

HPLC Application

ID No.: 19714

Benzodiazepines from urine using Strata-X-Drug N on a Kinetex C18 2.6µm, 50x2.1mm

Column: Kinetex[®] 2.6 µm C18 100 Å, LC Column 50 x 2.1 mm, Ea

Dimensions: 50 x 2.1 mm ID

Order No: 00B-4462-AN

Elution Type: Gradient

Eluent A: 0.1% Formic acid

Eluent B: 0.1% Formic acid in methanol

| Gradient Profile: | Step No. | Time (min) | Pct A | Pct B |
|-------------------|----------|------------|-------|-------|
| | 1 | 0 | 65 | 35 |
| | 2 | 4 | 5 | 95 |
| | 3 | 4.01 | 65 | 35 |
| | 4 | 7 | 65 | 35 |

Flow Rate: 300 µL/min

Col. Temp.: ambient

Detection: Mass Spectrometer (MS) @ amu (ambient)

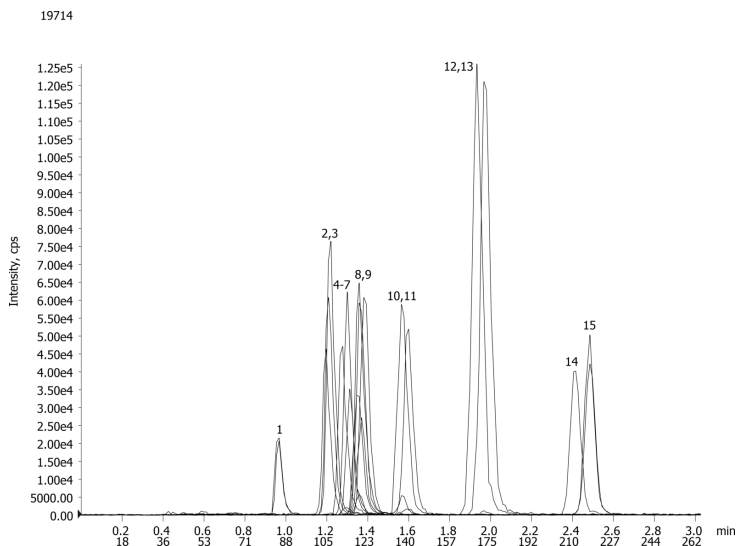
Detector Info: <a target="_blank"

href="https://sciex.com/products/mass-spectrometers?utm_campaign=2019%20application%20search&utm_source=phenomenex&utm_medium=referral">SCIEX<

Source: Cur-11, CAD-12, IS-4500, Tem-450, NEB-12

Dwell: 25ms

Polarity: Positive



Products used in this application:



Benzodiazepines from urine using Strata-X-Drug N on a Kinetex C18 2.6 μ m, 50x2.1mm

ANALYTES:

- 1 alpha-Hydroxyalprazolam
- 2 Oxazepam-d5
- 3 Oxazepam
- 4 Alprazolam-d5
- 5 Alprazolam
- 6 Nordiazepam-d5
- 7 Nordiazepam
- 8 Lorazepam-d4
- 9 Lorazepam
- 10 Clonazepam-d4
- 11 Clonazepam
- 12 Temazepam-d5
- 13 Temazepam
- 14 Diazepam-d5
- 15 Diazepam



Sample Preparation Details

for HPLC Application ID No.: 19714

Benzodiazepines from urine using Strata-X-Drug N on a Kinetex C18 2.6µm, 50x2.1mm

PRODUCT DESCRIPTION:

Strata™-X-Drug N 100 µm Polymeric Reversed Phase, 100 mg / 6 mL, Tubes , 30/Pk

Order No.: 8B-S129-ECH

SOLID PHASE EXTRACTION (SPE) PROCEDURE:

Note: The solvent volumes shown below are for a 100 mg bed mass.

The solvent volumes will need to be adjusted for a smaller or larger bed mass.

Condition:

Load:

Wash:

Dry:

10 minutes @ full vacuum

Elute:

Final Prep and Analysis:

Evaporate eluant under a stream of N2 gas at 50°C. Reconstitute with 1mL of 35% methanol

Inject: 0 µL on HPLC Mass Spectrometer (MS) @ amu (ambient)

| ANALYTES: | Spiked Conc. (ng/mL) | Log P | pKa | % Rec | %RSC (n=0) |
|---------------------------|-------------------------|-------|-----|-------|---------------|
| 1 alpha-Hydroxyalprazolam | 300 | | | 92.4 | |
| 2 Oxazepam-d5 | 300 | | | 100 | |
| 3 Oxazepam | 300 | | | 105.3 | |
| 4 Alprazolam-d5 | 300 | | | 100 | |
| 5 Alprazolam | 300 | | | 98.4 | |
| 6 Nordiazepam-d5 | 300 | | | 100 | |
| 7 Nordiazepam | 300 | | | 103.7 | |
| 8 Lorazepam-d4 | 300 | | | 100 | |
| 9 Lorazepam | 300 | | | 101.1 | |
| 10 Clonazepam-d4 | 300 | | | 100 | |
| 11 Clonazepam | 300 | | | 99.6 | |
| 12 Temazepam-d5 | 300 | | | 100 | |
| 13 Temazepam | 300 | | | 104.6 | |
| 14 Diazepam-d5 | 300 | | | 100 | |
| 15 Diazepam | 300 | | | 101.1 | |

Note: This method is designed as a convenient starting point for further investigation and can be tailored to meet your extraction goals. Call your local Phenomenex Representative for assistance in method development and optimization techniques.

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