Toxichem Krimtech 2011;78(Special Issue):306

Identification of 4-Methylamphetamine in a seized Amphetamine Mixture

Folker Westphal[1], Thomas Schäfer[2], Lothar Zechlin[3], Stefanie Stoll[2]

[1] State Bureau of Criminal Investigation Schleswig-Holstein, Section Narcotics/Toxicology, Mühlenweg 166, D-24116 Kiel, Germany
[2] Federal Bureau of Criminal Investigation, KT 1, Äppelallee 65, D-65173 Wiesbaden, Germany
[3] State Bureau of Criminal Investigation Rheinland-Pfalz, Valenciaplatz 1-7, D-55118 Mainz, Germany

Key words: 4-methylamphetamine, structure elucidation, NMR, GC-MS

Abstract

In 2010 a new designer drug 4-methylamphetamine was detected in an amphetamine mixture. The structure was elucidated by GC-MS after electron ionization (EI) and chemical ionization (CI) with methane as reagent gas, product ion spectrometry (EI-MS/MS with argon as collision gas under normalized conditions) of the immonium ion, and by NMR spectroscopy. Additionally, the acetyl, the trifluoroacetyl, the heptafluorobutyryl, and the formyl derivatives of 4-methylamphetamine have been prepared and measured on GC-MS.

1. Introduction

In 2010 a new designer drug was detected in an amphetamine mixture. The seized mixture of an off-white powder contained besides amphetamine, caffeine, di-(phenylisopropyl)amine (DPIA) and some by-products an other amphetamine type compound **1** (Fig. 1).

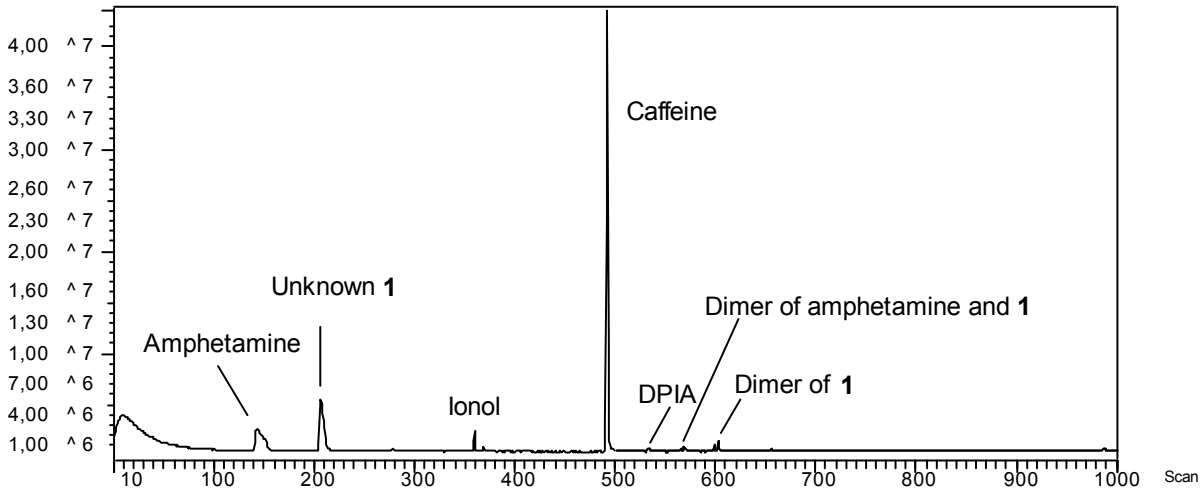


Fig. 1. GC-MS total ion chromatogram (TIC) of the alkaline diethyl ether extract [1].

Searching of the EI-MS-spectrum of **1** against the mass spectral library Designer Drugs [2] revealed 4-methylamphethamine as first hit. However, not all isomers were present in the library and no retention index was given. So, further measurements for structure elucidation had to be done.

2. Material and Methods

2.1. GC-MS analysis

For GC-MS analysis an alkaline diethyl ether extract was prepared. GC-MS spectra were recorded on a Finnigan TSQ 7000 triple stage quadrupole mass spectrometer coupled to a Thermo gas chromatograph (fused silica capillary column DB1, 30 m x 0.25 mm, film thickness 0.25 µm). The temperature program used consisted of an initial temperature of 80 °C (1 min) followed by a ramp to 280 °C at 15 °C/min finally held for 15 min. The carrier gas was helium (constant flow 1 ml/min). The ion source temperature was 150 °C, the electron ionization energy was 70 eV with an emission current of 400 µA. The scan time was 1s, and the scan range was m/z = 30 - 600. The spectrum after chemical ionization (CI) was recorded with methane as reagent gas under the same conditions. The scan range was m/z = 50 - 600 for the CI-MS spectrum. Product ion spectra were recorded with argon as collision gas in EI mode and normalization of the collision conditions with n-butylbenzene [3]. For derivatization the alkaline diethyl ether extract was divided into several portions, evaporated at room temperature under a gentle nitrogen stream and derivatized with methyl iodide, acetic acid anhydride, N-methyl-N-trimethylsilyltrifluoroacetamide (MSTFA), trifluoroacetic acid anhydride, heptafluorobutyric acid anhydride, and formylchloride in a sealed glass vial at 70 °C for 30 min. Derivates were reconstituted in diethyl ether or chloroform for GC-MS measurement.

