

SUPPORTING INFORMATION

Site-specific protein adducts of 4-hydroxy-2(E)-nonenal in human THP-1 monocytic cells:

Protein carbonylation is diminished by ascorbic acid

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Table S1: Mass spectrometry data of proteins identified in the gel regions (Figure 1A) that matched the immunoreactive bands in Figure 1B.

Table S2: Mass spectrometry data of proteins identified in the gel regions (Figure 2A) that matched the immunoreactive bands in Figure 2B.

Figures S1-S25: Tandem mass spectra of ARP-labeled HNE-peptide adducts.

Figure S26: Proposed CID fragmentation mechanisms of ARP-labeled HNE-peptide adducts.

Table S1. Mass spectrometry data of proteins identified in the gel regions (Figure 1A) that matched the immunoreactive bands in Figure 1B.

Region	Protein	Mass	pI	Sequence	Peptide	Mascot
No.				Coverage	No.	Score
3	Heat shock protein HSP 90β (HS90B_HUMAN)	83,212	4.97	18%	12	537
	Matching peptides			Start-End	Mass	Score
	IDIIPNPQER			73 - 82	1194.69	38
	ADHGEPIGR			169 - 177	951.47	29
	EDQTEYLEER			187 - 196	1311.58	60
	YIDQEELNK			276 - 284	1151.57	43
	TKPIWTR			285 - 291	901.55	42
	NPDDITQEEYGEFYK			292 - 306	1847.86	61
	ALLFIPR			331 - 337	829.56	38
	GVVDSIDLPLNISR			379 - 392	1513.85	55
	FYEAFSK			429 - 435	891.44	41
	LGIHEDSTNR			439 - 448	1141.58	38
	SIYYITGESK			482 - 491	1160.60	38
	EQVANSAFVER			492 - 502	1249.65	54
4	Heat shock cognate 71kDa (HSP7C_HUMAN)	70,854	5.37	40%	19	1307
	VEIANDQGNR			26 - 36	1228.60	33
	TTPSYVAFTDTER			37 - 49	1487.77	54
	NQVAMNPTNTVFDK Oxidation (M)			57 - 71	1665.78	77
	RFDDAVVQSDMK Oxidation (M)			77 - 88	1426.62	25
	SFYPEEVSSMVLTK Oxidation (M)			113 - 126	1632.82	87
	EIAEAYLGK			129 - 137	993.53	41
	TVTNAVVTVPAYFNDSQR			138 - 155	1982.03	112
	DAGTIAGLNVLR			160 - 171	1199.68	57
	IINEPTAAAIAYGLDK			172 - 187	1659.89	120

	STAGDTHLGGEDFDNR			221 - 236	1691.70	108
	MVNHFIAEFK Oxidation (M)			237 - 246	1251.66	56
	FEELNADLFR			302 - 311	1253.62	82
	SQIHDIVLVGGSTR			329 - 342	1481.87	90
	LLQDFFNKG			349 - 357	1081.55	64
	LLQDFFNKG Deamidation (N)			349 - 357	1082.54	49
	SINPDEAVAYGAAVQAAILSGDK			362 - 384	2260.20	81
	QTQTFTTYSNQPGLIQVYEGER			424 - 447	2774.38	33
	NSLESYAFNMK Oxidation (M)			540 - 550	1319.64	51
	NQTAEKEEFEHQK			584 - 597	1745.78	87
6-1	Protein disulfide isomerase A3 (PDIA3_HUMAN)	56,747	5.98	34%	14	691
	LAPEYEEAATR			63 - 73	1191.62	73
	YGVSGYPTLK			95 - 104	1084.61	36
	QAGPASVPLR			131 - 140	995.57	27
	DASIVGFFDDSFSEAHSEFLK			153 - 173	2348.37	66
	LNFAVASR			297 - 304	877.53	34
	TFSHELSDFGLESTAGEIPVVAIR			306 - 329	2575.80	44
	FVMQEEFSR			336 - 344	1172.62	47
	FLQDYFDGNLK			352 - 362	1359.85	73
	VVVAENFDEIVNNENK			380 - 395	1833.08	37
	MDATANDVPSPYEV			434 - 448	1664.86	63
	MDATANDVPSPYEV.G Oxidation (M)			434 - 448	1680.81	32
	GFPTIYFSPANK			449 - 460	1341.86	60
	ELSDFISYLQR			472 - 482	1370.93	73
	EATNPPVIEEKPK			483 - 496	1579.91	26
6-2	Adenylyl cyclase-associated protein 1 (CAP1_HUMAN)	51,823	8.27	30%	18	1276
	LEAVSHTSDMHR			18 - 29	1382.65	77
	LEAVSHTSDMHR Oxidation (M)			18 - 29	1398.61	29

	HAEMVHTGLK			72 - 81	1122.61	92
	HAEMVHTGLK Oxidation (M)			72 - 81	1138.52	76
	ALLVTASQCQQPAENK			85 - 100	1700.92	26
	ALLVTASQCQQPAENK Propionamide (C)			85 - 100	1771.97	76
	LSDLLAPISEQIK			101 - 113	1426.97	119
	EMNDAAMFYTNR			156 - 167	1462.72	94
	EMNDAAMFYTNR Oxidation (M)			156 - 167	1478.68	36
	EFHTTGLAWSK			199 - 209	1276.71	92
	EFHTTGLAWSK Oxidation (W)			199 - 209	1292.69	46
	KEPAVLELEGK			317 - 327	1212.73	91
	EPAVLELEGK			318 - 327	1084.63	78
	VENQENVSNLVIEDTELK			331 - 348	2073.26	107
	QVAYIYK			349 - 355	884.49	34
	VPTISINK			405 - 412	871.52	29
	SSEMNVLIPTEGGDFNEFPVPEQFK			434 - 458	2811.90	87
	SSEMNVLIPTEGGDFNEFPVPEQFK Oxidation (M)			434 - 458	2827.87	87
6-3	Vimentin (VIME_HUMAN)	53,619	5.06	44%	17	1136
	MFGGPGTASR			14 - 23	980.47	35
	SLYASSPGGVYATR			37 - 50	1495.87	95
	VELQELNDR			105 - 113	1115.59	63
	ILLAELEQLK			130 - 139	1169.90	90
	LGDLYEEEMR			146 - 155	1254.64	84
	LQEEMLQR			189 - 196	1046.53	51
	EEAENTLQSFR			197 - 207	1323.70	78
	KVESLQEEIAFLK			223 - 235	1534.02	44
	VESLQEEIAFLK			224 - 235	1405.95	48
	NLQEAEEWYK			283 - 292	1309.73	72
	FADLSEAANR			295 - 304	1093.55	41
	EMEENFAVEAANYQDTIGR			346 - 364	2187.24	129
	EMEENFAVEAANYQDTIGR Oxidation (M)			346 - 364	2203.19	29

	LQDEIQNMKEEMAR			365 - 378	1734.96	39
	ISLPLPNFSSLNLR			411 - 424	1571.17	84
	ETNLDLPLVDTHSK			425 - 439	1669.06	42
	DGQVINETSQHHDDLE			451 - 466	1836.85	112
7	Tubulin alpha-1B chain (TBA1B_HUMAN)	50,120	4.93	23%	7	439
	TIGGGDDSFNTFFSETGAGK			41 - 60	2007.93	90
	AVFVDLEPTVIDEVR			65 - 79	1701.98	68
	QLFHPEQLITGK			85 - 96	1410.79	73
	EIIDLVLDR			113 - 121	1085.64	47
	NLDIERPTYTNLNR			216 - 229	1718.89	30
	IHFPLATYAPVISA EK			265 - 280	1757.02	69
	VGINYQPPTVVPGGDLAK			353 - 370	1825.04	62
8	α-Enolase (ENOA_HUMAN)	47,139	7.01	24%	5	341
	GNPTVEVDLFTSK			16 - 28	1406.75	74
	AAVPSGASTGIYEAL E LR			33 - 50	1805.02	46
	IGA EVYHNLK			184 - 193	1143.63	91
	VVIGMDVAASEFFR Oxidation (M)			240 - 253	1556.82	31
	YISPDQLADLYK			270 - 281	1425.74	99
9	Actin, cytoplasmic 1 (ACTB_HUMAN)	41,710	5.29	53%	19	1607
	AGFAGDDAPR			19 - 28	976.38	51
	AVFPSIVGR			29 - 37	945.58	57
	AVFPSIVGRPR			29 - 39	944.57	52
	HQGV MVGMGQK			40 - 50	1171.53	67
	IWHHTFYNELR			85 - 95	1515.81	81
	VAPEEHPVLLTEAPLNPK			96 - 113	1954.15	123
	TTGIVMDSGDGVTHTVPIYEGYALPHAILR			148 - 177	3183.91	141
	TTGIVMDSGDGVTHTVPIYEGYALPHAILR			148 - 177	3199.92	64
	Oxidation (M)					

	DLTDYLMK			184 – 191	998.53	36
	DLTDYLMK Oxidation (M)			184 - 191	1014.45	40
	GYSFTTTAER			197 – 206	1132.52	75
	LCYVALDFEQEMATAASSSSLEK			216 - 238	2493.49	144
	LCYVALDFEQEMATAASSSSLEK Oxidation (M)			216 - 238	2509.39	76
	SYELPDGQVITIGNER			239 - 254	1790.97	108
	KDLYANTVLSGGTTMYPGIADR			291 - 312	2343.21	123
	DLYANTVLSGGTTMYPGIADR			292 - 312	2215.27	180
	DLYANTVLSGGTTMYPGIADR Oxidation (M)			292 - 312	2231.21	27
	EITALAPSTMK			316 - 326	1161.62	50
	QEYDESGPSIVHR			360 - 372	1516.68	112
12-1	Carbonic anhydrase 2 (CAH2_HUMAN)	29,228	6.87	51%	10	770
	HNGPEHWHK			10 - 18	1141.56	52
	QSPVDIDHTAK			28 - 39	1311.71	74
	YDPSLKPLSVSYDQATSLR			40 - 58	2140.16	121
	ILNNGHAFNVEFDDSDQK Deamidation (NQ)			59 - 76	2063.97	104
	GGPLDGTYR			81 - 89	935.51	51
	YAAELHLVHWNTK			114 - 126	1581.86	111
	AVQQPDGLAVLGIFLK			133 - 148	1669.05	107
	VVDVLDSIK			159 - 167	987.61	58
	SADFTNFDPR			172 - 181	1169.56	62
	LNFNGEGEPEELMVDNWR Oxidation (M)			228 - 245	2165.09	30
12-2	14-3-3 protein β/α (1433B_HUMAN)	28,065	4.76	21%	4	209
	AVTEQGHELSNEER			30 - 43	1598.78	73
	EMQPTHPIR Oxidation (M)			161 - 169	1124.56	26
	TAFDEAIAELDTLNEESYK			196 - 214	2159.16	84
	DSTLIMQLLR Oxidation (M)			215 - 224	1205.66	26
13	Peroxiredoxin-6 (PRDX6_HUMAN)	25,019	6.00	21%	4	163

	PGGLLLGDVAPNFEANTTVGR			2 - 22	2098.18	27
	LPFPIIDDR			98 - 106	1085.62	73
	LSILYPATTGR			145 - 155	1191.71	34
	NFDEILR			156 - 162	906.50	29
17	Histone H2B type 1-M (H2B1M_HUMAN)	13,981	10.31	46%	6	336
	ESYSVYVYK			36 - 44	1137.49	72
	QVHPDTGISSK			48 - 58	1168.52	82
	AMGIMNSFVNDIFER 2 Oxidation (M)			59 - 73	1775.77	32
	LAHYNKR			81 - 87	901.43	25
	EIQTAVR			94 - 100	816.40	40
	LLLPGELAK			101 - 109	953.57	85

Table S2. Mass spectrometry data of proteins identified in the gel regions (Figure 2A) that matched the immunoreactive bands in Figure 2B.

