Supporting information

Table S1: Nano LC-MS/MS analysis of all bands extracted from TMBZ gels with KF707 W.T. and mutant strains grown in MSM medium with glucose or biphenyl as the sole carbon source. The Uniprot accession number, amino acids coverage, significant peptide modifications and xCorr are listed.

17 kDa	**Accession**	**Description and % coverage**		
	L8MT74	Cytochrome c_4 OS=*Pseudomonas pseudoalcaligenes* KF707 = NBRC 110670 GN=ppKF707C_770 PE=3 SV=1 – [L8MT74_PSEPS] – NCBI Cyt c_4 BAU71765 – **31.36%**		
	Sequence		Modification	XCorr
	DIEAVSSYIQGLH AAVcGAcHGPDGNSAAPNFPK TVLEmTGLLTNmSDQDmADLAAYFASQK		C4 – C7 M5 – M12 – M17	4.76 4.31 4.27
22 kDa	Accession	Description and % coverage		
	L8MRV9	cbb_3-type cytochrome *c* oxidase subunit OS=*Pseudomonas pseudoalcaligenes* KF707 = NBRC 110670 GN=ppKF707_18660 PE=4 SV=1 – [L8MRV9_PSEPS] _NCBI CcoO1 BAU7355 - **4 4.39%**		
	Sequence		Modification	XCorr
	YGHYSVAGESVWDHPFLWGSK TEmDALVAYLQVLGTAIK mPSYPWLVENK TALELEGR GKTEmDALVAYLQVLGTAIK KLGVPYTDDDIAGAR LGVPYTDDDIAGAR YSDDWHR		M3 M1 M5	6.88 4.94 3.76 2.96 2.95 2.86 2.71 2.03
22 kDa	Accession	Description and % coverage		
	L8MLE9	cbb_3-type cytochrome *c* oxidase subunit OS=*Pseudomonas pseudoalcaligenes* KF707 = NBRC 110670 GN=ppKF707_18700 PE=4 SV=1 – [L8MLE8_PSEPS] _NCBI CcoO2 BAU73558 - **44.33%**		
	Sequence		Modification	XCorr

	YGHYSVAGESVWDHPFLWGSK	M1	6.88
	ALQTLGVPYSEDDVAGAQAAVK	M3	5.44
	mPAYPWLVENTLDGKDTAAK	M5	5.12
	TEmDALVAYLQVLGTAVK	M1	4.62
	GKTEmDALVAYLQVLGTAVK		4.37
	mPAYPWLVENTLDGK		3.07
	YSDDWHR		2.03

29 kDa	Accession	Description and % coverage
	L8M8C7	Ubiquinol cytochrome *c* oxidoreductase, cytochrome c_1 subunit OS=*Pseudomonas pseudoalcaligenes* KF707 NBRC 110670 GN=ppKF707_9870 PE=4 SV=1 – [L8M8C7_PSEPS] – NCBI Cyt c_1 BAU72675 - **37.07%**

Sequence	Modification	XCorr
DLGIPEELmmENLVFTGAK	M9 – M10	3.64
IQNLVTFLAYSALPVK	M7	3.28
VFPNVGmPNVLVSLQGR		3.03
TWFGAAPPDLTLVAR		2.89
VRGNDWLYSYLR		2.81
GNDWLYSYLR		2.71
LTEAEFDEK		2.12

34 kDa	Accession	Descriprion and % coverage
	L8MQP2	cbb_3-type cytochrome *c* oxidase subunit OS=*Pseudomonas pseudoalcaligenes* KF707 = NBRC 110670 GN=ppKF707C_18680 PE=3 SV=1 – [L8MQP2_PSEPS] – NCBI CcoP2 BAU73559 - **52.41%**

Sequence	Modification	XCorr
mAAmPAWGEILGDSGVR	M1 – M2	4.25
VHILAAYVYSLSHK	M3	3.69
LPEGAQADLEAGK	M2	3.65
EVDKAEQQYGPIFAR	M7	3.63
QDLAGLKLPEGAQADLEAGK	M1	3.39
GLFPGYADGWTQVAQWQR	M4	3.37
NGQmPAQLEYLGEDK	M16	3.27
GAmGFPNLTDGDWR		3.10
AEQQYGPIFAR		3.05
LPEGAQADLEAGKK		2.96
GPTDQTmGHAFDGIEEYDNPLPK		2.88
mAAMPAWGEILGDSGVR		2.77
WGGAPDAIR		2.58
YSAmSVEEVAK		2.46
QDLAGLKLPEGAQADLEAGKK		2.35
YSAMSVEEVAKDPSAmK		2.29

35 kDa	Accession	Description and % coverage

	L8MMN9	*cbb*$_3$-type cytochrome *c* oxidase subunit OS=*Pseudomonas pseudoalcaligenes* KF707 = NBRC 110670 GN=ppKF707C_18640 PE=3 SV=1 – [L8MMN9_PSEPS] – NCBI CcoP1 BAU73555 - **46,30%**	
	Sequence	Modification	XCorr
	VHLLAAYVYSLSQKPAAEGEAK		5.69
	GTPAmGAPNLTHPSAFIYGSSFAQLQQTIR		4.68
	QGVmPAQENILGNDK	M5	3.13
	GAYGFPNLTDNDWR	M4	2.70
	QESTEETVGHSDGIEEYDNPLPK	M4	2.58
	HGVmPAWGEVLGEQGVR	M4	2.46
	ADKQYGPIFAK		2.33
	YAAmPIEEVAKDPQALK		2.31
	QYGPIFAK		2.07
37 kDa	Accession	Descriprion and % coverage	
	L8MR96	Cytochrome *c* oxidase subunit OS=*Pseudomonas pseudoalcaligenes* KF707 = NBRC 110670 GN=ppKF707C_5503 PE=3 SV=1 – [L8MR96_PSEPS] – NCBI CoxII BAU71737 - **38,7%**	
	Sequence	Modification	XCorr
	YLGDQEFFSNLATPSEQIHNK		5.73
	ADHLNIVFHGKPGTSmAAFGK		5.43
	DEHYLLEVDQPLVVPVGTK		5.40
	ALTDKEWTLDELVAR		5.12
	ADHLNIVFHGKPGTSMAAFGK	M16	4.79
	APKDEHYLLEVDQPLVVPVGTK	C3 – C7 – M14	4.75
	KDAIPGFVNESWTR	M14	4.17
	LKELTDKEWTLDELVAR	M5	4.16
	GQcTELcGKDHGFmPIVVEAK	M10	4.10
	DHGFmPIVVEAK		3.74
	NAWGNNTGDmVTPK		3.23
	EVLALKQAESQ		3.00
	EWTLDELVAR		2.96
	GQcTELcGK		2.18

