Fatal 4-MTA Intoxication: Development of a LC Tandem Mass Spectrometric Assay for Multiple Matrices

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Outline

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- Case history
- Systematic Drug Screening
- Analysis of 4-MTA
  - Isolation of the compounds
  - Chromatography
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- Validation of the method
- Distribution of 4-MTA
- Conclusions

Introduction

4-methylthioamphetamine (4-MTA)

- Street name: Flatliner
- New, non-neurotoxic serotonin-releasing agent ($\leftrightarrow$ MDMA)
- Only three fatal overdose cases reported
  - toxicity range (Elliot):
    - 0.2 – 0.6 mg/L blood : moderately toxic
    - 0.6 – 1.5 mg/L blood : severely toxic
    - > 1.5 mg/L blood: fatal

Case history

- 27-year old drug dealer ✞ after collapse and reanimation
- Examination:

External: skin lesions

Internal:

✓ Lungs: emphysema, congestion & edema
✓ Heart: no cardiac anomalies
✓ Brain: slight congestion & edema of the white matter

↳ A wide variety of specimens was taken

Systematic Drug Screening

- Blood & urine
  - ✓ EMIT: amphetamines, cannabinoids, caffeine & cotinine
  - ✓ HPLC-DAD, GC-MS, GC-NPD: 4-MTA, MDMA & caffeine
- LC-MS/MS on a variety of matrices
  - Detailed distribution study in various blood and tissue samples

Analysis of 4-MTA

Isolation of the compounds

- 1mL sample (blood, urine or tissue homogenate)
- Internal standardisation (phentermine)
- L/L extraction
  - pH 9.5 (K_2CO_3)
  - Hexane/ethyl acetate (70/30)
  - MeOH/HCl
  - Evaporation of organic phase

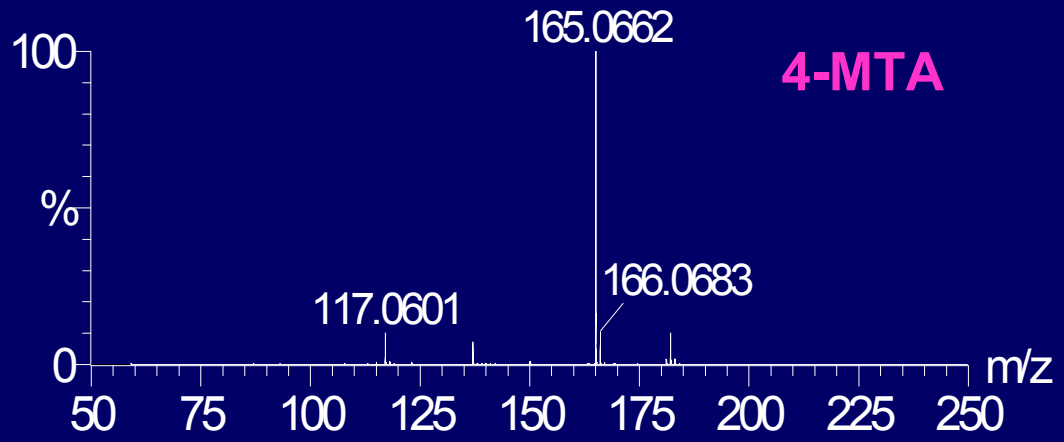
Chromatography

- Hypersil BDS phenyl column (narrow-bore)
- Gradient elution (NH_4Ac in H_20 & MeOH/AcCN)

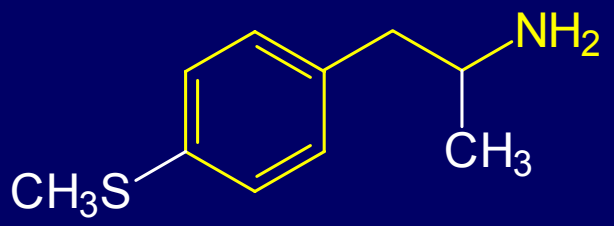
Mass spectrometry

- Q-TOF – ESI^+
- Parameter tuning

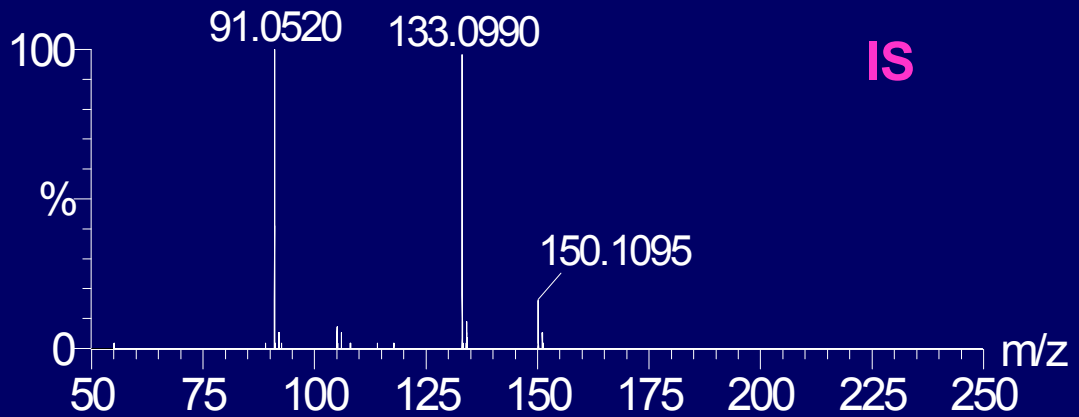
  ⇨ ☝ cone (20V) & collision energy (11-15eV)
- Quantification: reconstructed mass fragmentograms



