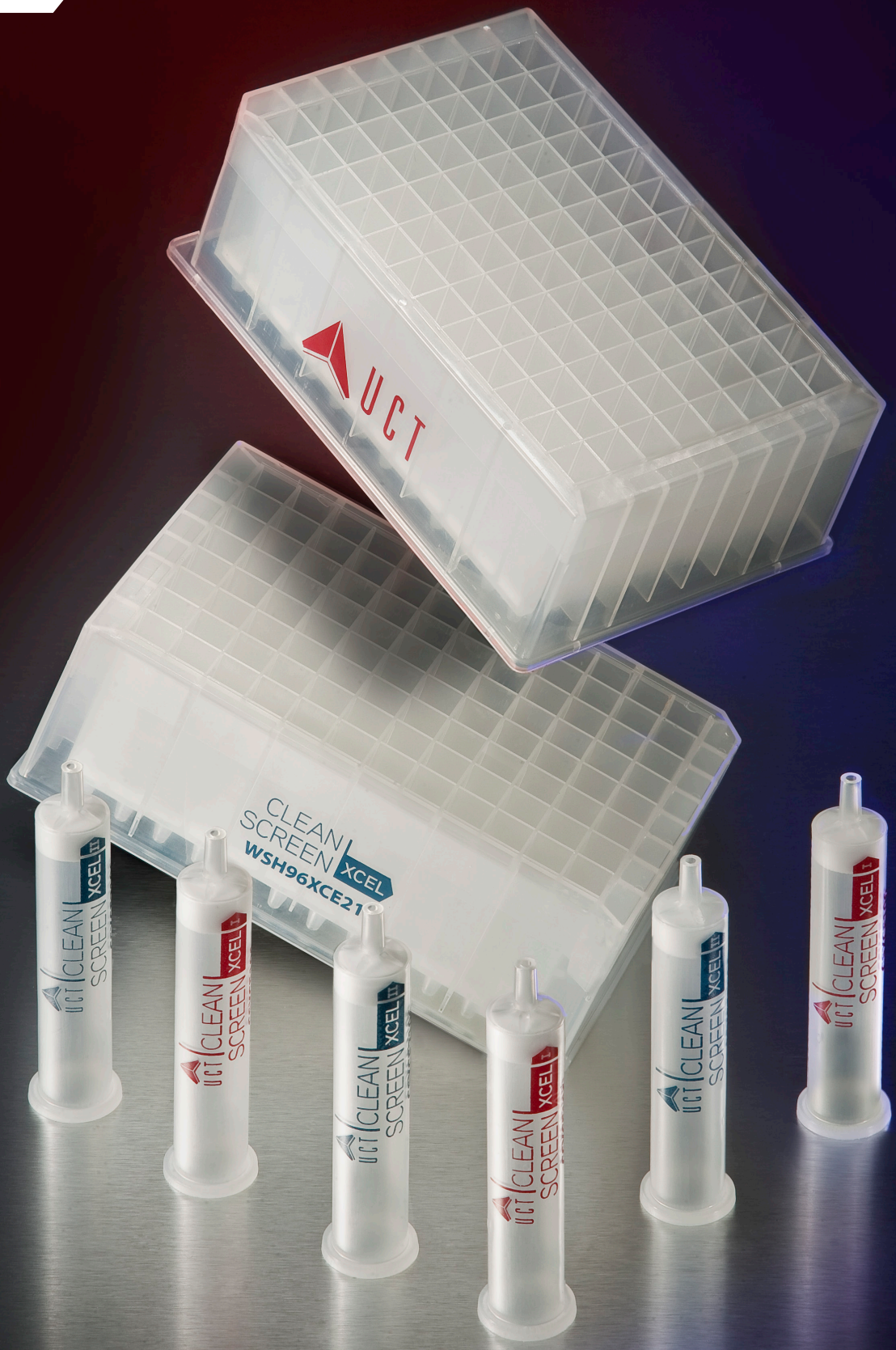
FORENSICS

UCT

CLEAN SCREEN XCEL®

EXCELLENCE JUST GOT BETTER



INNOVATION THROUGH CHEMISTRY

CLEAN SCREEN XCEL® COLUMNS

Clean Screen Xcel® solid phase extraction columns are designed to reduce the number of steps in the extraction. The result is a column that reduces sample prep times versus traditional SPE and minimizes the amount of solvent necessary. Additional advantages include reduced sample size and improved cleanliness and recovery versus dilute and shoot.

BENEFITS

- Sorbent conditioning step is eliminated
- Reduced sample size
- Increased sensitivity vs. dilute and shoot
- Decreased extraction steps
- Increased recovery values vs. dilute and shoot

CLEAN SCREEN XCEL® I:

Extracts a wide array of basic drugs including benzodiazepines and opiates.

Organic Loading = 12.4%
Surface Area = 500 m^2/g
Average Pore Size = 60 Å
Pore Volume = 0.77 cm^3/g

COLUMNS

Tube Volume (mL)	Sorbent Amount (mg)	Units per Pack	Part Number
1	130	100	CSXCE111
3	130	50	CSXCE103
6	130	50	CSXCE106
6	200	50	CSXCE206
10	130	50	ZSXCE010

WELL PLATES

Number of Wells	Sorbent Amount (mg)	Units per Pack	Extended Drip Tip	Part Number
48	130	1	NO	WSH48XCE11
96	80	1	YES	WSH96XCE108-LD
96	130	1	NO	WSH96XCE11
96	130	1	YES	WSH96XCE11-LD

CLEAN SCREEN XCEL® II:

Designed solely for the extraction of the THC metabolite.

Organic Loading = 16.7%
Surface Area = 500 m^2/g
Average Pore Size = 60 Å
Pore Volume = 0.77 cm^3/g