2.2. NMR analysis

NMR spectra were recorded with a Bruker Avance spectrometer operating at resonance frequencies of 500 MHz for ^{1}H-NMR-spectra. The mixture of compounds was previously separated on a preparative Waters LC-MS system, using a Waters Xbridge C18 column (5.0 µm, 19 mm x 150 mm) and a water/acetonitrile gradient. The eluents consisted of A (water + 0,05% formic acid) and B (acetonitrile). The gradient was as follows: initial A:B = 95:5 lineary to 25:75 in 10 min, then 0:100 until 14 min. The eluate was fractionated and collected. After evaporation some mg of the unknown compound were isolated. About 1 mg of the compound was dissolved in 500 µL perdeuterated water containing sodium acetate (1.9 ppm) as a quantitative standard (no quantification done). The following NMR-spectra were recorded using standard pulse programs at 300 K to obtain resonance frequencies of all proton- and carbon-atoms: one-dimensional (1D) ^{1}H-NMR, 2D-gradient selected ^{1}H,^{1}H-COSY, ^{1}H,^{13}C-HSQC and -HMBC. ^{1}H,^{13}C-HSQC and -HMBC correlate geminal and vicinal protons, carbon atoms with their directly attached protons, and carbon and proton atoms generally separated by three or two bonds, respectively. All spectra were referenced to TSP (trimethylsilyl propionic acid sodium salt).

3. Results and Discussion

Compound **1** was identified as 4-methylamphetamine (para-methylamphetamine).

GC-MS after chemical ionization (CI) with methane as reagent gas revealed a molecular weight of 149 amu showing strong losses of 17 amu from the fragments m/z = 150 ([M+H]+), 178 ([M+29]+), and 190 ([M+41]+) indicating a primary amine (Fig. 2). The mass spectrum after electron ionization (EI) was not identical with the isobaric cathinone and showed as base peak signal the fragment m/z = 44 (member of the immonium ion series) and minor fragments at m/z = 65, 77, 91, 105, 117, and 134 shifted by 14 amu in comparison to amphetamine indicating a methyl substitution in the aromatic moiety (Fig. 3).

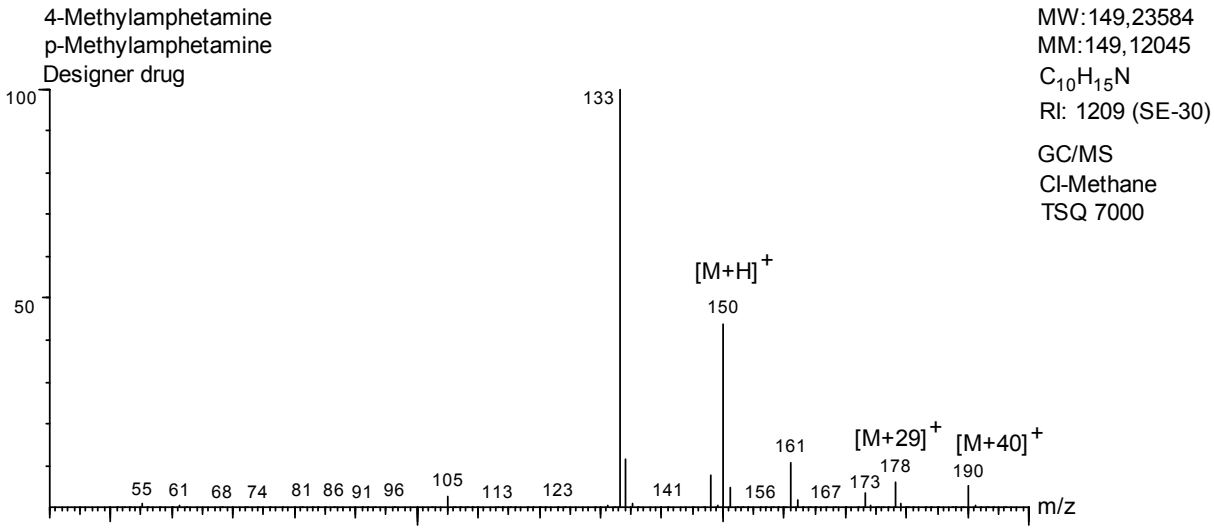


Fig. 2. MS of 4-methylamphetamine after CI.

