Data for the calculation of the molecular masses of peptides and proteins for use in mass spectrometry

Common MALDI Peptides

Angiotensin II	1046.5423	$C_{50}H_{71}N_3O_{12}$	DRVYIHPF
Angiotensin I	1296.6853	$C_{62}H_{89}N_{17}O_{14}$	DRVYIHPFHL
Glu-Fibrinopeptide	1570.6774	$C_{66}H_{95}N_{19}O_{26}$	EGVNDNEEGF FSAR
Renin Substrate	1758.9331	$C_{85}H_{123}N_{21}O_{20}$	DRVYIHPFHL LVYS
ACTH 18-39	2465.1989	$C_{112}H_{165}N_{27}O_{36}$	RPVKVYPNGA EDESA EAFPL EF
Insulin B	3494.6513	$C_{157}H_{232}N_{40}O_{47}S_2$	FVNQHLCGSH LVEAL YLVCG ERGFF YTPKA
Insulin	5730.6087	$C_{254}H_{377}N_{65}O_{75}S_6$	

GFP Fragments

Fragment	Mass	Fragment	Mass
y''_1	175.1195	b_1	130.0504
y''_2	246.1566	b_2	187.0719
y''_3	333.1886	b_3	286.1403
y''_4	480.2570	b_4	400.1832
y''_5	627.3254	b_5	515.2101
y''_6	684.3469	b_6	629.2531
y''_7	813.3895	b_7	758.2957
y''_8	942.4321	b_8	887.3383
y''_9	942.4321	b_9	944.3597
y''_{10}	1171.5020	b_{10}	1091.4281
y''_{11}	1285.5448	b_{11}	1238.4966
y''_{12}	1384.6133	b_{12}	1325.5286
y''_{13}	1441.6348	b_{13}	1396.5657
y''_n	-	b_n	-

[Glu[1]]-Fibrinopeptide B, Sigma F3261

Dissolve in H_2O (no TFA) to a concentration of 0.1 mg/ml.

NB. 0.1 mg/ml = 63.7 nmol/ml = 63.7 pmol/µL [as #mols = (amount in grams/molecular weight)]

Stock solution of 10 pmol/µl

Dilute 100 µL GFP with 537 µL (0.1 %TFA in H_2O).

LockMass solution 200 fmol/µl

Dilute 100 µL of Stock Solution with 400 µL (0.1%TFA in H_2O).

Immonium Ions[1]

Residue			Immonium Ion	Others
Glycine	Gly	G	30.03	
Alanine	Ala	A	44.05	
Serine	Ser	S	60.05	
Proline	Pro	P	70.07	
Valine	Val	V	72.08	41,55,69
Threonine	Thr	T	74.06	
Cysteine	Cys	C	76.02	
Isoleucine	Ile	I	86.10	44,72
Leucine	Leu	L	86.10	44,72
Asparagine	Asn	N	87.06	70
Aspartic acid	Asp	D	88.04	70
Glutamine	Gln	Q	101.07	84,112,129
Lysine	Lys	K	101.11	56,84,129
Glutamic acid	Glu	E	102.06	84,91
Methionine	Met	M	104.05	
Histidine	His	H	110.07	
Phenyl-alanine	Phe	F	120.08	91
Arginine	Arg	R	129.11	70,87,100,112
Tyrosine	Tyr	Y	136.08	91,107
Tryptophan	Trp	W	159.09	77,117,130,132

70,100,112,129	Arg may be present, but other acids can give these ions
70	If abundant, and no Arg present good indication of Pro present
84	If abundant a good indication of Lys present
86	Leu or Ile likely
110	His present
120, 91	Phe present, unless oxidized Met there
136, 91, 107	Tyr present
130, 159	Trp present

