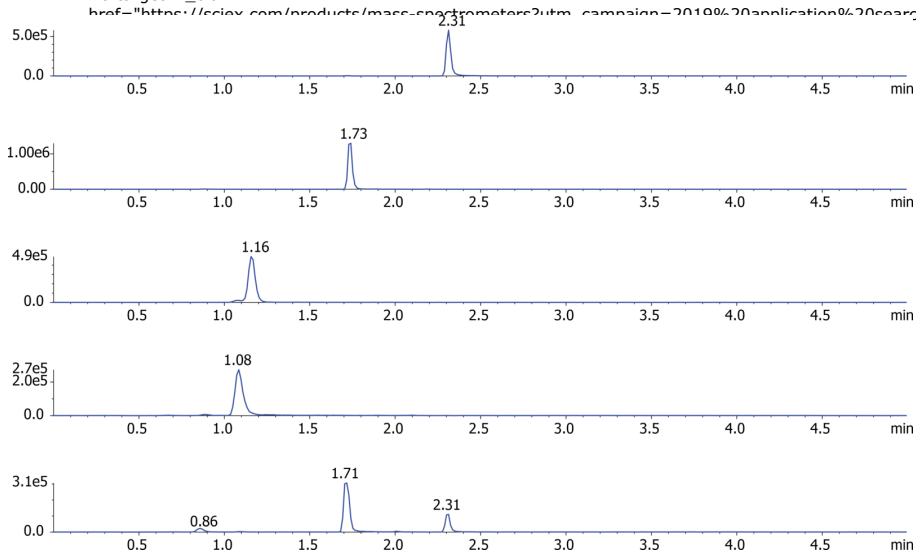


Nicotine and Metabolites by SPE using Gemini NX-C18, LC-MSMS_Fig.2**Column:** Gemini® 3 µm NX-C18 110 Å, LC Column 50 x 2 mm, Ea**Dimensions:** 50 x 2 mm ID**Order No:** 00B-4453-B0**Elution Type:** Gradient**Eluent A:** 20mM Ammonium Bicarbonate**Eluent B:** 100% Acetonitrile

Gradient Profile:	Step No.	Time (min)	Pct A	Pct B
	1	0	90	10
	2	3	25	75
	3	3.1	90	10
	4	5	90	10

Flow Rate: 500 µL/min**Col. Temp.:** 25 °C**Detection:** Tandem Mass Spec (MS-MS) @ (ambient)**Detector Info:** SCIEX<**Products used in this application:****ANALYTES:**

- 1 Nornicotine
Retention Time: 1.09 min
- 2 3-OH-Cotinine
Retention Time: 1.16 min
- 3 Anabasine
Retention Time: 1.71 min
- 4 Cotinine
Retention Time: 1.73 min
- 5 Nicotine
Retention Time: 2.31 min



Sample Preparation Details

for HPLC Application ID No.: 22035

Nicotine and Metabolites by SPE using Gemini NX-C18, LC-MSMS_Fig.2

PRODUCT DESCRIPTION:

StrataTM-X-C 33 μ m Polymeric Strong Cation, 60 mg / 3 mL, Tubes , 50/Pk

Order No.: 8B-S029-UBJ

SOLID PHASE EXTRACTION (SPE) PROCEDURE:

Note: The solvent volumes shown below are for a 60 mg bed mass.

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

Condition:

Load:

Wash:

Dry:

> 10" Hg for 5 min to remove residual water

Elute:

Final Prep and Analysis:

Reconstitution Solvent: 500 μ L Acetonitrile/20mM Ammonium bicarbonate (10:90)

Inject: 10 μ L on HPLC Tandem Mass Spec (MS-MS) @ (ambient)

ANALYTES:	Spiked Conc. (ng/mL)	Log P	pKa	% Rec	%RSC (n=0)
1 Nornicotine	0				
2 3-OH-Cotinine	0				
3 Anabasine	0				
4 Cotinine	0				
5 Nicotine	0				

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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For more information contact your Phenomenex Representative at info@phenomenex.com



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