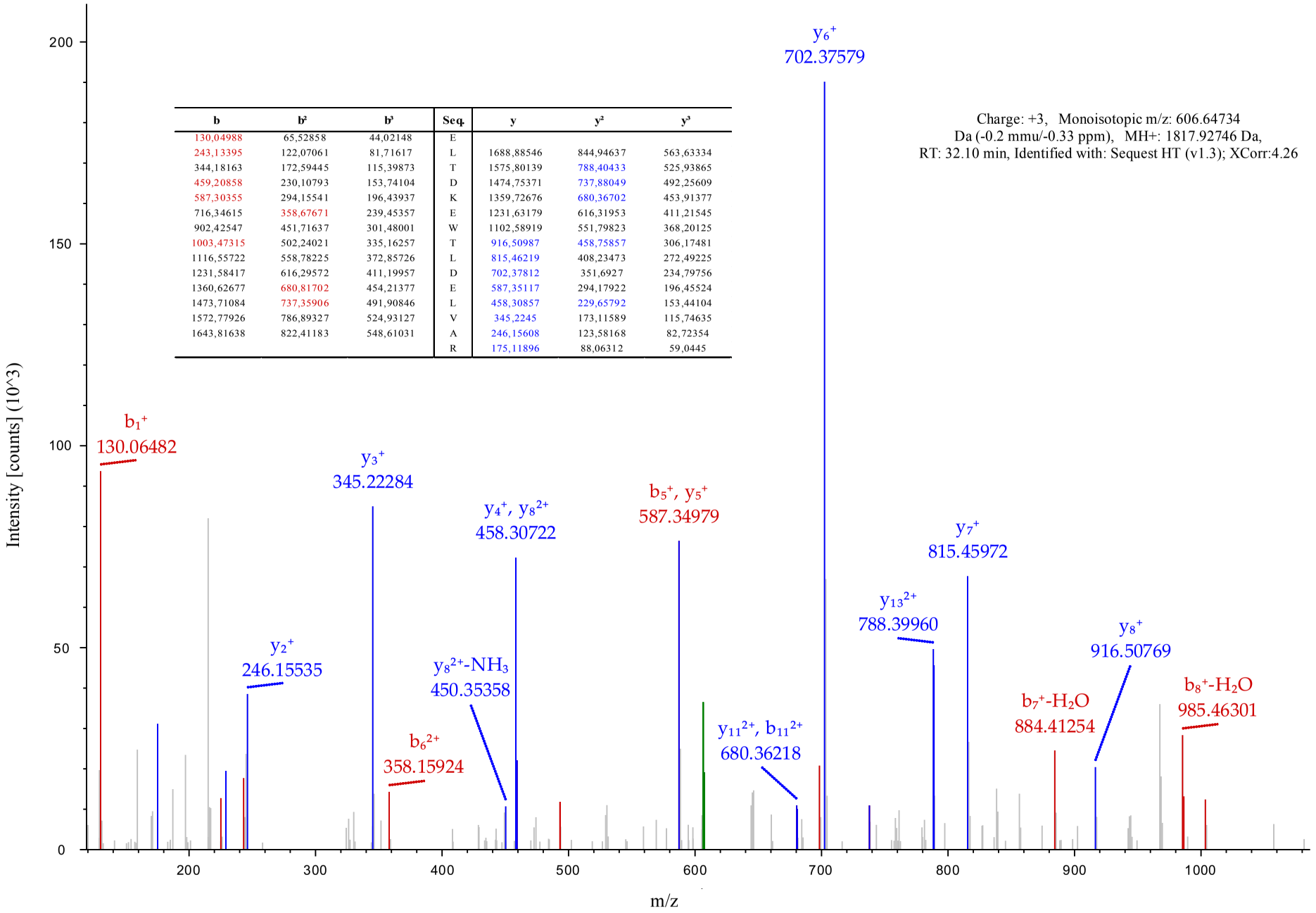
Figure S1: Nano *LC-MS/MS* fragmentation spectra found in the 37 kDa band, from cells grown with glucose (**A**) or with biphenyl (**B**), and belonging to CoxII subunit, Caa_3 SU II*c*.

A

ELTDKEWTLDELVAR

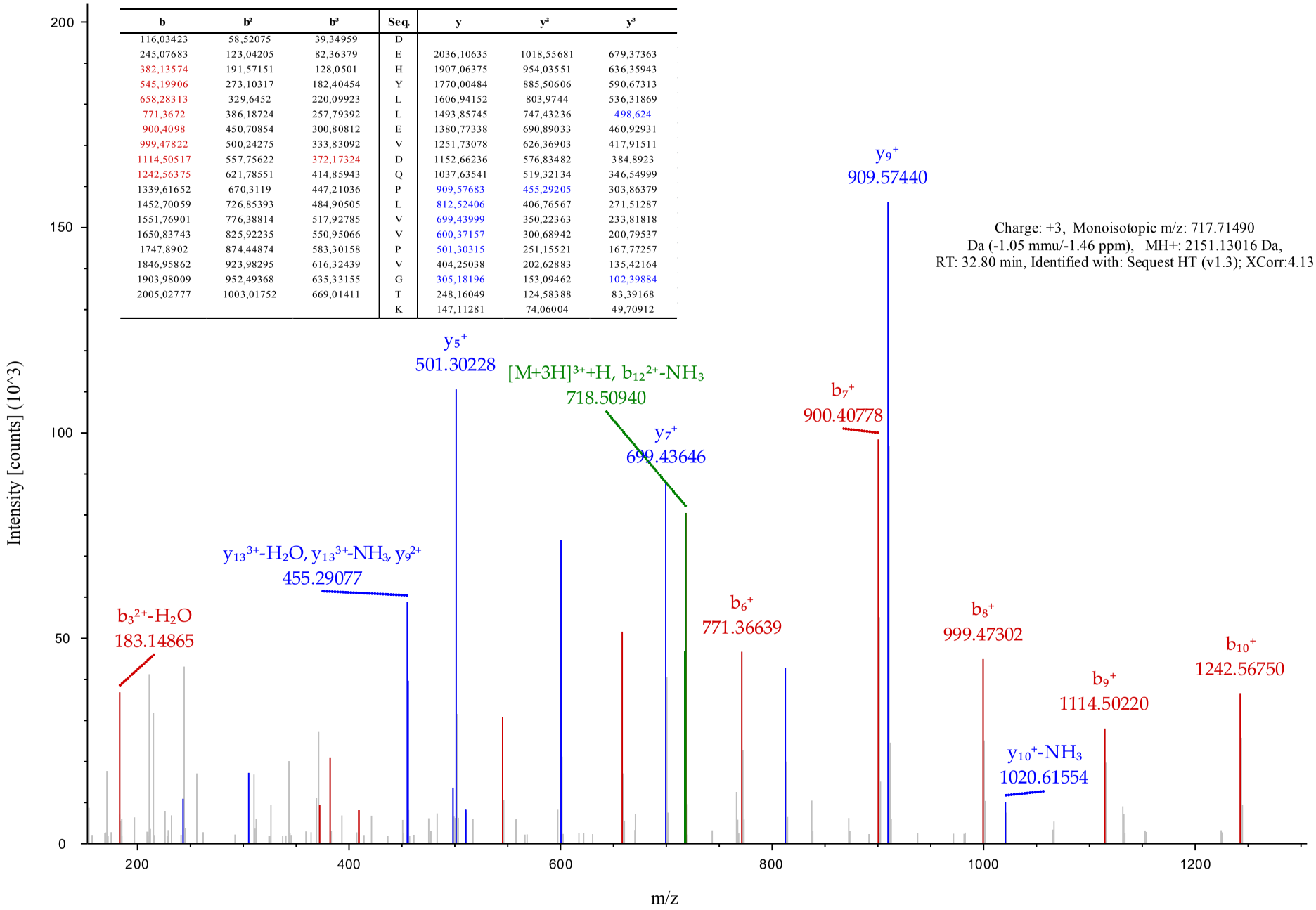
b	b²	b³	Seq.	y	y²	y³
130,04988	65,52858	44,02148	E			
243,13395	122,07061	81,71617	L	1688,88546	844,94637	563,63334
344,18163	172,59445	115,39873	T	1575,80139	788,40433	525,93865
459,20858	230,10793	153,74104	D	1474,75371	737,88049	492,25609
587,30355	294,15541	196,43937	K	1359,72676	680,36702	453,91377
716,34615	358,67671	239,45357	E	1231,63179	616,31953	411,21545
902,42547	451,71637	301,48001	W	1102,58919	551,79823	368,20125
1003,47315	502,24021	335,16257	T	916,50987	458,75857	306,17481
1116,55722	558,78225	372,85726	L	815,46219	408,23473	272,49225
1231,58417	616,29572	411,19957	D	702,37812	351,6927	234,79756
1360,62677	680,81702	454,21377	E	587,35117	294,17922	196,45524
1473,71084	737,35906	491,90846	L	458,30857	229,65792	153,44104
1572,77926	786,89327	524,93127	V	345,2245	173,11589	115,74635
1643,81638	822,41183	548,61031	A	246,15608	123,58168	82,72354
			R	175,11896	88,06312	59,0445

