EtG/EtS IN HAIR BY LC-MS/MS USING 200 MG CLEAN SCREEN® ETG EXTRACTION COLUMN

Part #

CSETG203 – CLEAN SCREEN® ETG 300 mg, 3 mL Tube

1. **PREPARE SAMPLE:**

   Into a clean glass tube add approx. 50-100 mg of decontaminated hair.

   Add 1 mL of D.I. H_2O, add internal standards*

   Vortex mix

   Incubate at 40 ^{0}C for 12 hours

   Centrifuge as appropriate

2. **CONDITION CLEAN SCREEN® EXTRACTION COLUMN:**

   1 x 3 mL CH_3OH containing 1% Formic Acid

   1 x 3 mL D.I. H_2O containing 1% Formic Acid

   NOTE: Aspirate at full vacuum or pressure

3. **APPLY SAMPLE:**

   Load at 1 to 2 mL/minute

4. **WASH COLUMN**

   1 x 3 mL D.I. H_2O:

   Dry column (**10 minutes** at full vacuum or pressure)

5. **ELUTE EtG/EtS ANALYTES:**

   2 x 3 mL CH_3OH containing 1% Formic Acid

   Collect eluate at 1 to 2 mL/minute

6. **DRY ELUATE:**

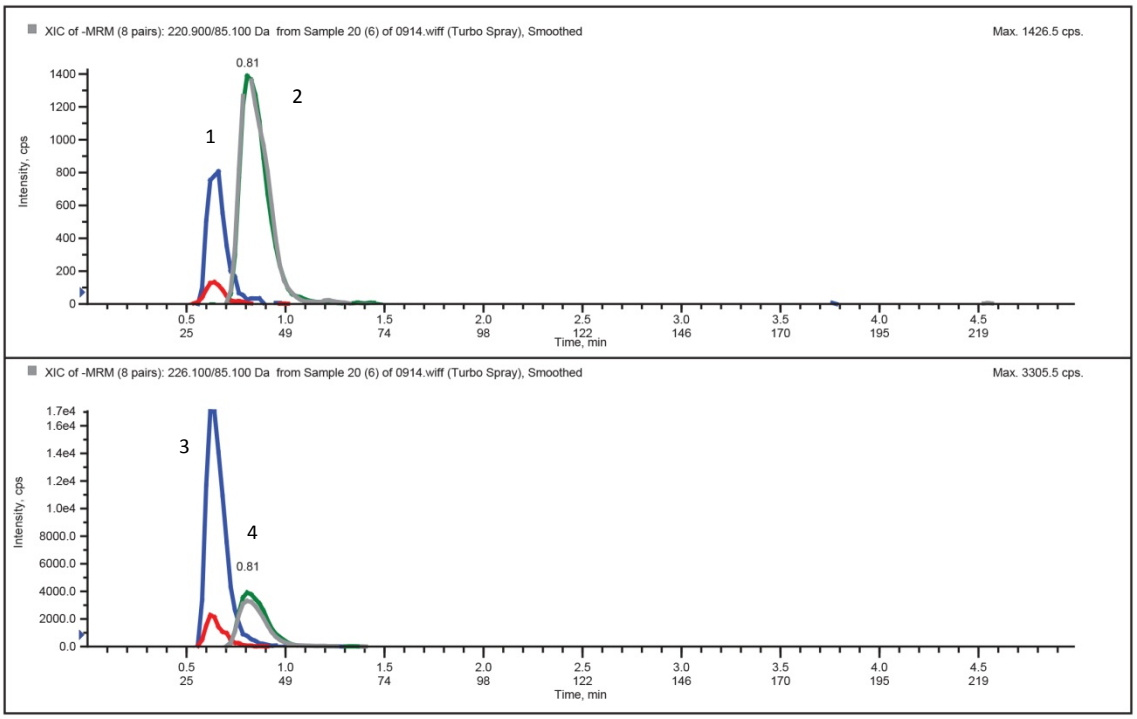
   Evaporate to dryness at < 40 °C

7. **RECONSTITUTE:**

   with 50-100 µL of Mobile Phase

INSTRUMENT CONDITIONS (LC-MS/MS):

CHROMATOGRAMS



Analyte	MRM Transitions		Relative Retention Time (min)
	Q1	Q3	
1.EtS	125.1	95.8	0.65
2.EtG	220.9	75.1	0.83
3.EtS D_5	130.1	97.8	0.63
4.EtG D_5	226.1	74.9	0.81

PARAMETERS

Mobile Phase A: 0.1% Formic Acid in D.I. H_2O

Mobile Phase B: 0.1% Formic Acid in Acetonitrile

Flow Rate: 0.35 mL/minute

Polarity: Negative

Injection Volume: 20 µL

LC Column: Diamond Hydride LC Column 100 x 2.1 mm (4µm)

Instrument: API 3200 Qtrap MS/MS with Shimadzu Prominence UFLC

Isocratic:

Time	%A	%B
0.00	50	50
5.00	50	50