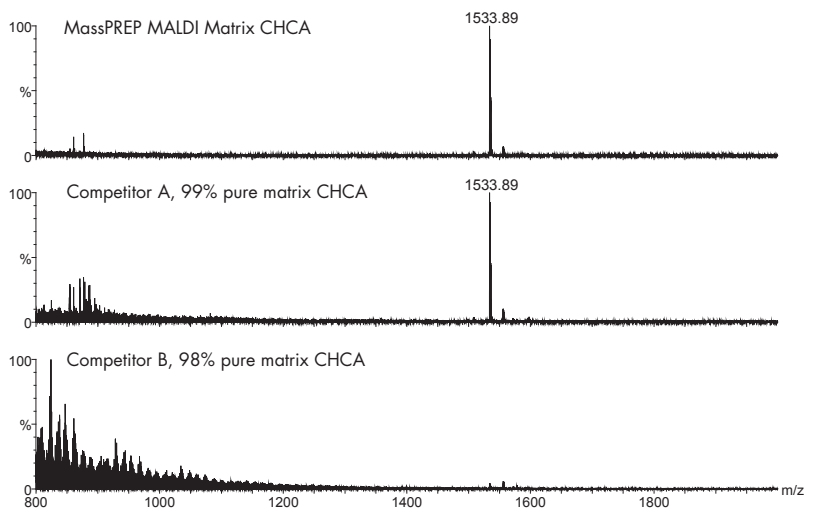
Waters MICROMASS MS TECHNOLOGIES

Waters® MassPREP™ MALDI Matrix

Waters® has developed a range of high-quality purified matrices designed to help optimize the desorption of sample from the target plate surface for maximum sensitivity in MALDI analysis. The matrices have been designed for scientists using a fully-supported, system solution from Waters with the goal of optimizing overall system performance. This specialty line of MassPREP™ consumables are suitable for all MALDI applications, including proteins, peptides, oligonucleotides, oligosaccharides, microbiology, and polymers.

MassPREP matrices

Description	Matrix
MassPREP MALDI Matrix CHCA	α - cyano - 4 - hydroxycinnamic acid
MassPREP MALDI Matrix SA	3,5 - dimethoxy - 4 - hydroxycinnamic acid (Sinapinic acid)
MassPREP MALDI Matrix DHB	2,5 - dihydroxybenzioc acid
MassPREP MALDI Matrix HPA	3 - hydroxypicolinic acid
MassPREP MALDI Matrix THAP	2,4,6 - trihydroxyacetophenone
MALDI Matrix Kit	CHCA, SA, DHB, HPA, THAP



Compared with other commercially available MALDI matrices, the MassPREP™ MALDI Matrix yields significantly cleaner spectra, enhancing data quality and sensitivity.

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MICROMASS® MS TECHNOLOGIES

Data for the calculation of the molecular masses of peptides and proteins for use in mass spectrometry

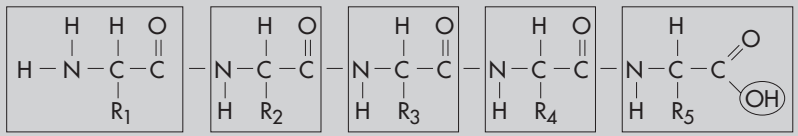
Peptide Mass Sufficiency

(Theoretical) Average Amino Acid 118.05670

Number of average amino acids	Theoretical Mass
1	118.057
2	236.113
3	354.170
4	472.227
5	590.283
6	708.340
7	826.397
8	944.454
9	1062.510
10	1180.567
11	1298.624
12	1416.680
13	1534.737
14	1652.794
15	1770.850
16	1888.907
17	2006.964
18	2125.021
19	2243.077
20	2361.134
21	2479.191
22	2567.247
23	2715.304
24	2833.361
25	2951.417

Amino acid masses sorted by weight

Amino Acid	Notation	Single	Mass in chain	Free weight	C terminal ion (Y'')	N terminal ion (B)
Glycine	Gly	G	57.0215	75.0320	76.0398	58.0293
Alanine	Ala	A	71.0372	89.0477	90.0555	72.0450
Serine	Ser	S	87.0321	105.0426	106.0504	88.0399
Proline	Pro	P	97.0528	115.0633	116.0711	98.0606
Valine	Val	V	99.0785	117.0790	118.0868	100.0763
Threonine	Thr	T	101.0477	119.0582	120.0660	102.0555
Cysteine	Cys	C	103.0093	121.0198	122.0276	104.0171
Isoleucine	Ile	I	113.0841	131.0946	132.1024	114.0919
Leucine	Leu	L	113.0841	131.0946	132.1024	114.0919
Asparagine	Asn	N	114.0430	132.0535	133.0613	115.0508
Aspartic acid	Asp	D	115.0270	133.0375	134.0453	116.0348
Glutamine	Gln	Q	128.0586	146.0691	147.0769	129.0664
Lysine	Lys	K	128.0950	146.1055	147.1133	129.1028
Glutamic acid	Glu	E	129.0427	147.0532	148.0610	130.0505
Methionine	Met	M	131.0406	149.0511	150.0589	132.0484
Histidine	His	H	137.0590	155.0695	156.0773	138.0668
Phenyl-alanine	Phe	F	147.0685	165.0790	166.0868	148.0763
Arginine	Arg	R	156.1012	174.1117	175.1195	157.1090
Tyrosine	Tyr	Y	163.0634	181.0739	182.0817	164.0712
Tryptophan	Trp	W	186.0794	204.0899	205.0977	187.0872
N-term (H)				1.0078	-	
C-term (OH)				17.0027	-	



Waters MICROMASS MS TECHNOLOGIES

PEG Calibration

Use PEG 200, 600, 1000, 2000.

