**LC/MS Analysis of Opioid Glucuronide Metabolites in Urine on Ascentis® Express 2.7 μm F5 after Solid Phase Extraction (SPE) using Supel™-Select HLB**

Sample pretreatment: β-glucuronidase hydrolysis

The workflow for the LC/MS analysis of native drugs and associated metabolites from urine is shown here. The convenient liquid form of beta-glucuronidase from Sigma was used for hydrolysis of the glucuronide metabolites. SPE using Supel-Select HLB yielded clean extracts. The Ascentis Express F5 columns gave highly efficient, well-resolved peaks and rapid separation. LC-MS Ultra CHROMASOLV solvents and additives provided trouble-free operation, and Cerilliant CRMs gave reliable quantification.

**Mobile phase**

- A: 5 mM ammonium formate, pH 4.2 (adjusted with formic acid)
- B: 5 mM ammonium formate in (95:5 acetonitrile:water) pH 5.2 (adjusted with formic acid)

**Gradient**

- 15% B held for 1 min, to 70% B in 2 min, held at 70% B for 7 min

**Flow rate**

- 0.4 mL/min

**Column temp.**

- 35°C

**Detector**

- ESI(+) TOF, full scan, m/z 100-1000

**Injection**

- 3 μL

**Sample**

- 300 ng/mL

**Application No.**

- G006299

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**Related Products**

**analytical column**

Ascentis Express F5, 2.7 Micron HPLC Column (Supelco 53569-U)

**eluent**

Acetonitrile (Sigma-Aldrich 34998)

Ammonium formate (Fluka 14266)

Water (Sigma-Aldrich 34877)

**SPE tube or plate**

Supel™-Select HLB SPE 96-well Plate (Supelco 575662-U)

**standard**

- Buprenorphine solution (Cerilliant B-044)
- Buprenorphine glucuronide solution (Cerilliant B-035)
- Codeine solution (Cerilliant C-015)
- Codeine-6-β-D-glucuronide solution (Cerilliant C-087)
- Hydrocodone solution (Cerilliant H-003)
- Hydrocodone solution (Cerilliant H-004)
- Hydromorphone solution (Cerilliant H-051)

**Related Products**

Morphine solution (Cerilliant M-030)

Morphine-3β-gluconuronic acid solution (Cerilliant M-031)

Nalbuphine solution (Cerilliant N-059)

Nalbuphine glucuronide solution (Cerilliant N-045)

Oxydodecaine solution (Cerilliant O-002)

Oxymorphone-O-D solution (Cerilliant O-003)

Oxymorphone-3β-gluconuronic acid solution (Cerilliant O-030)

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**Figure:**

Counts vs. Acquisition Time (Min) for Opioid Drugs and Opioid Glucuronides.