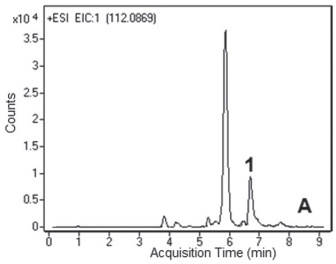
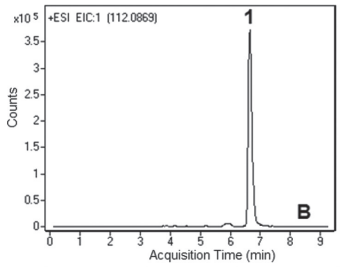
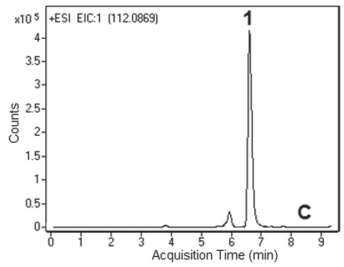
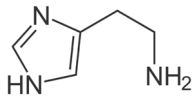
Histamine in Tuna

LC-MS method without derivatization









Histamine

Sample Preparation: Canned tuna was purchased from a local supermarket. Three tuna samples were prepared. The unspiked SPE sample was prepared by homogenising 5 g of tuna and 50 mL of DI H_2O / 0.1% FA in a Waring blender for 10 min at 13,500 rpm. The mixture was then centrifuged at 4000 g for 20 min. The supernatant was refrigerated (20 °C) for 10 min, treated by adding dropwise 3 M ammonia to a pH of 11.0, 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 H_2O / 0.1 % FA for direct analyses. The spiked SPE samples were prepared by homogenising 5.0 g of tuna, 50 mL of DI H_2O / 0.1% FA, and 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.).

Method Conditions

Column: Cogent Diamond Hydride™, 4µm, 100Å

Catalog No.: 70000-15P-2

Dimensions: 2.1 x 150 mm

Solvents: A: 50% DI H_2O / 50% 2-propanol / 0.1% formic acid
B: Acetonitrile / 0.1% formic acid

Gradient:

time (min.)	%B
0	80
5	10
7	10
8	80

Post Time: 3 min

Injection vol.: 1 microL

Flow rate: 0.4 mL/min

Detection: ESI – POSG - Agilent 6210 MSD TOF mass spectrometer

Peak: 1. Histamine (112.0869 m/z)

t_0: 0.9 min

Discussion

A small amount of histamine was found in a tuna sample (Figure A) after an extraction procedure and analysis using the Cogent Diamond Hydride column and MS detection. In Figure B, a tuna 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 tuna sample at a level of 1.0 mg/L.

The figures show that the identification of histamine by mass or retention time is not affected by the tuna matrix or extracted material. The histamine content in the tuna sample was determined based on the calibration curve and it was determined to be 320 ± 4 ng/grams of tuna (with a %RSD of 0.2 for n=5). The developed protocol after validation can be used for the analysis of this polar compound in a variety of food matrices.

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