



● Introducing a Novel
Bench Top Orbitrap for Highly
Selective Peptide/Protein
Quantitation

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European Field Marketing

Q Exactive™

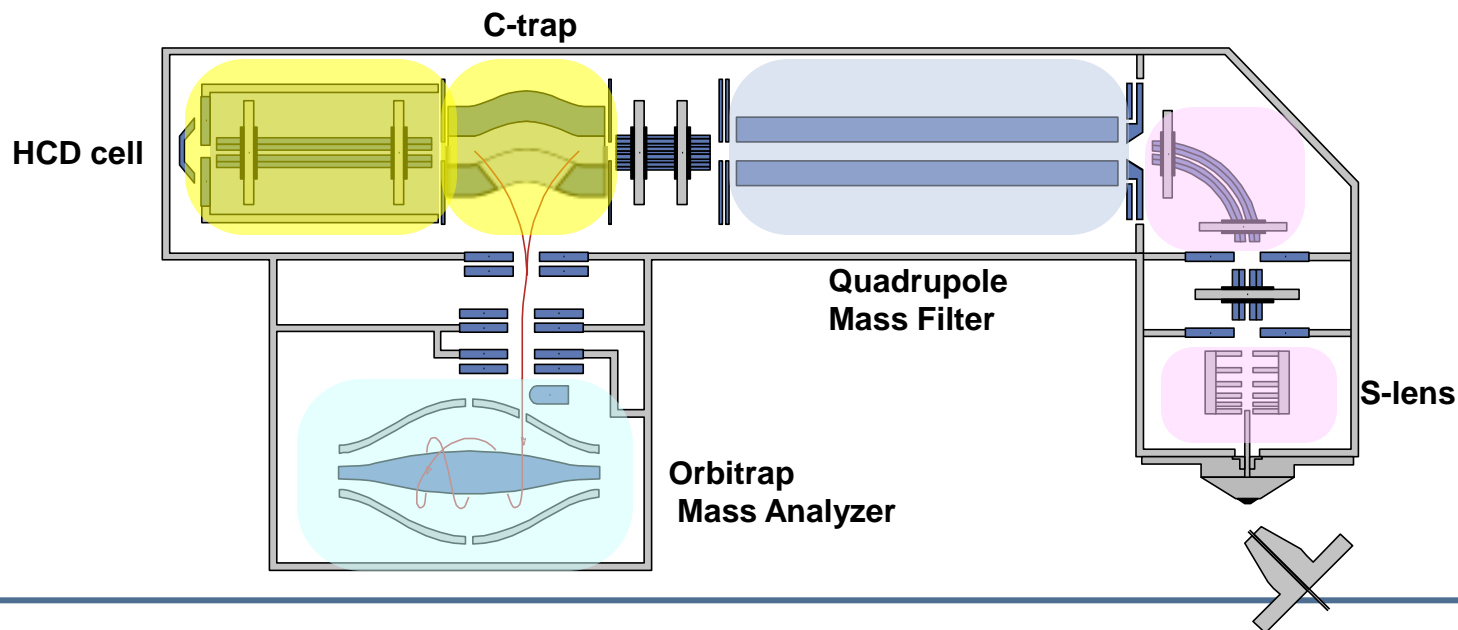


Q Exactive is the first benchtop LC-MS/MS Orbitrap instrument in the market

- **Q Exactive** can be seen as a complementary extension of the product line based on the Exactive™
- **Technological Advancements**
 - Quadrupole for precursor ion selection (MS/MS and SIM)
 - More sensitive ion source (S-Lens)
 - Higher scan speed (advanced signal processing)

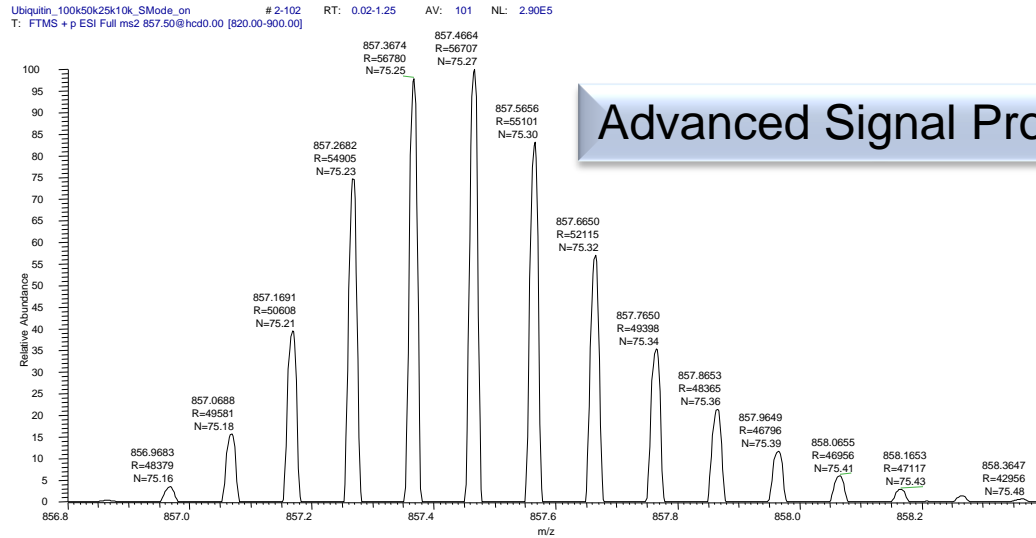
Q Exactive™ Hardware Innovations

- Quadrupole mass filter
- Advanced signal processing for Orbitrap data processing
- Predictive automatic gain control (pAGC) and parallel filling & detection
- C-trap directly interfaced to HCD (like in LTQ Orbitrap Velos)
- Possibility of multiple fills for spectrum multiplexing
- S-lens for higher transmission (like in LTQ Orbitrap Velos) with rugged optics

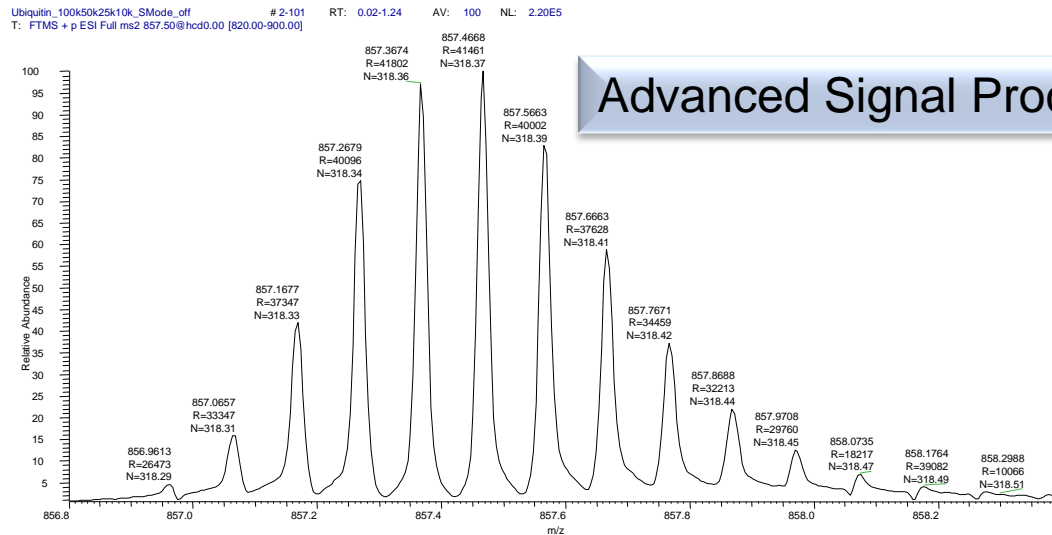


Increase of Resolution by Advanced Signal Processing

Ubiquitin, $[MH]^{10+}$



Advanced Signal Processing **on**

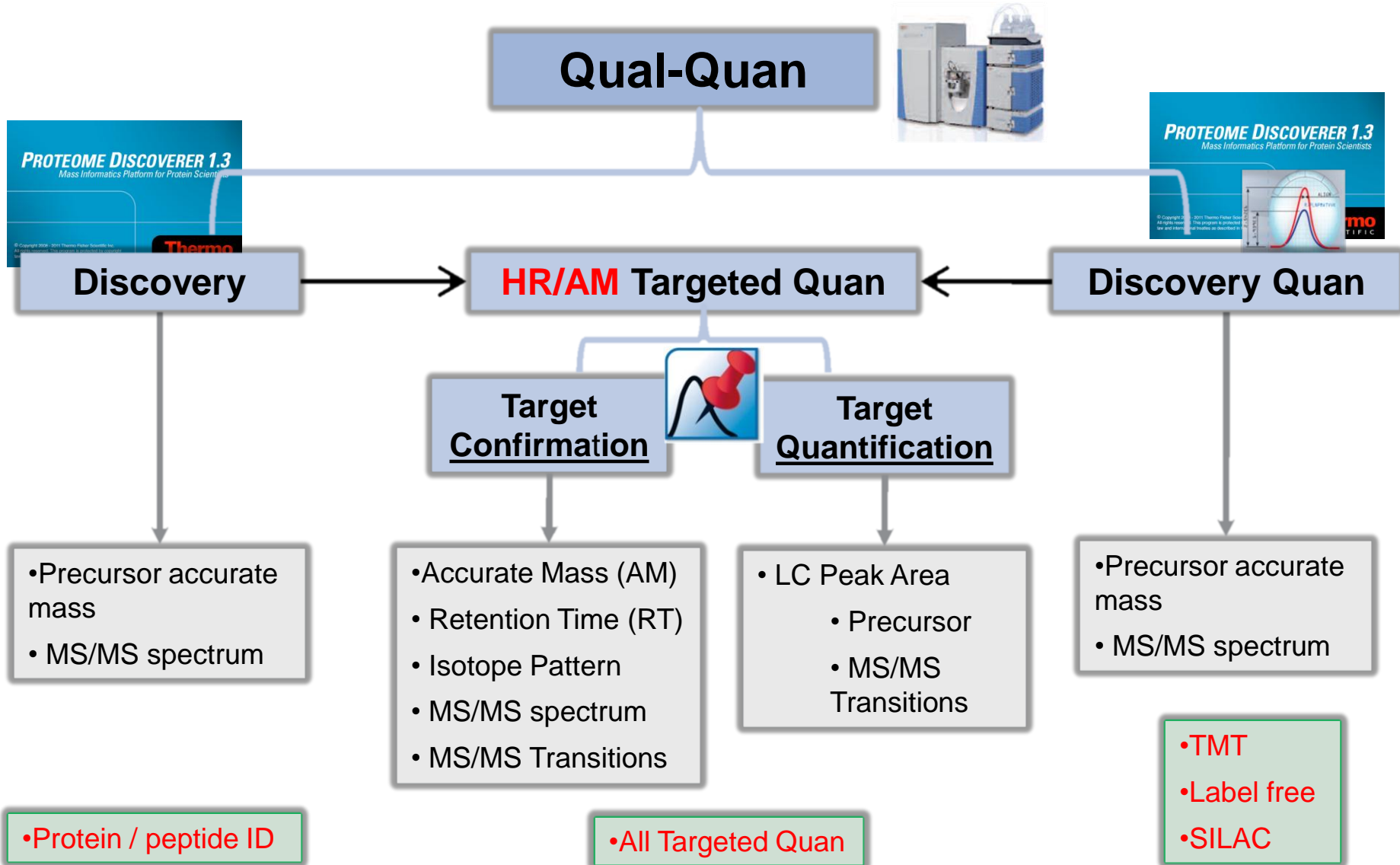


Advanced Signal Processing **off**

Feature and their provided Benefit

Feature	Benefit	
S-Lens	Higher ion transmission and duty cycle	
Quadrupole	“real MS/MS”	
Advanced Signal Processing	Higher Acquisition speed (UHPLC compatibility)	
Advanced Signal Processing	on	
Predictive AGC	Resolution at m/z 200	Max. scan speed (Hz)
Fast pos/neg switching	17.500 ▼	12
Spectrum Multiplexing	35.000 ▼	7
	70.000 ▼	3
All Ion Fragmentation	140.000 ▼	1.5
Parallel filling/detection	Faster scan speed/duty cycle	