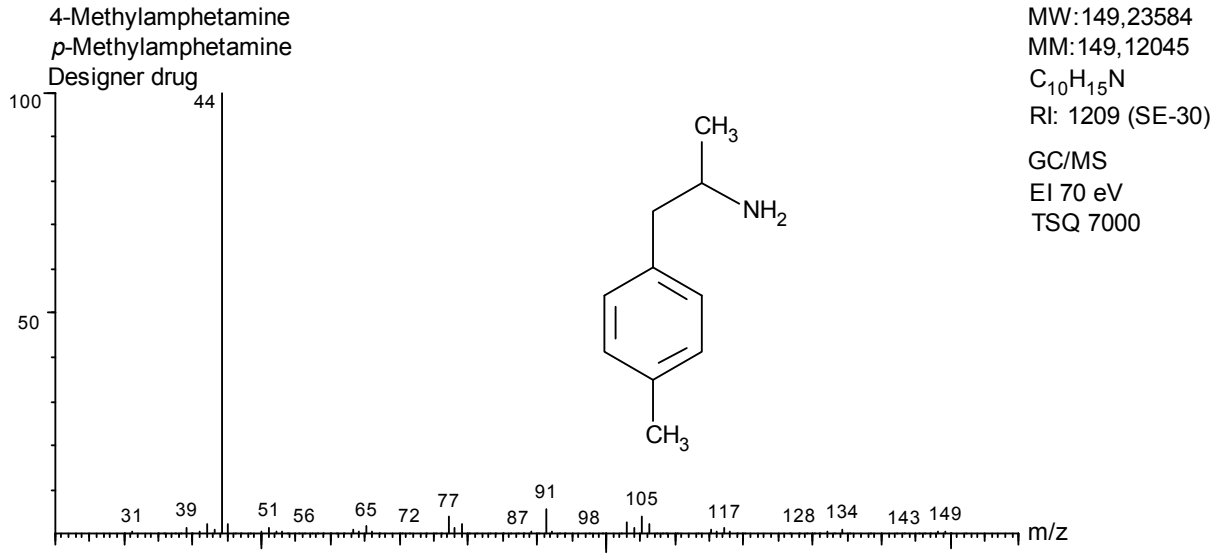


Fig. 3. MS of 4-methylamphetamine after EI.

The structure of the amino moiety was elucidated by product ion spectrometry (EI-MS/MS with argon as collision gas under normalized conditions) of the immonium ion m/z = 44 to be a N-unsubstituted immonium ion with an alpha-methyl-substituted carbon atom (Fig. 4, I + II). The second possible structure of the immonium (Fig. 4, III) is clearly ruled out by its different product ion spectrum [4,5]. The results of the GC-MS elucidation for compound **1** were consistent with an amphetamine bearing a methyl group in the aromatic moiety.