4-MTA



Precursor mass 182.1
Product mass 165



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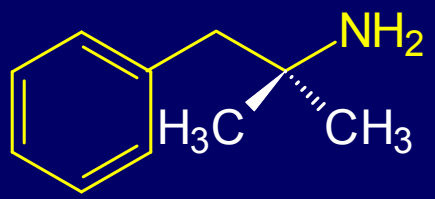
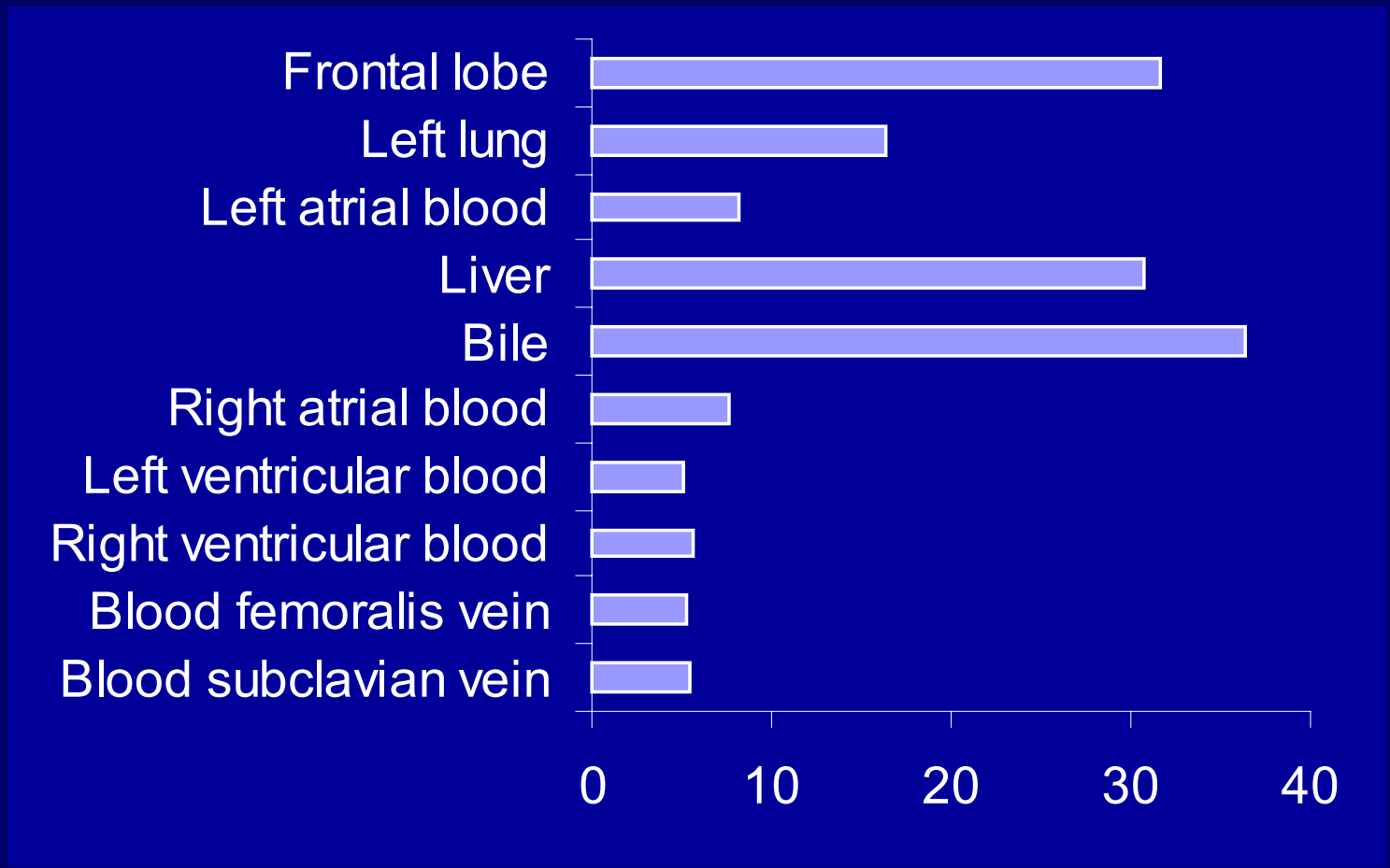


Precursor mass 150.1
Product masses 91 + 133.1

Method Validation Parameters (4-MTA)

	Blood	Urine	Tissue
Linearity			
Range (µg/L)	5-2500	5-2500	125-2500
Intercept	0.0049	0.0361	-0.0463
Slope	0.0030	0.0032	0.0026
R^2	0.9992	0.9992	0.9987
Total Reproducibility (n=5) CV%	<14%	<15%	<9%
LOD (µg/L)	2.5	2.5	50
LLQ (µg/L)	5	5	125
ULQ (µg/L)	2500	2500	2500

Distribution of 4-MTA (mg/L or mg/kg)



Conclusions

- Developed LC-MS/MS method
  - Selective
  - Sensitive (LOD 2.5 µg/L, except for 4-MTA in tissue homogenate)
  - Uncomplicated sample pretreatment
- Case:
  - Extensively examined
  - Interesting for cases without blood available
  - Contribute to distribution studies