From Discovery to Quantification - do it all on the Q Exactive



Challenges with IgG Characterization

ERBITUX HEAVY CHAIN

QVQLKQSGPG LVQPSQSLSI TCTVSGFLT N YGVHWVRQSP GKGLELGVIW SGGNTDYNT P
FTSRLSINKD NSKSQVFFKM NSLQSNDAI YYCARALTY Y DYEFAYWGQG TLVTVSAAST
KGPSVFPLAP SSKSTSGGTA ALGCLVKDYF PEPVTVSWNS GALTSGVHTF PAVLQSSGL
YSLSSVVTVP SSSLGTQTYI CNVNHKVDKR VEPKSCDKT HTCPPCPAP ELLGGPSVF
LFPPKDTLM ISRTPEVTC VVVDVSHED PEVKFNWYV DGVEVHNAK TKEEQYNST YRVVSVLT
VLHQDWLNGK EYKCKVSNKA LPAPIEKTI SKAKGQPRE PQVYTLPPS RDELTKNQV
SLTCLVKGF YPSDIAVEW ESNGQPENN YKTTTPVLD SDGSFFLYS KLTVDKSRW
QQGNVFC SVM HEALHNHYT QKSLSLSPG K

ERBITUX LIGHT CHAIN

DILLTQSPVI LSVSPGERV SFSCRASQSI GTNIHWYQQR TNGSPRLLI KYASESISG IPSRFSGSGS
GTDFTLSINS VESEDIADYY CQQNNNWPTT FGAGTKLEL KRTVAAPSV FIFPPSDEQ
LKSGTASVVC LLNFPYPREA KVQWKVDNA LQSGNSQES VTEQDSKDS TYSLSSTLT
LSKADYEKH KUYACEVTH QGLSSPVTK SFNRGA

Challenges with IgG Characterization

ERBITUX HEAVY CHAIN

QVQLKQSGPG LVQPSQSLSI TCTVSGFLTN YGVHWVRQSP GKGLELGVIW SGGNTDYNTPT
FTSRLSINKD NSKSQVFFKM NSLQSNDAI YYCARALTYT DYEFAYWGG TLVTVSAAST
KGPSVFPLAP SSKSTSGGTA ALGCLVKDYF PEPVTVSWNS GALTSGVHTF PAVLQSSGL
YLSSTVTVTP SSSLGTQTYI CNVNHKVDKR VEPKSCDKT HTCPPCPAP ELLGGPSVF
LFPPKDTLM ISRTPEVTC VVVDVSHED PEVKFNWYV DGVEVHNAK TKEEQYNST YRVVSVLT
VLHQDWLNGK EYKCKVSNKA LPAPIEKTI SKAKGQPRE PQVYTLPPS RDELTKNQV
SLTCLVKGF YPSDIAVEW ESNGQPENN YKTTTPVLD SDGSFFLYS KLTVDKSRW
QQGNVFCSVM HEALHNHYT QKSLSLSPG K

ERBITUX LIGHT CHAIN

DILLTQSPVI LSVSPGERV SFSCRASQSI GTNIHWYQQR TNGSPRLLI KYASESISG IPSRFSGSGS
GTDFTLSINS VESEDIADYY CQQNNNWPTT FGAGTKLEL KRTVAAPSV FIFPPSDEQ
LKSGTASVVC LLNMFYPREA KVQWKVDNA LQSGNSQES VTEQDSKDS TYSLSSTLT
LSKADYEKH KVYACEVTH QGLSSPVTK SFNRGAK

mAb Peptide Mapping Challenges for LC-MS

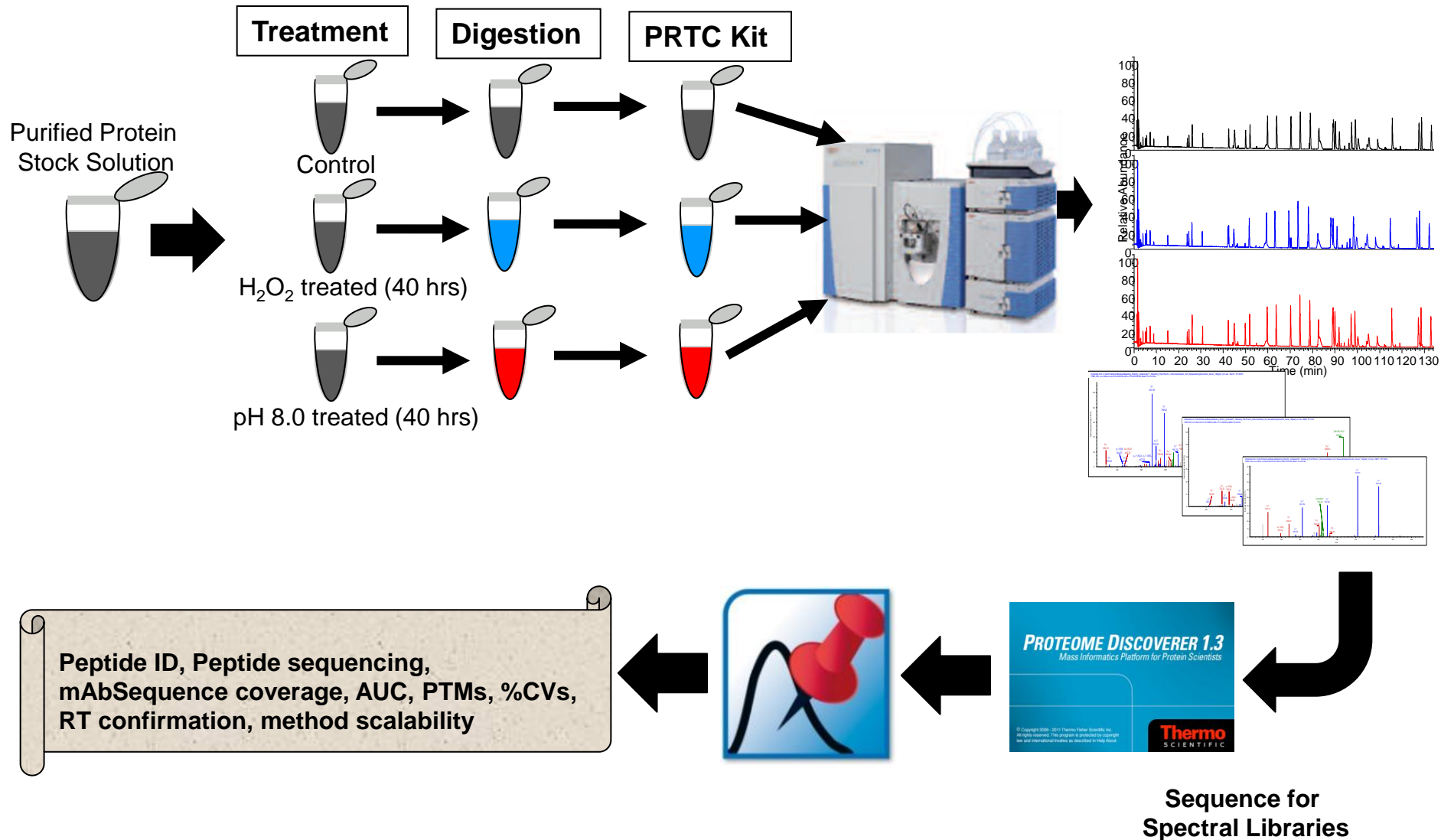
Erbix in silico Digestion

From-to	Sequence	[M+H] ⁺	HF	MALDI TOF	ESI TOF	From-to	Sequence	[M+H] ⁺	HF	MALDI TOF	ESI TOF
1-5	QVQLK	615.3824	7.47			291-294	TKPR	500.3122	1.45		
6-38	QSGPGLVQPSQSLStcTVSGFLTN YGVHWVR	3476.732	52.95			295-303	EEQYNSTYR	1189.512	12.09		
39-43	QSPGK	516.2776	1.96			304-319	VVSVLTVLHQDWLNGK	1808.006	3 42.38		
44-66	GLELGVWISGGNTDYNTPTFSR	2385.155	45.70			320-322	EYK	439.2187	0.00		
67-71	LSINK	574.3558	8.99			323-324	cK	249.1124	0.00		
72-75	DNSK	463.2147	2.37			325-328	VSNK	447.2561	2.72		
76-81	SQVFFK	755.4086	22.87			329-336	ALPAPIEK	838.5032	19.30		
82-97	MNSLQSNDAIYYcAR	1907.826	29.74			337-340	TISK	448.2765	3.44		
98-123	ALTYDYEFAYWQGTLVTVSAAS TK	2905.396	52.33			341-342	AK	218.1499	0.00		
124-135	GPSVFPLAPSSK	1186.647	29.57			343-346	GQPR	457.2517	2.72		
136-149	STSGGTAALGcLVK	1322.662	27.79			347-357	EPQVYTLPPSR	1286.674	25.16		
150-212	DYFPEPVTVSWNSGALTSVHTFP AVLQSSGLYSLSSVTVPSSSLGTQ TYIcNVNHK	6190.037	59.16			358-362	DELTK	605.314	8.94		
213-215	VDK	361.2081	0.00			363-372	NQVSLTcLVK	1162.613	6 28.24		
213-216	VDKR	516.3001	1.02			373-394	GFYPSDIAVEWESNGQPENNYK	2545.134	3 42.03		
217-220	VEPK	472.2765	2.74			395-411	TTPPVLDSDGSFFLYSK	1873.921	8 45.34		
221-224	ScDK	510.1864	2.55			412-416	LTVDK	575.3399	7.61		
225-250	THTcPPcPAPELLGGPSVFLFPPK	3622.281	47.31			417-418	SR	574.331	0.00		
251-257	DTLMISR	835.4342	21.18			419-441	WQQGNVFcSVMHEALHNHYT QK	2716.221	9 34.86		
258-276	TPEVTcVVVDVSHEDPEVK	2141.014	32.65			442-449	SLSLSPGK	788.4512	16.87		
277-290	FNWYVDGVEVHNAK	1677.802	34.34								

Automated mAb Quanfirmation using the Q Exactive and Pinpoint

- Sample preparation
- Qual/Quan scheme in 1 experiment (Quanfirmation)
- Detecting, verifying, and quantifying deamidated peptides (and other modified peptides)
- Automating the process through Pinpoint
- Determining the degree of modification per peptide
- QC analysis using Peptide Retention Time Calibration (PRTC) kit
- Conclusions

