Underivatized MMA and Succinic Acid in Human Urine on Gemini C18, 3µm, 100x3 mm

Column: Gemini® 3 µm C18 110 Å, LC Column 100 x 3 mm, Ea
Dimensions: 100 x 3 mm ID
Order No: 00D-4439-Y0
Elution Type: Gradient
Eluent A: 0.1% Formic Acid in DI H2O
Eluent B: 0.1% Formic Acid + 10 mM Ammonium Formate in MeOH

Gradient Profile:
- **Step No.**: 1
  - **Time (min)**: 0
  - **Pct A**: 85
  - **Pct B**: 15
- **Step No.**: 2
  - **Time (min)**: 1.5
  - **Pct A**: 5
  - **Pct B**: 95
- **Step No.**: 3
  - **Time (min)**: 2.5
  - **Pct A**: 5
  - **Pct B**: 95
- **Step No.**: 4
  - **Time (min)**: 2.51
  - **Pct A**: 85
  - **Pct B**: 15
- **Step No.**: 5
  - **Time (min)**: 4.5
  - **Pct A**: 85
  - **Pct B**: 15

Flow Rate: 700 µL/min
Col. Temp.: 40 °C
Detection: Tandem Mass Spec (MS-MS) @ (ambient)

Detection Info: - SecurityGuard™ Guard Cartridge System extends column lifetime.

- SecurityGuard Cartridges, Gemini C18 4 x 2.0mm ID, 10/Pk Part No.: AJ0-7596
- Holder Part No.: KJ0-4282

Analyst Note: SecurityGuard™ Guard Cartridge System extends column lifetime.

ANALYTES:

1. Succinic acid (117.0-->72.9)
   - Retention Time: 1.16 min

2. Methylmalonic acid (MMA) 117.0-->72.9; 117.0-->54.9
   - Retention Time: 1.48 min
Underivatized MMA and Succinic Acid in Human Urine on Gemini C18, 3u, 100x3 mm

**PRODUCT DESCRIPTION:**
Strata™-X-AW 33 µm Polymeric Weak Anion, 30 mg / 1 mL, Tubes , 100/Pk

Order No.: 8B-S038-TAK

**SOLID PHASE EXTRACTION (SPE) PROCEDURE:**

**Note:** The solvent volumes shown below are for a 30 mg bed mass.
The solvent volumes will need to be adjusted for a smaller or larger bed mass.

**Condition:**

**Load:**
Into individually labeled 1.5 mL conical micro-centrifuge tubes combine 0.5 mL 25 mM Ammonium formate, 50 uL IS and 100 uL blank, standard, or sample

**Wash:**

**Dry:**
Dry under high vacuum for 5-10 min

**Elute:**

**Final Prep and Analysis:**
This method is for the analysis of underivatized MMA.
Inject: 10 µL on HPLC Tandem Mass Spec (MS-MS) @ (ambient)

**ANALYTES:**

<table>
<thead>
<tr>
<th></th>
<th>Spiked Conc. (ng/mL)</th>
<th>Log P</th>
<th>pKa</th>
<th>% Rec</th>
<th>%RSC (n=0)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Succinic acid</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>2</td>
<td>Methylmalonic acid</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

**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.