Charge: +3, Monoisotopic m/z: 606.64734
Da (-0.2 mmu/-0.33 ppm), MH+: 1817.92746 Da,
RT: 32.10 min, Identified with: Sequest HT (v1.3); XCorr:4.26



DEHYLLEVDQPLVVPVGTK

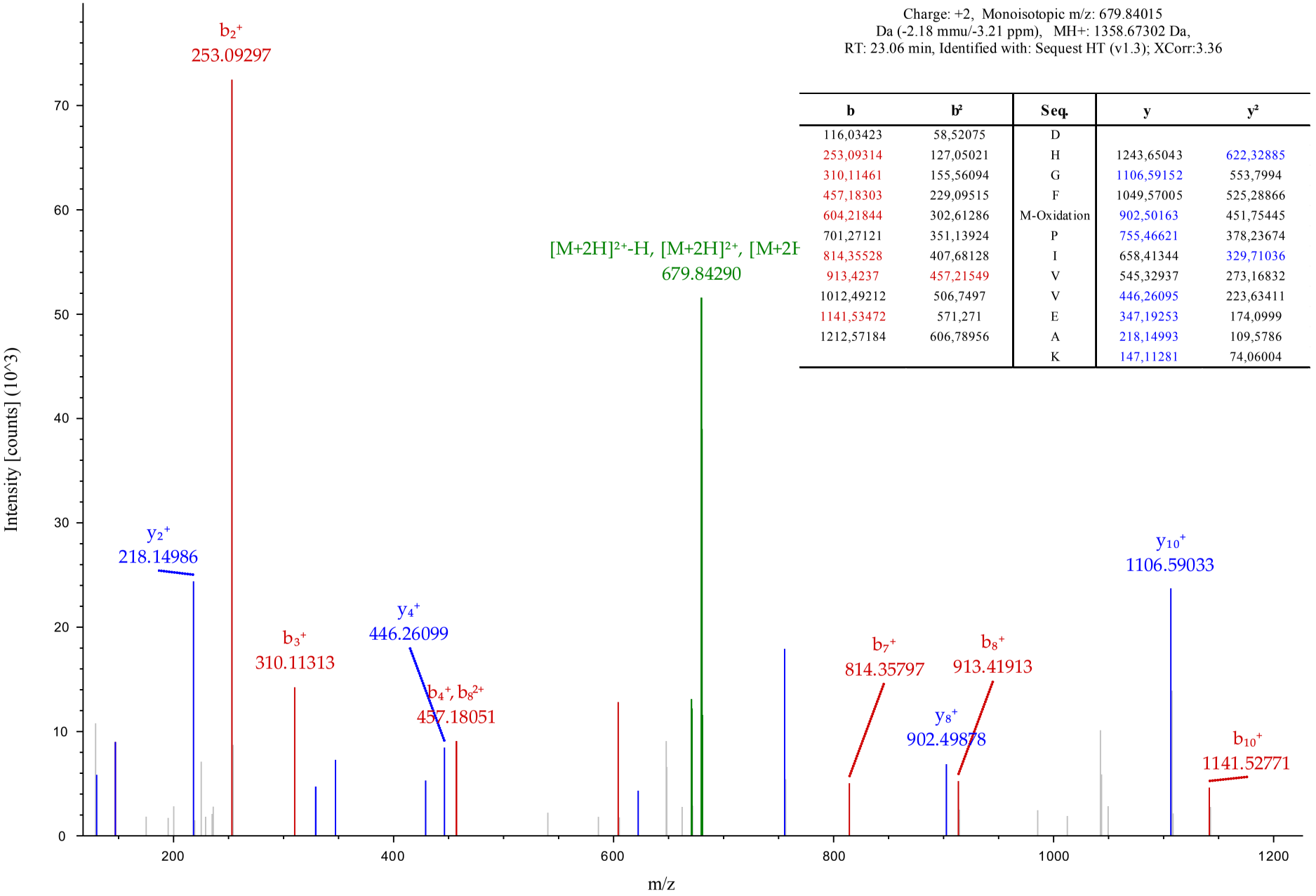
b	b^2	b^3	Seq.	y	y^2	y^3
116,03423	58,52075	39,34959	D			
245,07683	123,04205	82,36379	E	2036,10635	1018,55681	679,37363
382,13574	191,57151	128,0501	H	1907,06375	954,03551	636,35943
545,19906	273,10317	182,40454	Y	1770,00484	885,50606	590,67313
658,28313	329,6452	220,09923	L	1606,94152	803,9744	536,31869
771,3672	386,18724	257,79392	L	1493,85745	747,43236	498,624
900,4098	450,70854	300,80812	E	1380,77338	690,89033	460,92931
999,47822	500,24275	333,83092	V	1251,73078	626,36903	417,91511
1114,50517	557,75622	372,17324	D	1152,66236	576,83482	384,8923
1242,56375	621,78551	414,85943	Q	1037,63541	519,32134	346,54999
1339,61652	670,3119	447,21036	P	909,57683	455,29205	303,86379
1452,70059	726,85393	484,90505	L	812,52406	406,76567	271,51287
1551,76901	776,38814	517,92785	V	699,43999	350,22363	233,81818
1650,83743	825,92235	550,95066	V	600,37157	300,68942	200,79537
1747,8902	874,44874	583,30158	P	501,30315	251,15521	167,77257
1846,95862	923,98295	616,32439	V	404,25038	202,62883	135,42164
1903,98009	952,49368	635,33155	G	305,18196	153,09462	102,39884
2005,02777	1003,01752	669,01411	T	248,16049	124,58388	83,39168
			K	147,11281	74,06004	49,70912



DHGFM*PIVVEAK
M5-Oxidation (15.99492 Da)