COLUMNS

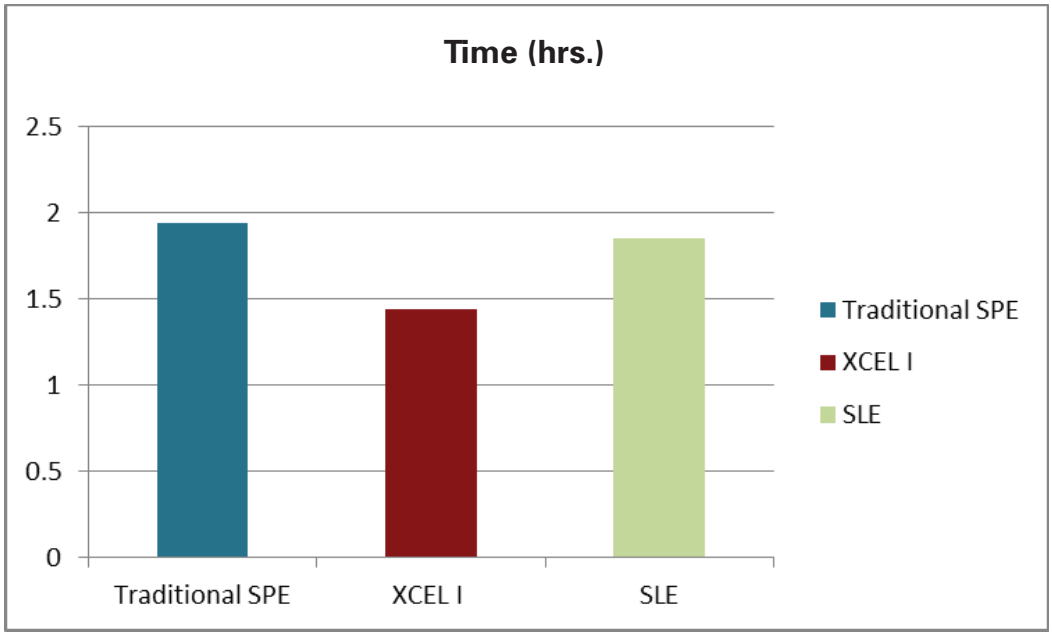
Tube Volume (mL)	Sorbent Amount (mg)	Units per Pack	Part Number
1	130	100	CSXCE211
3	130	50	CSXCE2103
6	130	50	CSXCE2106
6	200	50	ZSXCE2010
10	130	50	ZSXCE010

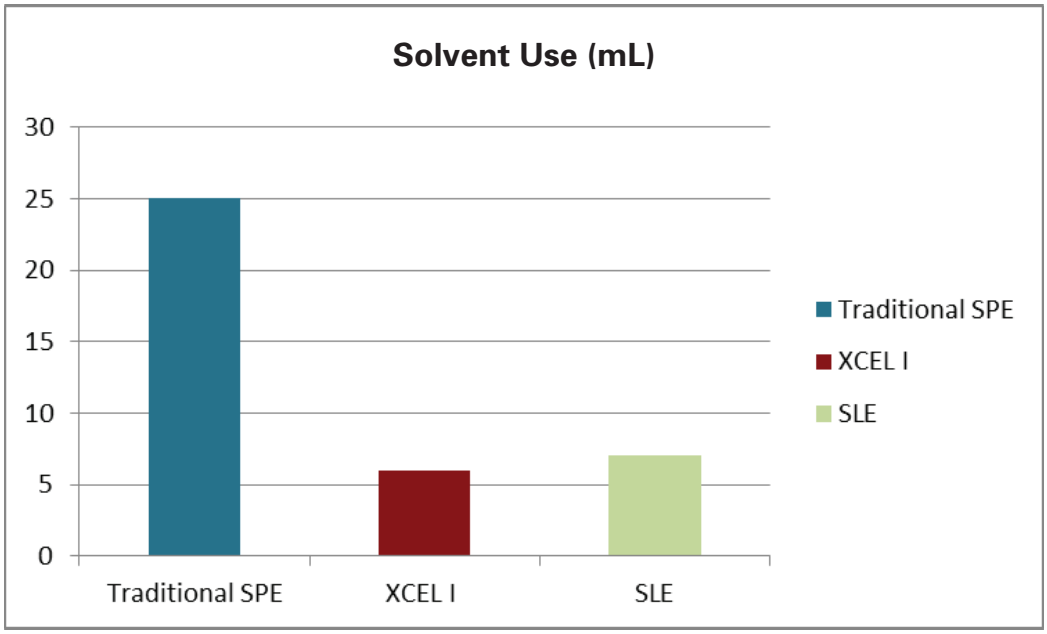
WELL PLATES

Number of Wells	Sorbent Amount (mg)	Units per Pack	Extended Drip Tip	Part Number
48	130	1	NO	WSH48XCE211
96	80	1	YES	WSH96XCE208-LD
96	130	1	NO	WSH96XCE211

CLEAN SCREEN XCEL®

uses less solvent and has shorter prep times than either traditional SPE or SLE extraction techniques.





SCREENING METHOD FOR 121 ACIDIC, NEUTRAL AND BASIC DRUG ANALYTES IN PLASMA, SERUM, URINE, OR TISSUE BY LC-MS/MS

Part #

CSXCE111 – CLEAN SCREEN XCEL® I 130 mg, 1 mL Tube
BETA-GLUC-10 – SELECTRAZYME® Beta-glucuronidase
SLDA50ID21-5UM – SELECTRA® DA HPLC Column, 50 x 2.1 mm, 5 µm
SLDAGDC21-5UM - SELECTRA® DA 5 µm Guard Cartridge
SLGRDHLDR - Guard Cartridge Holder

CLEAN SCREEN XCEL® I Columns

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
   NOTE: See Hydrolysis step if required

   Hydrolysis (for urine samples only): To 1-2 mL of urine sample, add 1 mL of acetate buffer (pH 5.0) containing 5,000 units/mL Selectrazyme®ββ-glucuronidase.
   Optionally, add 1 mL of acetate buffer and 25-50 µL of concentratedββ-glucuronidase.
   Vortex and heat for 1-2 hours at 65°C.
   (Hydroxylamine can be added to sample here if oxime derivative is preferred.) Allow sample to cool

2. **APPLY SAMPLE:**
   Load sample directly to column without any preconditioning
   Pull sample through at a rate of 1-2 mL/ minute
   Dry column thoroughly under full vacuum or positive pressure for 1 minute

3. **WASH 1 – ACIDIC & NEUTRAL COMPOUNDS (FRACTION 1):**
   Add 1 x 1 mL of DI H_2O
   Apply pressure to column for ~1 minute (either vacuum (10mm Hg) or positive pressure(~80-100psi). This ensures that the entire sample and any residual is pulled through to waste
   Add 1 x 1 mL of 0.1M Acetic Acid
   Apply pressure to column for ~1minute (either vacuum (10mm Hg) or positive pressure(~80-100psi).
   Add 1 x 2 mL Hexane to remove residual aqueous phase
   Dry column (5 minutes at full vacuum or pressure)

4. **ELUTION 1 – ACIDIC & NEUTRAL COMPOUNDS (FRACTION 1):**
   Add 1 x 1 mL Ethyl Acetate: Hexane (50:50) Collect eluate at 1 to 2 mL/minute

5. **DRY ELUTE:**
   Evaporate fraction to complete dryness under stream of dry air or nitrogen at ~ 35 °C Reconstitute with 100 µL of Mobile Phase

6. **WASH 2 - BASIC COMPOUNDS (FRACTION 2):**
   Add 1 x 1 mL of 2% Acetic Acid/98% Methanol
   Dry column 5 minutes at full vacuum (10mm Hg) or positive pressure (~80-100 psi)

7. **ELUTION 2 - BASIC COMPOUNDS (FRACTION 2):**
   1 x 1 mL of CH_2Cl_2/ IPA/ Ammonium Hydroxide (78/20/2).