Prepare each PEG in 50/50 Acetonitrile/H_2O at 10 mg/mL.

Mix all 4 in equal volume.

Add 20% NaI (2 mg/ml 50/50 2-propanol/H_2O).

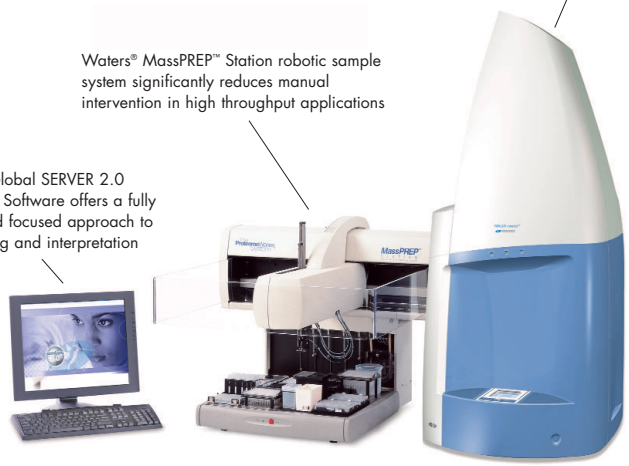
Spot 1:1 with Alpha cyano matrix (5 mg/mL).

The addition of NaI then produces sodiated PEG ions.

Masses follow sequence of $[(C_2H_4O)n+H_2O+Na]$

173.079	1185.682	2198.285
217.105	1229.708	2242.311
261.131	1273.734	2286.337
305.158	1317.761	2330.364
349.184	1361.787	2374.390
393.210	1405.813	2418.416
437.236	1449.839	2462.442
481.262	1493.865	2506.468
525.289	1537.892	2550.495
569.315	1581.918	2594.521
613.341	1625.944	2638.547
657.367	1669.970	2682.573
701.394	1713.996	2726.599
745.420	1758.023	2770.626
789.446	1802.049	2814.652
833.472	1846.075	2858.678
877.498	1890.101	2902.704
921.525	1934.128	2946.731
965.551	1978.154	2990.757
1009.577	2022.180	3034.783
1053.603	2066.206	3078.809
1097.629	2110.232	3122.835
1141.656	2154.259	3166.862

Waters® MALDI-TOF Mass Spectrometry Systems for Proteomics



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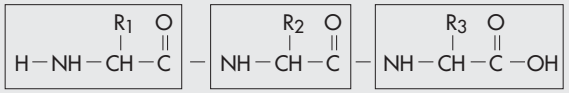
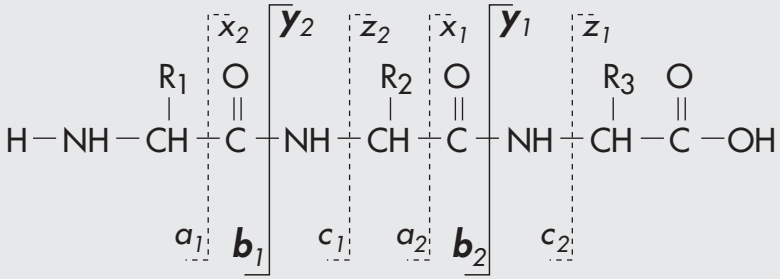
Data for the calculation of the molecular masses of peptides and proteins for use in mass spectrometry

Example of a 3 Amino Acid Peptide

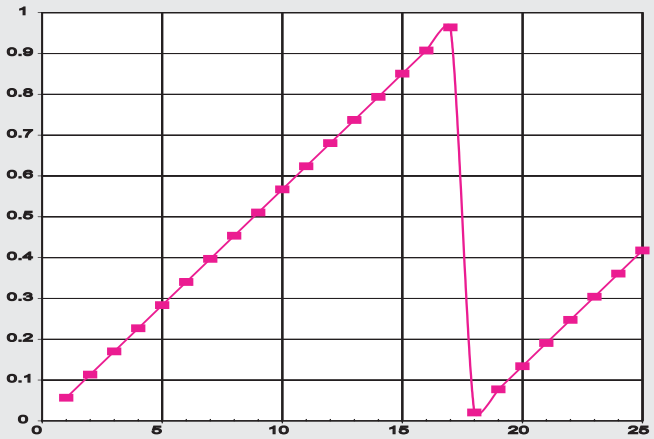


Amino-terminal residue N-term → Carboxyl-terminal residue C-term

Amino Acid Fragmentation Notation[2]



