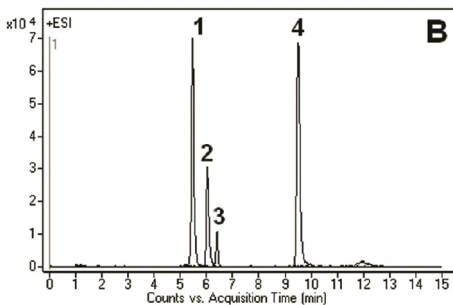
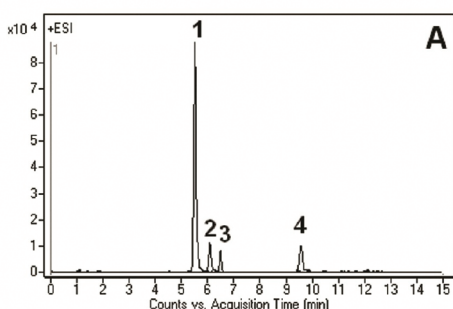
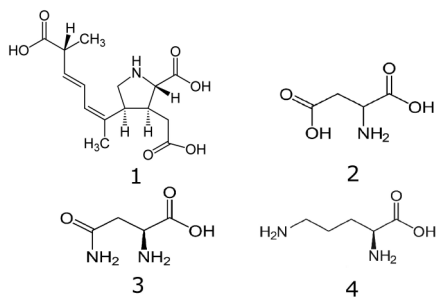


Domoic Acid, Amino Acids, & Ornithine in Cell Extracts
LC-MS analysis without analyte derivatization



Note: Domoic acid, a compound known for some 30 years, was found to have neurological effects when ingested by humans. Upon ingestion, it causes diarrhea, dizziness, seizures, permanent loss of short-term memory and sometimes death.

Method Conditions

Column: Cogent Diamond Hydride™, 4µm, 100A
Catalog No.: 70000-15P-2
Dimensions: 2.1 x 150 mm
Solvents: A: 50% DI H₂O/ 50% MeOH/ 0.1% formic acid
 B: Acetonitrile/ 0.1% formic acid
Gradient:

time (min.)	%B	time (min.)	%B
0	95	10	20
7	20	11	95

Post Time: 5 min
Injection Vol.: 1 microL
Flow Rate: 0.4 mL/min
Detection: ESI – POS - Agilent 6210 MSD TOF mass spectrometer
Sample: Methanolic extracts of proprietary cell cultures A and B, isolated by filtration
Peak:
 1. Domoic acid 312.1442 m/z (M +H)⁺
 2. L-aspartic acid 134.0453 m/z (M +H)⁺
 3. L-asparagine 133.0608 m/z (M +H)⁺
 4. Ornithine 133.0972 m/z (M +H)⁺
t₀: 0.9 min

Discussion

The potent toxin domoic acid (DA) is a highly polar compound which is poorly retained on most reversed phase columns. When a Diamond Hydride™ column was used, not only DA was retained but also two isobaric compounds (L-asparagine and ornithine) were retained and separated. In addition, the peak obtained for aspartic acid was very symmetrical. From these chromatograms, it is clear that the two cell extracts show differences in the ratios between the analyzed compounds (Figure A vs. B). The presented method can be used in studies of cell extracts. In addition, it is robust and reproducible. Cogent columns require a very short equilibration time between gradient runs. The developed method based on ANP-HPLC and MS detection **does not require derivatization** of DA, amino acids or ornithine.

Cat. No.	Description
70000-15P-2	Diamond Hydride™ HPLC Column, 100A, 4µm, 2.1 x 150 mm