8. **DRY ELUTE:**
   Evaporate fraction to complete dryness under stream of dry air or nitrogen at ~ 35 °C. Take care not to overheat or over evaporate. Certain compounds are heat labile, such as the amphetamines and phencyclidine. Reconstitute with 100 µL of Mobile Phase

NOTES:

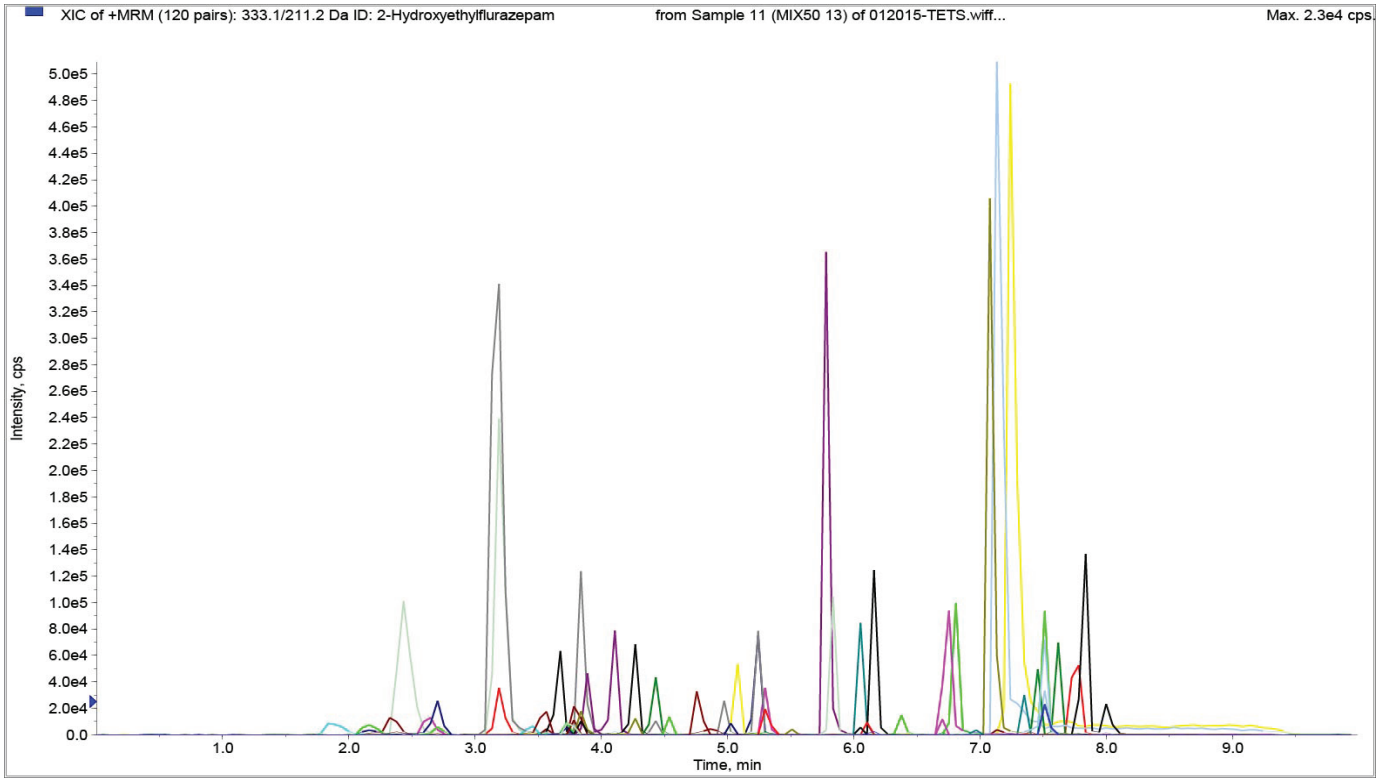
(1) Fraction 1 (Acid Neutrals) and Fraction 2 (Bases) can be combined together if need be. This is not generally recommended as the Acid/ Neutral fraction tends to be dirtier than the Basic one, so for more effective results, keep fractions separate.

(2) A keeper solvent such as DMF can be used to prevent the volatilization of amphetamines and phencyclidine. Use 30-50 µL of high purity DMF in the sample (Fraction 2) before evaporation.

(3) A 1% HCl in CH_3OH solution has been used to prevent volatization by the formation of the hydrochloric salt of the drugs. Add 1 drop of the solution prior to evaporating then continue to dryness.

(4) The hexane wash step can be removed if user is looking to analyze for Parent THC.

(5) To extract the benzodiazepine group at higher recovery, following the elution of the Acidic/Neutral drugs, a second elution can be done prior to moving on to the second wash phase. The second elution solvent would consist of 98% Ethyl Acetate/ 2% Ammonium Hydroxide.



HPLC Conditions

Instrument: Shimadzu HPLC 20-AD
Detector: AB Sciex API 3200 Qtrap MS/MS
LC Column: UCT Selectra® DA HPLC Column 50 x 2.1mm, 5 µm
Ionization mode: ESI+
Injection Volume: 20 µL
Flow Rate: 0.6 mL/minute
Mobile Phase A: 0.1% formic acid in water
Mobile Phase B: 0.1% formic acid in MeOH

Gradient:

Time	%A	%B
0.00	90	10
0.50	90	100
4.00	60	40
7.50	15	85
8.50	10	90
8.51	90	10
10.00	STOP	

Limits of detection for each compound class†

Representative Compounds	LOD Range (ng/mL)	Representative Compounds	LOD Range (ng/mL)
Ampehatmine/Methamphetamine	10	Benzodiazepines (24)	5-10
MDMA/MDA/MDEA	5-10	Fentanyl/Norfentanyl	1-5
Opiates (9)*	2	Carisoprodol/Meprobamate	25-100
Methadone/EDDP	5-20	Cathinones/Phenethylamines (10)	10-20
Sympathomimetic Amines	20	Anti-Psychotics (7)	10-50
Meperidine/Normeperidine	20	Z Drugs/Insomina Treatment (3)	20-25
Cocaine/BE/EME	5	Pain Management Compounds (14)	2-20
Tricyclic Antidepressants (16)	5-50	Caffeine/Theobromine/Theophylline	20-50
Gabapentin/Pregabalin	50	PCP/Ketamine/Norketamine	5-10
Acetaminophen	50	Anti-histamines	10-20

( )* - Number of compounds analyzed in this class.