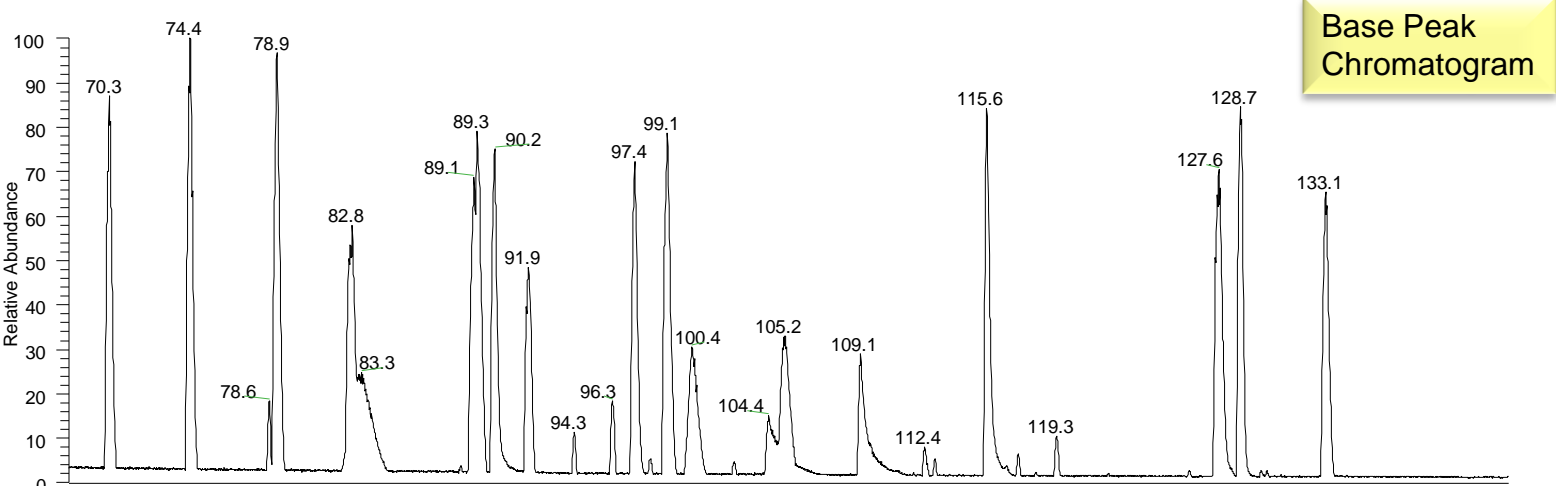
Experimental



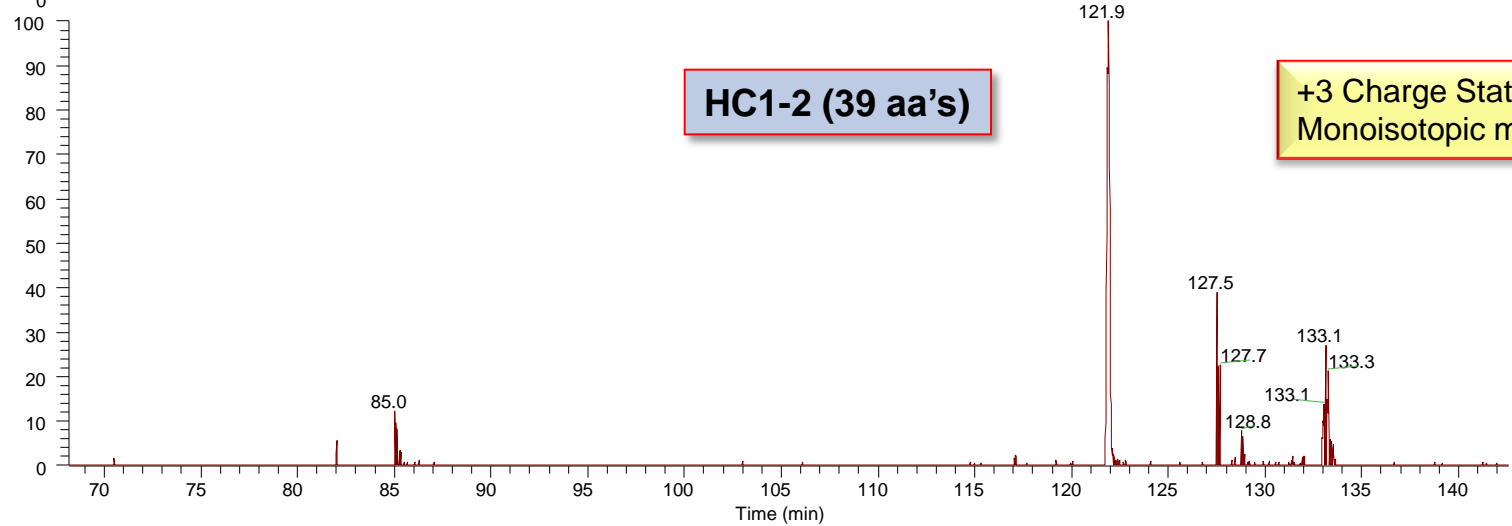
Peptide Retention Time Calibration (PRTC) Kit

#	Sequence	Observed Mass (z=2)	Hydrophobicity Factor (HF)
1	SSAAPPPPPR	493.7	7.57
2	GISNEGQNASIK	613.3	15.50
3	HVLTSIGEK	496.3	15.52
4	DIPVPKPK	451.3	17.65
5	IGDYAGIK	422.7	19.15
6	TASEFDSAIAQDK	695.8	25.88
7	SAAGAFGPESLR	586.8	25.24
8	ELGQSGVDTYLQTK	773.9	28.37
9	GLILVGGYGTR	558.3	32.18
10	GILFVGSGVSGGEEGAR	801.4	34.52
11	SFANQPLEVVYSK	745.4	34.96
12	LTILEELR	498.8	37.30
13	NGFILDGFPR	573.3	40.42
14	ELASGLSFPVGFK	680.4	41.19
15	LSSEAPALFQFDLK	787.4	46.66

HC1-2 Data Extraction – Pulling Information Out at the MS-Level



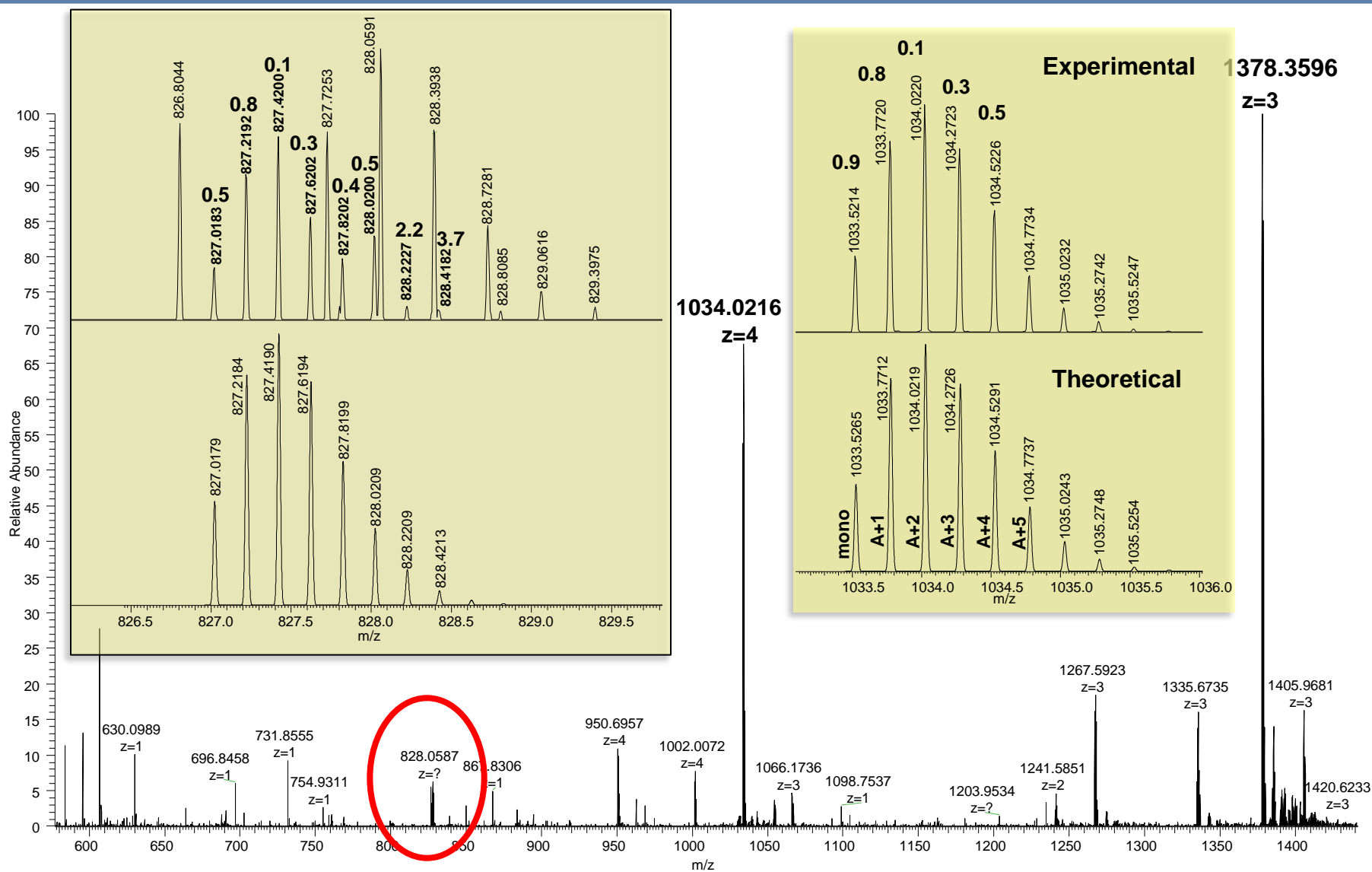
Base Peak Chromatogram



HC1-2 (39 aa's)

+3 Charge State
Monoisotopic m/z = 1378.0260

Orbitrap Data Acquisition Benefits for Large Peptides



Targeted Peptide Verification using HR/AM MS-Level Data

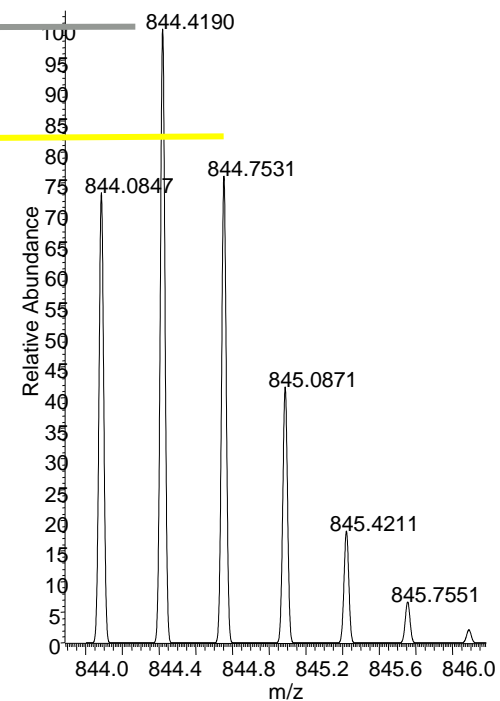
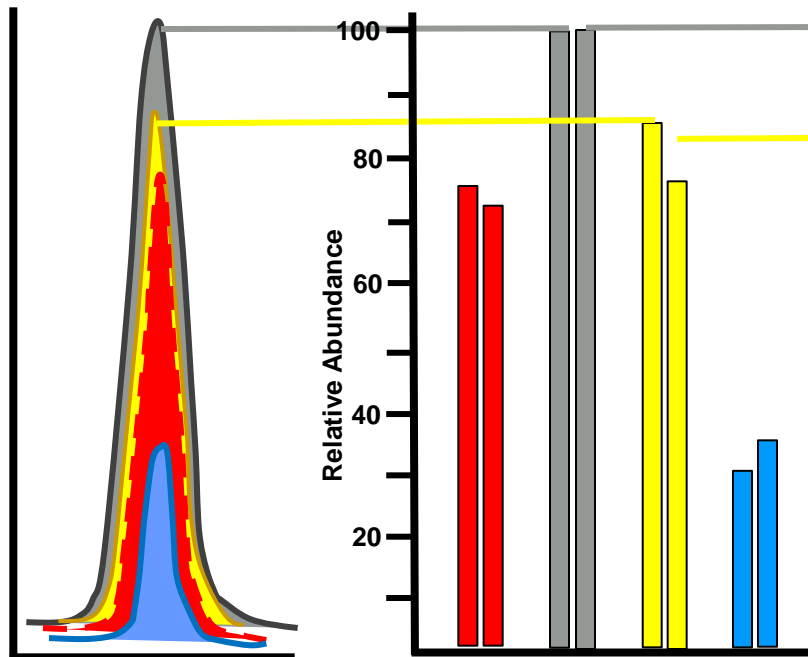
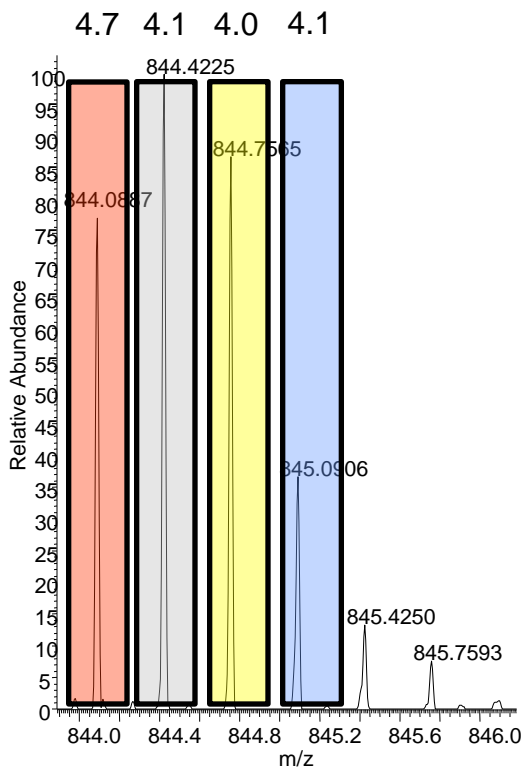
Experimental