Region	Protein	Mass	pI	Sequence	Peptide	Mascot
No.				Coverage	No.	Score
1	Ribonuclease inhibitor (RINI_HUMAN)	49,941	4.71	42 %	15	1090
	Matching peptides			Start-End	Mass	Score
	WAELLPLLQQCQVVR Carbamidomethyl (C)			20 - 34	1853.14	33
	LDDCGLTEAR Carbamidomethyl (C)			35 - 44	1149.54	61
	VNPALAE LNLR			54 - 64	1209.73	63
	SNELGDVG VHCVLQGLQTPSCK 2 Carbamidomethyl (C)			65 - 86	2398.23	104
	LSLQNCCLTGAGCGVLSSTLR Phospho (ST)			90 - 110	2176.19	25
	LSLQNCCLTGAGCGVLSSTLR 3 Carbamidomethyl (C)			90 - 110	2267.21	123
	ELTVSNNDINEAGVR			174 - 188	1630.81	106
	DSPCQLEALK Carbamidomethyl (C)			196 - 205	1160.57	64
	LESCGVTS DNCR 2 Carbamidomethyl (C)			206 - 217	1397.54	61
	LGDVGMAELCPGLLHPSSR Carbamidomethyl (C), Oxidation (M)			239 - 257	2025.01	89
	TLWIWECGITAK Carbamidomethyl (C)			260 - 271	1477.88	71
	ELSLAGNELGDEGAR			288 - 302	1530.77	127
	SCSFTAACCSHFSSVLAQNR 3 Carbamidomethyl (C)			322 - 341	2290.04	35
	ELCQGLGQPGSVLR Carbamidomethyl (C)			360 - 373	1513.84	74
	ELCQGLGQPGSVLR Carbamidomethyl (C), Deamidation (NQ)			360 - 373	1514.82	54
2	Protein disulfide-isomerase (PDIA1_HUMAN)	57,081	4.76	57 %	30	2141
	KSNFAEALAAHK			31 - 42	1158.56	86
	SNFAEALAAHK			32 - 42	1286.63	97
	ALAPEYAK			58 - 65	862.45	53
	VDATEESDLAQQYGVR			82 - 97	1780.86	125
	EADDIVNWLK			121 - 130	862.46	102

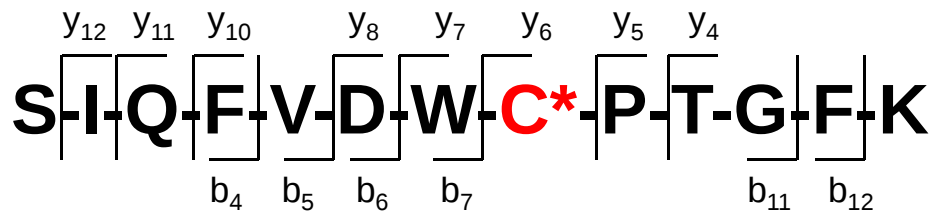
	QFLQAAEAIDDIPFGITSNSDVFSK			171 - 195	2713.54	110
	YQLDKDGVVLFK			196 - 207	1424.79	80
	DGVVLFK			201 - 207	777.44	31
	NNFEGEVTK			214 - 222	1037.48	32
	ENLLDFIK			223 - 230	991.58	76
	HNQLPLVIEFTEQTAPK			231 - 247	1965.13	164
	HNQLPLVIEFTEQTAPK Deamidation (NQ)			231 - 247	1966.13	41
	IFGGEIK			248 - 254	763.40	55
	THILLFLPK			255 - 263	1081.70	80
	ILFIFIDSDHTDNQR			286 - 300	1834.00	73
	ILEFFGLK			301 - 308	966.60	67
	LITLEEEMTK Oxidation (M)			317 - 326	1222.60	48
	YKPESEELTAER			327 - 338	1451.69	90
	ITEFCHR Carbamidomethyl (C)			339 - 345	962.44	49
	NFEDVAFDEK			376 - 385	1213.55	87
	NFEDVAFDEKK			376 - 386	1341.61	67
	QLAPIWDK			402 - 409	970.53	59
	QLAPIWDK Pyro-glu (N-term Q)			402 - 409	953.53	43
	LGETYKDHENIVIAK			410 - 424	1729.89	107
	DHENIVIAK			416 - 424	1038.52	72
	MDSTANEVEAVK Oxidation (M)			425 - 436	1309.58	39
	VHSFPTLK			437 - 444	928.48	45
	FFPASADR			445 - 452	910.41	44
	TVIDYNGER			453 - 461	1066.50	53
	FLESGGQDGAGDDDDLEDLEEAEPPDMEEDDDQK			469 - 502	3773.62	66
	Oxidation (M)					
3	Tubulin beta-2C (TBB2C_HUMAN)	49,799	4.79	21 %	8	453
	INVYYNEATGGK			47 - 58	1328.58	105
	AVLVDLEPGTMDSVR Oxidation (M)			63 - 77	1617.84	36
	AVLVDLEPGTMDSVR Me-ester (DE); Oxidation (M)			63 - 77	1631.85	33
	IMNTFSVVPSPK Oxidation (M)			163 - 174	1335.69	78

	FPGQLNADLR			242 - 251	1130.63	66
	ALTVPELTQQMFDAK Oxidation (M)			283 - 297	1707.88	72
	YLTVAAVFR			310 - 318	1039.67	34
	EVDEQMLNVQNK Deamidation (NQ), 2 Me-ester (DE)			325 - 336	1475.74	29
4	β-Actin (ACTB_HUMAN)	41,710	5.29	45 %	13	1026
	DDDIAALVVDNGSGMCK Carbamidomethyl (C), 3 Me- ester (DE), Oxidation (M)			2 - 18	1837.85	93
	AGFAGDDAPR			19 - 28	976.47	80
	AVFPSIVGR			29 - 37	945.61	63
	DSYVGDEAQSKR			51 - 62	1354.64	55
	YPIEHGIVTNWDDMEK Me-ester (DE), Oxidation (M)			69 - 84	1977.00	81
	VAPEEHPVLLTEAPLNPK			96 - 113	1954.16	135
	DLTDYLMK Oxidation (M)			184 - 191	1014.51	64
	GYSFTTTAER			197 - 206	1132.57	53
	LCYVALDFEQEMATAASSSSLEK Carbamidomethyl (C), Oxidation (M)			216 - 238	2566.29	54
	SYELPDGQVITIGNER			239 - 254	1790.97	91
	EITALAPSTMK Oxidation (M)			316 - 326	1177.64	52
	QEYDESGPSIVHR Pyro-glu (N-term Q)			360 - 372	1499.75	99
	QEYDESGPSIVHR			360 - 372	1516.74	106
5	14-3-3 protein β/α (1433B_HUMAN)	28,065	4.76	43%	9	754
	AVTEQGHELSNEER			30 - 43	1598.73	106
	NLLSVAYK			44 - 51	907.56	55
	VISSIEQK			63 - 70	903.51	52
	YLIPNATQPESK			106 - 117	1360.78	67
	YLSEVASGDNK			130 - 140	1182.57	92
	QTTVSNSQQAYQEAFEISK Pyro-glu (N-term Q)			141 - 159	2142.10	93
	QTTVSNSQQAYQEAFEISK			141 - 159	2159.15	144
	TAFDEAIAELDTLNEESYK			196 - 214	2159.15	109
	DSTLIMQLLR Oxidation (M)			215 - 224	1205.69	36
6	Keratin, type II cytoskeletal 1 (K2C1_HUMAN)	65,978	8.16	41 %	23	1714

	SLNNQFASFIDK			186 - 197	1383.74	96
	SLNNQFASFIDKVR			186 - 199	1638.93	81
	FLEQQNQVLQTK			200 - 211	1475.82	89
	WELLQQVDTSTR			212 - 223	1475.81	71
	THNLEPYFESFINNLR			224 - 239	1994.10	71
	NMQDMVEDYR			258 - 267	1300.57	51
	NMQDMVEDYR 2 Deamidation (NQ); Me-ester (DE)			258 - 267	1316.57	37
	NMQDMVEDYR Oxidation (M)			258 - 267	1316.56	37
	NKYEDEINKR			268 - 277	1308.68	41
	YEDEINKR			270 - 277	1066.54	60
	TNAENEFVTIK			278 - 288	1265.68	95
	TNAENEFVTIKK			278 - 289	1393.76	88
	SLDLDSIIAEVK			344 - 355	1302.77	111
	SKAEAESLYQSKYEELQITAGR			365 - 386	2501.33	29
	YEELQITAGR			377 - 386	1179.64	75
	IEISELNR			396 - 403	973.56	39
	QISNLQQSISDAEQR Pyro-glu (N-term Q)			418 - 432	1699.90	47
	QISNLQQSISDAEQR			418 - 432	1716.90	96
	LNDLEDALQQAK			444 - 455	1357.73	103
	LALDLEIATYR			473 - 483	1277.77	51
	TLLEGEESR			484 - 492	1033.54	38
	GGGGGGYGSGGSSYGSGGGSYGSGGGGGGGR			519 - 549	2383.98	189
	GSYGSGGSSYGSGGGSYGSGGGGGHGSYGSGSSSG			550 - 588	3312.35	119
	GYR					
7	Coronin-1A (COR1A_HUMAN)	50,994	6.25	255	12	848
	HVFGQPAK			13 - 20	883.43	62
	ADQCYEDVR Carbamidomethyl (C)			21 - 29	1155.41	49
	VSQTTWDSGFCAVNPK Carbamidomethyl (C)			30 - 45	1796.88	135
	EPVVTLEGHTK			122 - 132	1209.63	88
	DGGLICTSCR 2 Carbamidomethyl (C)			187 - 196	1138.48	35
	ILTTGFSR			234 - 241	894.52	45