†For the full application note with the complete compound list and LOD's contact your regional representative or download from our website, www.unitedchem.com:

SCREENING METHOD FOR 121 ACIDIC, NEUTRAL AND BASIC DRUG ANALYTES IN PLASMA, SERUM, URINE, OR TISSUE BY LC-MS/MS.

CARBOXY-THC IN URINE BY LC-MS/MS OR GC-MS USING CLEAN SCREEN XCEL® II EXTRACTION COLUMN

CLEAN SCREEN XCEL® II Columns

Part #

CSXCE2106 – CLEAN SCREEN XCEL® II 130 mg 6 mL Tube
SPHPHO6001-5 – Select pH Buffer Pouches 100 mM Phosphate pH 7.0
SLDA50ID21-5UM - Selectra® DA HPLC Column, 50 x 2.1 mm, 5 µm
SMSTFA-1-1 – SELECTRA-SIL® MSTFA w/ 1% TMCS
SBSTFA-1-1 – SELECTRA-SIL® BSTFA w/ 1% TMCS
SLDAGDC21-5UM - SELECTRA® DA 5 µm Guard Cartridge
SLGRDHLDR - Guard Cartridge Holder

1. PREPARE SAMPLE-ENZYME HYDROLYSIS OF GLUCURONIDES:

To 2 mL of urine add internal standard and 50 µL of
10 M NaOH
Mix/vortex
Hydrolyze for 15 minutes at 60-70 °C. Cool before proceeding
Adjust sample pH to 7.0 with 50 µL of 1:1 H_2O: Glacial Acetic Acid.
Add 200 µL pH 7.0 100mM Phosphate Buffer
(pH should be ~7.0)

2. APPLY SAMPLE:

Load at 1 to 2 mL/minute
Dry column (2 minutes at full vacuum or pressure)

3. WASH COLUMN:

1 x 2 mL Hexane
Dry Column at full vacuum or pressure for 10 minutes

4. ELUTE ANALYTE:

1 x 3 mL Hexane/ Ethyl Acetate/ Glacial Acetic Acid (49:49:2)
Collect eluate at 1 to 2 mL/minute

5. DRY ELUATE:

Evaporate to dryness at < 40 °C

6. RECONSTITUTE / DERIVATIZE:

- LC-MS/MS: Reconstitute sample in 100 µL of mobile phase
  Inject 20 µL.

- GC-MS: Dissolve residue in 50 µL of Ethyl Acetate and 50 µL
  MSTFA (with 1%TMCS)
  Overlay with N_2 and cap. Mix/vortex
  React 30 minutes at 70 °C; Cool and inject 1 µL

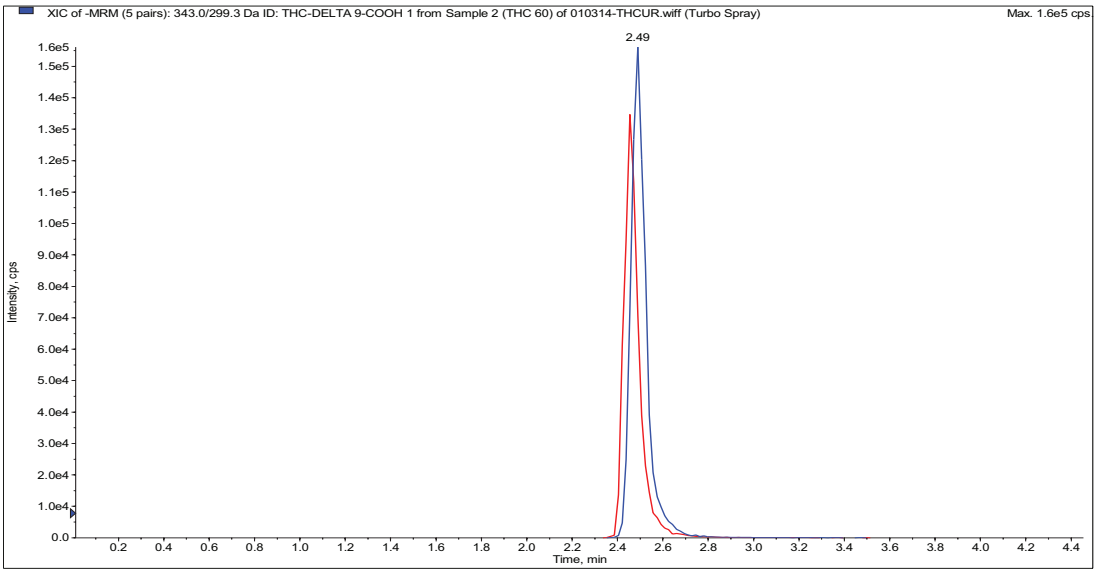
Alternate Derivatization

1. Form TMS Derivatives by adding 50 µL BSTFA w/ 1% TMCS and 50 µL of Ethyl Acetate; React 45 minutes at 70 °C

THC-delta-9-COOH
Comparison of Extraction Methods - XCEL II vs. Liquid/Liquid

Sample ID#	Liquid / Liquid Result (ng/mL)	2 Step XCEL I SPE Result (ng/mL)	Liquid / Liquid Area Response	2 Step XCEL I SPE Area Response
60 control	60.8	63.2	193689	521993
300 control	226	299	663859	1793767
Patient #1	157	171	366285	1210294
Patient #2	26	30.1	74689	283042
Patient #3	57	73	125662	497268
Patient #4	13.8	14	50238	168674
Patient #5	57.9	56.2	148767	469350
30 control	26.5	27	95809	215516

CARBOXY-THC



Analyte		MRM Transitions		Relative Retention Time (minutes)
		Q1	Q3	
1.	THC-DELTA 9-COOH D9	352	308	2.44
2.	THC-DELTA 9-COOH	343	299	2.49

HPLC Conditions

Instrument: Shimadzu Prominence UFLC
Detector: API 3200 Qtrap MS/MS
HPLC Column: SELECTRA® DA HPLC Column 50 x 2.1 mm 5 µm
Polarity: Negative
Reconstitute: 100 µL
Injection Volume: 20 µL
Flow Rate: 0.5 mL/minute
Mobile Phase A: 0.1% formic acid in water
Mobile Phase B: 0.1% formic acid in MeOH
Gradient:

Time	%A	%B
0.00	60	40
2.00	30	70
2.50	10	90
2.51	60	40
4.00	STOP	

SYMPATHOMIMETIC AMINES IN BLOOD, PLASMA/ SERUM, URINE, OR TISSUE BY LC-MS/MS CLEAN SCREEN XCEL® I EXTRACTION COLUMN

Select pH Buffer Pouches

Part #

CSXCE111 – CLEAN SCREEN XCEL® 130 mg, 1 mL Tube
SPHPHO6001-5 – Select pH Buffer Pouches 100 mM Phosphate pH 6.0
PFAA-0-1 – SELECTRA-SIL® PFAA
SPFPOH-1 – SELECTRA-SIL® PFPOH
SHFAA-0-1– SELECTRA-SIL® HFAA
SBSTFA-1-1 – SELECTRA-SIL® BSTFA w/ 1% TMCS
SLDA100ID21-3UM – SELECTRA® DA HPLC Column, 100 x 2.1 mm, 3 µm
SLDAGDC20-3UM - SELECTRA® DA 3 µm Guard Cartridge
SLGRDHLDR - Guard Cartridge Holder

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. APPLY SAMPLE:

Load sample directly to column without any preconditioning.
Pull sample through at a rate of 1-2 mL/ minute.
Dry column thoroughly under full vacuum or positive pressure for 1 minute.

