. The software was handled by the LKB 2130 data system.

Results and Discussion

For the detection of negative ions, ammonia was used as reagent gas. The following advantages were observed:

- (M−1)− is most frequently the base peak
- The fragmentation pattern is easily understood
- Few formations of cluster ions
- The absence of ion source contamination
- The filament is seldom destroyed by high pressure of the reagent gas

A disadvantage is that the ammonia can cause corrosion of the copper membrane in the vacuum valves in the reagent-gas inlet system. For this reason the ammonia is directly introduced after the vacuum valves. Negative ions are formed by interaction of electrons with NH3 molecules through three different mechanisms.

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\begin{align*}
\text{NH}_3 + e^- & \rightarrow \text{NH}_4^- & \text{resonance capture} \\
\text{NH}_3 + e^- & \rightarrow \text{NH}_2^- + \text{H}^+ & \text{dissociative resonance capture} \\
\text{NH}_3 + e^- & \rightarrow \text{NH}_2^- + \text{H}^+ + e^- & \text{ion-pair production}
\end{align*}
\]

Ionization of samples in the negative ionization mode will probably occur predominantly through secondary electron capture. However, charge-exchange reactions leading to the formation of sample ions A− from H− + AH → A− + H2 and
NH$_3^+$ + AH $\rightarrow$ A$^-$ + NH$_3$ may also play some role in the ionization process.

Figure 1 shows the ion intensity of the fragments from the reagent gas NH$_3$ as a function of the pressure in the ion source (the pressure was measured in the pumping line to the ion source). For an operating pressure of about 13.3 mPa the ion intensity of H$^+$ and NH$_3^-$ was fairly constant. However, NH$_3^-$ will dominate at a pressure about 13.3 mPa or higher, but the way in which this will influence the fragmentation process has not yet been studied.

Five central nervous system stimulating amines were studied by GC/MS with negative and positive ion chemical ionization. The NCI spectra of amphetamine, methylamphetamine, and phentermine in Figures 2-4 show (M-1)$^-$ as the base peak with an intense peak at m/z 91 and rearranged ions from the side chain at m/z 42 and 56. The PCI spectra have (M+1)$^+$ as the base peak. The side chain shows m/z 44 and 55 as intense peaks, which are base peaks when operating in the EI mode. A comparison of the PCI spectra of methylamphetamine and phentermine shows that it is difficult to differentiate them. The same situation is obtained when they are run in the EI mode. Thus the NCI mode of operation makes it possible to distinguish easily between these two compounds.

Figure 5 shows the NCI and PCI spectra of ephedrine, where (M-1)$^-$, m/z 107, 105, and 56 are of relatively high intensity in the NCI mode and (M+1)$^+$, m/z 148 (loss of water) and 58 in the PCI mode. Even in this case, the NCI mode is preferable to the PCI mode in the identification process. The structure and the fragmentation mechanism of some ions in the EI mass spectrum of ephedrine have been published recently (10). Of the five studied amines, ephedrine

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**Fig. 1.** The absolute intensity of H$^+$, NH$_3^-$, and NH$_3^+$ as a function of the ion source housing pressure

In all Figs. NH$_3$ was the reagent gas.

**Fig. 2.** (a) NCI mass spectrum of amphetamine; (b) PCI mass spectrum of amphetamine

**Fig. 3.** (a) NCI mass spectrum of methylamphetamine; (b) PCI mass spectrum of methylamphetamine

**Fig. 4.** (a) NCI mass spectrum of phentermine; (b) PCI mass spectrum of phentermine

**Fig. 5.** (a) NCI mass spectrum of ephedrine; (b) PCI mass spectrum of ephedrine
gives, in contrast to the others, a peak at (2M-1)⁻ (not shown in Figure 5a).

The NCI and PCI spectra of phenmetrazine are shown in Figure 6. The structure of the fragment m/z 107 in the NCI spectrum of phenmetrazine is probably identical to the fragment m/z 107 in the NCI spectrum of ephedrine. The peak at m/z 66 in this spectrum belongs to the nitrogen-containing part of the molecule, where three hydrogens are lost. One of these hydrogens is probably transferred to the oxygen simultaneously with the breakdown of the molecule.

A suggested fragmentation and explanation of the negatively charged nitrogen- or oxygen-containing fragments is shown in Scheme 1. In amphetamine and methylamphetamine the charge is probably on the carbon. Phentermine probably has the negative charge on the nitrogen, because the methyl groups often are intact. The side-chain fragment of ephedrine probably has the negative charge on the methyl substituted carbon and the aromatic fragment has the charge on the oxygen for m/z 107 and on the carbon for m/z 105. Phenmetrazine shows a similar oxygen-containing fragment,
Figure 10 shows a mass chromatogram where (M-1)- and (2M-1)- are shown for peak 4 (m/z 164 and 329) and peak 5 (m/z 150 and 301) taken from the gas chromatogram in Figure 7. The intensities of m/z 164 and 329 increase to a certain level, whereupon m/z 164 begins to decrease until m/z 329 has reached its maximum intensity. This phenomenon occurs due to high sample pressure. The (2M-1)-1 is formed from polar compounds and its intensity is enhanced by high sample pressure.

The sensitivity obtained for ephedrine when repetitive scanning was used was about 5 ng for a signal/noise ratio of 5/1. There are many unanswered problems in the negative ionization process. For instance, which hydrogen is lost for the peak (M-H)- which often is the base peak? The nitrogen fragment of the side chain loses two hydrogens, but which two is yet unknown.

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References