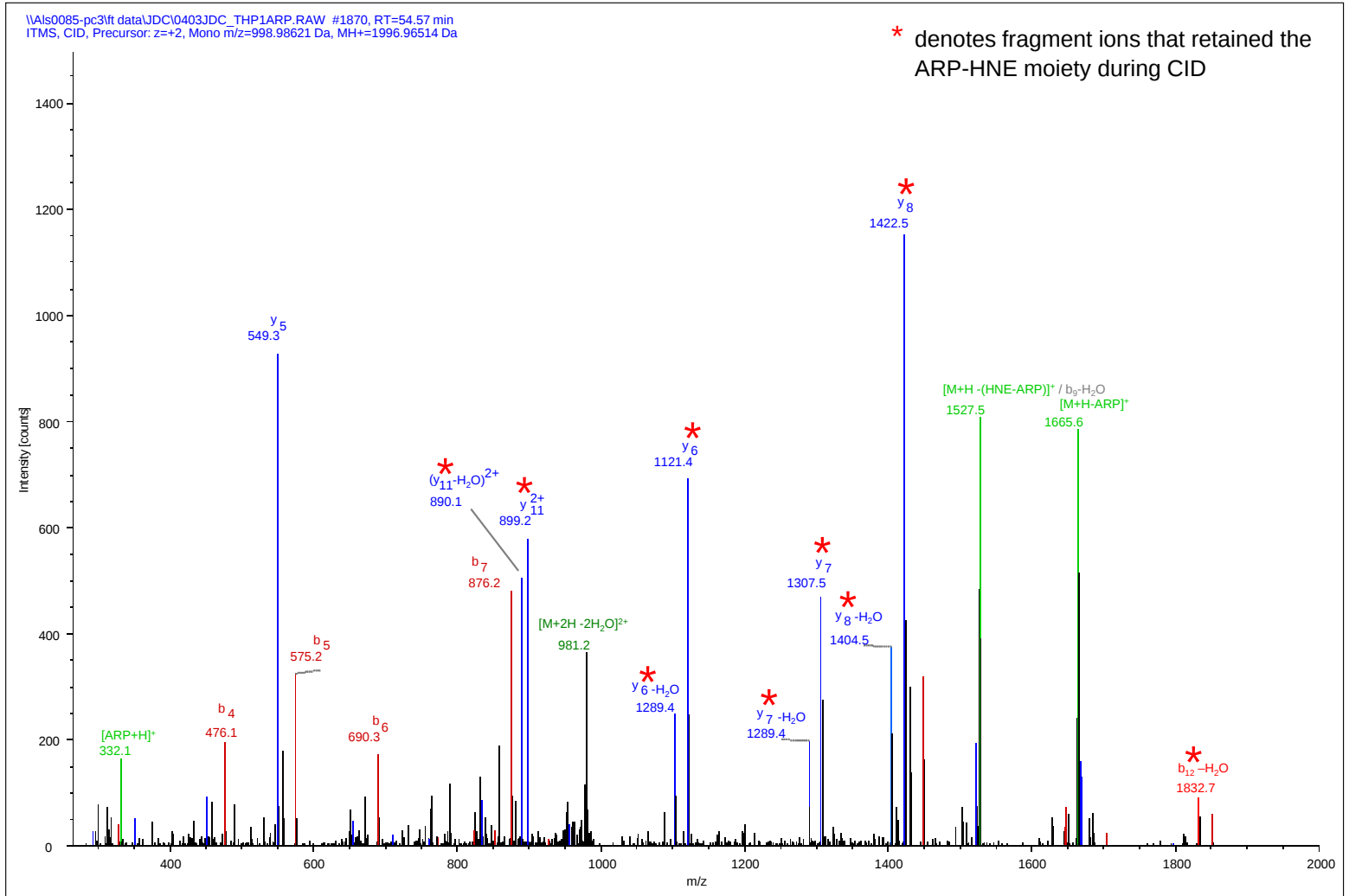
	QVALWDTK			246 - 253	960.49	68
	RCEPIAMTVPR Carbamidomethyl (C); Oxidation (M)			344 - 354	1345.63	27
	DAGPLLISLK			384 - 393	1026.61	89
	AAPEASGTPSSDAVSR			417 - 432	1502.67	83
	KLQATVQELQK			439 - 449	1285.74	99
	LQATVQELQK			440 - 449	1157.64	68
8	Stress-induced-phosphoprotein 1 (STIP1_HUMAN)	62,599	6.40	25 %	10	622
	LDPHNHVLYSNR					
	KAAALEFLNR			33 - 44	1464.64	79
	AAALEFLNR			78 - 87	1132.63	47
	ELIEQLR			79 - 87	1004.58	38
	ELDPTNMTYITNQAAVYFEK Oxidation (M)			154 - 160	900.51	26
	DAIHFYNK			253 - 272	2364.19	108
	LAYINPDLALEEK			318 - 325	1007.45	45
	DCEECIQLEPTFIK 2 Carbamidomethyl (C)			352 - 364	1488.82	91
	LILEQMQK Oxidation (M)			416 - 429	1781.85	67
	DPQALSEHLK			506 - 513	1018.50	39
				514 - 523	1137.52	82
9	α-Enolase (ENOA_HUMAN)	47,139	7.01	33%	9	617
	EIFDSR			10 - 15	766.35	28
	GNPTVEVDLFTSK			16 - 28	1406.72	87
	AAVPSGASTGIYEALER			33 - 50	1805.00	81
	IGAENVYHNLK			184 - 193	1143.58	91
	VVIGMDVAASEFFR Oxidation (M)			240 - 253	1556.80	57
	YDLDFK			257 - 262	800.38	28
	YISPDQLADLYK			270 - 281	1425.76	79
	VNQIGSVTESLQACK Carbamidomethyl (C)			344 - 358	1633.80	135
	YNQLLR			407 - 412	806.43	31
10	Purine nucleoside phosphorylase (PNPH_HUMAN)	32,097	6.45	40 %	9	493
	NTAEWLLSHTK			12 - 22	1299.71	57
	LTQAQIFDYGEIPNFPR			42 - 58	2009.05	39

	LVFGFLNGR			68 - 76	1022.64	35
	LVFGFLNGR Deamidation (NQ)			68 - 76	1023.62	35
	DHINLPGFSGQNPLR			134 - 148	1664.87	47
	FPAMSDAYDR Oxidation (M)			159 - 168	1188.52	36
	LGADAVGMSTVPEVIVAR Oxidation (M)			212 - 229	1801.02	67
	VFGFSLITNK			235 - 244	1125.69	69
	ANHEEVLAAGK			255 - 265	1138.61	108
11	Carbonic anhydrase 2 (CAH2_HUMAN)	29,228	6.87	55 %	12	1153
	HNGPEHWHK			10 - 18	1141.54	60
	QSPVDIDHTAK Pyro-glu (N-term Q)			28 - 39	1294.62	91
	QSPVDIDHTAK			28 - 39	1311.65	81
	YDPSLKPLSVSYDQATSLR			40 - 58	2140.18	149
	ILNNGHAFNVEFDDSDK			59 - 76	2062.94	149
	GGPLDGTyr			81 - 89	935.45	62
	YAAELHLVHWNTK			114 - 126	1581.82	115
	AVQQPDGLAVLGIFLK			133 - 148	1669.02	120
	VVDVLDSIK			159 - 167	987.57	86
	SADFTNFDPR			172 - 181	1169.56	72
	EPISVSSEQVLK			213 - 224	1315.75	98
	LNFNGEGEPEELMVDNWR Oxidation (M)			228 - 245	2165.00	70



TBA1B_Human

A.

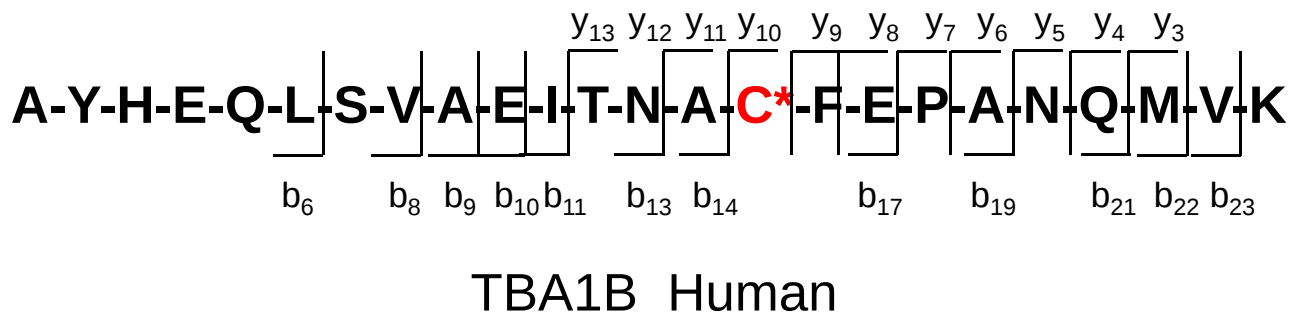


B.

