Opiates, Opioids and Benzodiazepines, Amphetamines & Illicit Drug Forensic Analysis by LC/MS

Julie Cichelli, PhD
Agilent Technologies
Application Engineer

April 29, 2014
Agenda

- A method for the rapid analysis of over 65 analytes in a single LC/MS analysis run
- Simplified method development through the use of Dynamic MRM (dMRM) and databases
- Qualitative and Quantitative data analysis
- Customized Reporting
Targeted Analysis of Over 65 Analytes for Toxicology

- An extensive screen and quantification in 5 to 6 minutes
- Internal standard corrected quantification
- Multi-point calibration curve covering a wide dynamic range
- Secondary qualifier ion for each analyte
- Simple sample preparation:

  1. Enzyme/Acid
  2. Hydrolyze
  3. Dilute
  4. Analyze
Chromatographic Separation of Over 65 Analytes

Isobaric Mass Ions: different ions that have identical mass
Poroshell 120 Columns for HPLC and UHPLC

Poroshell 120 is a high efficiency, high resolution column choice for enhancing productivity in LC and LC/MS

Poroshell 120 Columns have:

- 80-90% efficiency of sub-2µm columns
- ~40-50% lower pressure
- 2x efficiency of 3.5µm (totally porous)
- A 2µm frit to reduce clogging
- A 600 bar pressure limit for HPLC or UHPLC

- The superficially porous particle is 2.7µm with a solid core (1.7µm) and porous outer layer with a 0.5µm diffusion path
Poroshell 120 Performance After 3000 Injections

- Dilute and shoot sample preparation
- Analytes covering a wide range of retention times show excellent reproducibility

<table>
<thead>
<tr>
<th>Analyte</th>
<th>%RSD (RT)</th>
<th>Analyte</th>
<th>%RSD (RT)</th>
<th>Analyte</th>
<th>%RSD (RT)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Morphine</td>
<td>0.7</td>
<td>Meperidine</td>
<td>0.4</td>
<td>Triazolam</td>
<td>0</td>
</tr>
<tr>
<td>Codeine</td>
<td>0.4</td>
<td>zolpidem</td>
<td>0.3</td>
<td>Naltrexone</td>
<td>0.1</td>
</tr>
<tr>
<td>hydrocodone</td>
<td>0.4</td>
<td>Fentanyl</td>
<td>0.1</td>
<td>chlordiazepoxide</td>
<td>0.1</td>
</tr>
<tr>
<td>MDMA</td>
<td>0.3</td>
<td>EDDP</td>
<td>0.1</td>
<td>Desmethyl diazepam</td>
<td>0.1</td>
</tr>
<tr>
<td>norFentanyl</td>
<td>0.2</td>
<td>Nitrazepam</td>
<td>0.1</td>
<td>Buprenorphine</td>
<td>0.3</td>
</tr>
<tr>
<td>Heroin</td>
<td>0.2</td>
<td>Propoxephine</td>
<td>0.1</td>
<td>Cocaethylene</td>
<td>0.2</td>
</tr>
<tr>
<td>Methyl Phenidate</td>
<td>0.2</td>
<td>Buprenorphine</td>
<td>0.3</td>
<td>11-nor-9-carboxy-delta9-thc</td>
<td>0</td>
</tr>
</tbody>
</table>
The Need for Dynamic MRM (dMRM)

- Nearly 200 MRM transitions for analytes, qualifier ions, and internal standards
- Monitoring all MRM transitions over the entire run results in poor quantification due to short dwell times and long cycle times
- dMRM only monitors each transition during the appropriate retention time window
Easy LC/MS Method Creation and Customization

A Database containing over 2500x compounds each with multiple MRM transitions ensures fast method creation for development.
Qualitative Screening
Find By MRM
Quantitative Analysis
Batch at a Glance
Data Review with Compounds at a Glance
Quantitative Analysis
Calibration Curves

**Carisoprodol**
- $R^2 = 0.996$

**EPPD**
- $R^2 = 0.998$

**Propoxyphene**
- $R^2 = 0.996$

**Fenfluramine**
- $R^2 = 0.999$

For Forensic Use.
Customizable Reporting
Conclusions

• Agilent makes it easy to create custom analytical panels for the measurement of Opiates, Opioids, Benzodiazepines, Amphetamines & Illicits
• Panels can be comprehensive or focused to suit your needs
• Dynamic MRM functionality, automatic optimization and MRM transition Database help make method development straightforward
• Results can be achieved with a simple Dilute & Shoot sample preparation
• Agilent’s MassHunter software is optimized for your data review and reporting workflow