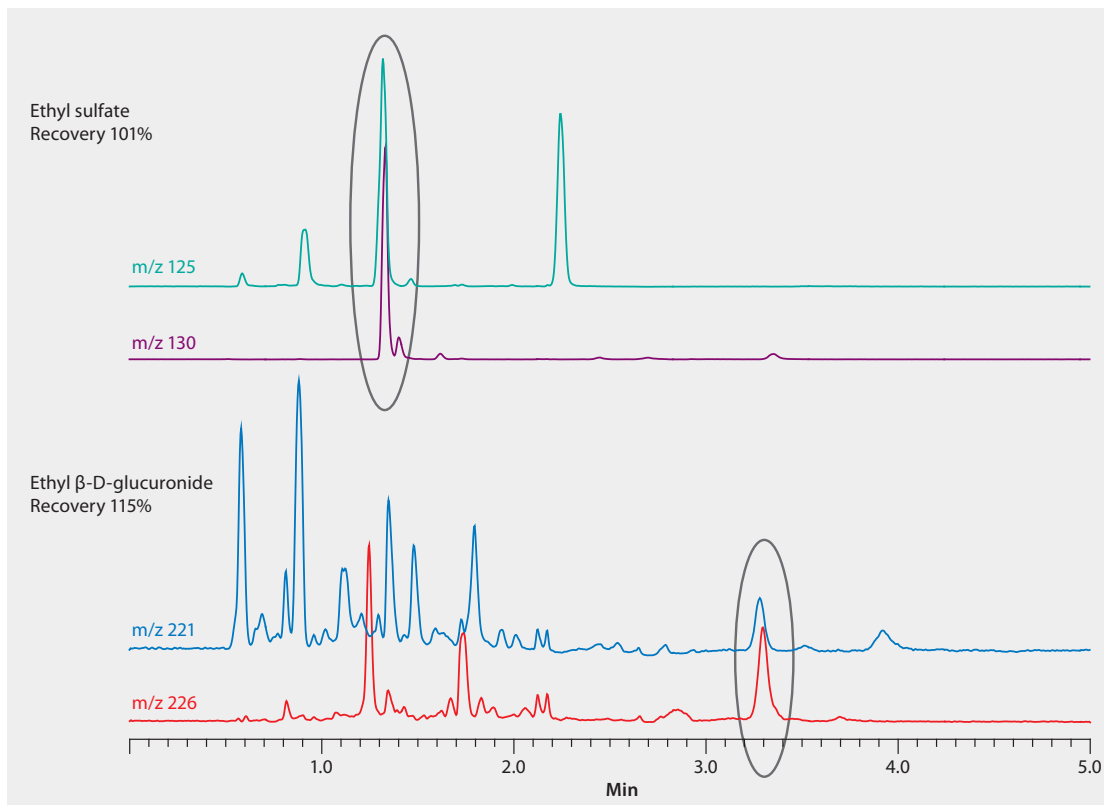


## LC/MS Analysis of Ethanol Metabolites in Urine on Ascentis® Express OH5 after Dilution with Deuterated Internal Standards

Ethyl sulphate (EtS) and ethyl glucuronide (EtG) are direct ethanol metabolites and may indicate recent alcohol consumption. The two compounds differ in their pathways for formation and degradation. Being polar compounds, they are poorly retained by C18 phases, and elute early in the chromatogram along with matrix compounds. This leads to poor or unreliable quantification. The method shown here uses HILIC mode on an Ascentis Express OH5 column to retain both analytes well, making it very likely to be more robust and reliable, as well as highly MS-friendly. Cerilliant CRMs provided reliable quantification. The internal standard was needed for accurate quantitation. External calibration resulted in a large excess of sulfate.

|                 |   |
|-----------------|---|
| market focus    | Clinical; Forensics and Toxicology  |
| column          | Ascentis Express OH5, 10 cm x 2.1 mm I.D., 2.7 µm particles (53757-U)   |
| mobile phase    | [A] 5 mM ammonium formate and 0.1% formic acid in 95:5, acetonitrile:water;<br>[B] 5 mM ammonium formate and 0.1% formic acid in 80:20, acetonitrile:water      |
| gradient        | 0 to 100% B in 1 min, held at 100% B for 4 min, to 0% B, held for 5 min   |
| flow rate       | 0.4 mL/min  |
| pressure        | 1450 psi (100 bar)  |
| column temp.    | 30 °C   |
| detector        | MS, ESI(-), 125.0 m/z, 126.1 m/z, 221.1 m/z, 226.2 m/z  |
| injection       | 5 µL  |
| sample          | urine spiked with ethyl sulfate and ethyl-β-D-glucuronide, both at 50 ng/mL, diluted 20:1 in acetonitrile containing deuterated internal standards at 100 ng/mL |
| Application No. | <b>G1006518</b>   |



**Related Products**

- accessory**  
Millex® syringe filter units, disposable, PTFE ([Sigma Z227544](#))
- analytical column**  
Ascentis® Express OH5, 2.7 Micron HPLC Column ([Supelco 53757-U](#))
- mobile phase component**  
Ammonium formate ([Fluka 70221](#))  
Formic acid ([Fluka 56302](#))

**Related Products**

- solvent**  
Acetonitrile ([Fluka 14261](#))  
Water ([Fluka 14263](#))
- standard**  
Ethyl-beta-D-glucuronide-d5, 1.0 mg/mL ([Cerilliant E-063](#))  
Ethyl-β-D-glucuronide ([Cerilliant E-015](#))  
Ethyl sulfate sodium salt ([Cerilliant E-064](#))  
Ethyl-d5 sulfate sodium salt ([Cerilliant E-066](#))