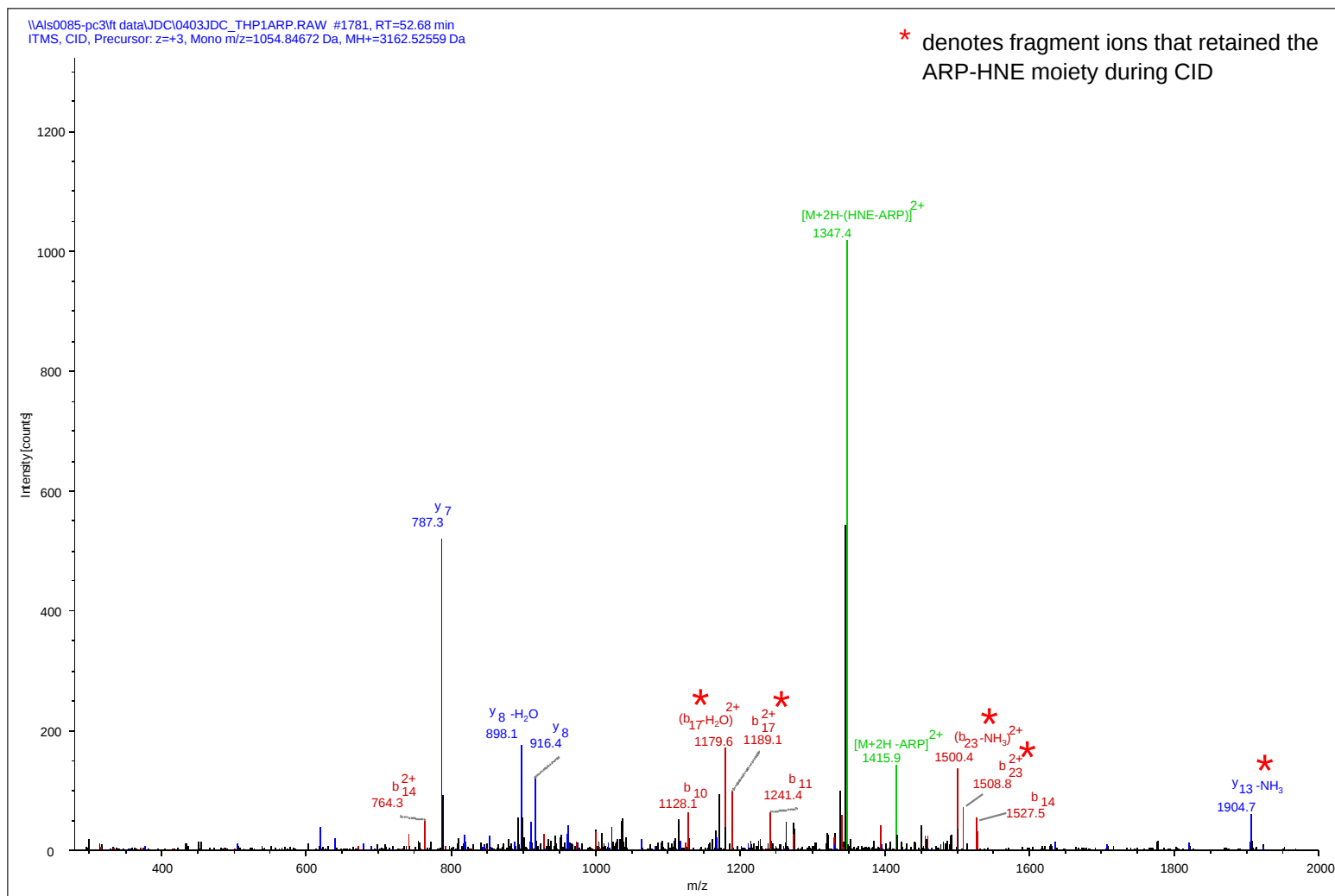
Mascot search result for the MS/MS spectrum depicted above:

#	b	b ⁺⁺	b ⁺	b ⁺⁺⁺	b ⁰	b ⁰⁺⁺	Seq.	y	y ⁺⁺	y ⁺	y ⁺⁺⁺	y ⁰	y ⁰⁺⁺	#
1	88.04	44.52			70.03	35.52	S							13
2	201.12	101.07			183.11	92.06	I	1909.93	955.47	1892.91	946.96	1891.92	946.47	12
3	329.18	165.09	312.16	156.58	311.17	156.09	Q	1796.85	898.93	1779.82	890.42	1778.84	889.92	11
4	476.25	238.63	459.22	230.12	458.24	229.62	F	1668.79	834.90	1651.76	826.39	1650.78	825.89	10
5	575.32	288.16	558.29	279.65	557.31	279.16	V	1521.72	761.37	1504.70	752.85	1503.71	752.36	9
6	690.35	345.68	673.32	337.16	672.34	336.67	D	1422.65	711.83	1405.63	703.32	1404.64	702.83	8
7	876.43	438.72	859.40	430.20	858.41	429.71	W	1307.63	654.32	1290.60	645.80	1289.62	645.31	7
8	1448.67	724.84	1431.64	716.33	1430.66	715.83	C	1121.55	561.28	1104.52	552.76	1103.54	552.27	6
9	1545.72	773.37	1528.70	764.85	1527.71	764.36	P	549.30	275.16	532.28	266.64	531.29	266.15	5
10	1646.77	823.89	1629.74	815.38	1628.76	814.88	T	452.25	226.63	435.22	218.12	434.24	217.62	4
11	1703.79	852.40	1686.77	843.89	1685.78	843.39	G	351.20	176.10	334.18	167.59			3
12	1850.86	925.93	1833.83	917.42	1832.85	916.93	F	294.18	147.59	277.15	139.08			2
13							K	147.11	74.06	130.09	65.55			1

Figure S1. (A) MS/MS spectrum and (B) Full scan mass spectrum of the $[M+2H]^{2+}$ ion of the ARP-HNE modified peptide SIQFVDWC*PTGFK; monoisotopic m/z_{calc} 998.9866; accuracy $\Delta(m/z) = -0.4$ ppm.

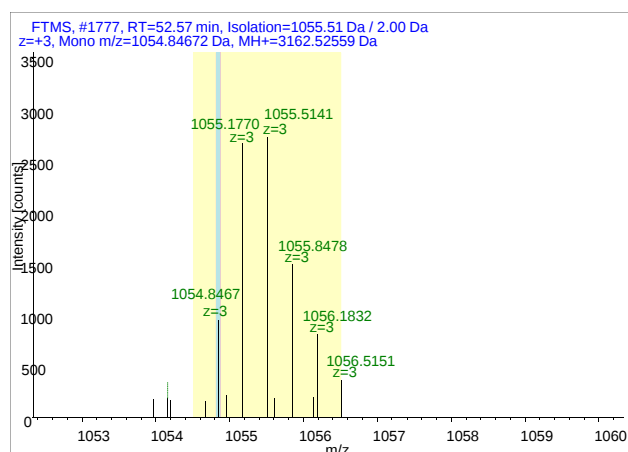


A.



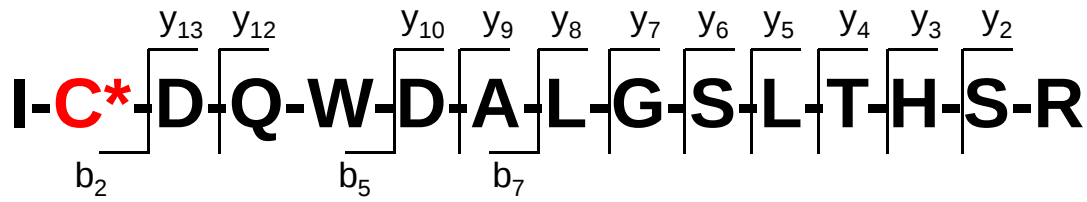
B.

Mascot search result for the MS/MS spectrum depicted above:



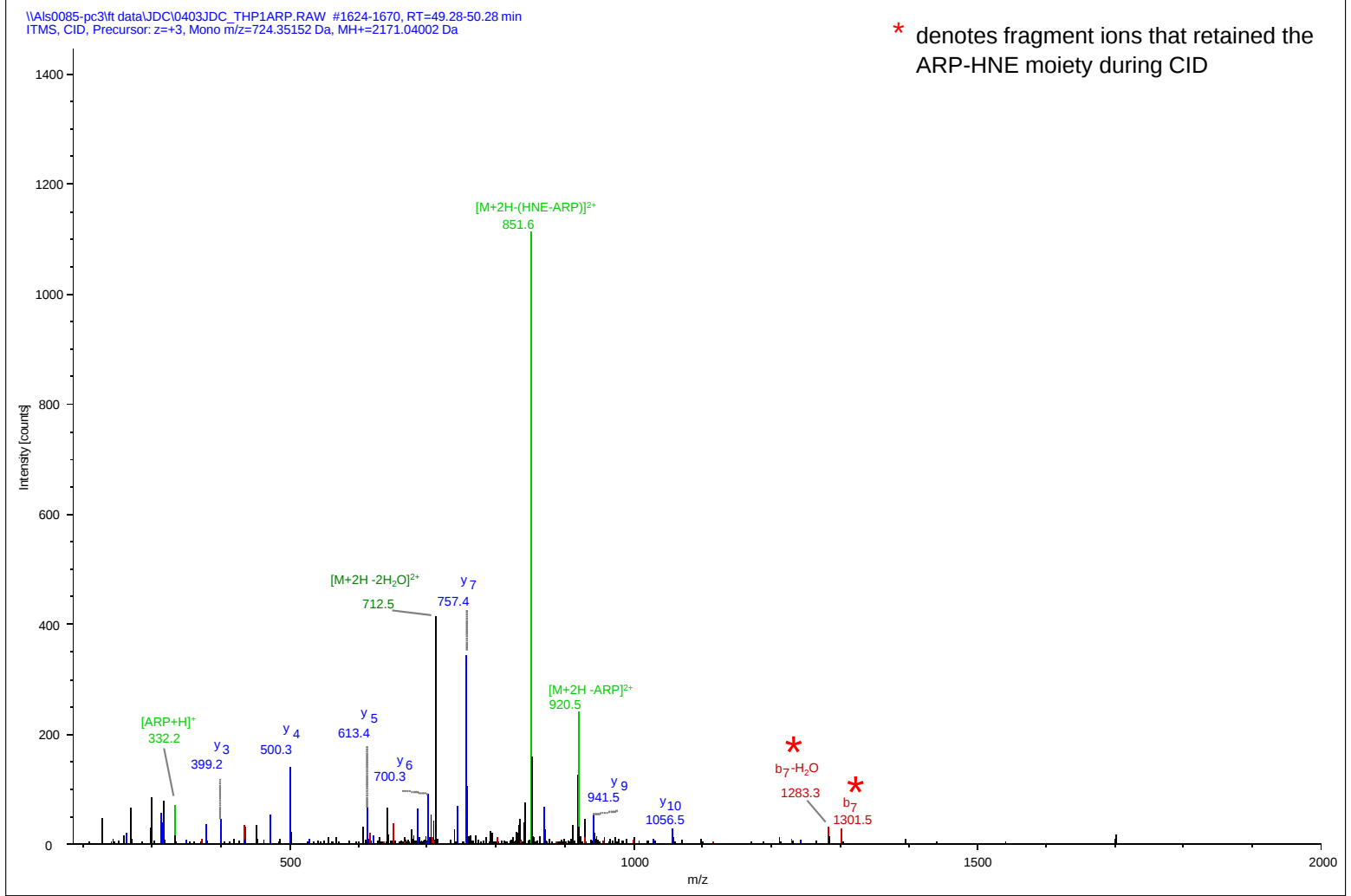
#	b	b ⁺⁺	b ⁺	b ⁺⁺⁺	b ⁰	b ⁰⁺⁺	Seq	y	y ⁺⁺	y ⁺	y ⁺⁺⁺	y ⁰	y ⁰⁺⁺	#
1	72.04	36.53					A							24
2	235.11	118.06					Y	3091.47	1546.24	3074.44	1537.72	3073.46	1537.23	23
3	372.17	186.59					H	2928.41	1464.71	2911.38	1456.19	2910.39	1455.70	22
4	501.21	251.11			483.20	242.10	E	2791.35	1396.18	2774.32	1387.66	2773.34	1387.17	21
5	629.27	315.14	612.24	306.62	611.26	306.13	Q	2662.30	1331.66	2645.28	1323.14	2644.29	1322.65	20
6	742.35	371.68	725.33	363.17	724.34	362.67	L	2534.25	1267.63	2517.22	1259.11	2516.23	1258.62	19
7	829.38	415.20	812.36	406.68	811.37	406.19	S	2421.16	1211.08	2404.13	1202.57	2403.15	1202.08	18
8	928.45	464.73	911.43	456.22	910.44	455.72	V	2334.13	1167.57	2317.10	1159.05	2316.12	1158.56	17
9	999.49	500.25	982.46	491.74	981.48	491.24	A	2235.06	1118.03	2218.03	1109.52	2217.05	1109.03	16
10	1128.53	564.77	1111.51	556.26	1110.52	555.76	E	2164.02	1082.52	2147.00	1074.00	2146.01	1073.51	15
11	1241.62	621.31	1224.59	612.80	1223.61	612.31	I	2034.98	1017.99	2017.95	1009.48	2016.97	1008.99	14
12	1342.66	671.84	1325.64	663.32	1324.65	662.83	T	1921.90	961.45	1904.87	952.94	1903.89	952.45	13
13	1456.71	728.86	1439.68	720.34	1438.70	719.85	N	1820.85	910.93	1803.82	902.41	1802.84	901.92	12
14	1517.74	764.38	1510.72	755.86	1509.73	755.37	A	1706.81	853.91	1689.78	845.39	1688.80	844.90	11
15	2099.99	1050.50	2082.96	1041.98	2081.98	1041.49	C	1635.77	818.39	1618.74	809.87	1617.76	809.38	10
16	2247.06	1124.03	2230.03	1115.52	2229.05	1115.03	F	1063.52	532.27	1046.50	523.75	1045.51	523.26	9
17	2376.10	1188.55	2359.07	1180.04	2358.09	1179.55	E	916.46	458.73	899.43	450.22	898.45	449.73	8
18	2473.15	1237.08	2456.13	1228.57	2455.14	1228.07	P	787.41	394.21	770.39	385.70			7
19	2544.19	1272.60	2527.16	1264.09	2526.18	1263.59	A	690.36	345.68	673.33	337.17			6
20	2658.23	1329.62	2641.21	1321.11	2640.22	1320.61	N	619.32	310.17	602.30	301.65			5
21	2786.29	1393.65	2769.26	1385.14	2768.28	1384.64	Q	508.28	253.14	488.25	244.63			4
22	2917.33	1459.17	2900.31	1450.66	2899.32	1450.16	M	377.22	189.11	360.20	180.60			3
23	3016.40	1508.70	2999.37	1500.19	2998.39	1499.70	V	246.18	123.59	229.15	115.08			2
24							K	147.11	74.06	130.09	65.55			1