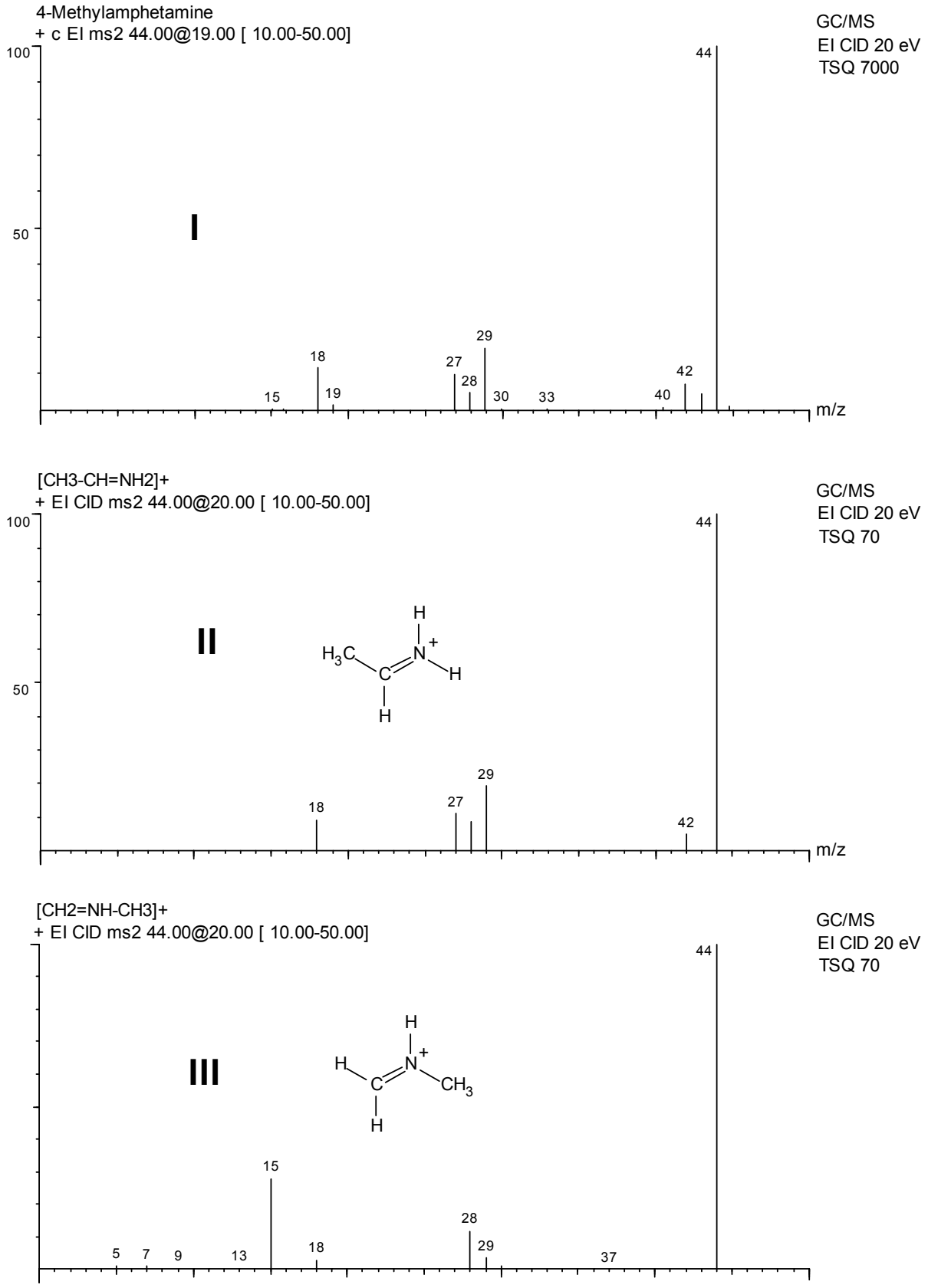


Fig. 4. Product ion spectrum of the immonium ion m/z = 44 of compound **1** (I) and corresponding data base [4] entries (II and III).

To clear the position of the methyl group NMR spectroscopic measurements were necessary. Before NMR measurement compound **1** was isolated from the mixture by preparative LC/MS. NMR measurements in deuterated water showed clearly a para substitution pattern (two doublets at 7.22 ppm and 7.27 ppm in ^{1}H-NMR, Fig. 5) with the corresponding ^{13}C-signals at 132.3 and 132.5 ppm, respectively. The additional methyl group in the aromatic ring resonated at 2.34 ppm as a singlet in the ^{1}H-spectrum with a chemical shift of 22.9 ppm for the corresponding ^{13}C-signal (see HSQC-spectrum in Fig. 7). The assignment of the quarternary carbon atoms was done by a Hetero-Multiple-Bond-Correlation (HMBC, not shown). Chemical shifts for all atoms are shown in Fig. 6.

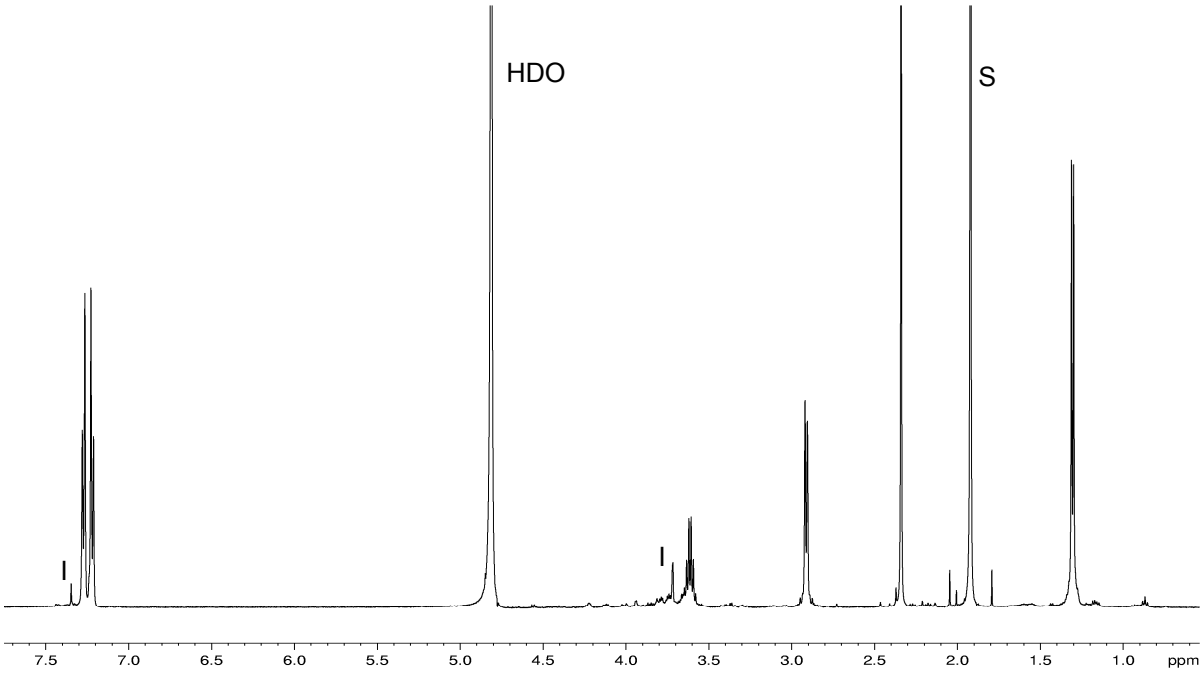
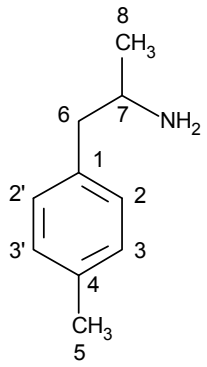


Fig. 5. 1H-NMR spectrum of 4-methylamphetamine after purification, S = standard (sodium acetate), I = Impurity.

