

# APPLICATIONS



## LC/MS/MS Analysis of Synthetic Cathinones (Bath Salts) from Urine and Whole Blood using a Kinetex<sup>®</sup> 2.6 μm C18 HPLC Column and Strata<sup>®</sup>-X-Drug B Solid Phase Extraction (SPE)

Daniel Spurgin and Dr. Jeff Layne  
Phenomenex, Inc., 411 Madrid Ave., Torrance, CA 90501 USA

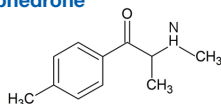
### Overview

Cathinone is a naturally occurring beta-ketone amphetamine analogue found in the leaves of the *Catha edulis* (khat) plant which is native to the Horn of Africa and the Arabian Peninsula.<sup>1</sup> Over the past decade, synthetic cathinones more commonly known as “bath salts” have emerged as a sought after psychoactive recreational drug with effects similar to that of methamphetamines, cocaine, or 3,4-methylenedioxyamphetamine (MDMA). This application illustrates the separation of 5 common synthetic cathinones using a Kinetex 2.6 μm C18 HPLC column, resulting in sharp, narrow peaks and specificity in both urine and whole blood. Kinetex core-shell columns deliver significantly improved chromatographic resolution, providing sub-2 μm performance but at backpressures that are compatible with conventional HPLC instruments.

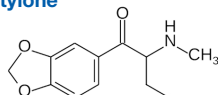


**Figure 1.**  
Chemical Structures of Cathinones

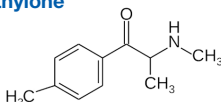
#### Mephedrone



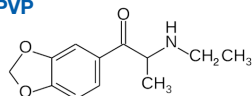
#### Butylone



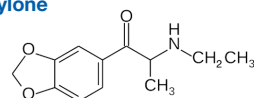
#### Methylone



#### α-PVP



#### Ethylone



### Experimental Conditions

#### LC/MS/MS Conditions

**LC Column:** Kinetex 2.6μm C18  
**Dimensions:** 50 x 4.6 mm  
**Part No.:** 00B-4462-E0  
**Mobile Phase:** A: 0.1% Formic Acid in Water  
B: 0.1% Formic Acid in Acetonitrile

Gradient: Time (min)	% B
0	5
4	95
4.1	5
6	5

**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)

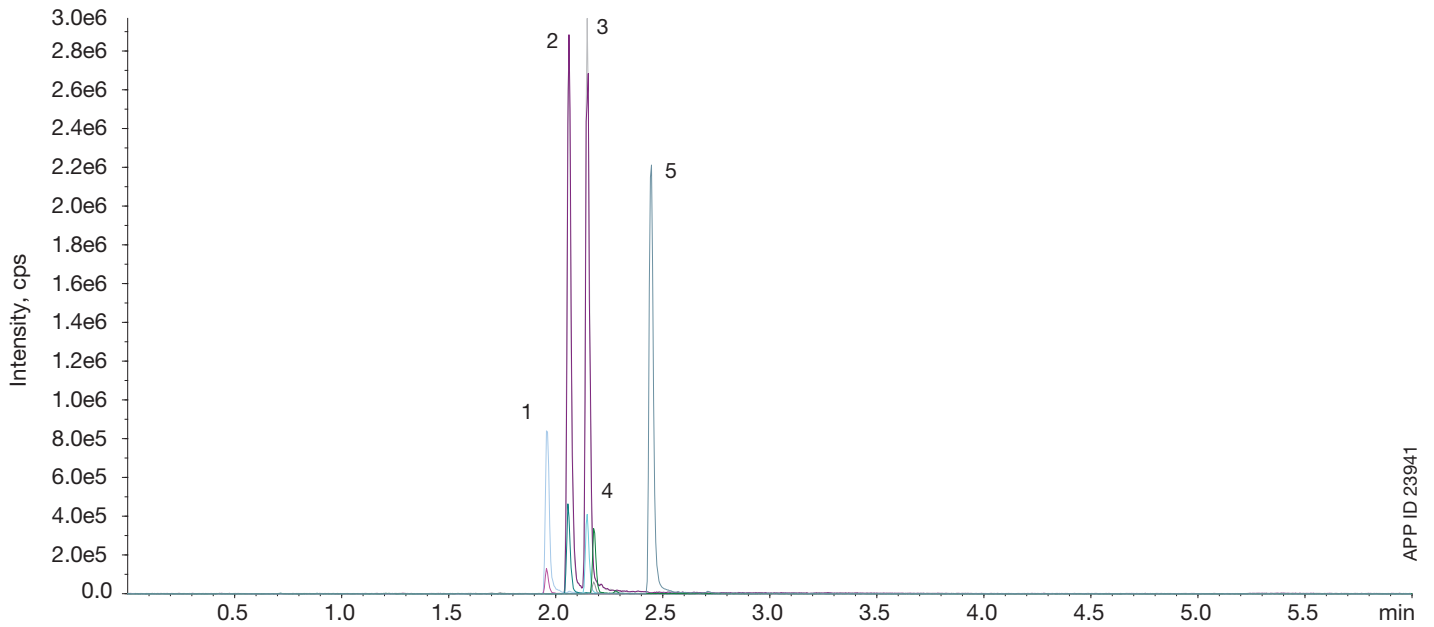
<b>Cartridge:</b>	Strata-X-Drug B
<b>Part No.:</b>	8B-S128-UCH
<b>Condition:</b>	1 mL each of methanol, DI water, and 100 mM sodium acetate
<b>Load:</b>	Load pretreated sample
<b>Weak Wash:</b>	2 mL of 100 mM sodium acetate (pH 5.0)
<b>Strong Wash:</b>	1 mL of methanol
<b>Dry Down:</b>	2 minutes at >10"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

