LC/MS/MS Analysis of Synthetic Cathinones (Bath Salts) from Urine and Whole Blood using a Luna® Omega 1.6 µm C18 UHPLC Column and Strata®-X-Drug B Solid Phase Extraction (SPE)

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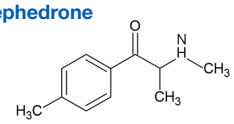
Overview

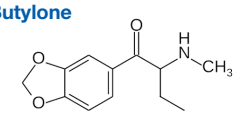
Bath Salts also known as synthetic cathinones have recently emerged as popular drugs of abuse. Synthetic cathinones are phenylalkylamines derivatives and are often called bk-amphetamines due to a ketone attached at the beta position on the amino alkyl chain which is attached to the phenyl ring.[1] Similar to phenylethylamines like MDMA, they may possess both amphetamine-like properties and the ability to modulate serotonin, causing distinct psychoactive effects.[2] This application illustrates the separation of 5 common synthetic cathinones using a Luna Omega 1.6 µm C18 UHPLC column. Luna Omega UHPLC columns contain a unique silica which is modified using a proprietary, post-synthetic thermal treatment process to provide extraordinary mechanical strength and significantly greater inertness than traditional fully porous and hybrid materials. This greatly minimizes any secondary interactions that negatively affect peak shape, allowing for greater method accuracy.

Figure 1.
Chemical Structures of Cathinones

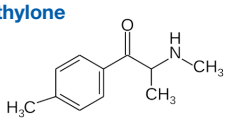
Mephedrone



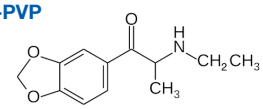
Butylone



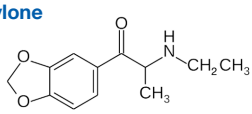
Methylone



α-PVP



Ethylone



Experimental Conditions

LC/MS/MS Conditions

LC Column: Luna Omega 1.6 µm C18
Dimensions: 50 x 2.1 mm
Part No.: 00B-4742-AN
Mobile Phase: A: 0.1% Formic Acid in Water
B: 0.1% Formic Acid in Acetonitrile
Gradient:

Time (min)	% B
0	10
4	95
4.1	10
6	10

Flow Rate: 700 µL/min
Injection Volume 1 µL
Temperature: 22 °C
Detection: MS/MS
Detection System: API 4000™ (SCIEX)
Analytes: 1. Methylone
2. Ethylone
3. Butylone
4. Mephedrone
5. α-PVP