Chemical shifts of compound **1**:



4-Methylamphetamine

Atom	δ [ppm]	Atom	δ [ppm]
		C-1	136.0
H-2/2‘	7.22 (d, $^{3}J_{2,3}$ = 7.8 Hz)	C-2	132.3
H-3/3‘	7.27 (d, $^{3}J_{3,2}$ = 7.8 Hz)	C-3	132.5
		C-4	140.4
H-5	2.34 (s)	C-5	22.9
H-6	2.91 (d, $^{3}J_{6,7}$ = 7.2 Hz)	C-6	42.5
H-7	3.61 (tq, $^{3}J_{7,6}$ = 7.2 Hz, $^{3}J_{7,8}$ = 6.6 Hz)	C-7	52.0
H-8	1.30 (d, $^{3}J_{8,7}$ = 6.6 Hz)	C-8	20.3

Fig. 6. NMR results of compound **1.**

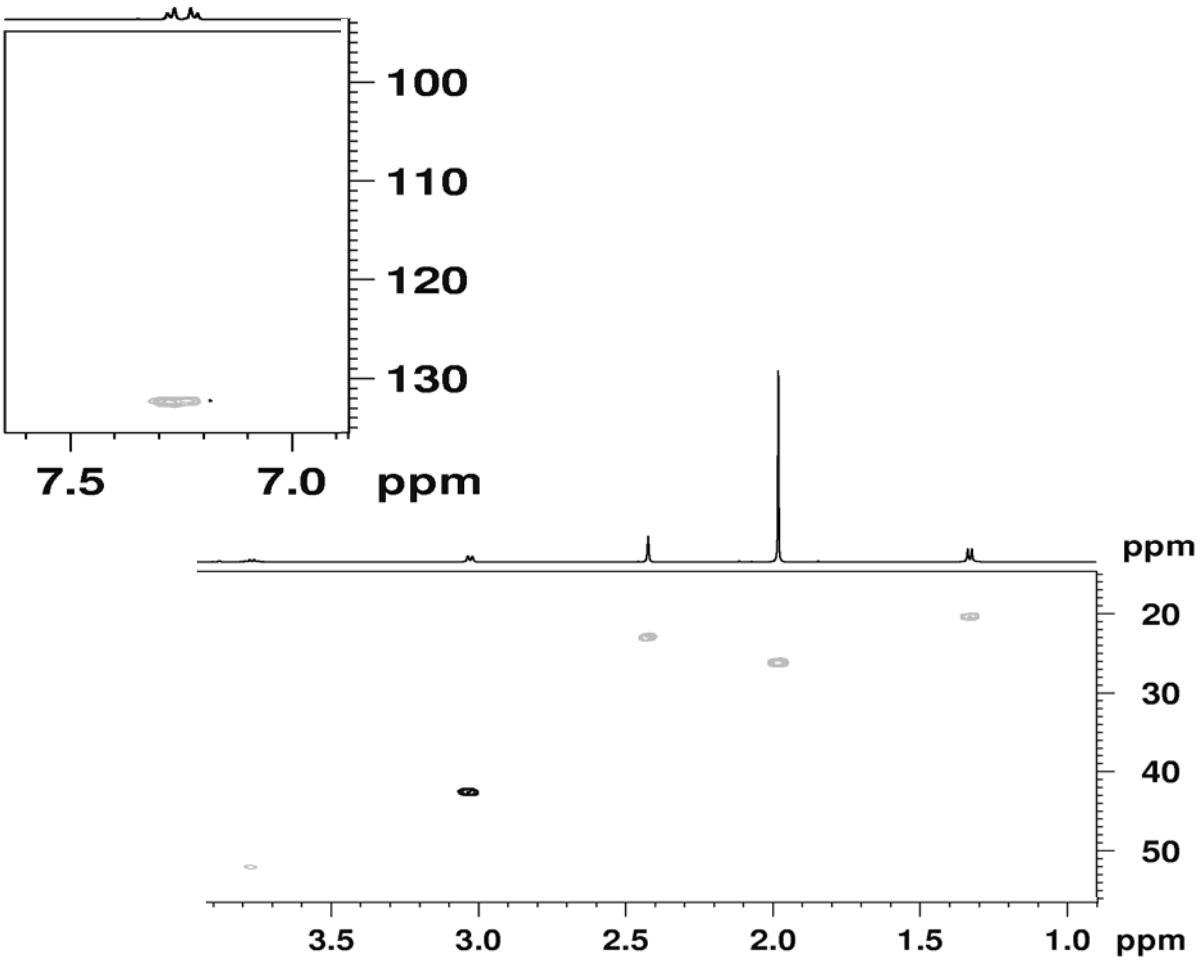


Fig. 7. Aromatic and aliphatic region of the HSQC-spectrum of 4-methylamphetamine.

Besides the dimer of amphetamine (di-(phenylisopropyl)amine, DPIA) which is built as a common by-product during amphetamine synthesis, the dimer of 4-methylamphetamine and even the mixed dimer of amphetamine and 4-methylamphetamine was detected (Fig. 8) indicating the synthesis of amphetamine and 4-methylamphetamine in the same batch. Because of the existence of two chiral centres in the molecules diasteromers of each dimer can be detected.