Mass Excess vs Mass



Enzyme Autolysis Peaks

Bovine Trypsin P00760

	M	[M+H]+	Sequence	
T15	905.497	**906.505**	NKPGVYTK	221-228
T11	1019.496	**1020.504**	APILSDSSCK	160-169
T9	1152.566	**1153.574**	SSGTSYPDVLK	146-156
T4	2162.049	**2163.057**	LGEDNINVVEGNEQFISASK	70-89
T5	2272.152	**2273.160**	SIVHPSYNSNTLNNDIMLIK	90-109
T5-6	2513.331	**2514.339**	SIVHPSYNSNTLNNDIMLIKL K	90-111

Porcine Trypsin P00761

	M	[M+H]+	Sequence	
T8	841.502	**842.510**	VATVSLPR	108-115
T7	1044.556	**1045.564**	LSSPATLNSR	98-107
T5	2210.097	**2211.105**	LGEHNIDVLEGNEQFINAAK	58-77
T6	2282.173	**2283.181**	IITHPNFNGNTLDNDIMLIK	78-97

Keratin Human P04264

	M	[M+H]+	Sequence	
T41	1178.593	**1179.601**	YEELQITAGR	376-385
T57	1276.703	**1277.711**	LALDLEIATYR	472-482
T18	1382.683	**1383.691**	SLNNQFASFIDK	185-196
T32-33	1392.725	**1393.733**	TNAENEFVTIKK	277-288
T20	1474.778	**1475.786**	FLEQQNQVLQTK	199-210
T55-56	1522.781	**1523.789**	LLRDYQELMNTK	460-471
T49	1715.844	**1716.852**	QISNLQQSISDAEQR	417-431
T17-18	1880.963	**1881.971**	EQIKSLNNQFASFIDK	181-196
T22	1992.969	**1993.977**	THNLEPYFESFINNLR	223-238
T22-23	2149.071	**2150.078**	THNLEPYFESFINNLRR	223-239
T60	2226.844	**2227.851**	GGGGGYGSGGSSYGSGGGSYGSGGGGGG	518-548

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Waters® Q-Tof™ Mass Spectrometry Systems for Proteomics

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- Patented Post Translational Modification (PTM) discovery software
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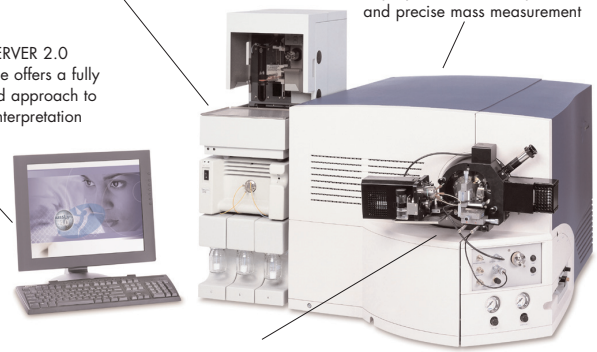

Waters Mass Spectrometry Systems for identification of proteins and PTM, feature the Waters CapLC Pump and Autosampler and the Waters family of Q-Tof Mass Spectrometers.

ProteinLynx™ Global SERVER 2.0 Bio-Informatics Software

ProteinLynx™ Global SERVER 2.0 is Waters total protein Mass-Informatics™ platform - featuring a new and comprehensive range of integrated tools for proteomics project management, relative protein quantitation, identification and characterization, all exploiting the specificity of exact mass data.

Using ProteinLynx Global SERVER 2.0 with Waters MS systems in your laboratory will:

- Reduce ambiguous protein identification by MS
- Increase confidence in your results
- Automatically distill useful knowledge from complex datasets
- Reduce raw data overload in your laboratory
- Overcome lack of resources for manual data interpretation

Peptide sequence from BSA	Lockspray™ corrected data (MassSeq™ 10 ppm tolerance)	Nominal mass data (MassSeq™ 100 ppm)
FKDLGEEHFK	FKDLGEEHFK	FKDLGEE RKK
LCVLHEKTPVSEK	LCVLHEKTPVSEK	LCVL RLLLRRLD
CCTESLVNR	CCTESLVNR	CCTESL KKL
KVPQVSTPTLVEVSR	KVPQVSTPTLVEVSR	KVPQVSTPTLVEVSR
LVNELTEFAK	LVNELTEFAK	LVNEL HTLPK
GFQNALLVR	(FG) QNALLVR	No result
YLYELAR	YLYELAR	YLYELAR

Green: Confident assignment
Red: Incorrect assignment
Yellow: Ambiguous assignment

De novo sequencing of peptides is significantly enhanced through the specificity of exact mass MS/MS. The table demonstrates how greater confidence in sequence assignment is achieved with improved mass accuracy. Results are shown for a digest of Bovine Serum Albumin using ProteinLynx Global SERVER 2.0 Software.

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[1]

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[2]

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