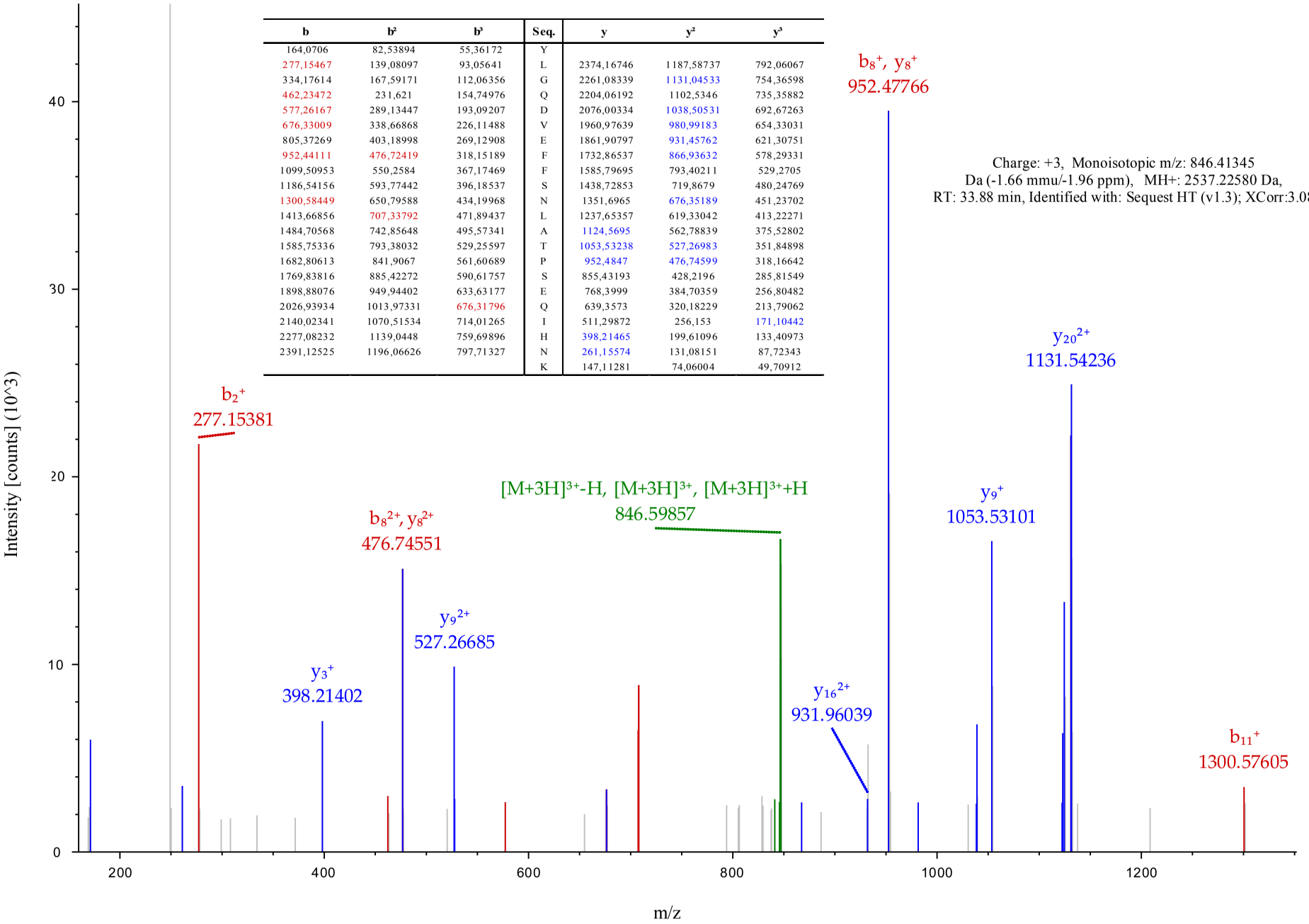
Charge: +2, Monoisotopic m/z: 679.84015
Da (-2.18 mmu/-3.21 ppm), MH+: 1358.67302 Da,
RT: 23.06 min, Identified with: Sequest HT (v1.3); XCorr:3.36

b	b²	Seq.	y	y²
116,03423	58,52075	D		
253,09314	127,05021	H	1243,65043	622,32885
310,11461	155,56094	G	1106,59152	553,7994
457,18303	229,09515	F	1049,57005	525,28866
604,21844	302,61286	M-Oxidation	902,50163	451,75445
701,27121	351,13924	P	755,46621	378,23674
814,35528	407,68128	I	658,41344	329,71036
913,4237	457,21549	V	545,32937	273,16832
1012,49212	506,7497	V	446,26095	223,63411
1141,53472	571,271	E	347,19253	174,0999
1212,57184	606,78956	A	218,14993	109,5786
		K	147,11281	74,06004



YLGQDVEFFSNLATPSEQIHNK

b	b²	b³	Seq.	y	y²	y³
164,0706	82,53894	55,36172	Y			
277,15467	139,08097	93,05641	L	2374,16746	1187,58737	792,06067
334,17614	167,59171	112,06356	G	2261,08339	1131,04533	754,36598
462,23472	231,621	154,74976	Q	2204,06192	1102,5346	735,35882
577,26167	289,13447	193,09207	D	2076,00334	1038,50531	692,67263
676,33009	338,66868	226,11488	V	1960,97639	980,99183	654,33031
805,37269	403,18998	269,12908	E	1861,90797	931,45762	621,30751
952,44111	476,72419	318,15189	F	1732,86537	866,93632	578,29331
1099,50953	550,2584	367,17469	F	1585,79695	793,40211	529,2705
1186,54156	593,77442	396,18537	S	1438,72853	719,8679	480,24769
1300,58449	650,79588	434,19968	N	1351,6965	676,35189	451,23702
1413,66856	707,33792	471,89437	L	1237,65357	619,33042	413,22271
1484,70568	742,85648	495,57341	A	1124,5695	562,78839	375,52802
1585,75336	793,38032	529,25597	T	1053,53238	527,26983	351,84898
1682,80613	841,9067	561,60689	P	952,4847	476,74599	318,16642
1769,83816	885,42272	590,61757	S	855,43193	428,2196	285,81549
1898,88076	949,94402	633,63177	E	768,3999	384,70359	256,80482
2026,93934	1013,97331	676,31796	Q	639,3573	320,18229	213,79062
2140,02341	1070,51534	714,01265	I	511,29872	256,153	171,10442
2277,08232	1139,0448	759,69896	H	398,21465	199,61096	133,40973
2391,12525	1196,06626	797,71327	N	261,15574	131,08151	87,72343
			K	147,11281	74,06004	49,70912

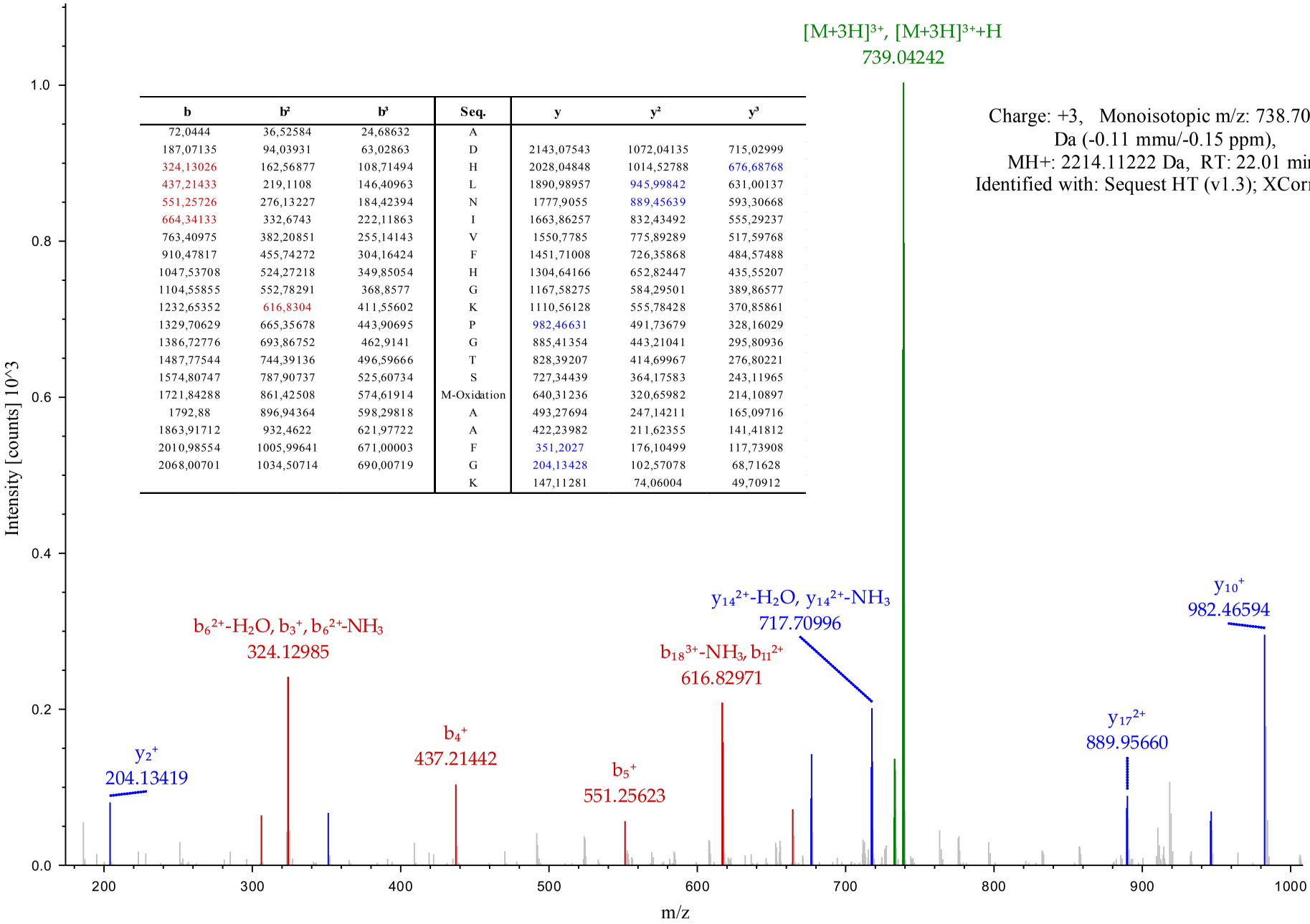


B

ADHLNIVFHGKPGTSM*AAFGK

*M16-Oxidation (15.99492 Da)

