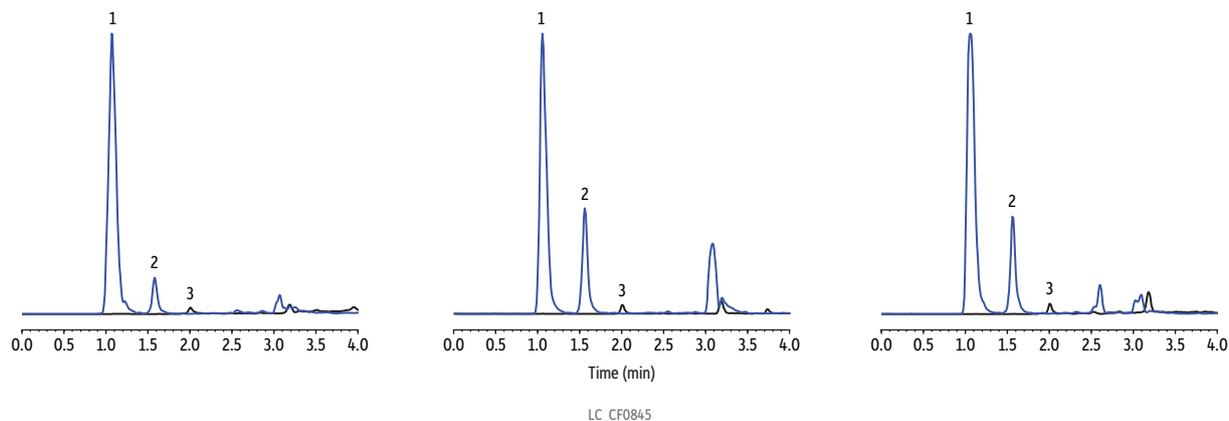


EtG and EtS in Urine on Force Biphenyl (100 mm x 3 mm, 3 µm)



Peaks	tr (min)	Precursor	Product 1	Product 2
1. Urinary interference	1.06	125.0	80.1	-
2. EtS	1.56	125.0	96.8	80.1
3. EtG	2.01	221.1	75.1	85.2

Column Force Biphenyl (cat.# 962931E)
 Dimensions: 100 mm x 3 mm ID
 Particle Size: 3 µm
 Pore Size: 100 Å
 Guard Column: Force Biphenyl EXP guard cartridge 5 mm, 3 mm ID, 3 µm (cat.# 962950253)
 Temp.: 30 °C

Standard/Sample
 Ethyl sulfate sodium salt (EtS) standard (cat.# 34103)
 Ethyl-beta-D-glucuronide (EtG) standard, 1000 µg/mL, methanol, 1 mL/ampul (cat.# 34101)

Diluent: Water
 Conc.: 50 ng/mL in urine
 Inj. Vol.: 10 µL

Mobile Phase
 A: Water, 0.1% formic acid
 B: Methanol, 0.1% formic acid

Time (min)	Flow (mL/min)	%A	%B
0.00	0.6	100	0
3.00	0.6	5	95
4.00	0.6	2	98
4.01	0.6	100	0
6.00	0.6	100	0

Max Pressure: 427 bar
Detector Shimadzu 8060 MS/MS
 Ion Source: Electrospray
 Ion Mode: ESI-
 Mode: MRM
Instrument Shimadzu Nexera X2

Sample Preparation
 Urine (100 µL) was aliquoted to an autosampler vial (cat.#21143), fortified at 50 ng/mL with EtG and EtS, and water (900 µL) was added before the sample was capped (cat.#24498), vortexed for ~30 seconds, and 10 µL injected onto the LC-MS/MS for analysis.

Notes
 An Ultra Shield UHPLC PreColumn Filter 0.2 µm frit (cat.#25811) was installed before the guard cartridge.