Figure S3. (A) MS/MS spectrum and (B) Full scan mass spectrum of the $[M+3H]^{3+}$ ion of the ARP-HNE modified peptide AYHEQLSVAEITNAC*FEPANQMVK; monoisotopic m/z_{calc} 1054.8401; accuracy $\Delta(m/z) = 6.3$ ppm.

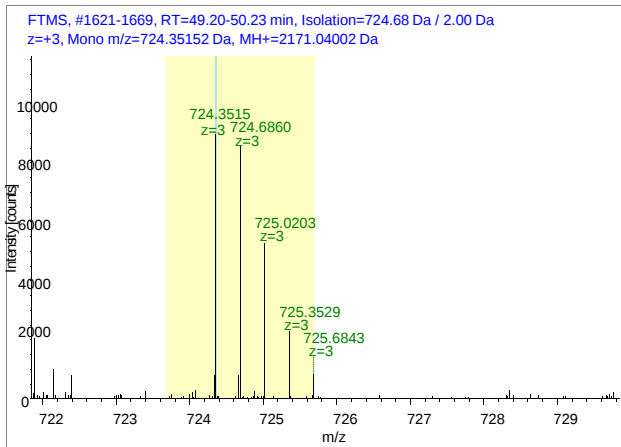


ACTN4_Human

A.



B.



Mascot search result for the MS/MS spectrum depicted above:

#	b	b ⁺⁺	b ⁺	b ⁺⁺⁺	b ⁰	b ⁰⁺⁺	Seq.	y	y ⁺⁺	y ⁺	y ⁺⁺⁺	y ⁰	y ⁰⁺⁺	#
1	114.09	57.55					I							15
2	686.34	343.67					C	2057.95	1029.48	2040.93	1020.97	2039.94	1020.47	14
3	801.36	401.19			783.35	392.18	D	1485.71	743.36	1468.68	734.84	1467.70	734.35	13
4	929.42	465.21	912.40	456.70	911.41	456.21	Q	1370.68	685.84	1353.65	677.33	1352.67	676.84	12
5	1115.50	558.25	1098.47	549.74	1097.49	549.25	W	1242.62	621.81	1225.60	613.30	1224.61	612.81	11
6	1230.53	615.77	1213.50	607.25	1212.52	606.76	D	1056.54	528.78	1039.52	520.26	1038.53	519.77	10
7	1301.57	651.29	1284.54	642.77	1283.55	642.28	A	941.52	471.26	924.49	462.75	923.51	462.26	9
8	1414.65	707.83	1397.62	699.32	1396.64	698.82	L	870.48	435.74	853.45	427.23	852.47	426.74	8
9	1471.67	736.34	1454.64	727.83	1453.66	727.33	G	757.40	379.20	740.37	370.69	739.38	370.20	7
10	1558.70	779.86	1541.68	771.34	1540.69	770.85	S	700.37	350.69	683.35	342.18	682.36	341.69	6
11	1671.79	836.40	1654.76	827.88	1653.78	827.39	L	613.34	307.17	596.32	298.66	595.33	298.17	5
12	1772.83	886.92	1755.81	878.41	1754.82	877.92	T	500.26	250.63	483.23	242.12	482.25	241.63	4
13	1909.89	955.45	1892.87	946.94	1891.88	946.45	H	399.21	200.11	382.18	191.60	381.20	191.10	3
14	1996.93	998.97	1979.90	990.45	1978.91	989.96	S	262.15	131.58	245.12	123.07	244.14	122.57	2
15							R	175.12	88.06	158.09	79.55			1

Figure S4. (A) MS/MS spectrum and (B) Full scan mass spectrum of the $[M+3H]^{3+}$ ion of the ARP-HNE modified peptide IC*DQWDALGSLTHSR; monoisotopic m/z_{calc} 724.3506, accuracy $\Delta(m/z) = 1.3$ ppm.