Sample Preparation

Pretreatment

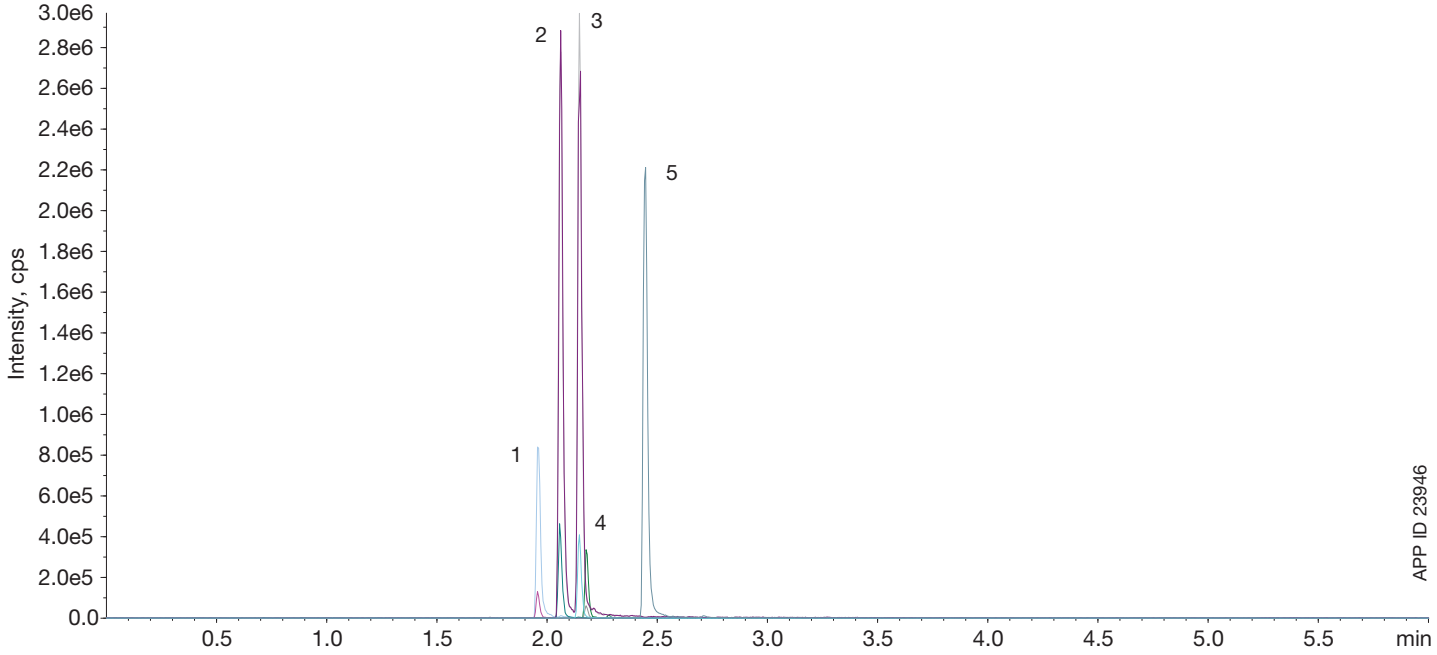
Urine	Add 2 mL of 100 mM sodium acetate buffer (pH 5.0) and 50 µL of internal standards (@10 ppm) to 2 mL of urine
Whole blood	1. Add 2 mL of ice cold methanol:acetonitrile (50:50) and 20 µL of internal standards (@10 ppm) and 2 mL of 100 mm sodium acetate to 1 mL of blood. 2. Centrifuge at 4700 rpm 10 °C for 5 min 3. Transfer supernatant for SPE

Solid Phase Extraction (SPE)

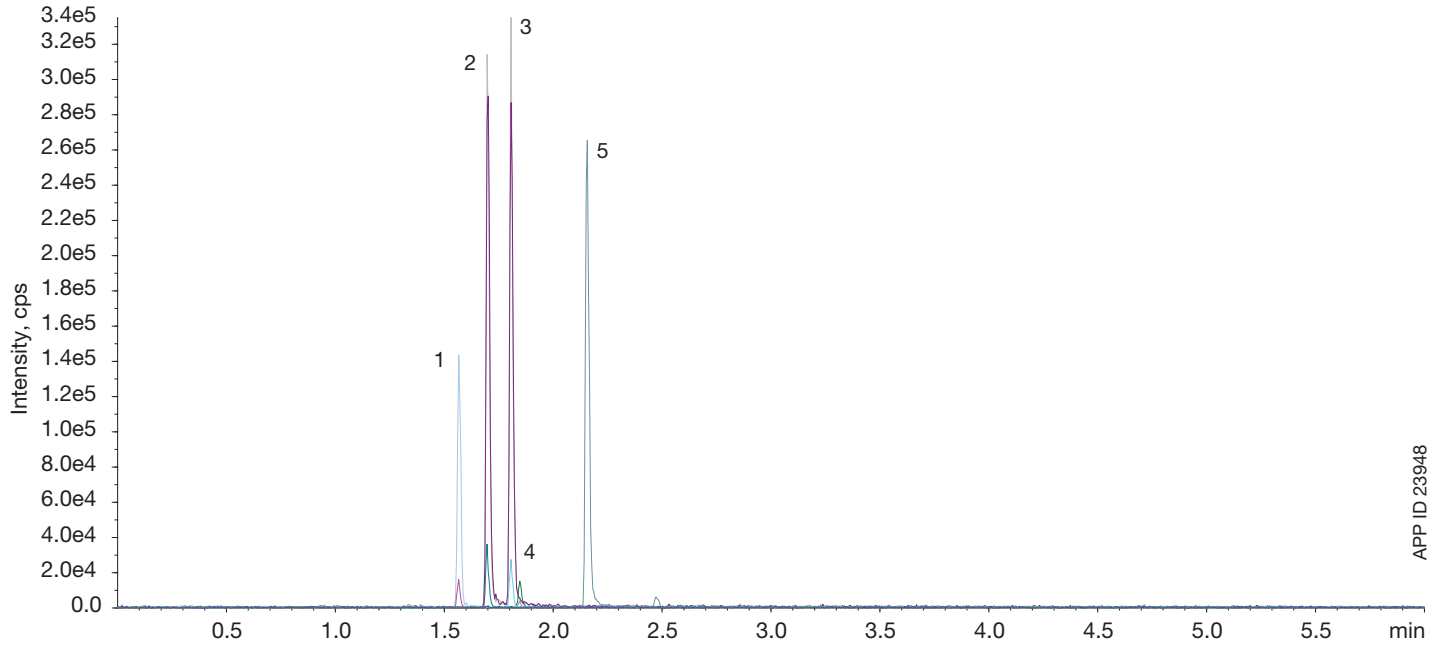
Cartridge:	Strata-X-Drug B
Part No:	8B-S128-UCH
Condition:	1 mL each of methanol, DI water, and 100 mm sodium acetate
Load:	Load pretreated sample
Weak Wash:	2 mL of 100 mM sodium acetate (pH 5.0)
Strong Wash:	1 mL of methanol
Dry Down:	2 minutes at >10" hg
Elute:	3 mL of ethyl acetate: IPA: ammonium hydroxide (70:20:10)
Evaporate:	to 500 µL and add 100 µL of HCl:methanol (20:80), evaporate to dryness under nitrogen
Reconstitute:	100 µL of methanol