3 WASH:

1 x 3 mL 98% Methanol: 2% Acetic Acid
Dry column thoroughly under full vacuum or positive pressure for a minimum of 5 minutes.

4. ELUTION:

1 x 3 mL CH_2Cl_2/ IPA/ NH_4OH (78:20:2)
Collect eluate at 1 to 2 mL/minute.

NOTE: Prepare elution solvent daily.
Add IPA/ NH_4OH, mix, then add CH_2Cl_2 (pH 11-12).

5. DRY ELUTE:

Add 50 µL of 1% HCl in CH_3OH to each tube
Evaporate fraction to complete dryness under stream of dry air or nitrogen at ~ 35 °C.

NOTE: A 1% HCl in CH_3OH solution has been used to prevent volatization by the formation of the hydrochloric salt of the drugs.

6. RECONSTITUTE / DERIVATIZE

- LC-MS/MS: Reconstitute sample in 100 µL of mobile phase Inject 5-20 µL.

- GC-MS: Fluoroacylate with PFPA (PFAA)
  Add 50 µL PFPA. Over lay with N_2 and cap
  *Improve derivatization by addition of PFPOH
  React 20 minutes at 70 °C. Evaporate to dryness <40 °C
  Reconstitute with 100 µL Ethyl Acetate.

NOTES:

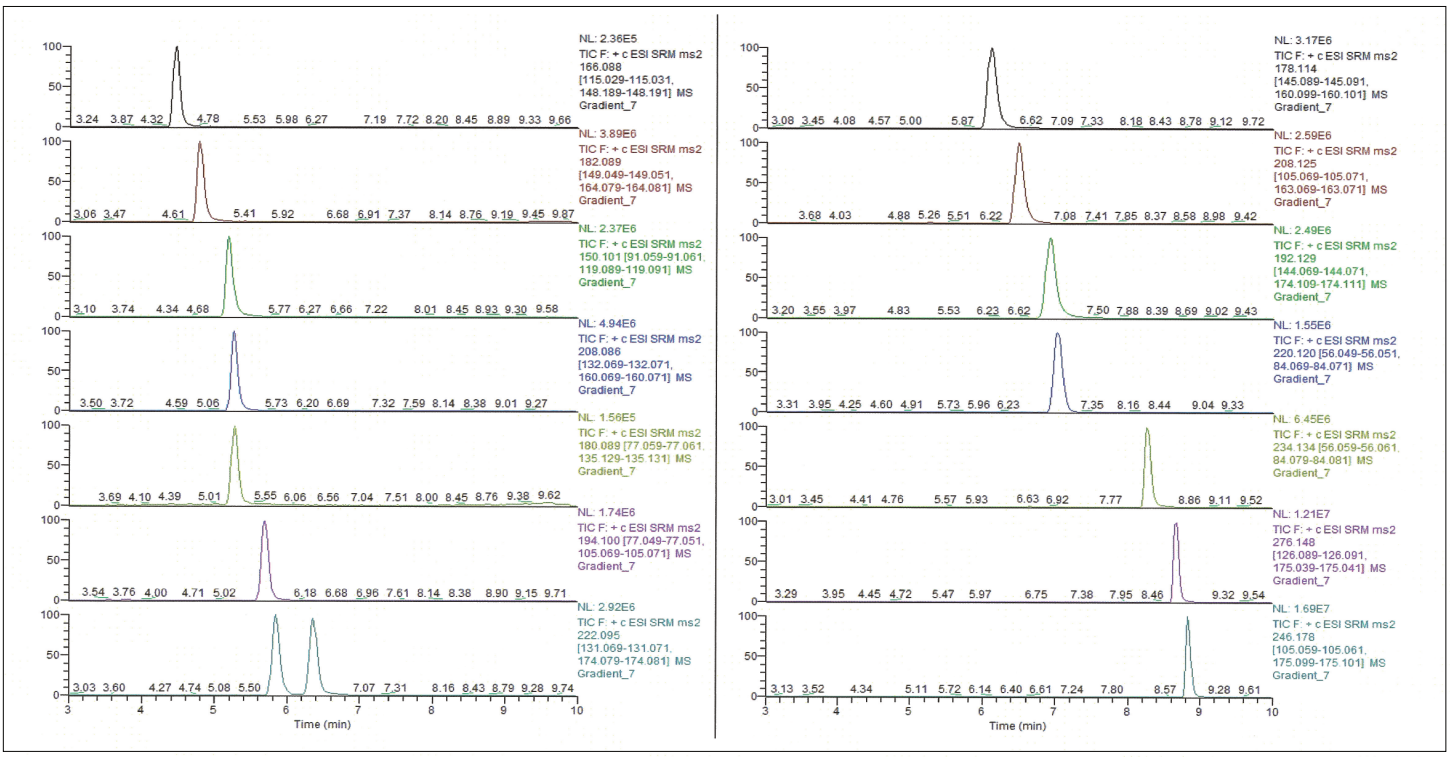
1. It is important to dry the column thoroughly to achieve the highest recovery of all compounds. Any residual moisture will slow down the drying of the elution solvents prior to derivatization for GC/MS analysis, if being used. Also, any residual moisture could reduce the reactivity of the derivatization agent resulting in low GC/MS sensitivity and inconsistent results.