SMGGKEDLIWELLNQAQEHFGK
 +3 CS = C₁₁₂H₁₇₅N₃₀O₃₅S₁

Theoretical

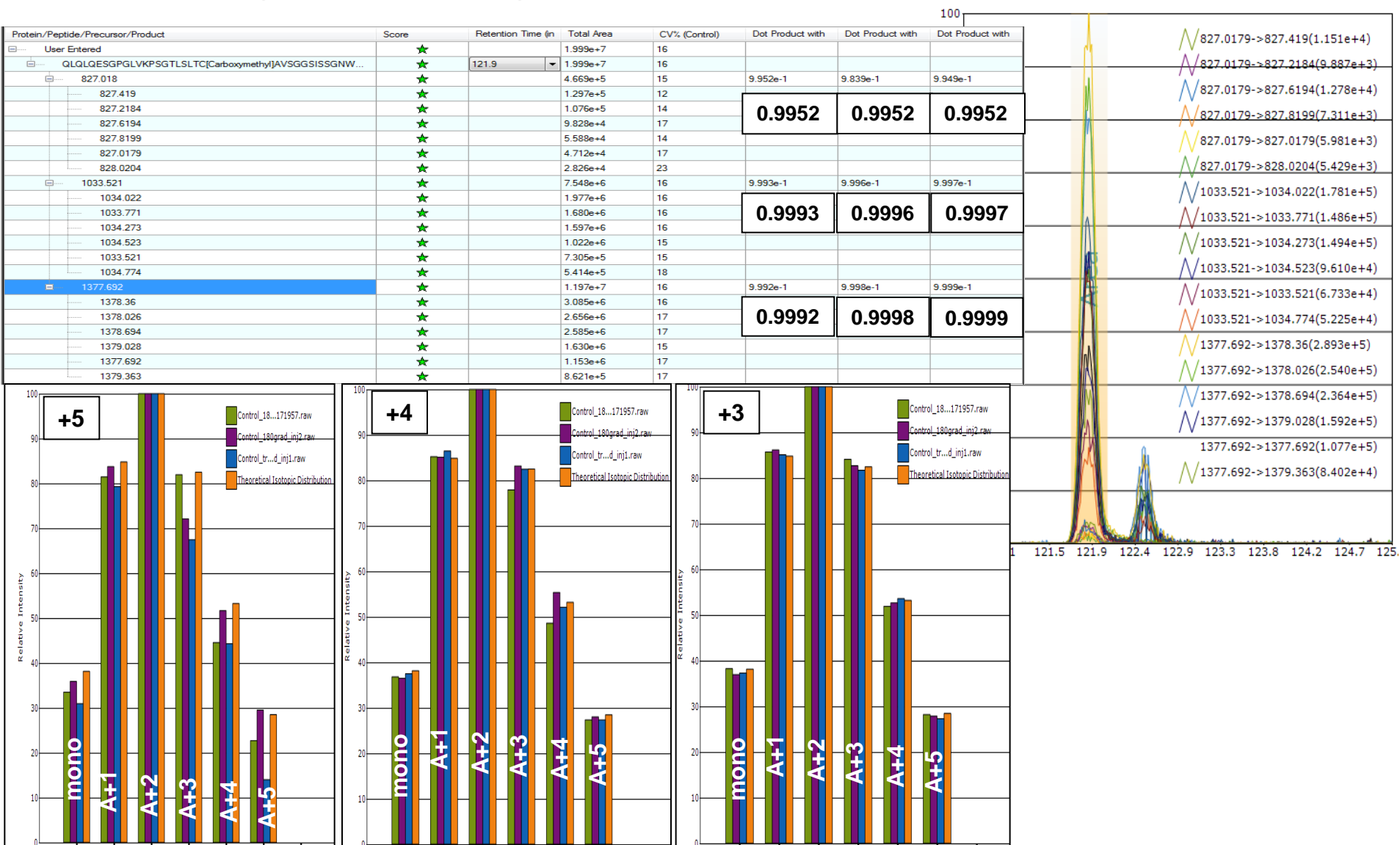
Mass Spectral Data

XICs

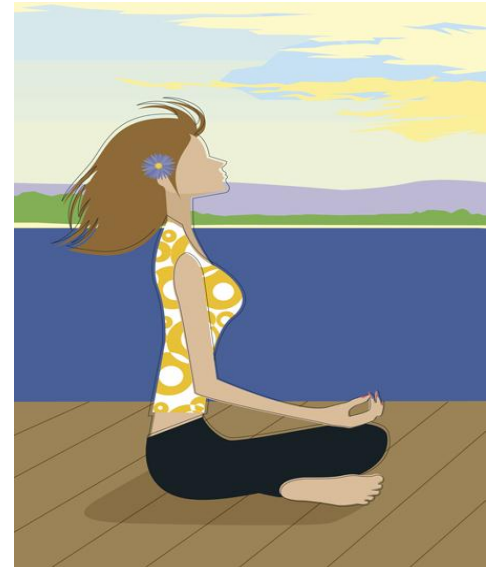
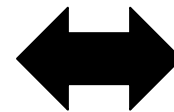


$$\rho = 1/n-1 \sum_{i=1}^n [(X_i - X_a)/s_x) * (Y_i - Y_a)/s_y)] \quad \longrightarrow \quad -1 \leq \rho \leq 1$$

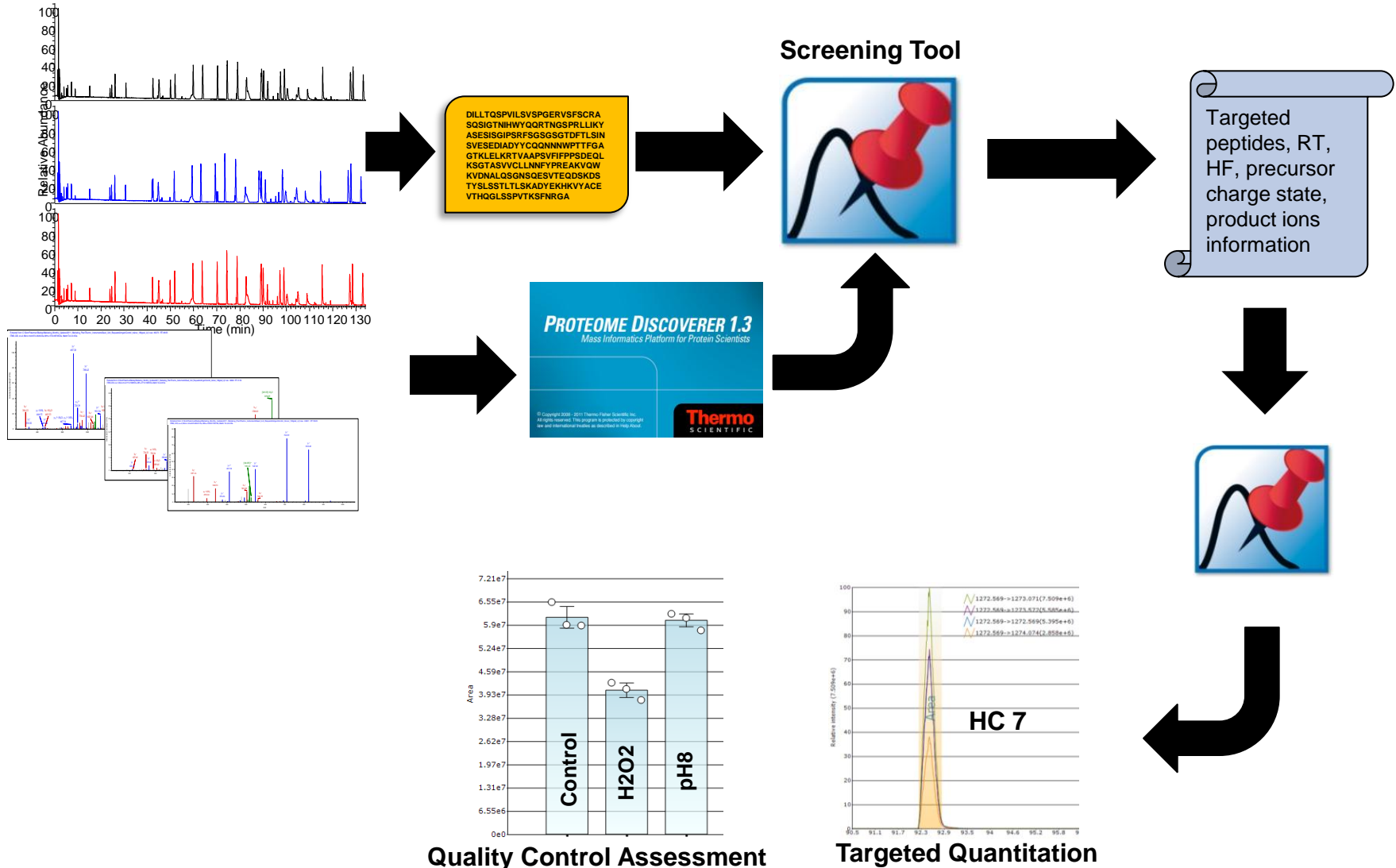
Handling Large Peptides in Pinpoint – Taking Advantage of Charge States and Isotopes



Intra- and Inter-sample Targeted Quantitation

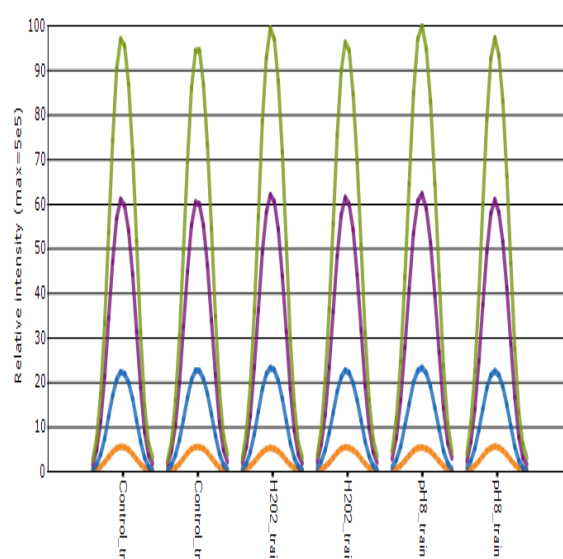
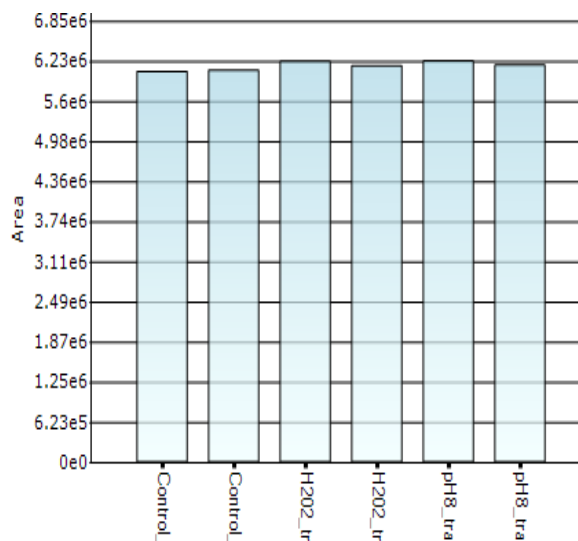