Q1	Q2	Analyte	Retention Time (min)
208.1	160.2	Methylone	1.56
222.3	174.1	Ethylone	1.69
222.3	174.1	Butylone	1.80
178.1	160.2	Mephedrone	1.84
232.4	91	α-PVP	2.15

Extracted Ion Chromatagram of Bath Salts in Urine



Extracted Ion Chromatagram of Bath Salts in Whole Blood



guarantee

If the Phenomenex products in this technical note do not provide at least an equivalent separation as compared to other products of the same particle size, phase, and dimensions, return the product with comparative data within 45 days for a FULL REFUND.

Luna® Omega Ordering Information

1.6 µm Minibore Columns (mm)					SecurityGuard™ ULTRA Cartridges‡
Phases	**30 x 2.1**	**50 x 2.1**	**100 x 2.1**	**150 x 2.1**	**3/pk**
Polar C18	00A-4748-AN	00B-4748-AN	00D-4748-AN	00F-4748-AN	AJ0-9505
PS C18	00A-4752-AN	00B-4752-AN	00D-4752-AN	00F-4752-AN	AJ0-9508
C18	00A-4742-AN	00B-4742-AN	00D-4742-AN	00F-4742-AN	AJ0-9502
					for 2.1 mm ID

‡ SecurityGuard ULTRA Cartridges require holder, Part No.: AJ0-9000

Strata®-X-Drug B Ordering Information

Format	Sorbent Mass	Part Number	Unit
Tube			
	10 mg	8B-S128-AAK	1 mL (100/box)
	30 mg	8B-S128-TAK	1 mL (100/box)
	30 mg	8B-S128-TBJ	3 mL (50/box)
	60 mg	8B-S128-UBJ	3 mL (50/box)
	60 mg	8B-S128-UCH	6 mL (30/box)
	60 mg	8B-S128-UCL	6 mL (200/bag)
Giga™ Tube			
	100 mg	8B-S128-EDG	12 mL (20/box)
96-Well Plate			
	10 mg	8E-S128-AGB	2 Plates/box
	30 mg	8E-S128-TGB	2 Plates/box
	60 mg	8E-S128-UGB	2 Plates/box

Reference:

1 *Zaitsu K, Katagi M, Tatsuno M et al. Recently abused b-keto derivates of 3,4-methylenedioxyphenylalkylamines: a review of their metabolisms and toxciological analysis. Forensic Toxicol 2011.*

2. *Prosser J, Nelson L. The Toxicology of Bath Salts: A Review of Synthetic Cathinones. J. Med. Toxicol. 2012*

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