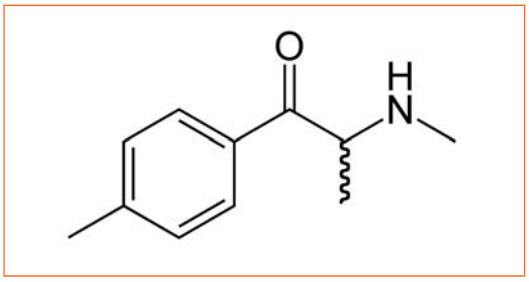
Extraction of Mephedrone From Biological Fluids Using EVOLUTE® CX

Application Note

Introduction

This application note describes the extraction of Mephedrone from biological fluids using EVOLUTE CX.



This method demonstrates the extraction of Mephedrone (MCAT) from a range of biological fluids including plasma, urine and whole blood. This extraction is achieved through cation exchange using EVOLUTE CX, a mixed-mode resin-based SPE sorbent. Analytes were quantified using LC-MS/MS and recoveries were all above 90%.

Figure 1. Structure of Mephedrone

EVOLUTE CX mixed-mode resin-based SPE sorbent extracts a wide range of basic analytes from biological fluids and other aqueous matrices using a generic procedure which minimizes method development time. EVOLUTE CX removes matrix components such as proteins, salts, non-ionizable interferences and phospholipids, delivering cleaner extracts with reproducible recoveries for accurate quantitation.

Analytes

Mephedrone.

EVOLUTE CX Configuration

EVOLUTE CX 25 mg 96-well plate, part number 601-0025-P01.

EVOLUTE CX Procedure

Sample pre-treatment:	Dilute 100 µL of sample (plasma, urine, whole blood) 1:3 (v/v) with 50 mM ammonium acetate buffer (pH 6).
Whole blood pre-treatment:	Sonicate 1 mL blood sample for 10 minutes in 50 mM ammonium acetate buffer (pH 6) followed by centrifugation at 5,500 rpm for 10 minutes. Retain supernatant and discard cellular debris (pellet).
Condition:	Methanol (1 mL).
Equilibration:	50 mM ammonium acetate (pH 6) (1 mL).
Sample loading:	Pre-treated sample (400 µL).
Interference wash 1:	50 mM ammonium acetate (pH 6) (1 mL).
Interference wash 2:	Methanol (1 mL).
Analyte elution:	5% ammonium hydroxide in methanol (1 mL).
Post Extraction:	Evaporate eluate to dryness and reconstitute in 200 µL of water/methanol (80/20 (v/v)) prior to analysis.

HPLC Conditions

Instrument: Waters Acquity UPLC (Waters Assoc., Milford, MA, USA).

Column: Acquity UPLC BEH C18 column (1.7µm, 50 x 2.1 mm id) (Waters Assoc., Milford, MA, USA).

Mobile Phase: **A**: 0.1% formic acid aq and **B**:0.1% formic acid/MeOH at a flow rate of 0.5 mL/min.

Gradient: Isocratic 60% **A** and 40% **B**.

Injection Volume: 5 µL.

Column Temperature: 35 °C.

Mass Spectrometry Conditions

Instrument: Premier XE triple quadrupole mass spectrometer (Waters Assoc., Manchester, UK) equipped with an electrospray interface for mass analysis.

Desolvation Temperature: 450 °C

Ion Source Temperature: 150 °C

Collision Gas Pressure: 3.46 x 10-3 mbar

Scan Function	Analyte	MRM Transition	Cone Voltage (V)	Collision Energy (eV)
1	Mephedrone (quant)	178.1 > 160.0	35	12
2	Mephedrone (qual)	178.1 > 145.0	35	19

Table 1. MRM transitions for Mephedrone

Results

All results show recoveries above 90% with RSDs below 10%, Figure 2. shows an example mass chromatogram of Mephedrone (10 ng/mL) from whole blood.

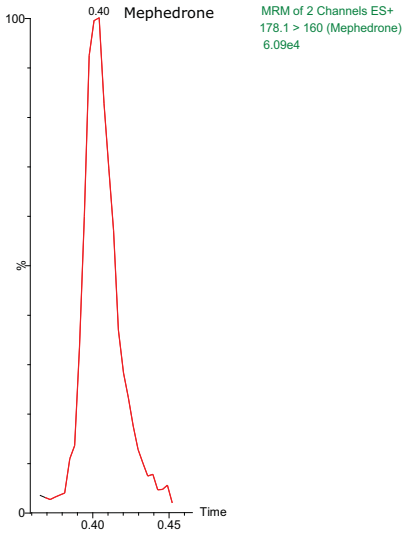


Figure 2. Example mass chromatogram showing Mephedrone at 10 ng/mL from whole blood.

References

This application note is based on the poster 'Mephedrone: Evaluation of Extraction using Mixed-Mode Cation Exchange SPE with UPLC-MS/MS Analysis.', R Jones et al, presented at TIAFT, Bonn, Germany, Aug 31—Sep 2, 2010.

Ordering information

Part number	Description	Quantity
601-0025-P01	EVOLUTE CX 25 mg 96-well plate	1

NORTH AMERICA
Main Office: +1 704 654 4900
Toll Free: +1 800 446 4752
Fax: +1 704 654 4917
Order Tel: +1 704 654 4900
press (4) at the auto attendant
Order Fax: +1 434 296 8217
ordermailbox@biotage.com
1-pointsupport@biotage.com

EUROPE
Main Office: +46 18 56 5900
Fax: +46 18 59 1922
Order Tel: +46 18 56 57 10
Order Fax: +46 18 56 57 05
order@eu.biotage.com

Japan
Tel: +81 3 5627 3123
Fax:+81 3 5627 3121
jp_order@biotage.com

Distributors
To locate a distributor please visit our Web site at www.biotage.com

Biotage | IST

www.biotage.com