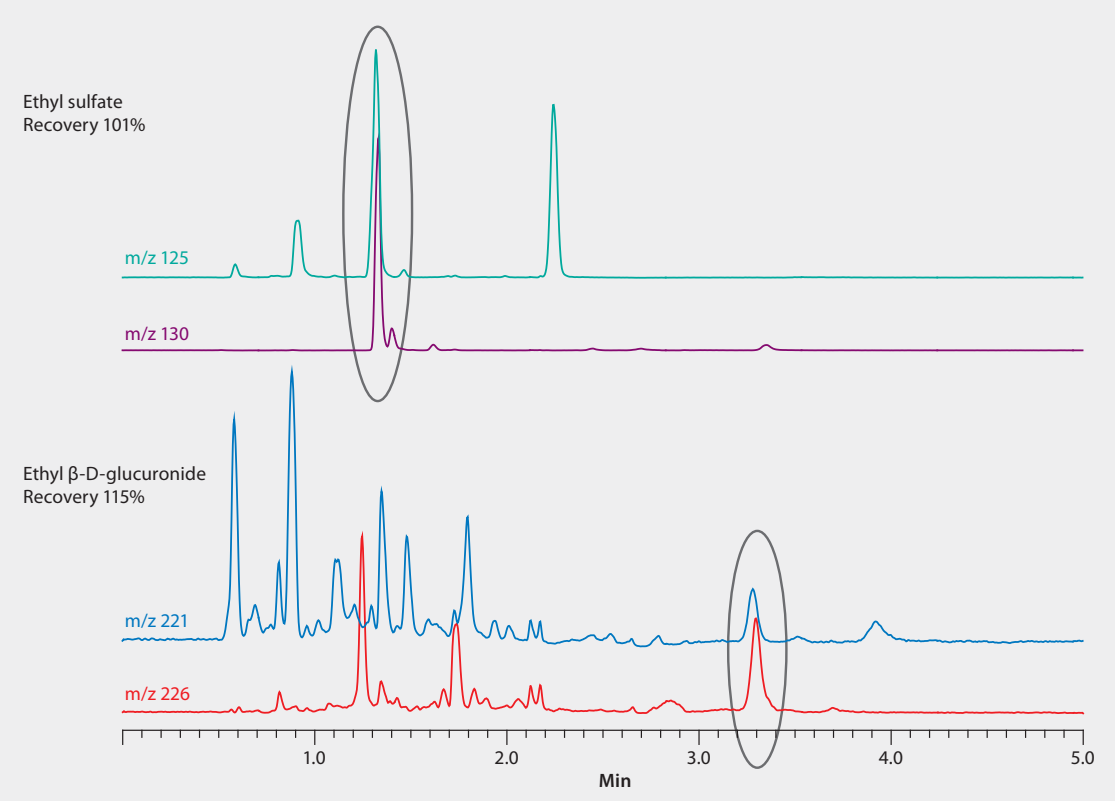
LC/MS Analysis of Ethanol Metabolites in Urine on Ascentis® Express OH5 after Dilution with Deuterated Internal Standards

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; [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-ß-D-glucuronide, both at 50 ng/mL, diluted 20:1 in acetonitrile containing deuterated internal standards at 100 ng/mL
Application No.	G1006518



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-d_5 sulfate sodium salt (Cerilliant E-066)