Data Processing Workflow



PRTC Kit Analysis

Protein/Peptide/Precursor/Product	Score	CV% (All files)	Retention Time (in)	Total File Area	Total File Area	Total File Area	Total File Area
PRTC kit	★	1		8.349e+7	8.218e+7	8.516e+7	8.373e+7
SSAAPPPPPR[HeavyR]	★	2	15.55	3.170e+6	3.166e+6	3.273e+6	3.333e+6
GISNEGQNASIK[HeavyK]	★	2	16.81	6.216e+6	6.272e+6	6.504e+6	6.385e+6
HVLTSIGEK[HeavyK]	★	1	19.59	7.255e+6	7.325e+6	7.294e+6	7.126e+6
DIPVPKPK[HeavyK]	★	1	20.96	3.912e+6	3.886e+6	4.002e+6	3.923e+6
IGDYAGIK[HeavyK]	★	1	21.37	3.778e+6	3.739e+6	3.865e+6	3.764e+6
TASEFDSAIAQDK[HeavyK]	★	1	23.33	6.064e+6	6.084e+6	6.219e+6	6.154e+6
SAAGAFGPESLR[HeavyR]	★	2	25.23	5.561e+6	5.404e+6	5.601e+6	5.488e+6
ELGQSGVDTYLQTK[HeavyK]	★	1	27.89	3.823e+5	3.696e+5	3.838e+5	3.788e+5
GLILVGGYGTR[HeavyR]	★	3	31.1	4.012e+6	3.942e+6	4.080e+6	3.958e+6
GILFVSGVSGGEEGAR[HeavyR]	★	2	31.41	6.941e+6	6.878e+6	7.205e+6	6.946e+6
SFANQPLEVVYSK[HeavyK]	★	1	31	1.039e+7	1.008e+7	1.052e+7	1.034e+7
LTILEELR[HeavyR]	★	4	34.43	5.198e+6	4.871e+6	5.290e+6	4.803e+6
NGFILDGFPR[HeavyR]	★	7	30.36	2.335e+6	2.179e+6	2.346e+6	2.622e+6
ELASGLSFPVGFK[HeavyK]	★	1	37.13	1.039e+7	1.039e+7	1.060e+7	1.046e+7
LSSEAPALFQFDLK[HeavyK]	★	2	39.08	7.887e+6	7.590e+6	7.973e+6	8.048e+6



Filename	Score	Total File Area	File Retention Time	Peak Start	Peak End	FWHM
Control_tr...d...	★	6.064e+6	23.389	23.28	23.50	0.12
Control_tr...d...	★	6.084e+6	23.304	23.19	23.42	0.13
H202_train.....	★	6.219e+6	23.3325	23.22	23.44	0.12
H202_train.....	★	6.154e+6	23.272	23.16	23.38	0.13
pH8_train.....	★	6.226e+6	23.3245	23.22	23.43	0.12
pH8_train.....	★	6.164e+6	23.293	23.18	23.41	0.12

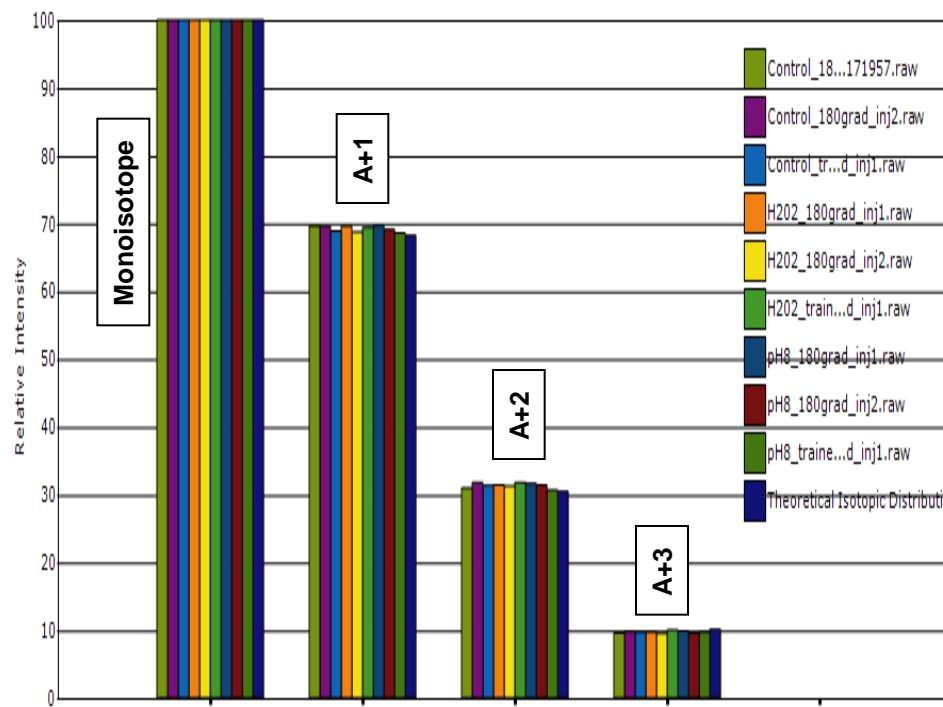
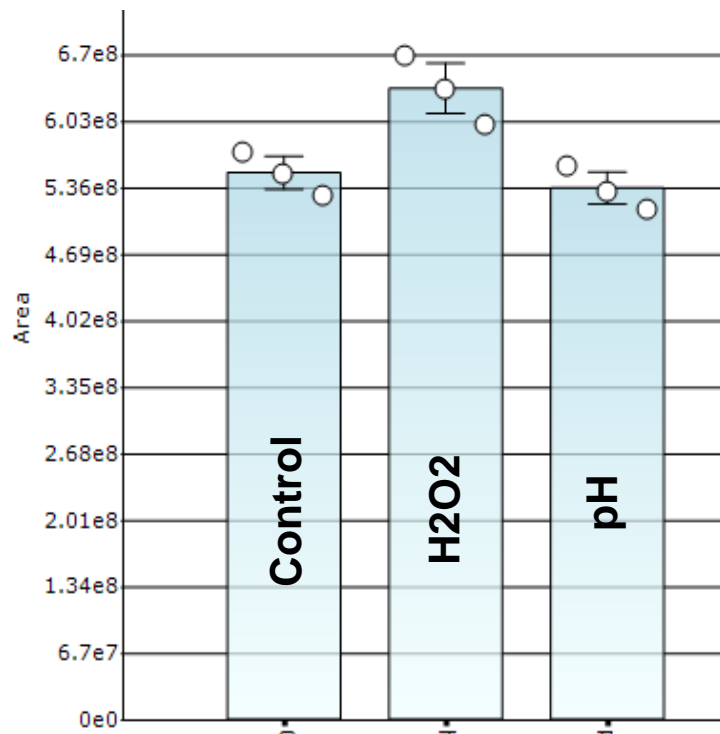
Heavy Chain Analysis

Annotation	From-To	Peptide Facets	RT (min)	AUC Control	AUC H2O2	AUC pH8	CV% Control	CV% H2O2	CV% 2 pH8	AUC Ratio H2O2	AUC Ratio2 pH
HC1	1-39	N-terminus, 3Q, 1N, 1C	121.94	1.67E+07	1.73E+07	4.18E+06	16	18	13	1.04	0.25
HC2	40-44	1Q	4.87	2.26E+07	3.00E+07	2.48E+07	17	20	24	1.33	1.10
HC3	45-65	2N	89.69	9.43E+08	1.09E+09	8.55E+08	3	5	3	1.16	0.91
HC4-5	66-74	Missed cleavage	27.48	1.64E+07	1.99E+07	1.61E+07	5	8	8	1.21	0.98
HC7	77-98	1Q, 2N, 1C	104.92	1.82E+08	1.74E+08	1.80E+08	6	23	6	0.96	0.98
HC8	99-117	1N, 1Q	88.43	3.03E+08	3.46E+08	3.04E+08	3	6	3	1.14	1.00
HC9	118-129	1C	79.49	5.51E+08	6.36E+08	5.36E+08	3	4	3	1.15	0.97
HC10	130-143	1C	70.66	4.93E+08	5.50E+08	4.71E+08	3	5	5	1.12	0.96
HC11	144-206	4N, 1C, 2Q	127.2	2.08e09	2.38E+09	2.02E+09	5	8	7	1.14	0.97
HC12	207-209	Small peptide	2.35	2.89E+07	3.67E+07	3.05E+07	5	9	11	1.27	1.06
HC12-13	207-213	Missed cleavage	52.52	2.07E+07	2.34E+07	2.16E+07	6	1	5	1.13	1.04
HC13	210-213	Small peptide	5.22	4.39E+07	5.58E+07	4.78E+07	6	6	9	1.27	1.09
HC14-15	214-240	2 Missed cleavages, 4C	99.86	7.15E+08	8.11E+08	6.45E+08	2	4	4	1.13	0.90
HC15	215-240	1 Missed cleavage, 4C	104.84	4.99E+08	5.69E+08	4.50E+08	3	5	4	1.14	0.90
HC16	241-247	Met	50.49	1.83E+08	3.88E+07	1.76E+08	3	6	3	0.21	0.96
HC17	248-280	1Q, 1N, 1C	109.7	6.18E+08	6.96E+08	6.11E+08	5	6	5	1.13	0.99
HC19	285-293	1Q, 1N	51.49	2.18E+06	2.65E+06	1.58E+06	4	4	4	1.21	0.73
HC20	294-309	1Q, 1N	99.7	1.16E+09	1.32E+09	1.15E+09	3	5	5	1.14	0.99
HC21	310-312	EYK	4.03	3.75E+07	4.65E+07	4.10E+07	2	6	7	1.24	1.09
HC22-23	313-318	1N, 1C	6.78	4.04E+06	5.15E+06	4.29E+06	3	10	11	1.28	1.06
HC23	315-318	1N	2.1	4.17E+07	5.38E+07	4.42E+07	10	11	12	1.29	1.06
HC24	319-326		51.44	2.21E+08	2.51E+08	2.19E+08	4	5	3	1.14	0.99
HC25	327-330		5.75	5.84E+07	7.26E+07	6.32E+07	2	8	9	1.24	1.08
HC26-27	331-332	Missed cleavage, 1Q	3.49	1.50E+07	1.86E+07	1.55E+07	2	10	6	1.24	1.03
HC28	337-347	1Q	60.41	4.85E+08	5.36E+08	4.83E+08	4	5	5	1.11	1.00
HC29	348-352	Met	8.98	4.55E+07	2.68E+07	4.61E+07	3	3	4	0.59	1.01
HC30	353-362	1Q, 1N, 1C	74.88	4.93E+08	5.69E+08	4.98E+08	4	5	5	1.15	1.01
HC31	363-384	1Q, 3N	92.46	3.44E+08	3.87E+08	2.75E+08	4	5	5	1.12	0.80
HC32	385-401	Met	96.84	5.34E+08	1.51E+08	5.36E+08	4	4	4	0.28	1.00
HC33	402-406		24.09	7.37E+07	8.90E+07	7.64E+07	2	7	6	1.21	1.04
HC35	409-431	Met, 3Q, 2N	83.52	1.00E+09	5.17E+08	9.88E+08	3	5	4	0.52	0.99
HC36	432-439	C-terminus	45.99	2.39E+06	2.81E+06	2.50E+06	2	3	3	1.18	1.05