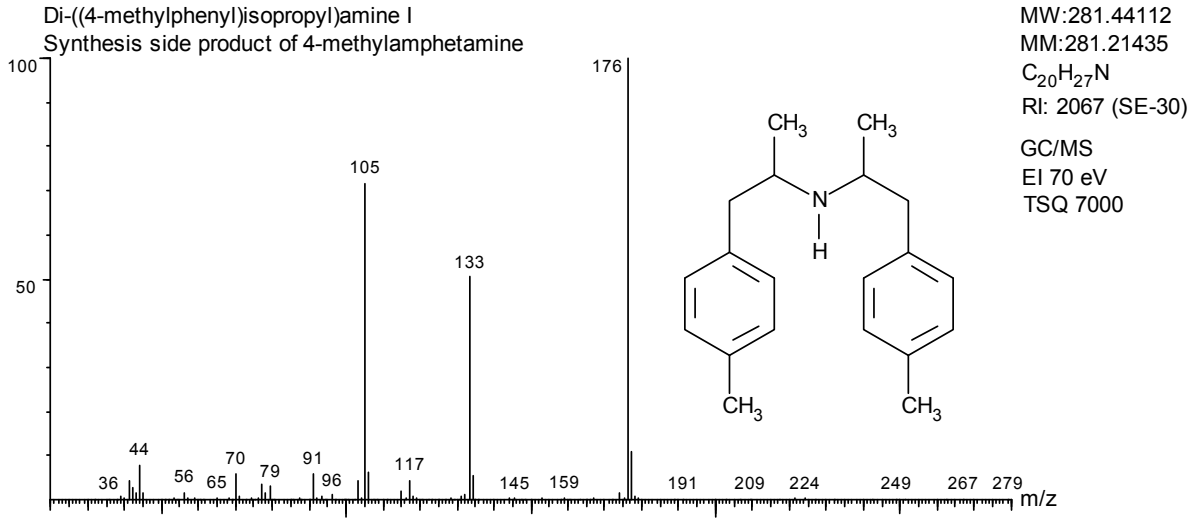


Fig. 8. Synthesis side products of 4-methylamphetamine (continued on next page).

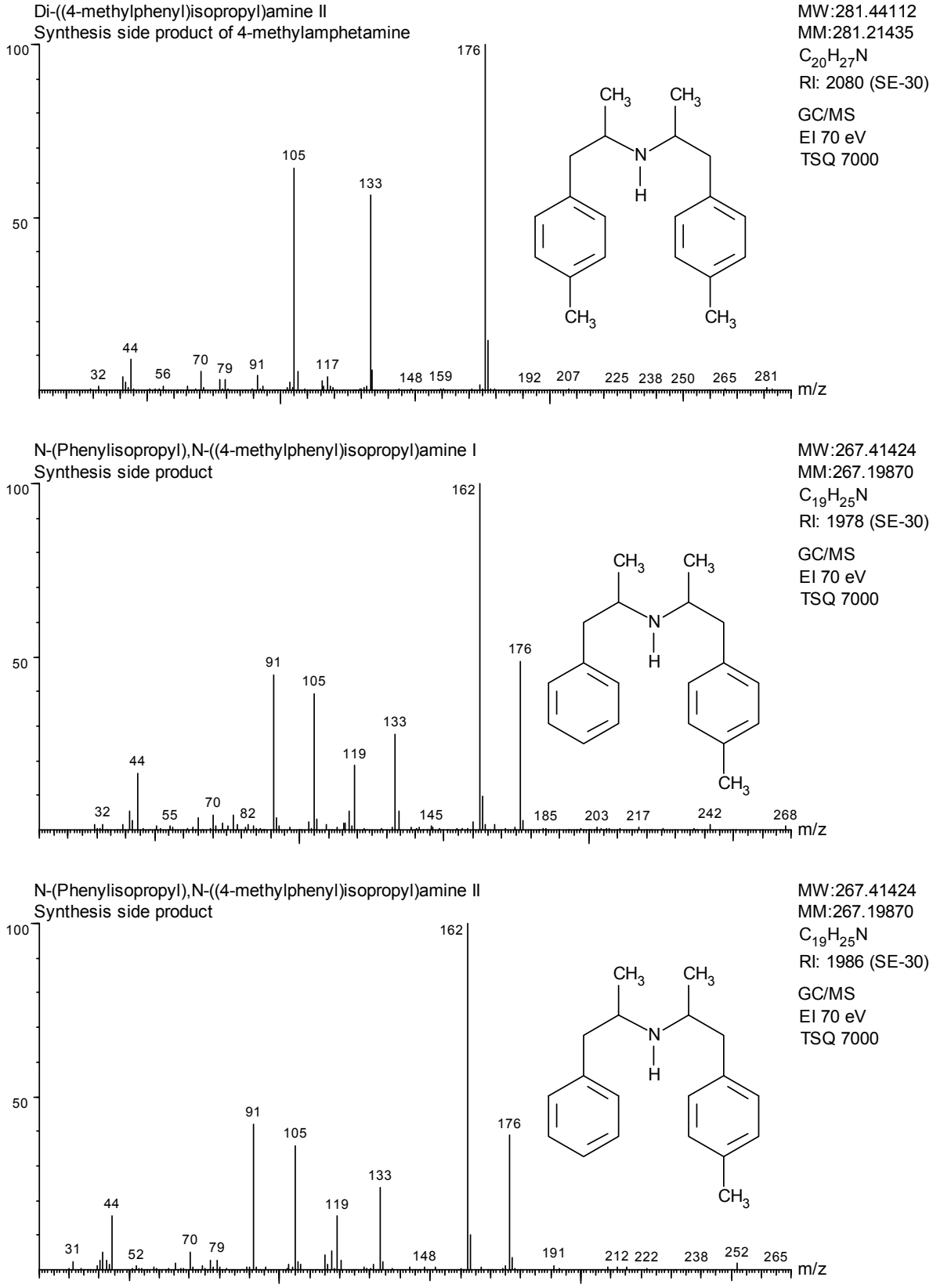


Fig. 8 (continued). Synthesis side products of 4-methylamphetamine.