b	b²	b³	Seq.	y	y²	y³
72,0444	36,52584	24,68632	A			
187,07135	94,03931	63,02863	D	2143,07543	1072,04135	715,02999
324,13026	162,56877	108,71494	H	2028,04848	1014,52788	676,68768
437,21433	219,1108	146,40963	L	1890,98957	945,99842	631,00137
551,25726	276,13227	184,42394	N	1777,9055	889,45639	593,30668
664,34133	332,6743	222,11863	I	1663,86257	832,43492	555,29237
763,40975	382,20851	255,14143	V	1550,7785	775,89289	517,59768
910,47817	455,74272	304,16424	F	1451,71008	726,35868	484,57488
1047,53708	524,27218	349,85054	H	1304,64166	652,82447	435,55207
1104,55855	552,78291	368,8577	G	1167,58275	584,29501	389,86577
1232,65352	616,8304	411,55602	K	1110,56128	555,78428	370,85861
1329,70629	665,35678	443,90695	P	982,46631	491,73679	328,16029
1386,72776	693,86752	462,9141	G	885,41354	443,21041	295,80936
1487,77544	744,39136	496,59666	T	828,39207	414,69967	276,80221
1574,80747	787,90737	525,60734	S	727,34439	364,17583	243,11965
1721,84288	861,42508	574,61914	M-Oxidation	640,31236	320,65982	214,10897
1792,88	896,94364	598,29818	A	493,27694	247,14211	165,09716
1863,91712	932,4622	621,97722	A	422,23982	211,62355	141,41812
2010,98554	1005,99641	671,00003	F	351,2027	176,10499	117,73908
2068,00701	1034,50714	690,00719	G	204,13428	102,57078	68,71628
			K	147,11281	74,06004	49,70912



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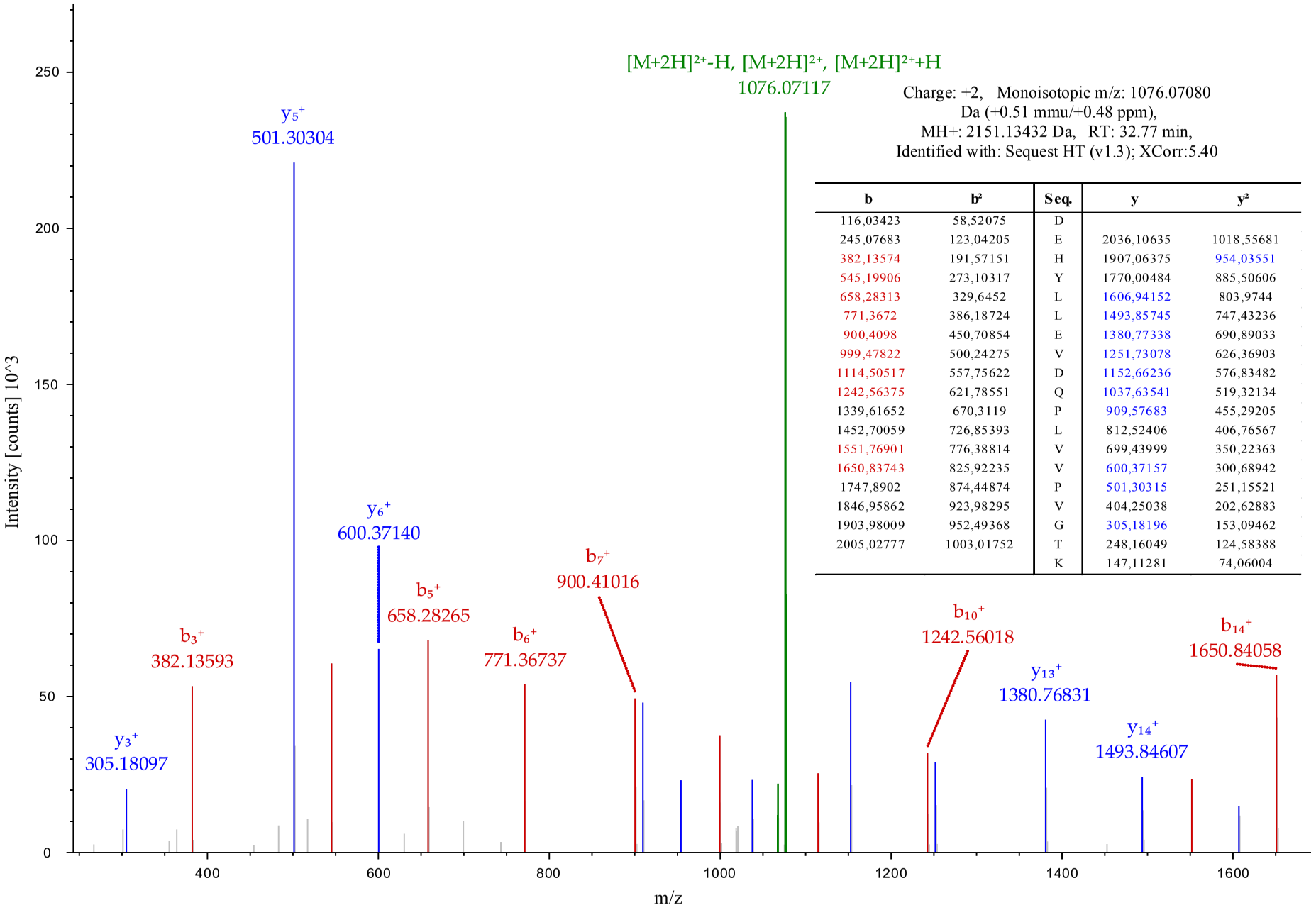
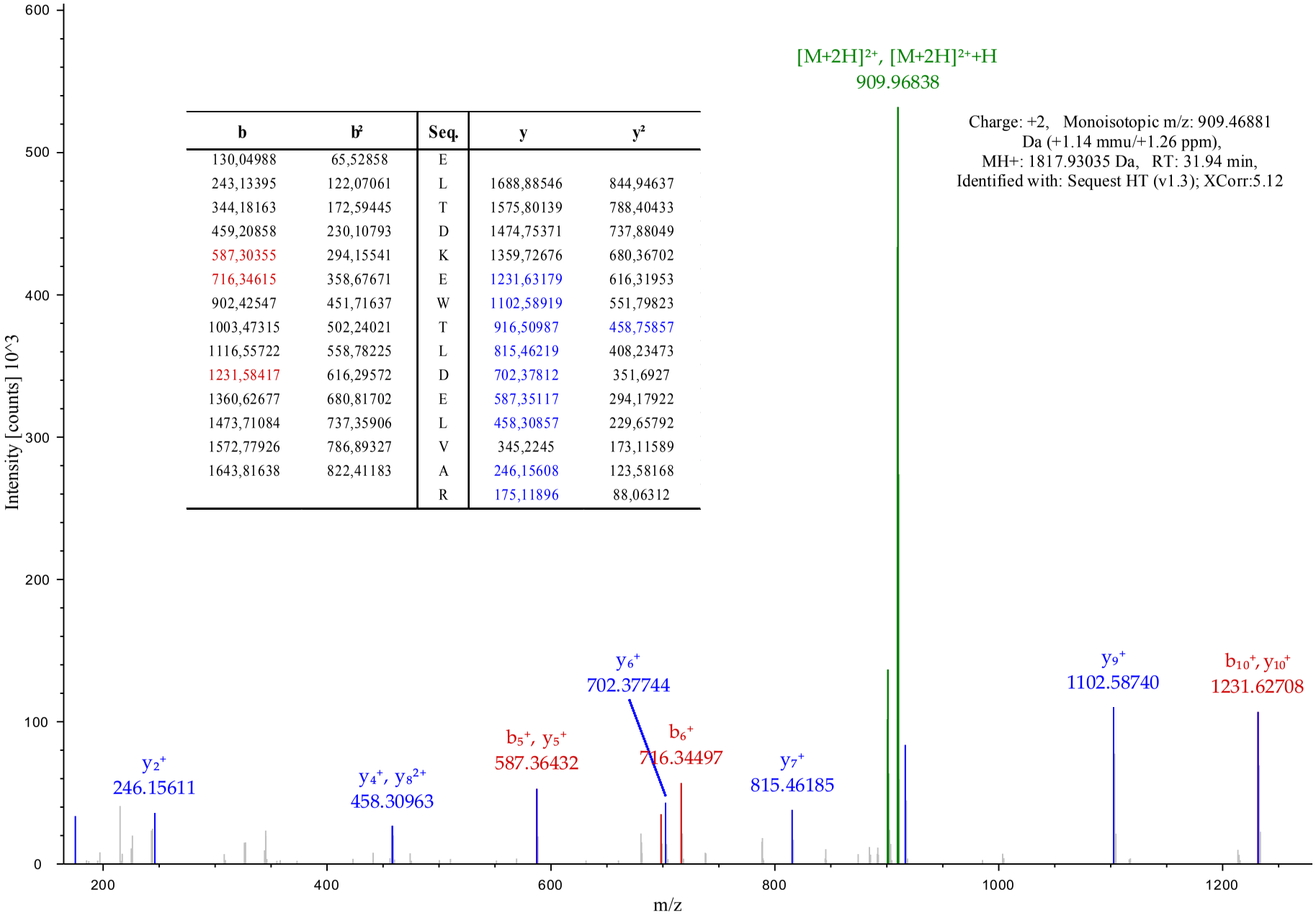


