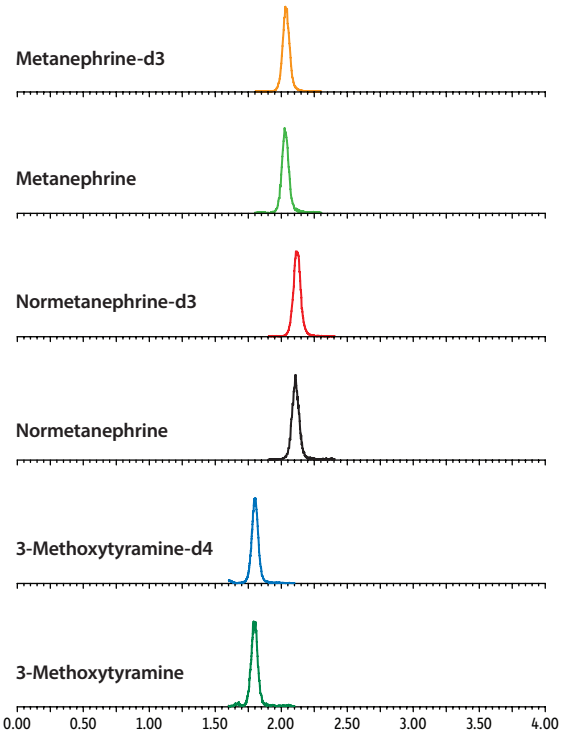
Metanephrine, Normetanephrine, and 3-Methoxytyramine in Human Plasma on Raptor HILIC-Si



LC_CF0689

Peaks	t_R (min)	Conc. (pg/mL)	Precursor Ion	Product Ion
1. 3-Methoxytyramine-d4 (IS)	1.80	400	155.07	122.93
2. 3-Methoxytyramine	1.80	50	151.00	119.00
3. Metanephrine-d3 (IS)	2.03	200	183.00	151.15
4. Metanephrine	2.03	50	179.94	148.22
5. Normetanephrine-d3 (IS)	2.11	400	169.00	136.96
6. Normetanephrine	2.11	50	166.00	134.02

Column
Dimensions: 50 mm x 2.1 mm ID
Particle Size: 2.7 µm
Pore Size: 90 Å
Temp.: 30 °C
Standard/Sample
Diluent: Mobile phase A:mobile phase B (10:90)
Inj. Vol.: 10 µL
Mobile Phase
A: Water, 100 mM ammonium formate, pH 3.0
B: Acetonitrile

Time (min)	Flow (mL/min)	%A	%B
0.00	0.3	10	90
5.00	0.3	10	90

Detector MS/MS
Ion Mode: ESI+
Mode: MRM
Instrument UHPLC
Sample Preparation Charcoal stripped human plasma was fortified with analytes at 50 pg/mL (0.25, 0.27, and 0.30 nmol/L for metanephrine, normetanephrine, and 3-methoxytyramine, respectively). Internal standard (IS) was prepared at 4 ng/mL for metanephrine-d3 and at 8 ng/mL for normetanephrine-d3 and 3-methoxytyramine-d4 in methanol. The plasma sample (200 µL) was mixed with 10 µL of IS solution and 600 µL of 50 mM ammonium acetate solution. The mixture was loaded in an EVOLUTE EXPRESS WCX 96-well plate (30 mg) and washed with 1 mL water and 1 mL methanol:acetonitrile (50:50). The elution was performed twice with 0.9 mL of 5% formic acid in methanol:acetonitrile (50:50) and evaporated to dryness at 55 °C under a gentle stream of nitrogen. Dried extract was reconstituted with 100 µL of diluent and injected (10 µL) for analysis.