Additionally, the formyl, the acetyl, the trifluoroacetyl, the heptafluorobutyryl, the trimethylsilyl and the methylated (p-methylmethamphetamine!) derivatives of 4-methylamphetamine have been prepared and measured on GC-MS (Fig. 9).

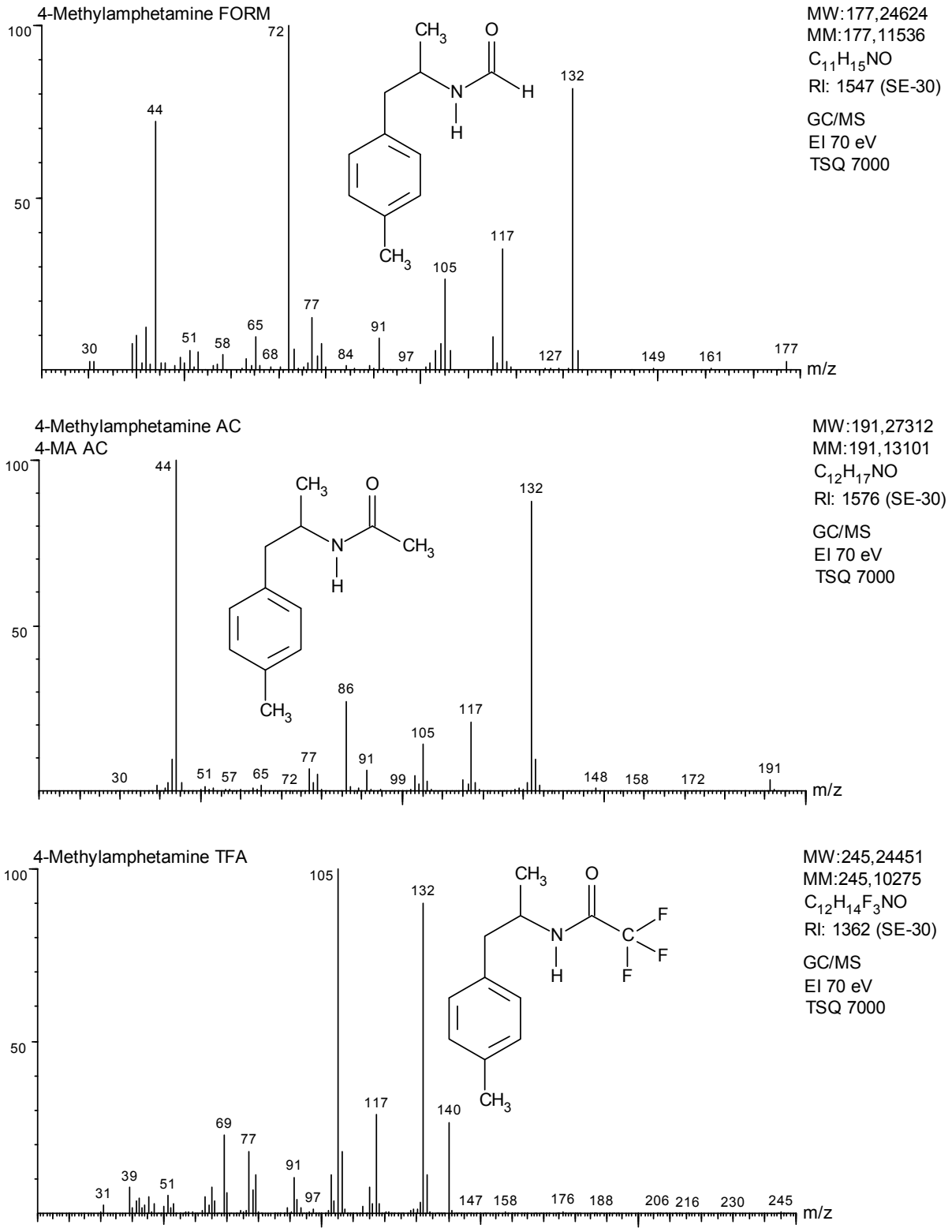


Fig. 9. Derivatives of 4-methylamphetamine.