Q1	Q2	Analyte	Retention Time (min)
208.1	160.2	Methylone	1.96
222.3	174.1	Ethylone	2.06
222.3	174.1	Butylone	2.15
178.1	160.2	Mephedrone	2.18
232.4	91	α-PVP	2.44



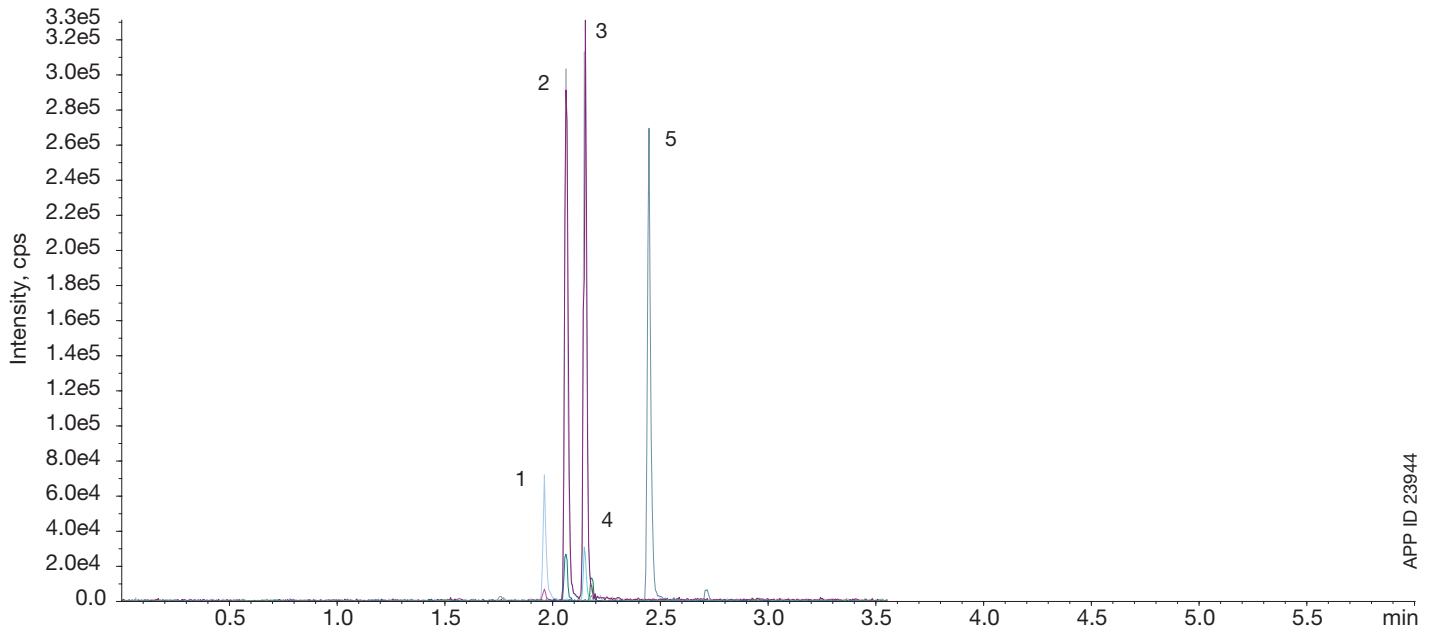
# APPLICATIONS

## Extracted Ion Chromatogram of Bath Salts in Urine



APP ID 23941

## Extracted Ion Chromatogram of Bath Salts in Whole Blood



APP ID 23944



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.

# APPLICATIONS









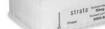

## Kinetex<sup>®</sup> Ordering Information

2.6 µm Analytical Columns (mm)						SecurityGuard <sup>™</sup> ULTRA Cartridges <sup>†</sup>
Phases	30 x 4.6	50 x 4.6	75 x 4.6	100 x 4.6	150 x 4.6	3/pk
F5	—	00B-4723-E0	—	00D-4723-E0	00F-4723-E0	AJ0-9320
Biphenyl	—	00B-4622-E0	—	00D-4622-E0	00F-4622-E0	AJ0-9207
XB-C18	—	00B-4496-E0	00C-4496-E0	00D-4496-E0	00F-4496-E0	AJ0-8768
C18	00A-4462-E0	00B-4462-E0	00C-4462-E0	00D-4462-E0	00F-4462-E0	AJ0-8768
C8	—	00B-4497-E0	00C-4497-E0	00D-4497-E0	00F-4497-E0	AJ0-8770
HILIC	—	00B-4461-E0	00C-4461-E0	00D-4461-E0	00F-4461-E0	AJ0-8772
Phenyl-Hexyl	—	00B-4495-E0	00C-4495-E0	00D-4495-E0	00F-4495-E0	AJ0-8774