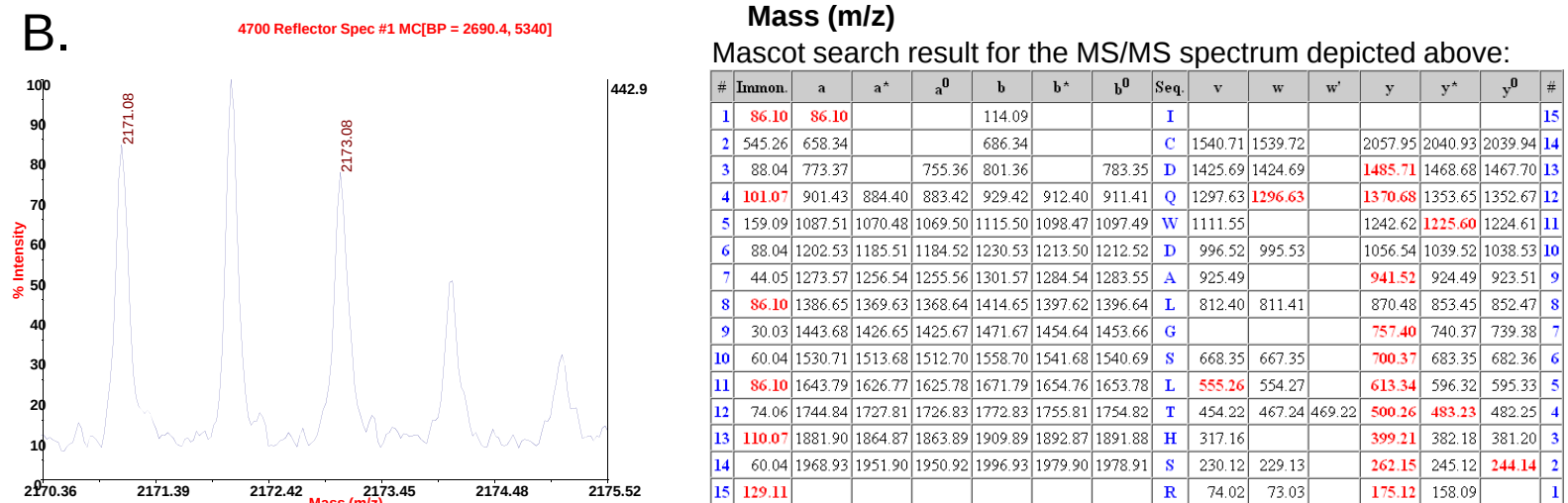
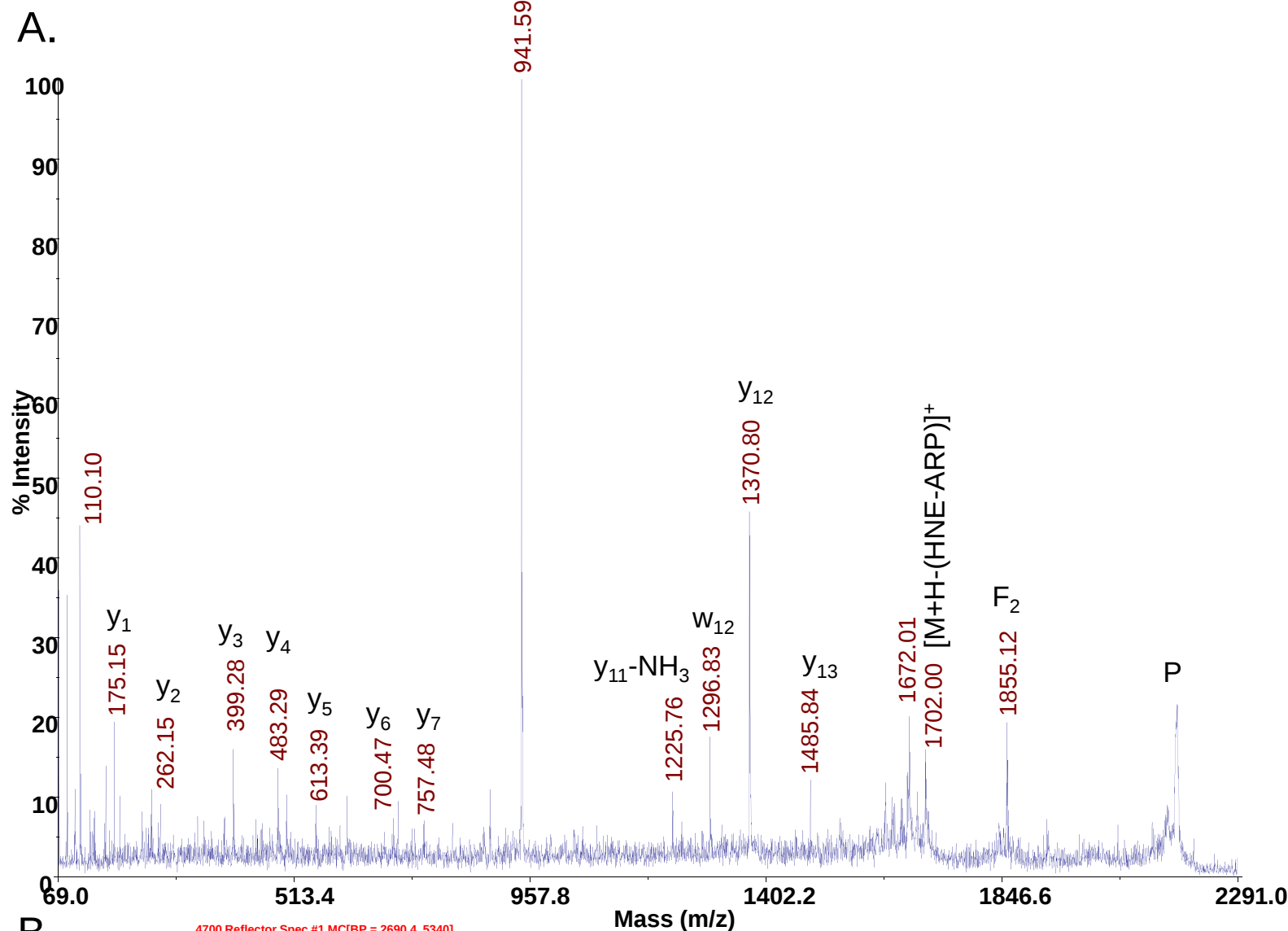
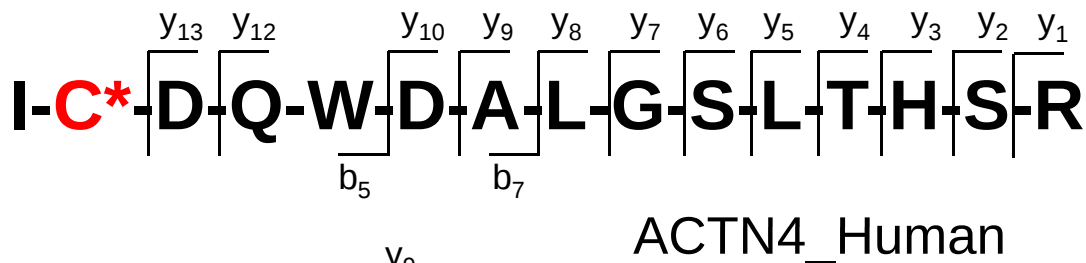
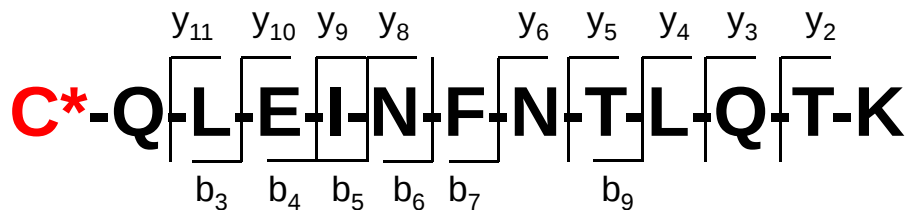
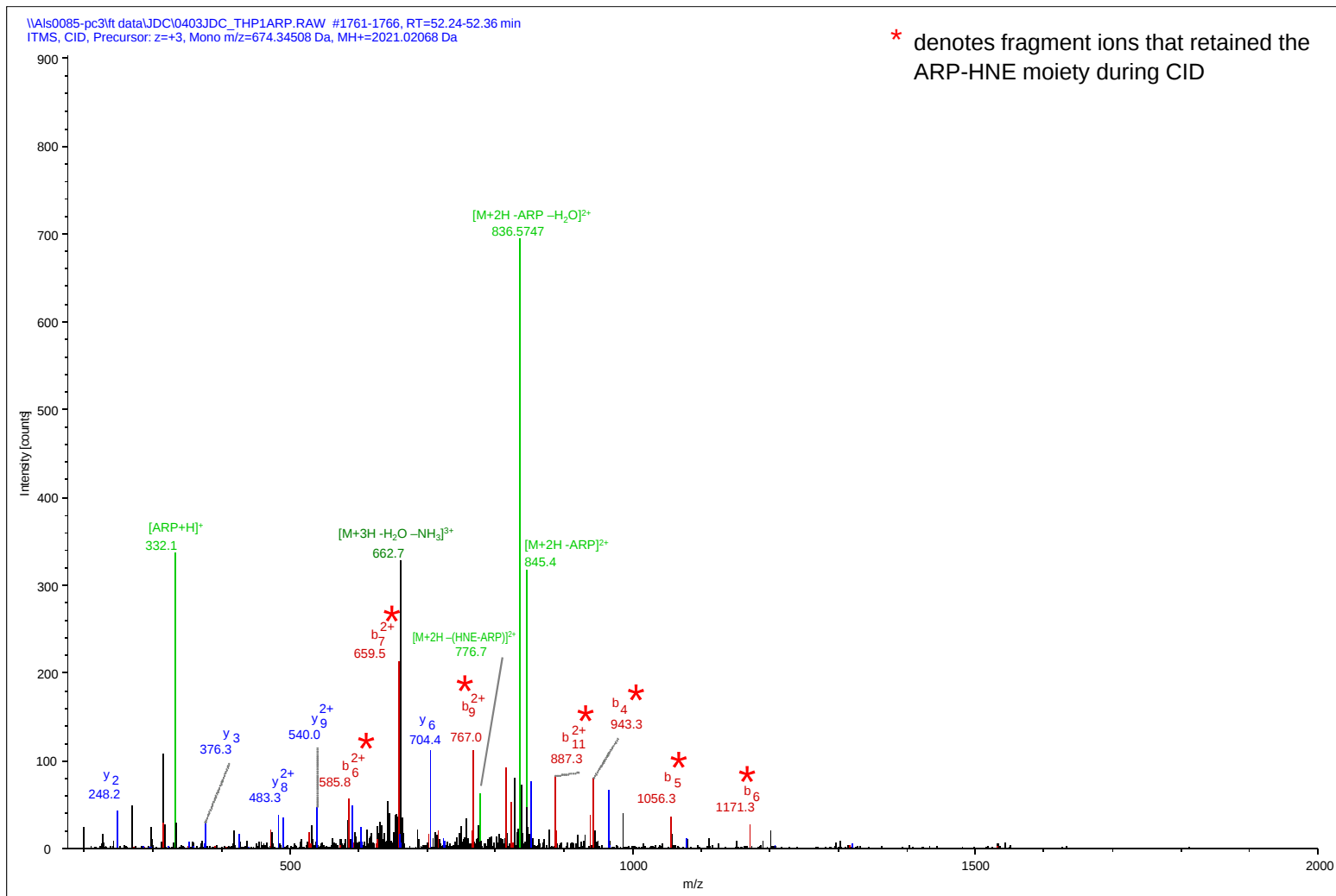


Figure S5. (A) MS/MS and (B) MS spectrum of the MH⁺ ion of the ARP-HNE modified peptide IC*DQWDALGSLTHSR obtained on the MALDI-TOF/TOF instrument; expected monoisotopic MH⁺ 2171.04 Da; mass accuracy Δm = 0.04 Da (20 ppm). P, precursor ion; F1 and F2, non-peptide fragment ions related to the ARP tag (see Figure S26).



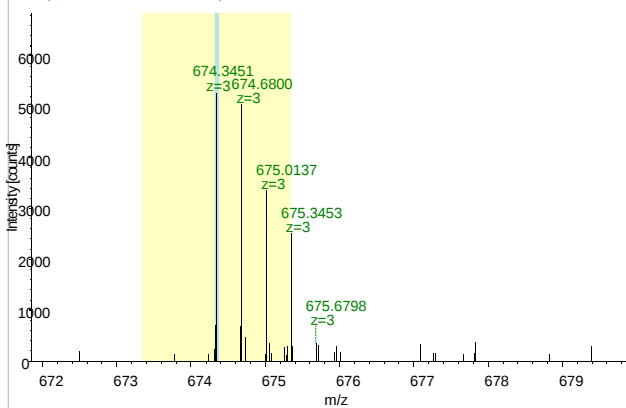
ACTN4_Human

A.



B.