2. **Sodium periodate can be added to sample during preparation if oxidation is preferred.**
   To 1 mL of 100 mM phosphate buffer (pH 6.0) add internal standard(s). Add 2 mL of urine and 1 mL 0.35 M sodium periodate.
   Mix/vortex
   Incubate at room temp.for 20 min.
   Sample pH should be 6.0 ± 0.5.
   Adjust pH accordingly with 100 mM monobasic or dibasic sodium phosphate
   Sample is now ready to be added to the extraction column

3. **Alternate Derivatization**
   1. Fluoroacylate with HFPA (HFAA)
   Add 50 µL HFPA. Over lay with N_2 and cap
   *Improved derivatization by addition of PFPOH
   React 20 minutes at 70 °C. Evaporate to dryness <40 °C
   Reconstitute with 100 µL Ethyl Acetate

   2. Form TMS Derivatives by adding 50 µL BSTFA w/ 1% TMCS and 50 µL of Ethyl Acetate; React 45 minutes at 70 °C

SYMPATHOMIMETIC AMINES



Analyte		MRM Transitions		Relative Retention Time (minutes)
		Q1	Q3	
1.	Epedrine	166.0	115.0	4.45
2.	Flephedrone	182.0	149.0	4.78
3.	Methamphetamine	150.1	91.1	5.18
4.	Methylone	208.0	132.0	5.26
5.	MDA	180.1	77.0	5.27
6.	MDMA	194.1	77.0	5.69
7.	Butylone	222.0	131.0	5.84
8.	Ethylone	222.0	131.0	6.36
9.	Mephedrone	178.1	145.0	6.10
10.	MDEA	208.1	105.0	6.48
11.	Methcathinone	192.1	144.0	6.93
12.	Ritalinic Acid	220.1	56.0	7.03
13.	Methylphenidate	234.1	56.0	8.26
14.	MDPV	276.1	126.0	8.67
15.	Pyrovalerone	246.1	105.0	8.83

HPLC Conditions

Instrument: Thermo Scientific™ Dionex™ Ultimate™ 3000 LC system
Detector: TSQ Vantage™ tandem mass spectrometer
Ionization mode: ESI+
HPLC column: UCT Selectra® DA, 100 × 2.1 mm, 3 µm (p/n: SLDA100ID21-3UM)
Guard column: UCT Selectra® DA, 10 × 2.0 mm, 3 µm, (p/n: SLDAGDC21-3UM)
Guard column holder: p/n: SLDGRDHLDR
Column temp.: 40 °C
Injection volume: 10 µL
Flow rate: 300 µL/min

Mobile Phase A: 0.1% formic acid in water

Mobile Phase B: 0.1% formic acid in MeOH

Gradient:

Time	%A	%B
0	98	2
1	65	35
5	65	35
7	0	100
10	0	100
10.2	98	2
15	98	2

EXTRACTION OF BENZODIAZEPINES FROM URINE, BLOOD, PLASMA/SERUM, TISSUE BY LC-MS/MS

Part #

CSXCE106 CLEAN SCREEN XCEL® I 130 mg, 6 mL Tube
SPHPHO6001-5 – Select pH Buffer Pouches 100 mM Phosphate pH 6.0
SMTBSTFA-1-1 – SELECTRA-SIL® MTBSTFA w/ 1% TBDMCS
SLDA100ID21-3UM – Selectra® DA HPLC Column 100 x 2.1mm 3 µm
SLDAGDC21-3UM - SELECTRA® DA 3 µm Guard Cartridge
SLGRDHLDR - Guard Cartridge Holder

SELECTRAZYME®
Beta-glucuronidase

1a. PREPARE SAMPLE (Blood, Plasma and Serum)

To 1 mL of 100 mM phosphate buffer (pH 6.0) add internal standards.
Add 1-2 mL of blood, plasma/ serum, 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.

1b. PREPARE SAMPLE (Urine) ENZYME HYDROLYSIS OF GLUCURONIDES

To 1-2 mL of urine sample, add 1 mL of acetate buffer (pH 5.0) containing 5,000 units/mL of Selectrazyme® ß-glucuronidase.
Optionally, add 1 mL of acetate buffer and 25-50 µL of concentrated ß-glucuronidase.
Vortex and heat for 1-2 hours at 65°C.
Allow sample to cool.
Do not adjust pH~ sample is ready to be added to the extraction column.

2. APPLY SAMPLE

Load sample directly to column without any preconditioning.
Pull sample through at a rate of 1-2 mL/ minute.
Dry column thoroughly under full vacuum or positive pressure for 1 minute.

3. WASH

1 x 3mL 100 mM phosphate buffer (pH6).
1 x 3 mL CH_2Cl_2
Dry column thoroughly under full vacuum or positive pressure for a minimum of 5-10 minutes.

4. ELUTION

1 x 3 mL Ethyl Acetate:NH_4OH (98:2)
Collect eluate at 1 to 2 mL/minute.
NOTE: Prepare elution solvent daily.

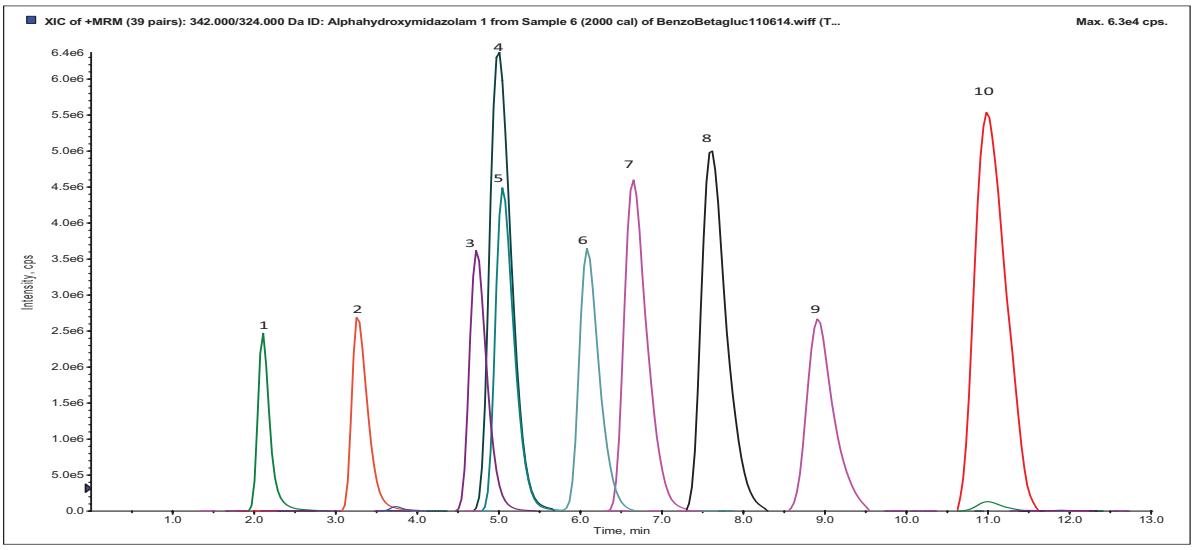
5. DRY ELUTE

Evaporate fraction to complete dryness under stream of dry air or nitrogen at ~ 35 °C.

