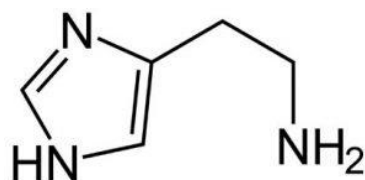
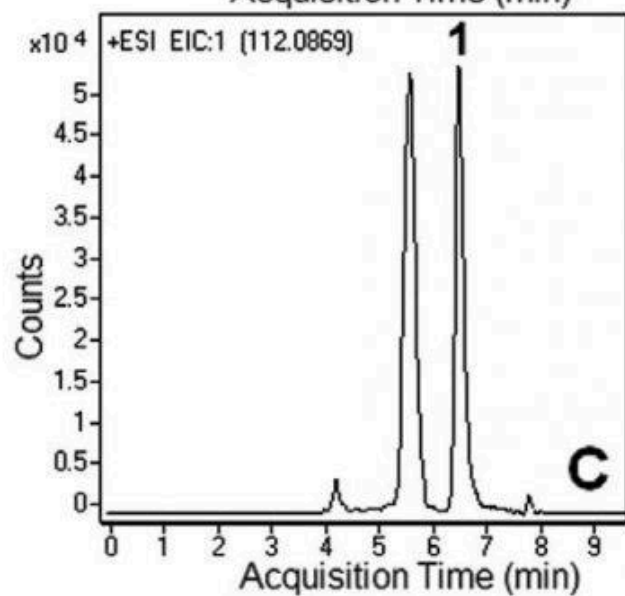
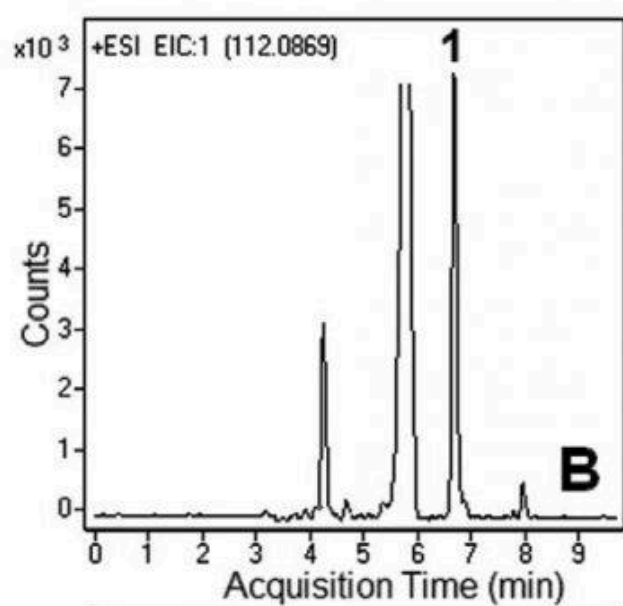
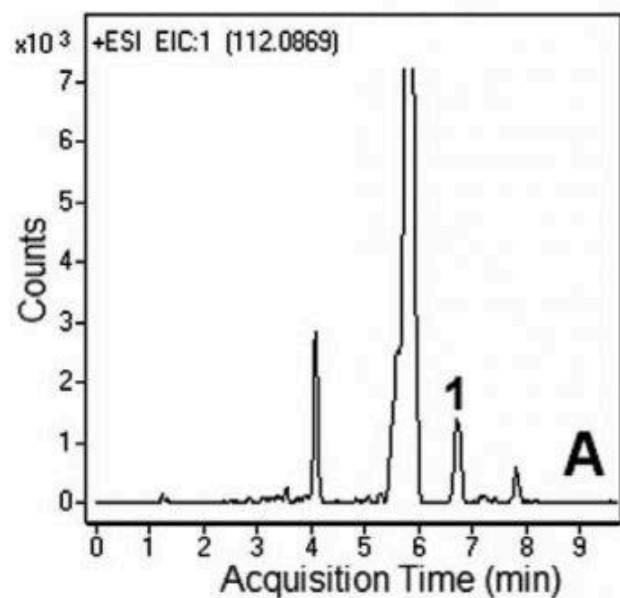


Histamine in Cheese Analyzed by LCMS - AppNote

No Derivatization Required for this Analysis of Histamine

A small but measurable amount of Histamine was found in a Cheese sample (*Figure A*) after an extraction procedure and analysis using the Cogent Diamond Hydride Column with MS detection. Two spiked Cheese samples were also analyzed. In *Figure B*, the Cheese sample was spiked before the extraction procedure at a level of 0.5 mg/L and *Figure C* shows the Histamine peak in a spiked extract from the Cheese sample at a level of 8.0 mg/L.

From the figures, it is obvious that the identification of Histamine by mass or retention time is not affected by the Cheese matrix or the extracted material. The Histamine content in the Cheese sample was determined based on a calibration curve and it was calculated to be 500 ± 5 ng/grams of Cheese (with a %RSD of 0.2 for n=5).



Peak:Histamine 112.0869 m/z [M+H]⁺**Method Conditions****Column:** Cogent Diamond Hydride™, 4µm, 100Å**Catalog No.:** [70000-15P-2](#)**Dimensions:** 2.1 x 150mm**Mobile Phase:**

A: DI Water / 50% 2-Propanol / 0.1% Formic Acid

B: Acetonitrile / 0.1% Formic Acid

Gradient:

| Time (Minutes) | %B |
|----------------|----|
| 0 | 80 |
| 5 | 10 |
| 7 | 10 |
| 8 | 80 |

Post Time: 5 minutes**Flow rate:** 0.4 mL/minute**Detection:** ESI – POS - Agilent 6210 MSD TOF Mass Spectrometer**Injection vol.:** 1µL

Sample Preparation: Grated Parmesan Cheese was purchased from a local supermarket. Three Cheese samples were prepared. The unspiked SPE sample was prepared by homogenizing 5g Cheese and 50 mL DI Water / 0.1% Formic Acid in a Waring blender for 10 minutes at 13,500 rpm. The mixture was then centrifuged at 4000 g for 20 minutes. The supernatant was refrigerated (20°C) for 10 min, treated by adding dropwise 3 M ammonia to a pH of 11.0, and then centrifuged at 1000 g for 5 min. The resulting supernatant was purified by Solid Phase Extraction (SPE) on a conditioned C18 sorbent and eluted with 2 mL of Methanol. After removal of the Methanol by Nitrogen gas, the extracted sample was re-dissolved in 2.0 mL of DI Water / 0.1 % Formic Acid for direct analyses. The spiked SPE samples were prepared by homogenizing 5.0 g of Cheese, 50 mL of DI Water / 0.1% Formic Acid, and an appropriate amount of 1 mg/mL Histamine stock solution in a Waring blender for 10 min at 13,500 rpm. Afterwards, the sample preparation was completed by following procedures for the unspiked SPE samples (i.e. centrifuge, SPE, etc.).

t₀: 0.9 minutes**Attachment****No 314 Histamine in Cheese by LC-MS pdf** 0.2 Mb [Download File](#)

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MicroSolv Technology Corporation
9158 Industrial Blvd. NE, Leland, NC 28451

Tel: (732) 380-8900
Fax: (910) 769-9435
Email: customers@mtc-usa.com
Website: www.mtc-usa.com