Stress-Free Peptides – Confirming Stable Peptides

Protein/Peptide/Precursor/Product	Score	Ratio of Groups	Retention Time (in)	Total Area	Total Area (H2O2)	Total Area (pH8)	CV% (Control)	CV% (H2O2)	CV% (pH8)
User Entered Control_180grad_inj1_1105131...	★	1.0 : 1.1 : 9.7e-1		7.855e+9	8.951e+9	7.604e+9	5	6	3
GLEWIGEISHSGTTNYPNLSK	★	1.0 : 1.2 : 9.1e-1	89.69	9.427e+8	1.091e+9	8.551e+8	3	5	3
NQFSLELTSVTAADTAVYYC[Carboxyme...	★	1.0 : 9.6e-1 : 9.8...	104.92	1.824e+8	1.742e+8	1.795e+8	6	23	6
NFDYWGGGLTVTSSASTK	★	1.0 : 1.1 : 1.0	88.43	3.032e+8	3.460e+8	3.038e+8	3	6	3
GPSVFLAPC[Carboxymethyl]SR	★	1.0 : 1.2 : 9.7e-1	79.49	5.509e+8	6.358e+8	5.355e+8	3	4	3
644.821	★	1.0 : 1.2 : 9.7e-1		5.509e+8	6.358e+8	5.355e+8	3	4	3
644.8214	★	1.0 : 1.2 : 9.7e-1		2.609e+8	3.013e+8	2.538e+8	3	4	3
645.3228	★	1.0 : 1.1 : 9.7e-1		1.818e+8	2.091e+8	1.764e+8	3	4	3
645.8234	★	1.0 : 1.2 : 9.7e-1		8.270e+7	9.576e+7	8.056e+7	3	4	4
646.324	★	1.0 : 1.2 : 9.7e-1		2.543e+7	2.959e+7	2.480e+7	3	4	3

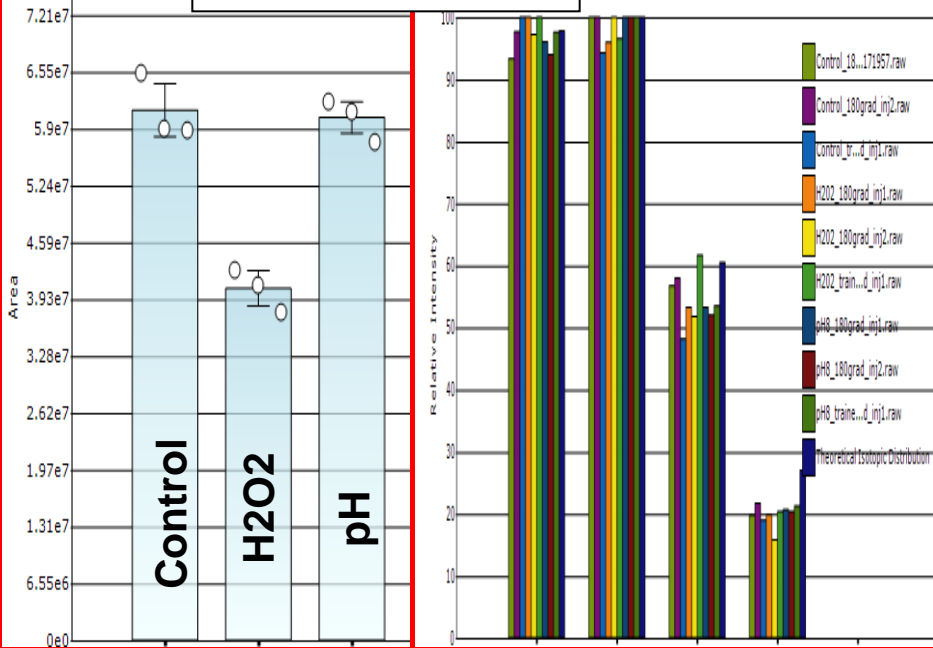
Filename	Score	File Retention Time	FWHM	Dot Product with Theoretical Isotopic Envelope
Control_18...171957r...	★	79.5545	0.32	9.998e-1
Control_180grad_inj2...	★	78.97701	0.35	9.997e-1
Control_tr...d_inj1.raw	★	78.39149	0.32	9.997e-1
H202_180grad_inj1.raw	★	78.93401	0.35	9.998e-1
H202_180grad_inj2.raw	★	78.14751	0.35	9.998e-1
H202_train...d_inj1.raw	★	78.2505	0.32	9.998e-1
pH8_180grad_inj1.raw	★	79.239	0.31	9.997e-1
pH8_180grad_inj2.raw	★	78.776	0.31	9.998e-1
pH8_train...d_inj1.raw	★	78.4555	0.31	9.998e-1



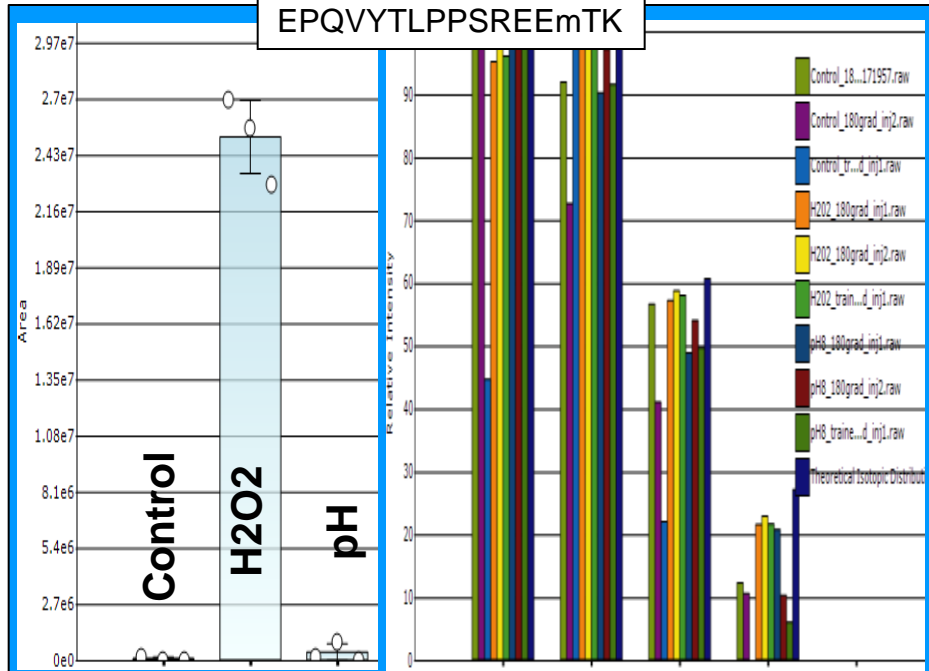
Pinpoint Data Processing Results – Met (Ox)

Protein/Peptide/Precursor/Product	Score	Ratio of Groups	Retention Time (in)	Total Area	Total Area (H2O2)	Total Area (pH8)	CV% (Control)	CV% (H2O2)	CV% (pH8)	Dot	Dot	Dot	Dot	Dot	Dot	Dot	Dot
VSVLTVVHQDWLNGK	★	1.0 : 1.1 : 9.9e-1	99.7	1.158e+9	1.320e+9	1.150e+9	3	5	5								
C[Carboxymethyl]KVSNK	★	1.0 : 1.3 : 1.1	6.78	4.036e+6	5.152e+6	4.291e+6	3	10	11								
TKGQPR	★	1.0 : 1.2 : 1.0	3.49	1.502e+7	1.863e+7	1.552e+7	2	10	6								
EPQVYTLPPSREEMTK	★	1.0 : 6.6e-1 : 9.9...	64.72	6.117e+7	4.061e+7	6.030e+7	5	5	3								
635.652	★	1.0 : 6.7e-1 : 9.9...		4.451e+7	2.967e+7	4.421e+7	5	5	4	9.999e-1	1.000e+0	9.997e-1	9.998e-1	9.997e-1	9.995e-1	9.999e-1	9.996e-1
952.975	★	1.0 : 6.6e-1 : 9.7...		1.666e+7	1.093e+7	1.609e+7	6	5	3	9.991e-1	9.985e-1	9.983e-1	9.976e-1	9.987e-1	9.995e-1	9.995e-1	9.995e-1
EPQVYTLPPSREEM[Oxid]TK	★	1.0 : 330.2 : 5.1	56.95	1.022e+5	3.373e+7	5.185e+5	76	7	103								
640.984	★	1.0 : 292.0 : 4.6		8.622e+4	2.518e+7	3.980e+5	79	7	101	9.942e-1	9.845e-1	9.442e-1	1.000e+0	9.999e-1	9.999e-1	9.990e-1	9.986e-1
960.972	★	1.0 : 536.4 : 7.5		1.594e+4	8.552e+6	1.204e+5	60	6	108	9.792e-1	9.611e-1	9.794e-1	9.986e-1	9.987e-1	9.985e-1	9.912e-1	9.909e-1

EPQVYTLPPSREEMTK



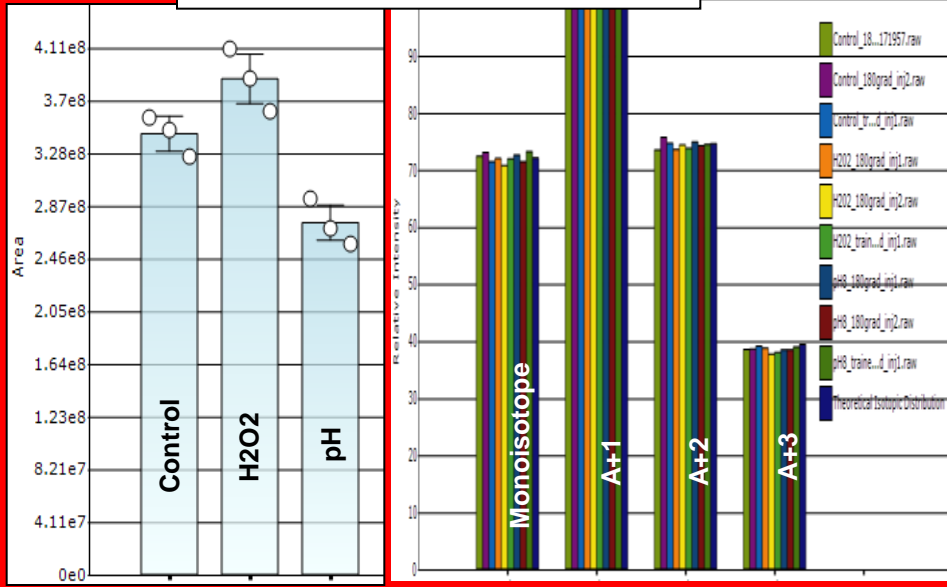
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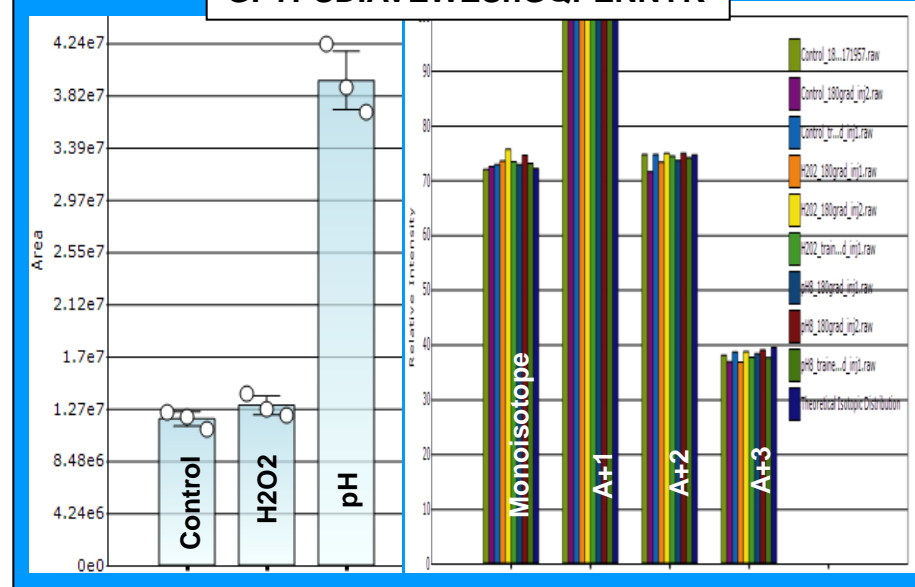
Pinpoint Data Processing Results - Deamidation