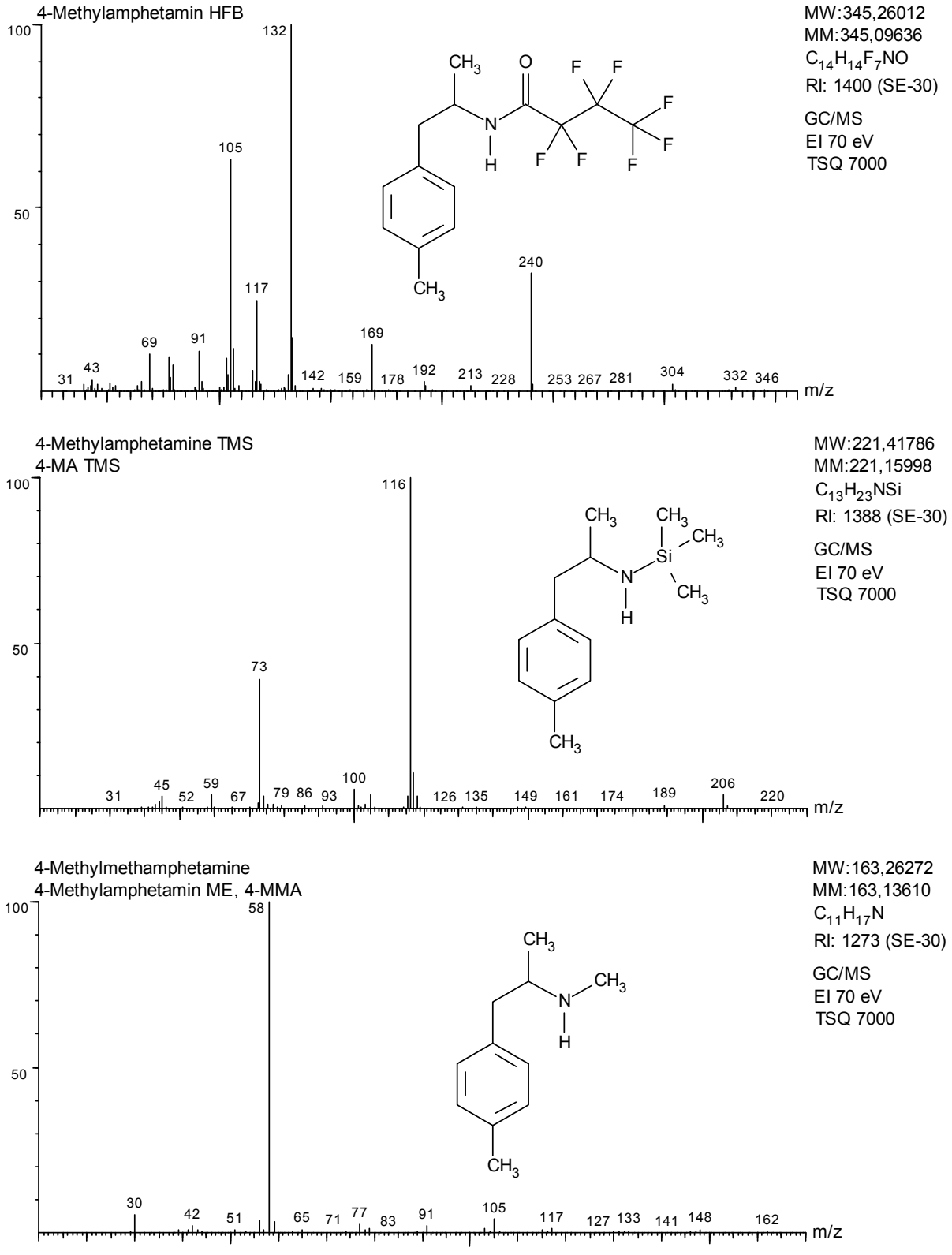


Fig. 9 (continued). Derivatives of 4-methylamphetamine.

4. Conclusion and Outlook

In a seized amphetamine mixture besides amphetamine, caffeine, and di-(phenylisopropyl)-amine (DPIA) the new designer drug 4-methylamphetamine and some of its synthesis by-products were detected. The structure of 4-methylamphetamine was elucidated by GC-MS, GC-MS/MS and NMR. Some derivatives have been prepared and their EI-GC-MS spectra were recorded. Meanwhile 4-methylamphetamine has been seized by various police organizations in Germany [6] and was already detected in serum samples of drivers in Germany [7].

5. References

[1] Structure formulas and mass spectra were generated and processed by Chemograph Plus Software, P. Rösner, DigiLab Software GmbH, www.chemograph.de.

[2] Rösner P, Junge Th, Westphal F, Fritschi G, Designer Drugs 2010, Wiley VCH.

[3] Dawson PH, Sun WF. A round robin on the reproducibility of standard operating conditions for the acquisition of library MS/MS spectra using triple quadrupols. Int. J. Mass Spectrom. Ion Proc. 1984;55:155-170.

[4] Junge Th, Rösner P, Westphal F. Product ion mass spectra of important organic ions, a free printed version can be ordered from the authors.

[5] Rösner P, Junge Th. Investigation of the alkylamino group of aliphatic an arylaliphatic amines by collision-induced dissociation mass spectra of $C_4H_{10}N^+$ immonium ions, J. Mass. Spectrom. 1996;31:1047-1053.

[6] State Bureaus of Criminal Investigation in Schleswig-Holstein, Hamburg, and Rheinland-Pfalz.

[7] Peters FT, Mattis P, Hoffmann K, Westphal F, Kießling G. Systematic screening for new designer drugs in routine sample extracts using full-scan GC-MS – experience from one year and identification of 4-methylamphetamine. Oral Presentation on the XVII. GTFCh-Symposium, Mosbach 2011 (see this proceeding).