



NARCOTICS/METABOLITES PANEL IN BLOOD, PLASMA/SERUM, URINE, TISSUE BY LC-MS/MS OR GC-MS CLEAN SCREEN® DAU EXTRACTION COLUMN

Part #

CSDAU – CLEAN SCREEN® DAU

SLDA50ID21-5UM – SELECTRA® HPLC Column 50 x 2.1 mm, 5 µm

1. PREPARE SAMPLE:

To 1 mL of 100 mM phosphate buffer (pH 6.0) add internal standards
Add 1-2 mL of blood, plasma/ serum, urine, or 1 g (1:4) tissue homogenate
Mix/vortex and let stand for 5 minutes
Add 2 mL of 100 mM phosphate buffer (pH 6.0). Mix/vortex
Sample pH should be 6.0 ± 0.5.
Adjust pH accordingly with 100 mM monobasic or dibasic sodium phosphate.
Centrifuge for 10 minutes at 2000 rpm and discard pellet

2. CONDITION CLEAN SCREEN® EXTRACTION COLUMN:

1 x 3 mL CH₃OH
1 x 3 mL D.I. H₂O
1 x 3 mL 100 mM phosphate buffer (pH 6.0)
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. H₂O
1 x 3 mL 100 mM acetic acid
1 x 3 mL CH₃OH
Dry column (5 minutes at full vacuum or pressure)

5. ELUTE NARCOTICS/METABOLITES:

1 x 3 mL CH₂Cl₂/ IPA/ NH₄OH (78:20:2 v/v)
Collect eluate at 1 to 2 mL/minute
or
1 x 3mL Ethyl Acetate/ IPA/ NH₄OH (78:20:2 v/v)

NOTE: Prepare elution solvent daily

Add IPA/ NH₄OH, mix, then add CH₂Cl₂ (pH 11-12)

6. DRY ELUATE:

Evaporate to dryness at < 40 °C

7. RECONSTITUTE / DERIVATIZE:

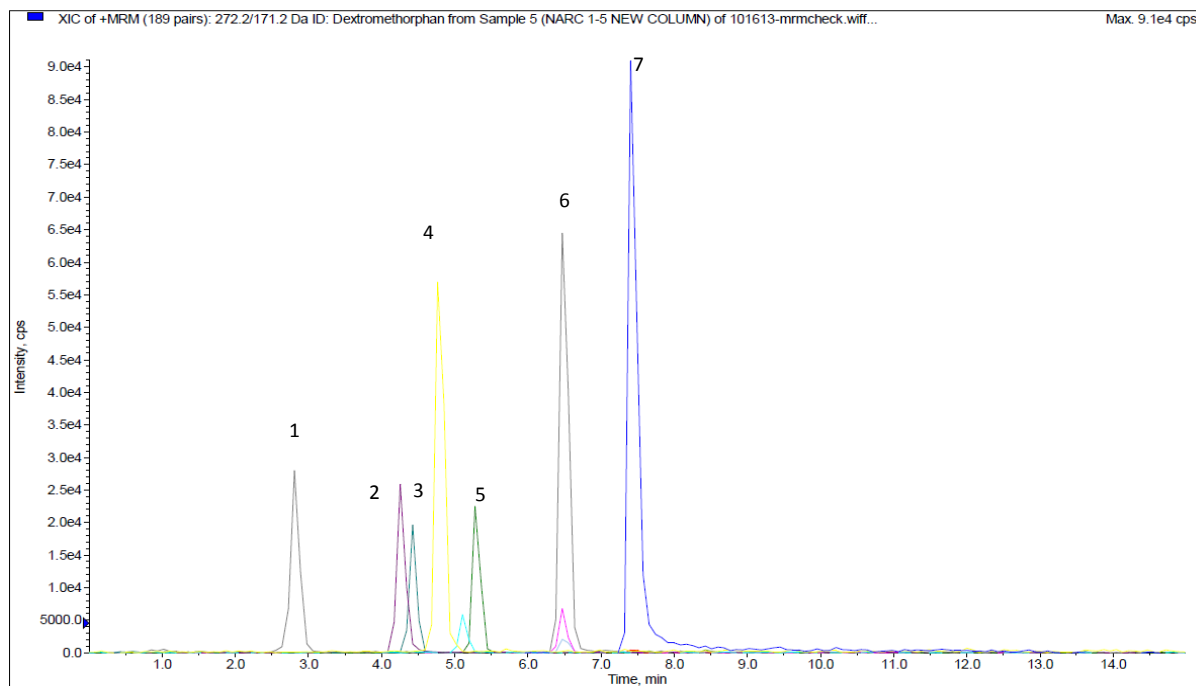
- **LC-MS/MS:** Reconstitute sample in 100 µL of mobile phase
Inject 20 µL.
- **GC-MS:** Dissolve residue in 100 µL of Ethyl Acetate

Alternate Derivatization

Dissolve residue in 50 µL of Ethyl Acetate and 50 µL of derivatizing reagent and react at 70 °C for 30 minutes; Cool and inject 1-2 µL

INSTRUMENT CONDITIONS (LC-MS/MS):

CHROMATOGRAM



Analyte	MRM Transitions		Relative Retention Time (min)
	Q1	Q3	
1. Naloxone	328.2	310.2	2.80
2. Norketamine	224.1	207.1	4.25
3. NorFentanyl	233.2	84.1	4.45
4. Tramadol	264.2	58.0	4.80
5. Normeperdine	234.1	91.2	5.10
6. Norbuprenorphine	414.2	187.1	6.50
7. DXM	272.2	171.2	7.42

PARAMETERS

Mobile Phase A: 0.1% Formic Acid in D.I. H₂O

Mobile Phase B: 0.1% Formic Acid in Methanol

Flow Rate: 0.5 mL/minute

Polarity: Positive

Injection Volume: 20 µL

LC Column: Selectra[®] DA HPLC Column 50 x 2.1 mm 5 µm

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

Gradient:

Time	%A	%B
0.00	80	20
0.50	80	20
12.00	10	90
12.01	80	20
15.00	STOP	