Protein/Peptide/Precursor/Product	Score	Ratio of Groups	Retention Time (in)	Total Area	Total Area (H2O2)	Total Area (pH8)	CV% (Control)	CV% (H2O2)	CV% (pH8)	Dot	Dot	Dot	Dot
NQVSLTC[Carboxymethyl]LVK	★	1.0 : 1.2 : 1.0	74.88	4.930e+8	5.685e+8	4.983e+8	4	5	5				
GFYPSDIAVEWESNGQPENNYK	★	1.0 : 1.1 : 8.0e-1	92.46	3.439e+8	3.866e+8	2.746e+8	4	5	5				
1272.569	★	1.0 : 1.1 : 8.0e-1		3.439e+8	3.866e+8	2.746e+8	4	5	5	1.000e+0	9.999e-1	1.000e+0	1.000e+0
1273.071 A+1	★	1.0 : 1.1 : 8.0e-1		1.208e+8	1.362e+8	9.624e+7	4	5	5				
1273.572 A+2	★	1.0 : 1.1 : 8.0e-1		8.953e+7	1.008e+8	7.194e+7	4	5	5				
1272.569 Mono	★	1.0 : 1.1 : 8.0e-1		8.692e+7	9.750e+7	6.915e+7	4	5	5				
1274.074 A+3	★	1.0 : 1.1 : 8.0e-1		4.667e+7	5.215e+7	3.723e+7	4	5	5				
GFYPSDIAVEWESN[Deamid]GQPENNYK	★	1.0 : 1.1 : 3.3	91	1.191e+7	1.301e+7	3.940e+7	5	6	6				
1273.061	★	1.0 : 1.1 : 3.3		1.191e+7	1.301e+7	3.940e+7	5	6	6	9.999e-1	9.994e-1	9.997e-1	9.999e-1
1273.563 A+1	★	1.0 : 1.1 : 3.3		4.213e+6	4.586e+6	1.382e+7	5	7	6				
1274.064 A+2	★	1.0 : 1.1 : 3.3		3.088e+6	3.390e+6	1.025e+7	5	6	6				
1273.061 Mono	★	1.0 : 1.1 : 3.3		3.035e+6	3.321e+6	1.005e+7	5	5	6				
1274.566 A+3	★	1.0 : 1.1 : 3.4		1.572e+6	1.718e+6	5.287e+6	5	5	6				

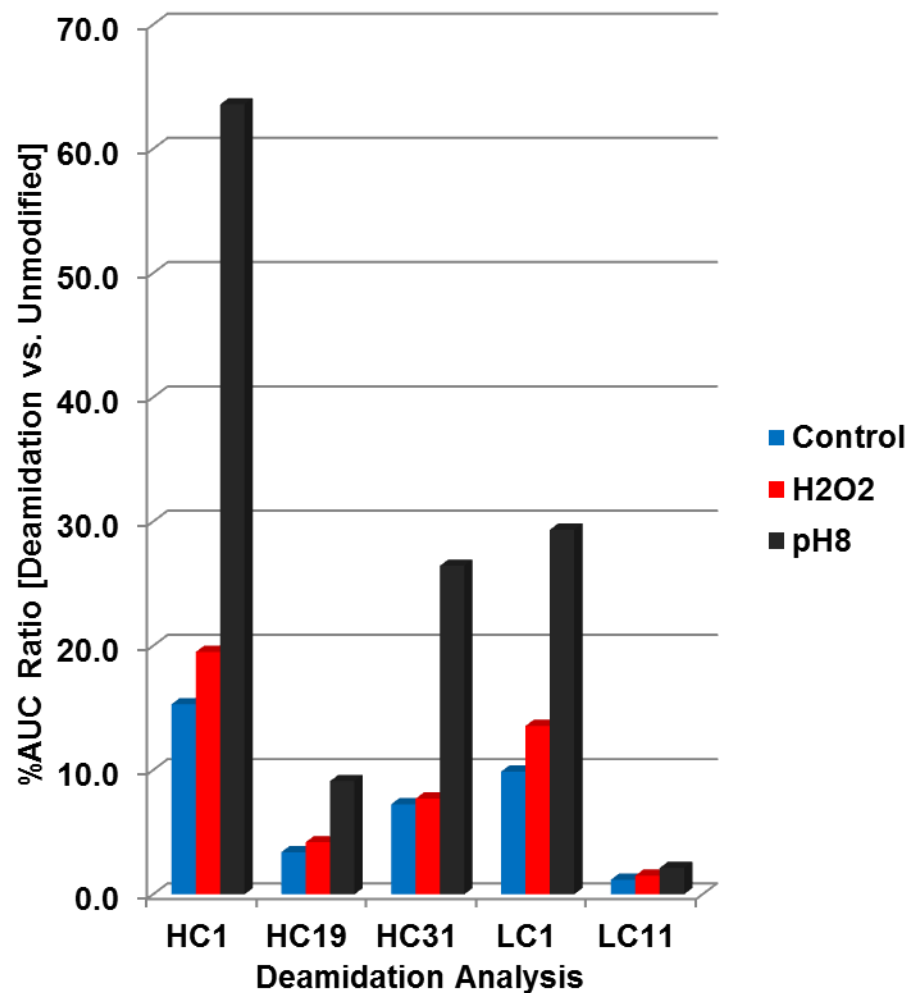
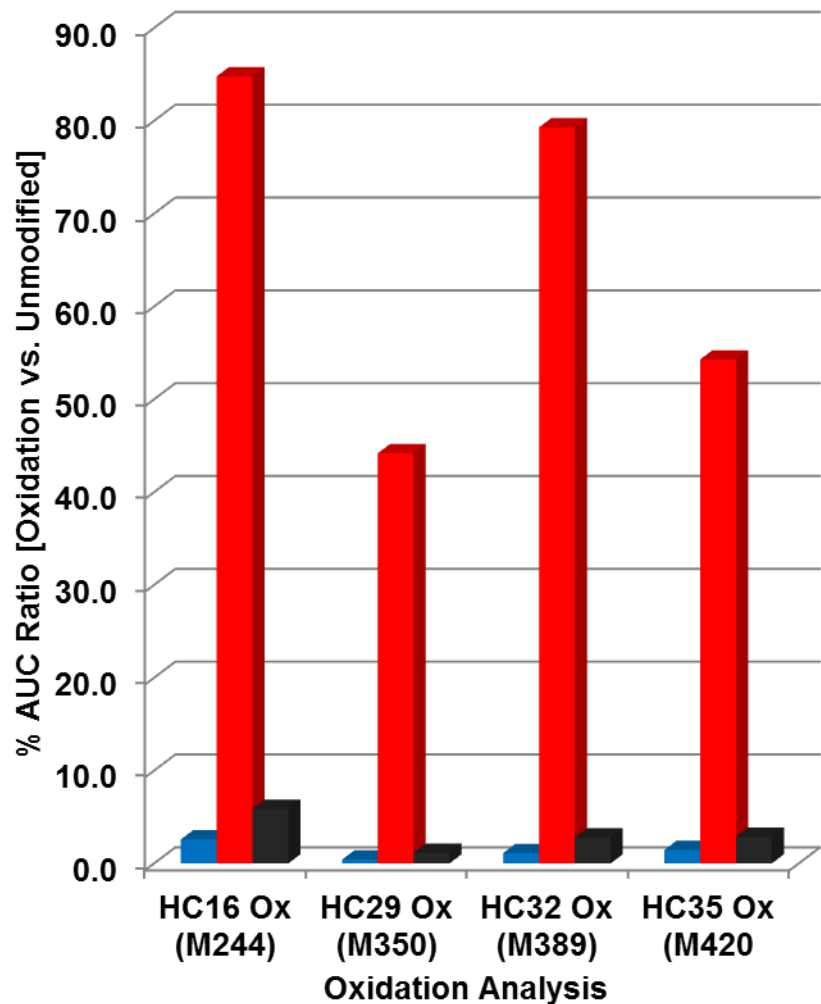
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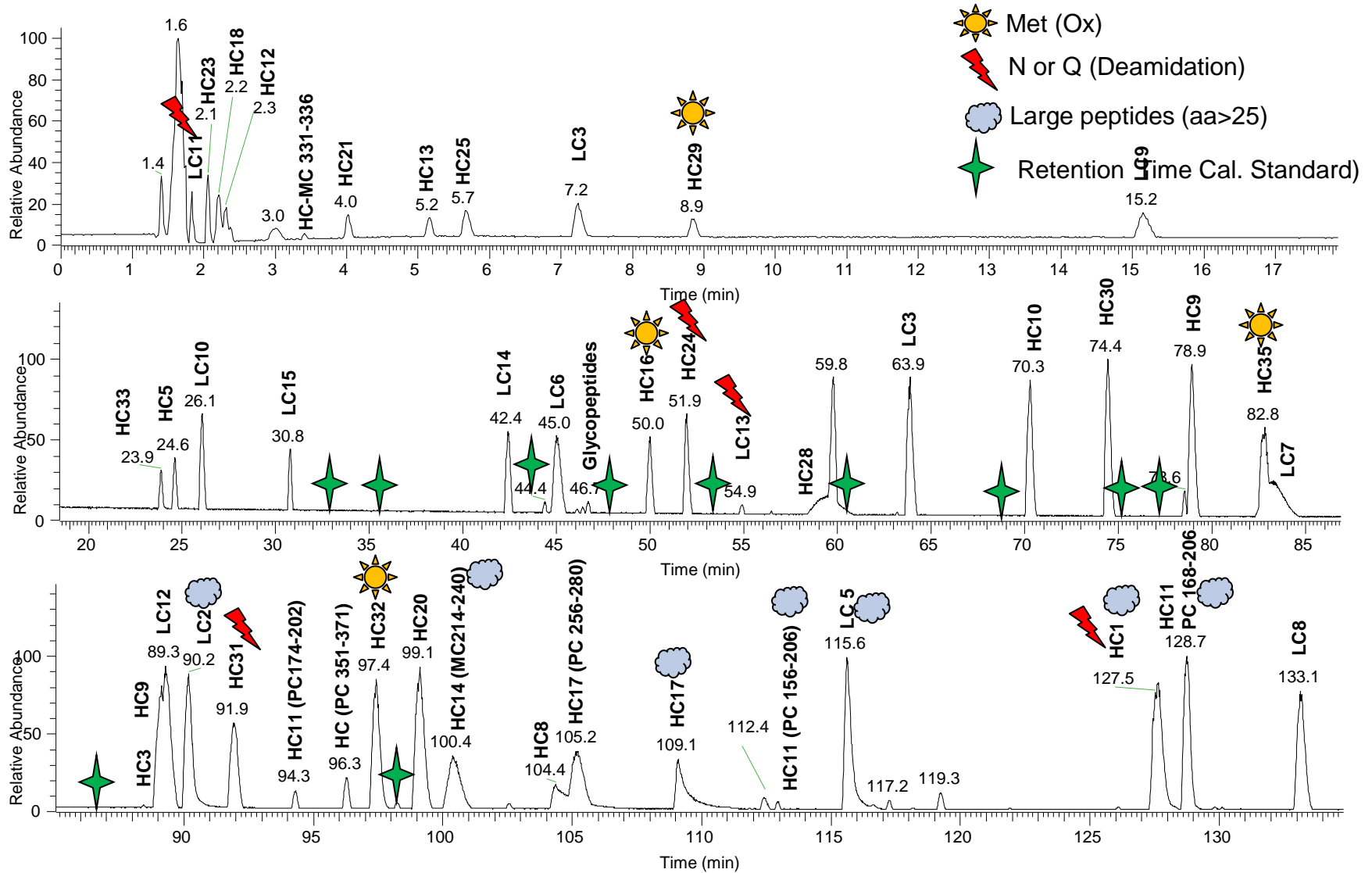
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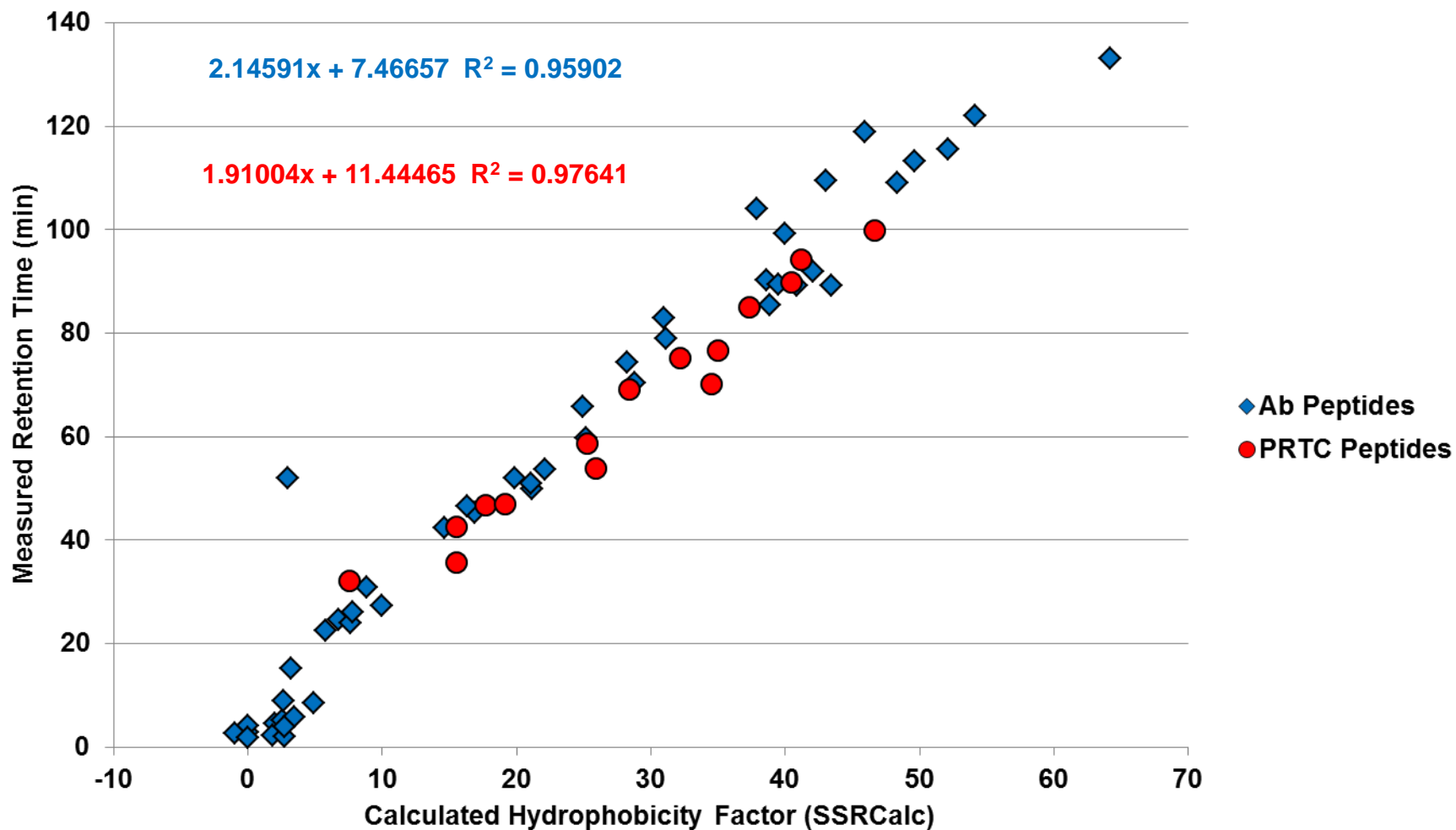
PTM Analysis – Intra- and Inter-Sample Analysis