6. RECONSTITUTE / DERIVATIZE

- LC-MS/MS: Reconstitute sample in 100 µL of mobile phase Inject 10-20 µL.
- GC-MS: Dissolve residue in 50 µL of ACN and 50 µL MTBSTFA w/ 1%TBDMCS
  Overlay with N_2 and cap. Mix/vortex
  React 30 minutes at 70 °C; Cool and inject 1-2 µL

BENZODIAZEPINES



Analyte		MRM Transitions		Relative Retention Time (minutes)	% Recovery (Urine)	% Recovery (Blood)
		Q1	Q3			
1.	7-Aminoclonazepam	286.0	222.3	2.10	93%	95%
2.	Midazolam	326.0	291.0	3.26	89%	89%
3.	Lorazepam	321.0	303.3	4.73	93%	76%
4.	Oxazepam	287.0	241.3	4.98	78%	96%
5.	Clonazepam	316.1	270.2	5.05	78%	78%
6.	Alpha-Hydroxy-Alprazolam	325.1	297.1	6.08	103%	82%
7.	Nordiazepam	271.0	140.1	6.65	101%	80%
8.	Temazepm	301.1	255.2	7.59	79%	68%
9.	Alprazolam	309.1	205.3	8.91	90%	89%
10.	Diazepam	285.1	193.2	10.98	100%	90%

HPLC Conditions

Instrument: Agilent 1200 Binary Pump SL
Detector: API 4000 Qtrap MS/MS
HPLC Column: SELECTRA® DA HPLC Column 100 x 2.1mm 3 µm
Polarity: Positive
Reconstitute: 100 µl
Injection Volume: 10 µL
Flow Rate: 0.3 mL/minute
Mobile Phase A: 0.1% formic acid in water
Mobile Phase B: 0.1% formic acid in MeOH
Gradient:

Time	%A	%B
0.0	40	60
8.00	40	60
8.01	5	95
9.01	5	95
9.50	40	60
13.00	STOP	

BATH SALTS IN BLOOD, PLASMA/SERUM, URINE, OR TISSUE BY LC-MS/MS OR GC-MS
CLEAN SCREEN XCEL® I EXTRACTION COLUMN

Part #

CSXCE111 – CLEAN SCREEN XCEL® 130 mg, 1 mL Tube
PFAA-0-1 – SELECTRA-SIL® PFAA
SPFPOH-1 – SELECTRA-SIL® PFPOH
SLDA100ID21-3UM – SELECTRA® DA HPLC Column 100 × 2.1 mm, 3 µm
SLDAGDC21-3UM - SELECTRA® DA 3 µm Guard Cartridge
SLGRDHLDR - Guard Cartridge Holder

SELECTRA-SIL®
Derivatizing Reagents

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. APPLY SAMPLE

Load sample directly to column without any preconditioning.
Pull sample through at a rate of 1-2 mL/ minute.
Dry column thoroughly under full vacuum or positive pressure for 1 minute.

3. WASH

1 x 3 mL 98% Methanol: 2% Acetic Acid
Dry column thoroughly under full vacuum or positive pressure for a minimum of 5 minutes.

4. ELUTION

1 x 3 mL CH_2Cl_2/ IPA/ NH_4OH (78:20:2)
Collect eluate at 1 to 2 mL/minute.
NOTE: Prepare elution solvent daily.
Add IPA/ NH_4OH, mix, then add CH_2Cl_2 (pH 11-12).

5. DRY ELUTE

Add 50 µL of 1% HCl in CH_3OH to each tube
Evaporate fraction to complete dryness under stream of dry air or nitrogen at ~ 35 °C.

NOTE: A 1% HCl in CH_3OH solution has been used to prevent volatization by the formation of the hydrochloric salt of the drugs.

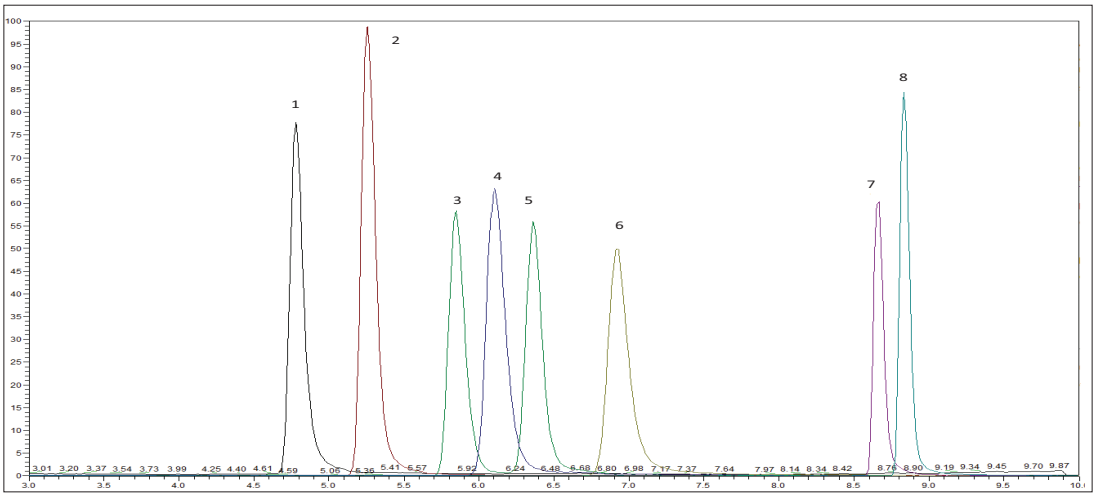
6. RECONSTITUTE / DERIVATIZE

- LC-MS/MS: Reconstitute sample in 100 µL of mobile phase Inject 5 µL.

- GC-MS: Fluoroacylate with PFPA (PFAA)
  Add 50 µL PFPA. Over lay with N_2 and cap
  *Improved derivatization by addition of PFPOH
  React 20 minutes at 70 °C. Evaporate to dryness <40 °C
  Reconstitute with 100 µL Ethyl Acetate

NOTE: It is important to dry the column thoroughly to achieve the highest recovery of all compounds. Any residual moisture will slow down the drying of the elution solvents prior to derivatization for GC/MS analysis, if being used. Also, any residual moisture could reduce the reactivity of the derivatization agent resulting in low GC/MS sensitivity.