b	b²	Seq.	y	y²
116,03423	58,52075	D		
245,07683	123,04205	E	2036,10635	1018,55681
382,13574	191,57151	H	1907,06375	954,03551
545,19906	273,10317	Y	1770,00484	885,50606
658,28313	329,6452	L	1606,94152	803,9744
771,3672	386,18724	L	1493,85745	747,43236
900,4098	450,70854	E	1380,77338	690,89033
999,47822	500,24275	V	1251,73078	626,36903
1114,50517	557,75622	D	1152,66236	576,83482
1242,56375	621,78551	Q	1037,63541	519,32134
1339,61652	670,3119	P	909,57683	455,29205
1452,70059	726,85393	L	812,52406	406,76567
1551,76901	776,38814	V	699,43999	350,22363
1650,83743	825,92235	V	600,37157	300,68942
1747,8902	874,44874	P	501,30315	251,15521
1846,95862	923,98295	V	404,25038	202,62883
1903,98009	952,49368	G	305,18196	153,09462
2005,02777	1003,01752	T	248,16049	124,58388
		K	147,11281	74,06004

ELTDKEWTLDELVA



b	b²	Seq.	y	y²
130,04988	65,52858	E		
243,13395	122,07061	L	1688,88546	844,94637
344,18163	172,59445	T	1575,80139	788,40433
459,20858	230,10793	D	1474,75371	737,88049
587,30355	294,15541	K	1359,72676	680,36702
716,34615	358,67671	E	1231,63179	616,31953
902,42547	451,71637	W	1102,58919	551,79823
1003,47315	502,24021	T	916,50987	458,75857
1116,55722	558,78225	L	815,46219	408,23473
1231,58417	616,29572	D	702,37812	351,6927
1360,62677	680,81702	E	587,35117	294,17922
1473,71084	737,35906	L	458,30857	229,65792
1572,77926	786,89327	V	345,2245	173,11589
1643,81638	822,41183	A	246,15608	123,58168
		R	175,11896	88,06312

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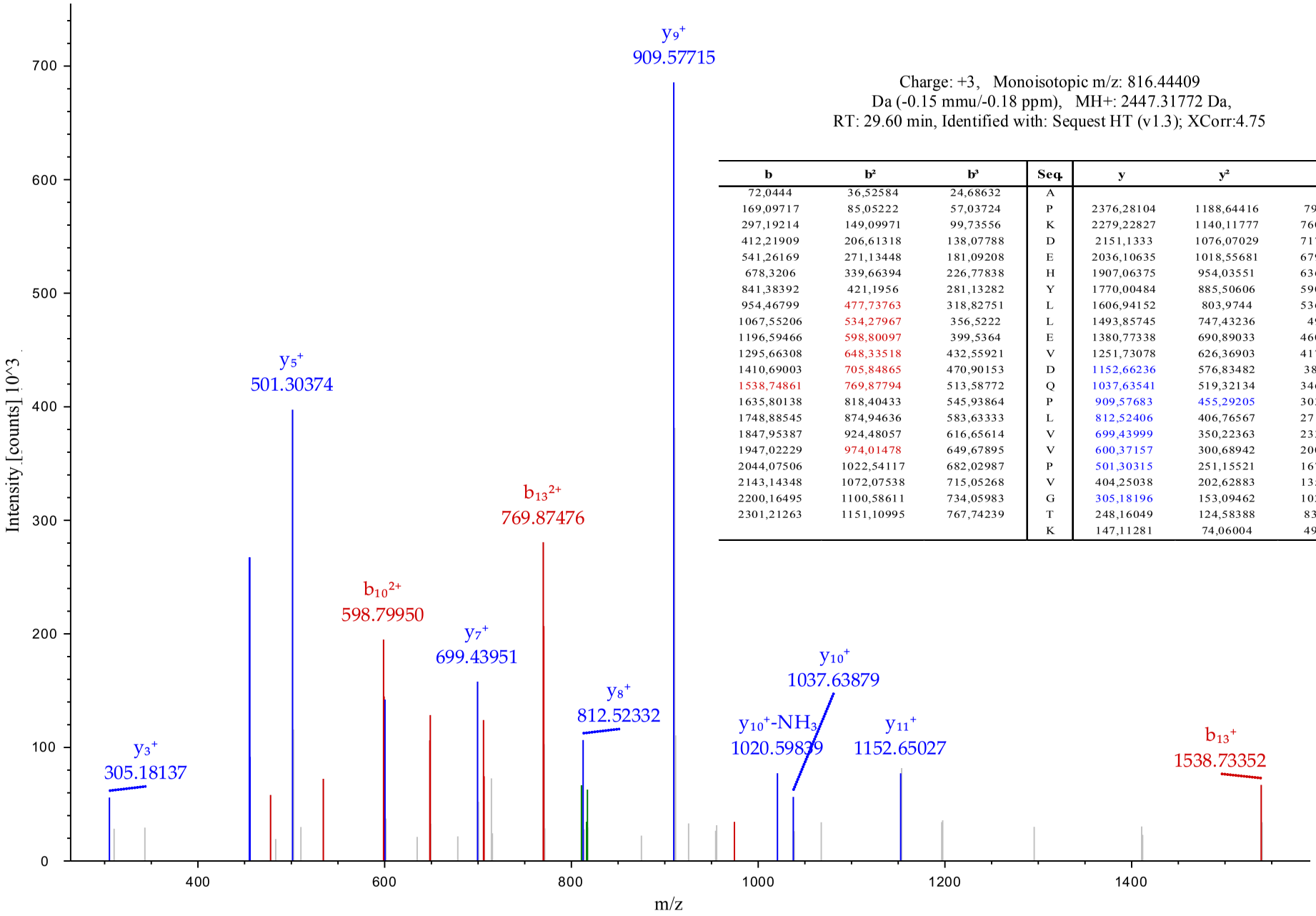


