

# HPLC Application

ID No.: 20906

## Nicotinic acid / Nicotinamide (10 ng/mL) in Human Plasma by Impact on Gemini 3µm C18 100x4.6mm

**Column:** Gemini® 3 µm C18 110 Å, LC Column 100 x 4.6 mm, Ea

**Dimensions:** 100 x 4.6 mm ID

**Order No:** 00D-4439-E0

**Elution Type:** Gradient

**Eluent A:** 0.1% formic acid

**Eluent B:** Methanol 100%

| Gradient Profile: | Step No. | Time (min) | Pct A | Pct B |
|-------------------|----------|------------|-------|-------|
|                   | 1        | 0          | 90    | 10    |
|                   | 2        | 2.5        | 10    | 90    |
|                   | 3        | 2.6        | 90    | 10    |
|                   | 4        | 4          | 90    | 10    |

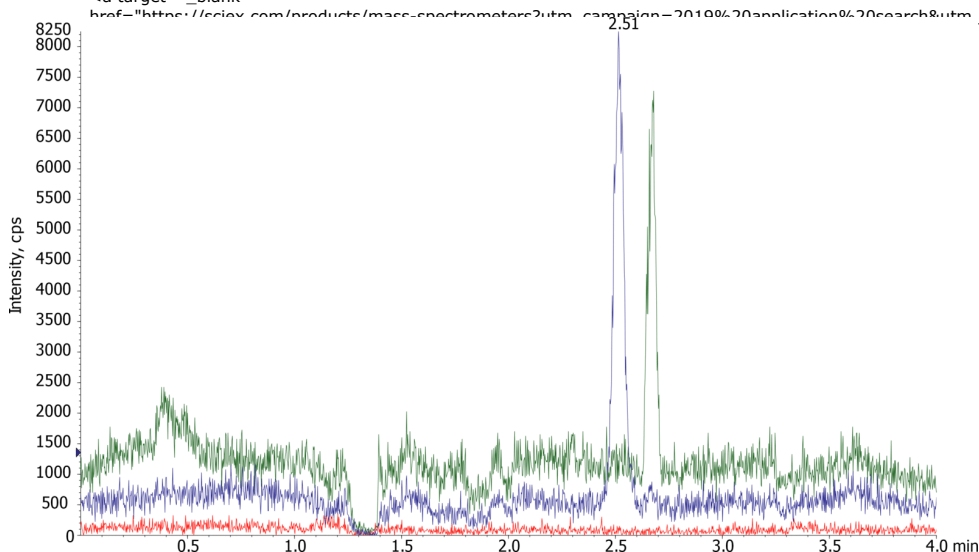
**Flow Rate:** 0.6 mL/min

**Col. Temp.:** ambient

**Detection:** Electrospray Mass Spec (ESMS) @ (ambient)

**Detector Info:**

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### ANALYTES:

- 1 Nicotinamide  
Retention Time: 2.51 min
- 2 Nicotinic acid  
Retention Time: 2.68 min



Products used in this application:



# Sample Preparation Details

for HPLC Application ID No.: 20906

## Nicotinic acid / Nicotinamide (10 ng/mL) in Human Plasma by Impact on Gemini 3 $\mu$ m C18 100x4.6mm

### PRODUCT DESCRIPTION:

Impact<sup>™</sup> Protein Precipitation, 2mL Square Well Filter Plate, 2/Pk

Order No.: CE0-7565

### SOLID PHASE EXTRACTION (SPE) PROCEDURE:

**Note:** The solvent volumes shown below are for a Proprietary bed mass.

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

#### Condition:

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#### Load:

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#### Wash:

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#### Dry:

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#### Elute:

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#### Final Prep and Analysis:

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Inject: 2  $\mu$ L on HPLC Electrospray Mass Spec (ESMS) @ (ambient)

| ANALYTES:        | Spiked Conc.<br>(ng/mL) | Log P | pKa | % Rec | %RSC<br>(n=0) |
|------------------|-------------------------|-------|-----|-------|---------------|
| 1 Nicotinamide   | 10                      |       |     |       |               |
| 2 Nicotinic acid | 10                      |       |     |       |               |

**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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