BATH SALTS



Analyte		MRM Transitions		Relative Retention Time (minutes)
		Q1	Q3	
1.	Flephedrone	182.1	164.2	4.78
2.	Methylone	208.1	160.1	5.26
3.	Ethylone	222.0	131.0	5.84
4.	Mephedrone	178.1	145.0	6.10
5.	Butylone	222.0	131.0	6.36
6.	Methcathinone	192.2	144.0	6.93
7.	MDPV	276.2	126.1	8.67
8.	Pyravalerone	246.2	105.2	8.83

HPLC Conditions

Instrument: Thermo Scientific™ Dionex™ Ultimate™ 3000 LC system
Detector: TSQ Vantage™ tandem mass spectrometer
Ionization mode: ESI+
HPLC column: Selectra® DA, 100 × 2.1 mm, 3 µm (p/n: SLDA100ID21-3UM)
Guard column: Selectra® DA, 10 × 2.0 mm, 3 µm, (p/n: SLDAGDC21-3UM)
Guard column holder: p/n: SLDGRDHLDR
Column temp.: 40 °C
Injection volume: 10 µL
Flow rate: 300 µL/min
Mobile Phase A: 0.1% formic acid in water
Mobile Phase B: 0.1% formic acid in MeOH
Gradient:

Time	%A	%B
0	98	2
1	65	35
5	65	35
7	0	100
10	0	100
10.2	98	2
15	98	2

EXTRACTION OF BASIC DRUGS AND METABOLITES FROM URINE/ BLOOD USING CLEAN SCREEN XCEL® I EXTRACTION COLUMN

Part #:
CSEXCE106 - CLEAN SCREEN XCEL® I 130 mg / 6 mL Tube
SPACE5001-5 - SELECT pH Buffer Pouch, 100mM acetate pH 5.0
SPHPHO6001-5 - SELECT pH Buffer Pouch, 100mM phosphate pH 6.0
BETA-GLUC-10 - Selectrzyme® beta-glucuronidase, 10 mL

1a. Sample Preparation (blood)

To 1-2 mL blood add 2 ml of 100mM phosphate buffer pH 6.0. Add appropriate volume and concentration internal standards. Sample is ready to be added to extraction column.

1b. Sample Preparation (urine hydrolysis))

To 1-2 mL urine sample add 500 µL of 100mM acetate buffer pH 5.0 containing 5,000 units/mL ß-glucuronidase. **Optionally**, add 500 µL of acetate buffer and 25 µL of concentrated ß-glucuronidase. Add appropriate volume and concentration internal standards. Vortex and heat for 1-2 hours at 65 °C. Allow sample to cool. **Do not adjust pH- sample is ready to be added to extraction column.**

2. Applying Sample to Column

Load sample directly to column without any preconditioning. Pull sample through at a rate of 1-2 mL/ minute. Dry column thoroughly under vacuum (10 mm Hg) or positive pressure (~ 80-100 psi) for 1 minute.

3a. Wash (Blood Only)

Wash sample with 2 mL of 100mM phosphate buffer pH 6.0.

3b. Wash (Urine and Blood)

Wash sample with 1 mL of 2% glacial acetic acid/ 98 % methanol. Dry column thoroughly under vacuum (10 mm Hg) or positive pressure (~ 80-100 psi) for a minimum of 5 minutes.

NOTE 1: It is important to dry the column thoroughly to achieve the highest recovery of all compounds. Any residual moisture will slow down the drying of the elution solvents prior to derivatization for GC/MS analysis. Also, any residual moisture could reduce the reactivity of the derivatization agent resulting in low GC/MS sensitivity.

4. Elution

Elute samples with ~1-2 mL CH_2Cl_2 / IPA / ammonium hydroxide (78/20/2)
Evaporate fraction to complete dryness under stream of dry air or nitrogen at ~ 35 °C.

(**Note:** For amphetamine group analysis, add 200 µL of 1% HCl in MeOH to eluate to minimize amphetamine group loss to volatilization.)

GC/MS Analysis

Derivatize compounds with appropriate derivatizing procedure or reconstitute in 100 µL ethyl acetate and inject 1-2 µL into the GC/ MS system for analysis.

LC/MS Analysis

Reconstitute in methanol or appropriate mobile phase.

Analyte	% Recovery	
	Urine	Blood
6-MAM	97	100
Amphetamine	112	104
Benzoylecgonine	54	58
Buprenorphine	92	92
Chlorpheniramine	85	94
Citalopram	99	95
Clonidine	85	98
Clozapine	98	94
Cocaine	103	96
Codeine	96	99
Cyclobenzaprine	102	92
Diphenhydramine	87	88
Doxepin	97	93
EDDP	85	88
Ephedrine	75	57
Fentanyl	88	89
Fluoxetine	97	92
Hydrocodone	98	94
Hydromorphone	91	91
Imipramine	97	97
MDA	95	84
MDEA	100	93
MDMA	99	90
Meperidine	82	95
Methadone	92	93
Methamphetamine	99	104
Morphine	92	85
Nalophrine	99	97
Naltrexone	99	90
Naxolone	99	94
Norfentanyl	84	85
Nortriptyline	79	89
Oxycodone	98	94
Oxymorphine	94	88
Paroxetine	90	92
Phencyclidine	90	93
Propoxyphene	90	95
Pseudoephedrine	79	56
Sertraline	75	70
Tramadol	89	81
Venlafaxine	102	93
Zolpidem	92	95

NOTES

PRICES AND TERMS

Our prices are subject to change without notice. The price in effect when we receive your order will apply. All prices are in US Dollars and are F.O.B. Terms of payment are net 30 days.

MINIMUM ORDERS

We welcome all orders, therefore, we do not have a minimum order requirement. When ordering, please include your purchase order number, complete "Ship To" and "Bill To" address, catalog number, quantity, and description of product(s). Also include your name and a phone number where you can be reached should we have any questions concerning your order.

SHIPMENTS

Normal processing is within 24 hours after receipt of an order. Unless special shipping requests have been made, our trained staff will send all orders by UPS Ground service. The appropriate shipping charges (freight & insurance costs) will be added to the invoice, unless otherwise instructed by the customer.

SPECIAL PRICING

We offer special pricing for volume purchases and standing orders. These discounts apply to bonded phase extraction column purchases only. Please call a sales representative for more information on special pricing qualifications.

RETURN POLICY

Our Quality Manager will handle all returns. Before returning merchandise, please call to obtain a return authorization number from the quality manager. We will need to know the reason for the return, date of purchase, purchase order number and invoice number in order to issue a return authorization number. Return merchandise must be received before a credit can be issued. Returns will not be accepted after 90 days. A restocking fee of 25% of the price paid, or a minimum of $25.00 (whichever is greater) will be charged on all returns.

WARRANTY

All products manufactured by UCT are guaranteed against defects in materials and workmanship for a period of 90 days after shipment. UCT will replace any items that prove to be defective during this time period.

The exclusive remedy requires the end user to first advise UCT of the defective product by phone or in writing. Secondly, the defective product must be returned within 30 days after proper approval from our Quality Manager. All returns must indicate the purchase order number, the lot number and the shipping date. UCT's total liability is limited to the replacement cost of UCT products.

This warranty does not apply to damage resulting from misuse.

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