for 4.6 mm ID

<sup>†</sup> SecurityGuard ULTRA Cartridges require holder, Part No.: AJ0-9000

## Strata<sup>®</sup>-X-Drug B Ordering Information

Format	Sorbent Mass	Part Number	Unit
<b>Tube</b>			
	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)
<b>Giga<sup>™</sup> Tube</b>			
	100 mg	8B-S128-EDG	12 mL (20/box)
<b>96-Well Plate</b>			
	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. Marinetti, L, Antonides, H. Analysis of Synthetic Cathinones Commonly Found in Bath Salts in Human Performance and Postmortem Toxicology: Method Development, Drug Distribution and Interpretation of Results. *Journal of Analytical Toxicology*, 2013.



### www.phenomenex.com

Phenomenex products are available worldwide. For the distributor in your country, contact Phenomenex USA, International Department at international@phenomenex.com

### Terms and Conditions

Subject to Phenomenex Standard Terms & Conditions, which may be viewed at [www.phenomenex.com/TermsAndConditions](http://www.phenomenex.com/TermsAndConditions).

### Trademarks

Kinetex and Strata are registered trademarks and SecurityGuard and Giga are trademarks of Phenomenex. API 4000 is a trademark of SCIEX Pte. Ltd. SCIEX is being used under license.

### Disclaimer

Kinetex EVO is patented by Phenomenex. U.S. Patent Nos. 7,563,367 and 8,658,038 and foreign counterparts.  
Strata-X is patented by Phenomenex. U.S. Patent No 7,119,145.

For RESEARCH USE ONLY. Not for use in diagnostic procedures.

© 2017 Phenomenex, Inc. All rights reserved.

### Australia

t: +61 (0)2-9428-6444  
f: +61 (0)2-9428-6445  
auinfo@phenomenex.com

### Austria

t: +43 (0)1-319-1301  
f: +43 (0)1-319-1300  
anfrage@phenomenex.com

### Belgium

t: +32 (0)2 503 4015 (French)  
t: +32 (0)2 511 8666 (Dutch)  
f: +31 (0)30-2383749  
beinfo@phenomenex.com

### Canada

t: +1 (800) 543-3681  
f: +1 (310) 328-7768  
info@phenomenex.com

### China

t: +86 400-606-8099  
f: +86 (0)22 2532-1033  
phen@agela.com

### Denmark

t: +45 4824 8048  
f: +45 4810 6265  
nordicinfo@phenomenex.com

### Finland

t: +358 (0)9 4789 0063  
f: +45 4810 6265  
nordicinfo@phenomenex.com

### France

t: +33 (0)1 30 09 21 10  
f: +33 (0)1 30 09 21 11  
franceinfo@phenomenex.com

### Germany

t: +49 (0)6021-58830-0  
f: +49 (0)6021-58830-11  
anfrage@phenomenex.com

### India

t: +91 (0)40-3012 2400  
f: +91 (0)40-3012 2411  
indiainfo@phenomenex.com

### Ireland

t: +353 (0)1 247 5405  
f: +44 1625-501796  
eirinfo@phenomenex.com

### Italy

t: +39 051 6327511  
f: +39 051 6327555  
italiainfo@phenomenex.com

### Luxembourg

t: +31 (0)30-2418700  
f: +31 (0)30-2383749  
nlinfo@phenomenex.com

### Mexico

t: 01-800-844-5226  
f: 001-310-328-7768  
tecnicomx@phenomenex.com

### The Netherlands

t: +31 (0)30-2418700  
f: +31 (0)30-2383749  
nlinfo@phenomenex.com

### New Zealand

t: +64 (0)9-4780951  
f: +64 (0)9-4780952  
nzinfo@phenomenex.com

### Norway

t: +47 810 02 005  
f: +45 4810 6265  
nordicinfo@phenomenex.com

### Puerto Rico

t: +1 (800) 541-HPLC  
f: +1 (310) 328-7768  
info@phenomenex.com

### Spain

t: +34 91-413-8613  
f: +34 91-413-2290  
espinfo@phenomenex.com

### Sweden

t: +46 (0)8 611 6950  
f: +45 4810 6265  
nordicinfo@phenomenex.com

### United Kingdom

t: +44 (0)1625-501367  
f: +44 (0)1625-501796  
ukinfo@phenomenex.com

### USA

t: +1 (310) 212-0555  
f: +1 (310) 328-7768  
info@phenomenex.com

### All other countries Corporate Office USA

t: +1 (310) 212-0555  
f: +1 (310) 328-7768  
info@phenomenex.com