Charge: +3, Monoisotopic m/z: 816.44409
Da (-0.15 mmu/-0.18 ppm), MH+: 2447.31772 Da,
RT: 29.60 min, Identified with: Sequest HT (v1.3); XCorr:4.75

b	b²	b³	Seq.	y	y²	y³
72,0444	36,52584	24,68632	A			
169,09717	85,05222	57,03724	P	2376,28104	1188,64416	792,7652
297,19214	149,09971	99,73556	K	2279,22827	1140,11777	760,41427
412,21909	206,61318	138,07788	D	2151,1333	1076,07029	717,71595
541,26169	271,13448	181,09208	E	2036,10635	1018,55681	679,37363
678,3206	339,66394	226,77838	H	1907,06375	954,03551	636,35943
841,38392	421,1956	281,13282	Y	1770,00484	885,50606	590,67313
954,46799	477,73763	318,82751	L	1606,94152	803,9744	536,31869
1067,55206	534,27967	356,5222	L	1493,85745	747,43236	498,624
1196,59466	598,80097	399,5364	E	1380,77338	690,89033	460,92931
1295,66308	648,33518	432,55921	V	1251,73078	626,36903	417,91511
1410,69003	705,84865	470,90153	D	1152,66236	576,83482	384,8923
1538,74861	769,87794	513,58772	Q	1037,63541	519,32134	346,54999
1635,80138	818,40433	545,93864	P	909,57683	455,29205	303,86379
1748,88545	874,94636	583,63333	L	812,52406	406,76567	271,51287
1847,95387	924,48057	616,65614	V	699,43999	350,22363	233,81818
1947,02229	974,01478	649,67895	V	600,37157	300,68942	200,79537
2044,07506	1022,54117	682,02987	P	501,30315	251,15521	167,77257
2143,14348	1072,07538	715,05268	V	404,25038	202,62883	135,42164
2200,16495	1100,58611	734,05983	G	305,18196	153,09462	102,39884
2301,21263	1151,10995	767,74239	T	248,16049	124,58388	83,39168
			K	147,11281	74,06004	49,70912

Figure S2: Solvent excluded surface of *Pp-caa*$_3$ model structure colored according to the electrostatic potential contoured from -10.0 (intense red) to +10.0 kT/*e* (intense blue) (where k is the Boltzmann constant, T is the absolute temperature, and *e* is the electron charge). The black circle indicates the conserved region found on the His282 side of SU II haem *c* pocket. The surface on the right panel is rotated by 180° around the vertical axis respect to the surface in the left panel.

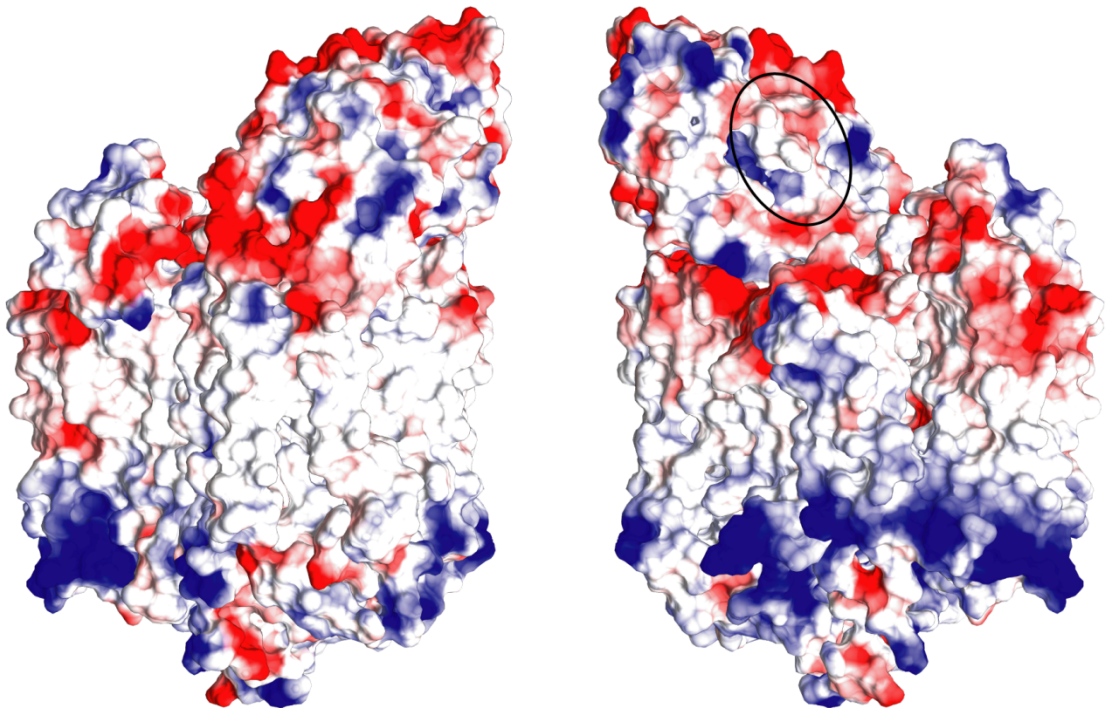
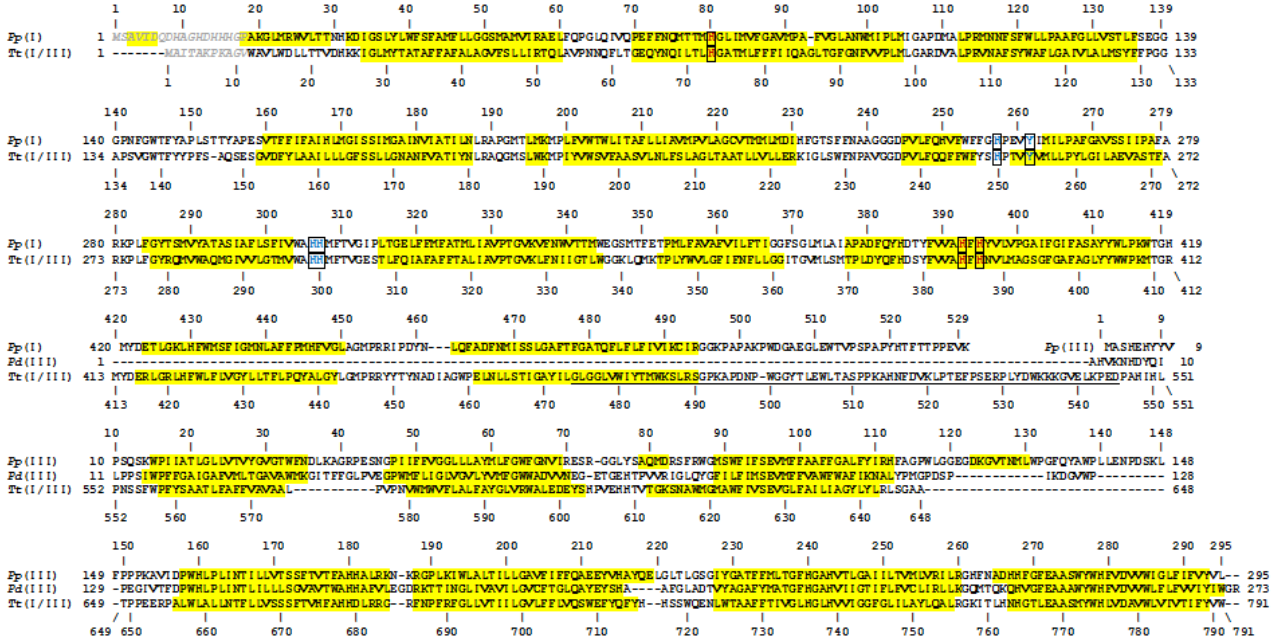


Figure S3: Sequence alignment of *Pp*-*caa*$_3$ SU I, II*c* and III [*Pp*(I), *Pp*(II*c*), and *Pp*(III), respectively] with the corresponding SU from *Pd*-*aa*$_3$ and *Tt*-*caa*$_3$. The colored secondary structure elements (α-helix, yellow; β-strand, cyan) are derived from the prediction performed by the PROMALS3D server. Iron and copper binding residues are bordered and colored in red and blue, respectively. Italicized gray residues were not modeled due to the absence of a template for such regions. The linker between SU I and III in *Tt*-*caa*$_3$ is underlined.