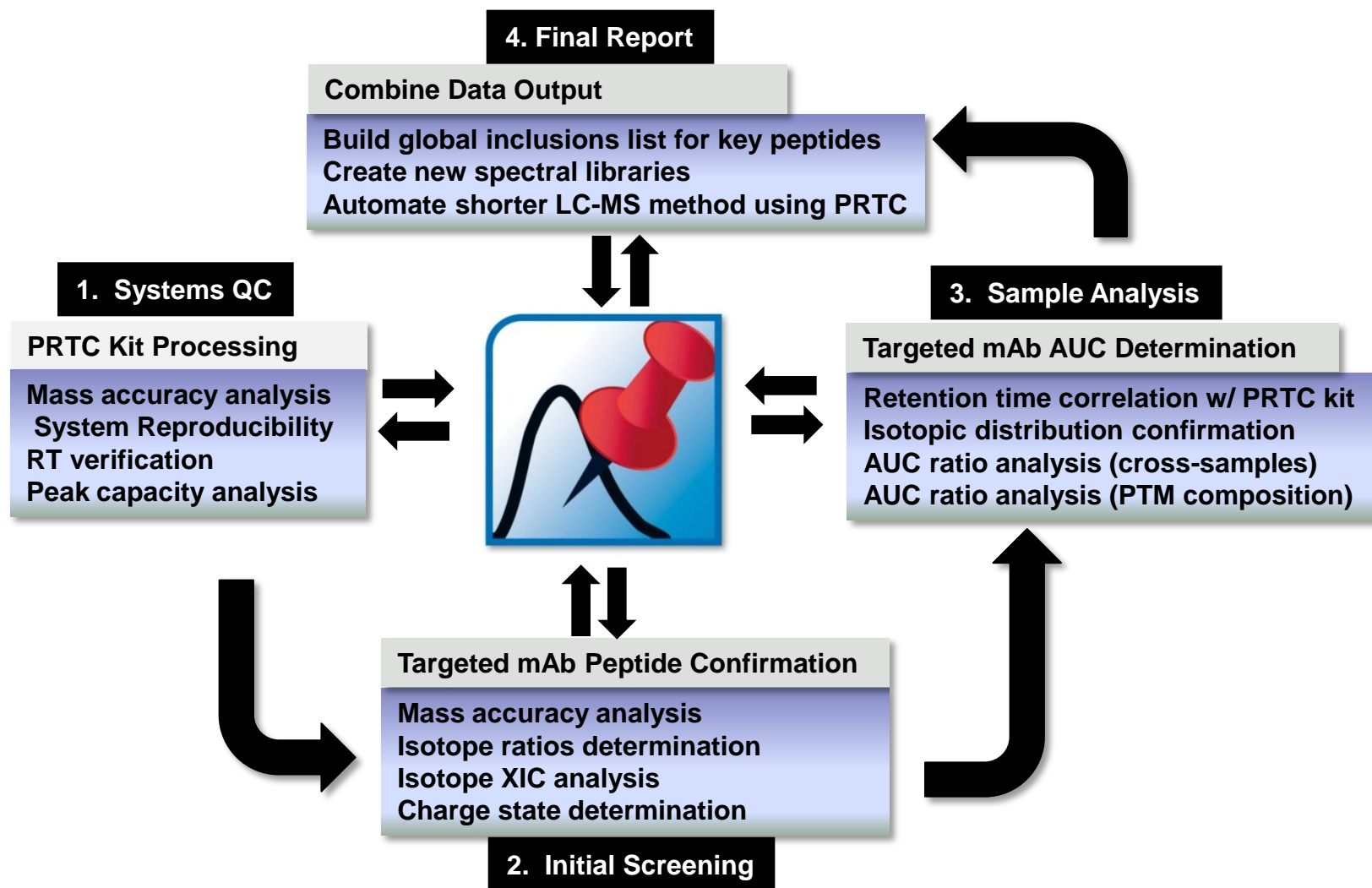
Peptide Mapping Results for the 180 min Method



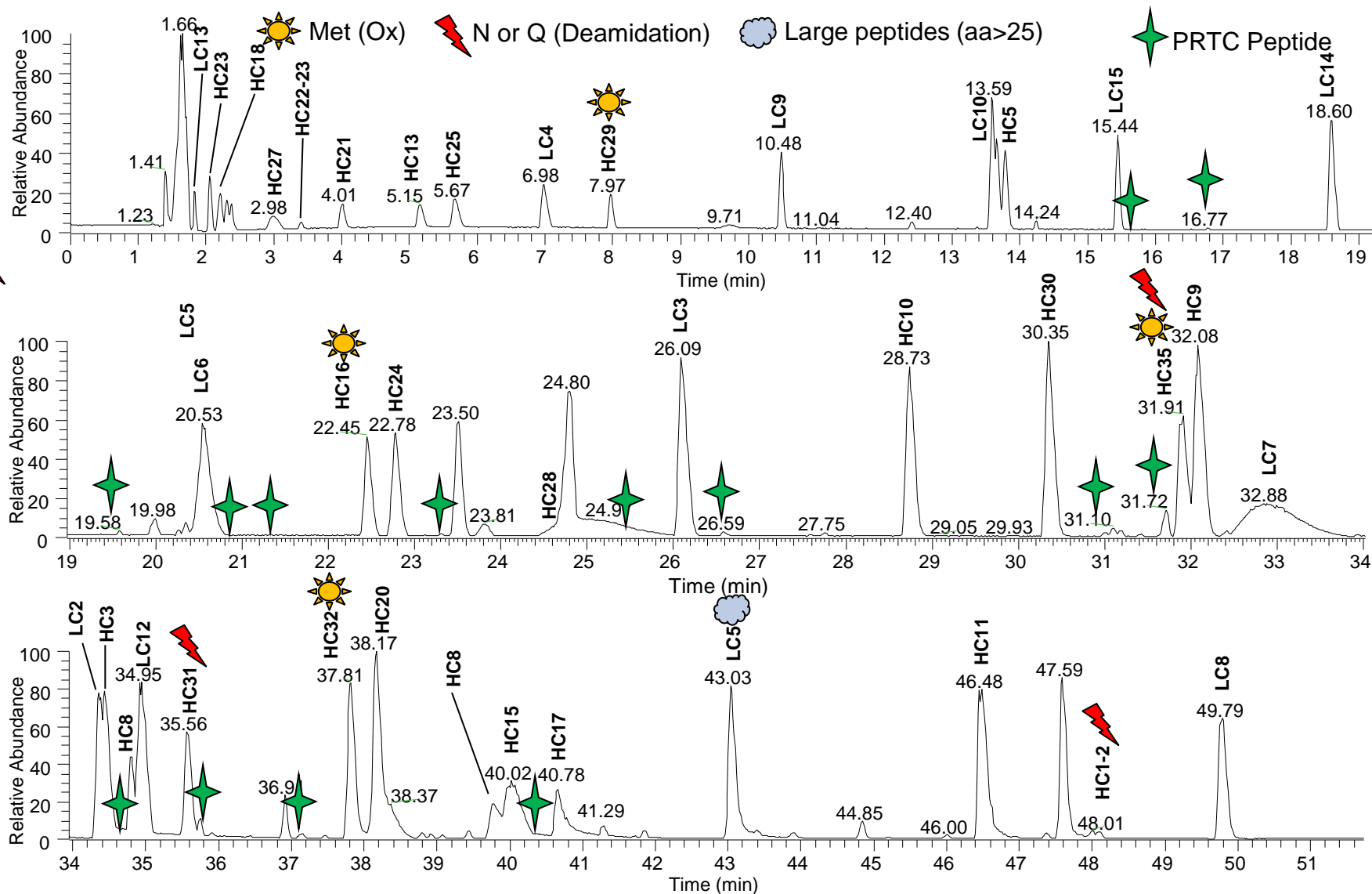
Retention Time Analysis – Comparison with Internal Standards



The Workflow



60 Minute Labeling



Conclusions

Targeted quantification (=confirmation + quantitation) analysis of mAb using the Q Exactive mass spectrometer maximizes characterization, verification, and quantification in 1 experiment.

- The high resolution (>30,000)/accurate mass data acquired over the entire mass range enabled automated and efficient data extraction for quantitation.
- The use of the PRTC kit enables LC-MS QC analysis as well as the potential for normalization across samples and technical replicates.
- Production of control and stressed samples produce a routine method for identifying active sites (modification sites) that can be incorporated into a tightly controlled method for production analysis.
- Q Exactive quantification routine enables the user to reprocess data if additional sequence features are needed for production analysis or characterization instead of having to re-acquire targeted data.
- Quantified 99.1% of the heavy chain and 100% of the light chain using the method and identified, quantified, and determined % composition of 9 different modified peptides.
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