BIOPHARMACEUTICALS

HALO



Separation of Amino Acids by HILIC/ Single Quadrupole Mass Detection

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Representative spectra of 21 standard amino acids separated on a HALO® Glycan column are shown. Amino acid isomers (e.g., Leu, IIe), as well as amino acids: Gln and Lys, are detected under the same mass channels (m/z 132.1 and 147.1, respectively). The elution order for the isomers is Leu first, followed by IIe. The target amino acid is specified with an asterisk (*) symbol in chromatograms showing multiple peaks (e.g., Cys, Thr, Asn, Glu, Asp).

PEAK IDENTITIES:

Amino Acid	Acronym	Monoisotopic mass ^{ref 2} [M]	SIR mass ^{ref 6} [M+H ⁺]
Alanine	Ala, A	89.1	90
Arginine	Arg, R	174.2	175
Asparagine	Asn, N	132.1	133
Aspartic acid	Asp, D	133.1	134
Cysteine	Cys, C	121.2	122
Cystine ref 7	n/a	240.3 ref 7	241
Glutamic acid	Glu, E	147.1	148
Glutamine	Gln, Q	146.2	147
Glycine	Gly, G	75.1	76
Histidine	His, H	155.2	156
Isoleucine	lle, I	131.2	132
Leucine	Leu, L	131.2	132
Lysine	Lys, K	146.2	147
Methionine	Met, M	149.2	150
Phenylalanine	Phe, F	165.2	166
Proline	Pro, P	115.1	116
Serine	Ser, S	105.1	106
Threonine	Thr, T	119.1	120
Tryptophan	Trp, W	204.2	205
Tyrosine	Tyr, Y	181.2	182
Valine	Val, V	117.1	118

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TEST CONDITIONS:

Column: HALO 90 Å Glycan, 2.7 µm, 2.1 x 100 mm Part Number: 92922-605 Mobile Phase A: 85/15 Acetonitrile/ 10 mM ammonium formate, 0.15% Formic Acid Mobile Phase B: 10 mM ammonium formate pH 3.0, 0.15% Formic Acid Gradient: Time %B 0.0 0.0 10.0 5.0 15.0 37.5 15.5 95.0 95.0 20.0 20.5 0.0 25.0 0.0 Flow Rate: 0.4 mL/min Column Temperature: 30 °C Sample Manager Temperature: 5 ± 3 °C **Injection Volume:** 1 µL **Data Courtesy of:** Neshita Kipuw, Pfenex Expression Technology

MS CONDITIONS:

Ionization Mode: ESI Polarity: Positive Full Scan: m/z 50-1000 Cone Voltage: 15 V Gas Flow Pressure: 70-100 psi



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