SU I/III Promals3D alignment



SU II Promals3D alignment

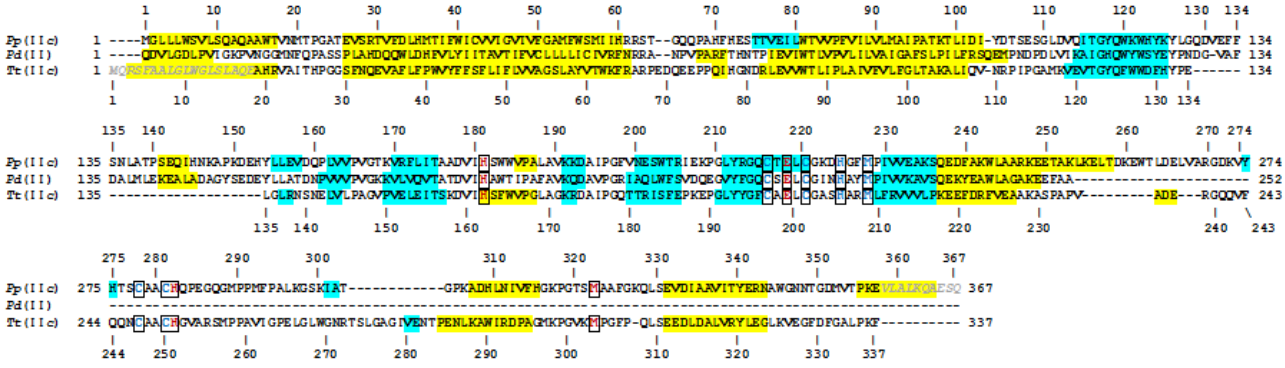


Table S2: Nano-LC-MS/MS analysis results for 37 kDa bands extracted from TMBZ gels from W.T. (grown with glucose or biphenyl as sole carbon source). In addition to the information in Table 2 (main manuscript), extracted peak areas for all the detected peptides as well as protein areas for the two bands are shown (Proteome Discoverer 1.4). For one peptide, ADHLNIVFHGKPGTSM*AAFGK, raw parent peak intensities are also given as an example.

				Glucose (Band 24)				Biphenyl (Band 28)			
Accession	Description	Σ# Unique Peptides	Σ# PSMs	# Peptides Band24	Σ# Spectral Matches Band24	Sequest Protein Area Band 24-Glucose		# Peptides Band28	Σ# Spectral Matches Band28	Sequest Protein Area Band 28-Biphenyl	
L8MR96	Cytochrome c oxidase subunit 2 OS=Pseudomonas pseudoalcaligenes KF707 = NBRC 110670 GN=ppKF707_5503 PE=3 SV=1 - [L8MR96_PSEPS]	14	33	8	10	8,250E7		13	23	1,879E9	
	Sequence	Modifications	MH+ [Da]	# Spectral matches Band24	XCorr Band24	Sequest Peptide Area Band 24 - Glucose		# Spectral matches Band28	XCorr Band28	Sequest Peptide Area Band 28 - Biphenyl	
	DHGFMPIVVEAK	M5(Oxidation)	1358,67716	2	3,36	4,122E7		3	3,35	1,983E9	
	DEHYLLEVDQPLVVPVGTK		2151,13432	2	4,13	3,866E7		2	5,40	1,760E9	
	ADHLNIVFHGKPGTSMAAFGK	M16(Oxidation)	2214,11222	1	2,65	3,966E7	4.3E5 = Parent Peak Intensity @ (m/z = 554.28, z=4)	2	5,43	1,681E9	7.0E6 = Parent Peak Intensity @ (m/z = 554.28, z=4)
	APKDEHYLLEVDQPLVVPVGTK		2447,31992	1	6,04	1,999E7		2	4,75	9,818E8	
	ELTDKEWTLDELVAR		1817,93035	1	4,26	1,303E7		2	5,12	1,844E9	
	NAWGNNTGDMVTPK	M10(Oxidation)	1520,67961	1	2,19	7,646E6		1	3,23	2,259E8	
	YLGQDVEFFSNLATPSEQIHNK		2537,23111	1	3,08	7,260E7		1	5,73	1,811E9	
	KDAIPGFVNESWTR		1619,81778					2	4,17	8,662E8	
	LKELTDKEWTLDELVAR		2059,10703					2	4,16	9,672E8	
	DAIPGFVNESWTR		1491,71977	1	2,35	1,337E8					
	GQCTELCGK	C3(Carbamidomethyl); C7(Carbamidomethyl)	1052,45012					1	2,18	2,522E7	
	EVLALKQAESQ		1215,65715					1	3,00	9,154E6	
	GQCTELCGKDHGFMPIVVEAK	C3(Carbamidomethyl); C7(Carbamidomethyl); M14(Oxidation)	2392,10904					1	4,10	1,651E8	
	ADHLNIVFHGKPGTSMAAFGK		2198,11733					1	4,79	2,278E8	
	DHGFMPIVVEAK		1342,68225					1	3,74	1,235E8	
	EWTLDELVAR		1231,63188					1	2,96	8,510E7	

Table S3: Primers for KF707 deletion mutant strains which were used in this study.

Target	Oligo nucleotide sequence (5'→3')
Δc_4	UPFor ACGTGTAAGCTTCCTTCACAGTTATTCGGCGCA UPRev AGGAGTAAGTATGGAACAGTAGCGGCGGTGGATGATCATC DOWNFor ACTGTTCCATACTTACTCCTACCCAAGGCGGTGATCGAT DOWNRev ATTTCGAATTCTCTCGTCCAGCCGCGCTAAAG
Δc_5	UPFor CGGGCTGGATCCGCGAGACCC UPRev ACATTGATCTTAATTGTACCTTCAGCCGGGAGCCGTCCG DOWNFor AGGTACAATTAAGATCAATGTCCTGCTTCCTCACCGGCTA DOWNRev ATCCCGAATTCTCACTCCCCTGCGCTACCAC
$\Delta c_4/c_5$	UPFor ATTAGTGGATCCAAGAGGACGGGGCGACGCAG UPRev TAGGAGTCAGAATGGTTCAGTTAGGGGTTCCACGGTTAAT DOWNFor ACTGAACCATTCTGACTCCTAAAGTAACACCCCTGCCTGC DOWNRev GGGCTGAATTCGATGTAGAACTTGCGCTCGGG

Figure S4: Cox activity in KF707 wild type (W.T.) and cyt c_4 or c_5 deletion strains (Table 1), visualized as a blue color using NADI staining (Marrs and Gest, 1973). The staining was performed after overnight growth on LB plates with 3 mL of a 1:1 mixture of 35 mM α-naphthol and 30 mM N,N-dimetyl-*p*-phenylenediamine (DMPD) dissolved in ethanol and water, respectively (Materials and Methods).