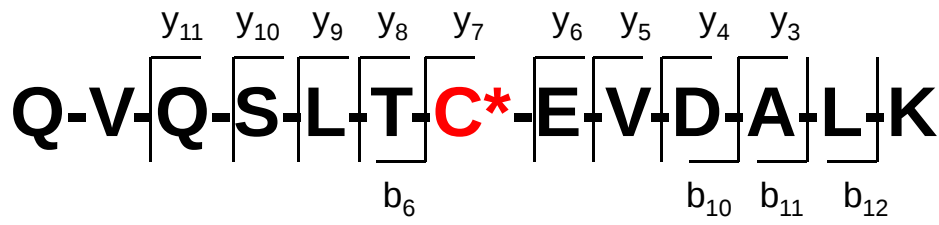
FTMS, #1759-1765, RT=52.18-52.31 min, Isolation=674.34 Da / 2.00 Da
 z=+3, Mono m/z=674.34508 Da, MH+=2021.02068 Da



Mascot search result for the fragment ion spectrum depicted above:

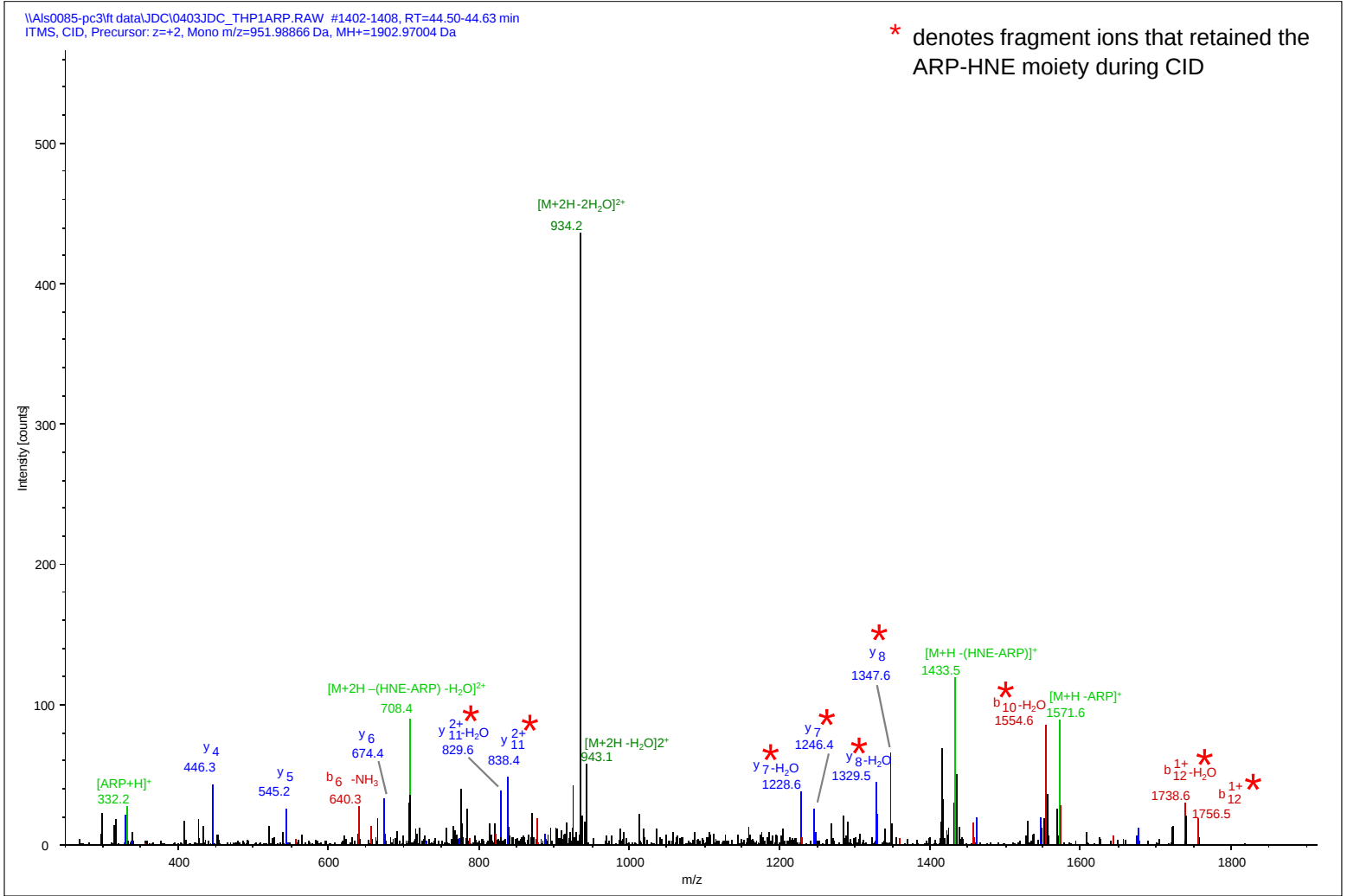
#	b	b ⁺⁺	b [*]	b ^{++*}	b ⁰	b ⁰⁺⁺	Seq.	y	y ⁺⁺	y [*]	y ^{++*}	y ⁰	y ⁰⁺⁺	#
1	573.25	287.13					C							13
2	701.31	351.16	684.28	342.65			Q	1448.77	724.89	1431.75	716.38	1430.76	715.89	12
3	814.39	407.70	797.37	399.19			L	1320.72	660.86	1303.69	652.35	1302.71	651.86	11
4	943.44	472.22	926.41	463.71	925.43	463.22	E	1207.63	604.32	1190.61	595.81	1189.62	595.31	10
5	1056.52	528.76	1039.50	520.25	1038.51	519.76	I	1078.59	539.80	1061.56	531.28	1060.58	530.79	9
6	1170.56	585.79	1153.54	577.27	1152.55	576.78	N	965.51	483.26	948.48	474.74	947.49	474.25	8
7	1317.63	659.32	1300.61	650.81	1299.62	650.31	F	851.46	426.23	834.44	417.72	833.45	417.23	7
8	1431.68	716.34	1414.65	707.83	1413.67	707.34	N	704.39	352.70	687.37	344.19	686.38	343.70	6
9	1532.72	766.87	1515.70	758.35	1514.71	757.86	T	590.35	295.68	573.32	287.17	572.34	286.67	5
10	1645.81	823.41	1628.78	814.89	1627.80	814.40	L	489.30	245.16	472.28	236.64	471.29	236.15	4
11	1773.87	887.44	1756.84	878.92	1755.86	878.43	Q	376.22	188.61	359.19	180.10	358.21	179.61	3
12	1874.91	937.96	1857.89	929.45	1856.90	928.96	T	248.16	124.58	231.13	116.07	230.15	115.58	2
13							K	147.11	74.06	130.09	65.55			1

Figure S6. (A) MS/MS spectrum and (B) Full scan mass spectrum of the $[M+3H]^{3+}$ ion of the ARP-HNE modified peptide C*QLEINFNTLQTK; monoisotopic m/z_{calc} 674.3447; accuracy $\Delta(m/z)$ = 0.6 ppm.

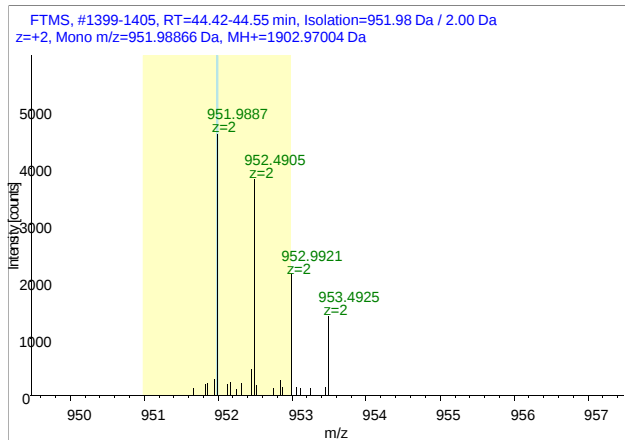


VIME_Human

A.



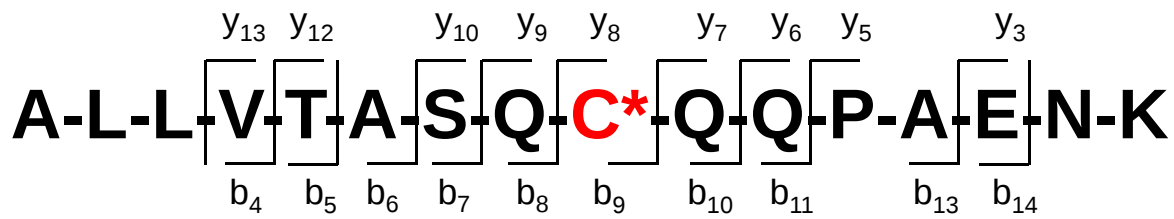
B.



Mascot search result for the MS/MS spectrum depicted above:

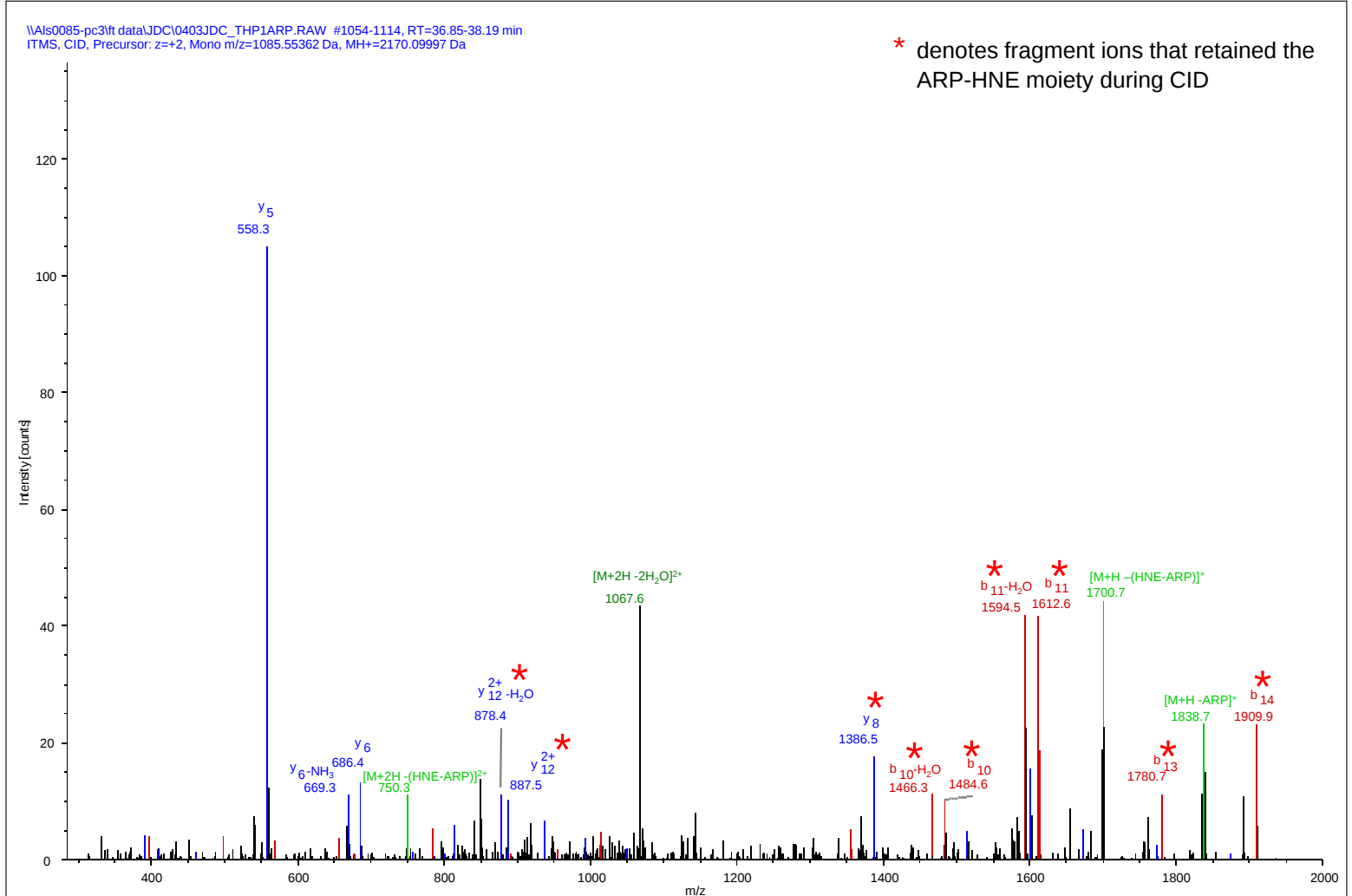
#	b	b ⁺⁺	b ⁺	b ⁺⁺	b ⁰	b ⁰⁺⁺	Seq.	y	y ⁺⁺	y ⁺	y ⁺⁺	y ⁰	y ⁰⁺⁺	#
1	129.07	65.04	112.04	56.52			Q							13
2	228.13	114.57	211.11	106.06			V	1774.91	887.96	1757.88	879.44	1756.90	878.95	12
3	356.19	178.60	339.17	170.09			Q	1675.84	838.42	1658.81	829.91	1657.83	829.42	11
4	443.22	222.12	426.20	213.60	425.21	213.11	S	1547.78	774.39	1530.75	765.88	1529.77	765.39	10
5	556.31	278.66	539.28	270.14	538.30	269.65	L	1460.75	730.88	1443.72	722.36	1442.74	721.87	9
6	657.36	329.18	640.33	320.67	639.35	320.18	T	1347.66	674.34	1330.64	665.82	1329.65	665.33	8
7	1229.60	615.30	1212.58	606.79	1211.59	606.30	C	1246.62	623.81	1229.59	615.30	1228.61	614.81	7
8	1358.64	679.83	1341.62	671.31	1340.63	670.82	E	674.37	337.69	657.35	329.18	656.36	328.68	6
9	1457.71	729.36	1440.69	720.85	1439.70	720.35	V	545.33	273.17	528.30	264.66	527.32	264.16	5
10	1572.74	786.87	1555.71	778.36	1554.73	777.87	D	446.26	223.63	429.23	215.12	428.25	214.63	4
11	1643.78	822.39	1626.75	813.88	1625.77	813.39	A	331.23	166.12	314.21	157.61			3
12	1756.86	878.93	1739.83	870.42	1738.85	869.93	L	260.20	130.60	243.17	122.09			2
13							K	147.11	74.06	130.09	65.55			1

Figure S9. (A) MS/MS spectrum and (B) Full scan mass spectrum of the $[M+2H]^{2+}$ ion of the ARP-HNE modified peptide QVQSLTC*EVDALK; monoisotopic m/z_{calc} 951.9868; accuracy $\Delta(m/z) = 2.0$ ppm.

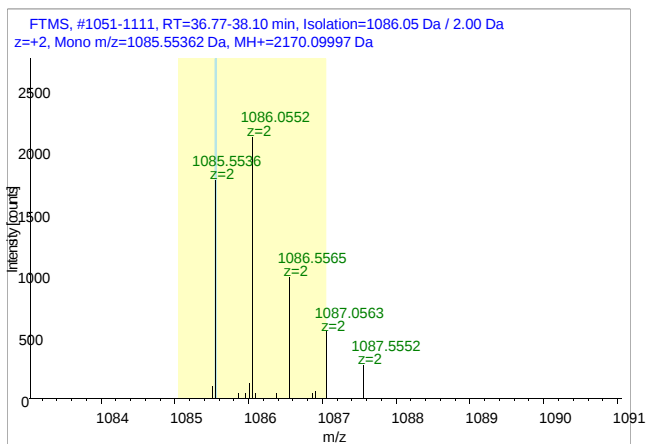


CAP1_Human

A.



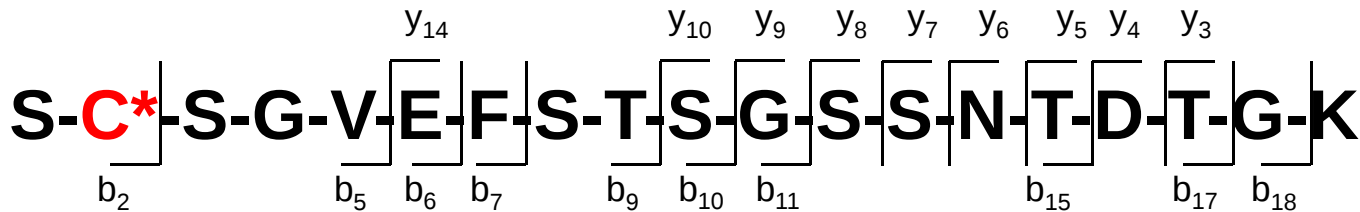
B.



Mascot search result for the MS/MS spectrum depicted above:

#	b	b ⁺⁺	b [*]	b ⁺⁺⁺	b ⁰	b ⁰⁺⁺	Seq.	y	y ⁺⁺	y [*]	y ⁺⁺⁺	y ⁰	y ⁰⁺⁺	#
1	72.04	36.53					A							16
2	185.13	93.07					L	2099.06	1050.03	2082.04	1041.52	2081.05	1041.03	15
3	298.21	149.61					L	1985.98	993.49	1968.95	984.98	1967.97	984.49	14
4	397.28	199.14					V	1872.89	936.95	1855.87	928.44	1854.88	927.95	13
5	498.33	249.67			480.32	240.66	T	1773.83	887.42	1756.80	878.90	1755.82	878.41	12
6	569.37	285.19			551.36	276.18	A	1672.78	836.89	1655.75	828.38	1654.77	827.89	11
7	656.40	328.70			638.39	319.70	S	1601.74	801.37	1584.71	792.86	1583.73	792.37	10
8	784.46	392.73	767.43	384.22	766.45	383.73	Q	1514.71	757.86	1497.68	749.34	1496.70	748.85	9
9	1356.70	678.85	1339.67	670.34	1338.69	669.85	C	1386.65	693.83	1369.62	685.32	1368.64	684.82	8
10	1484.76	742.88	1467.73	734.37	1466.75	733.88	Q	814.41	407.71	797.38	399.19	796.39	398.70	7
11	1612.82	806.91	1595.79	798.40	1594.81	797.91	Q	686.35	343.68	669.32	335.16	668.34	334.67	6
12	1709.87	855.44	1692.84	846.93	1691.86	846.43	P	558.29	279.65	541.26	271.13	540.28	270.64	5
13	1780.91	890.96	1763.88	882.44	1762.90	881.95	A	461.24	231.12	444.21	222.61	443.22	222.12	4
14	1909.95	955.48	1892.92	946.97	1891.94	946.47	E	390.20	195.60	373.17	187.09	372.19	186.60	3
15	2023.99	1012.50	2006.97	1003.99	2005.98	1003.50	N	261.16	131.08	244.13	122.57			2
16							K	147.11	74.06	130.09	65.55			1

Figure S10. (A) MS/MS spectrum and (B) Full Scan mass spectrum of the $[M+2H]^{2+}$ ion of the ARP-HNE modified peptide ALLVTASQC*QQPAENK; monoisotopic m/z_{calc} 1085.5534, accuracy $\Delta(m/z) = 0.2$ ppm.



A. VDAC2_Human

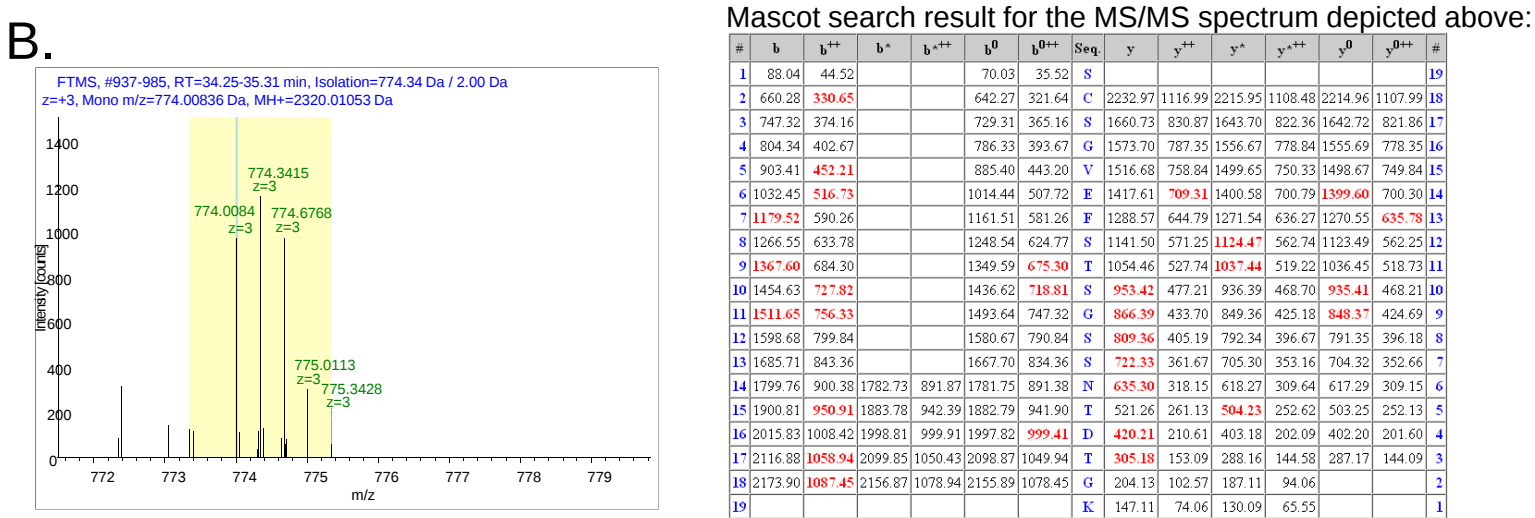
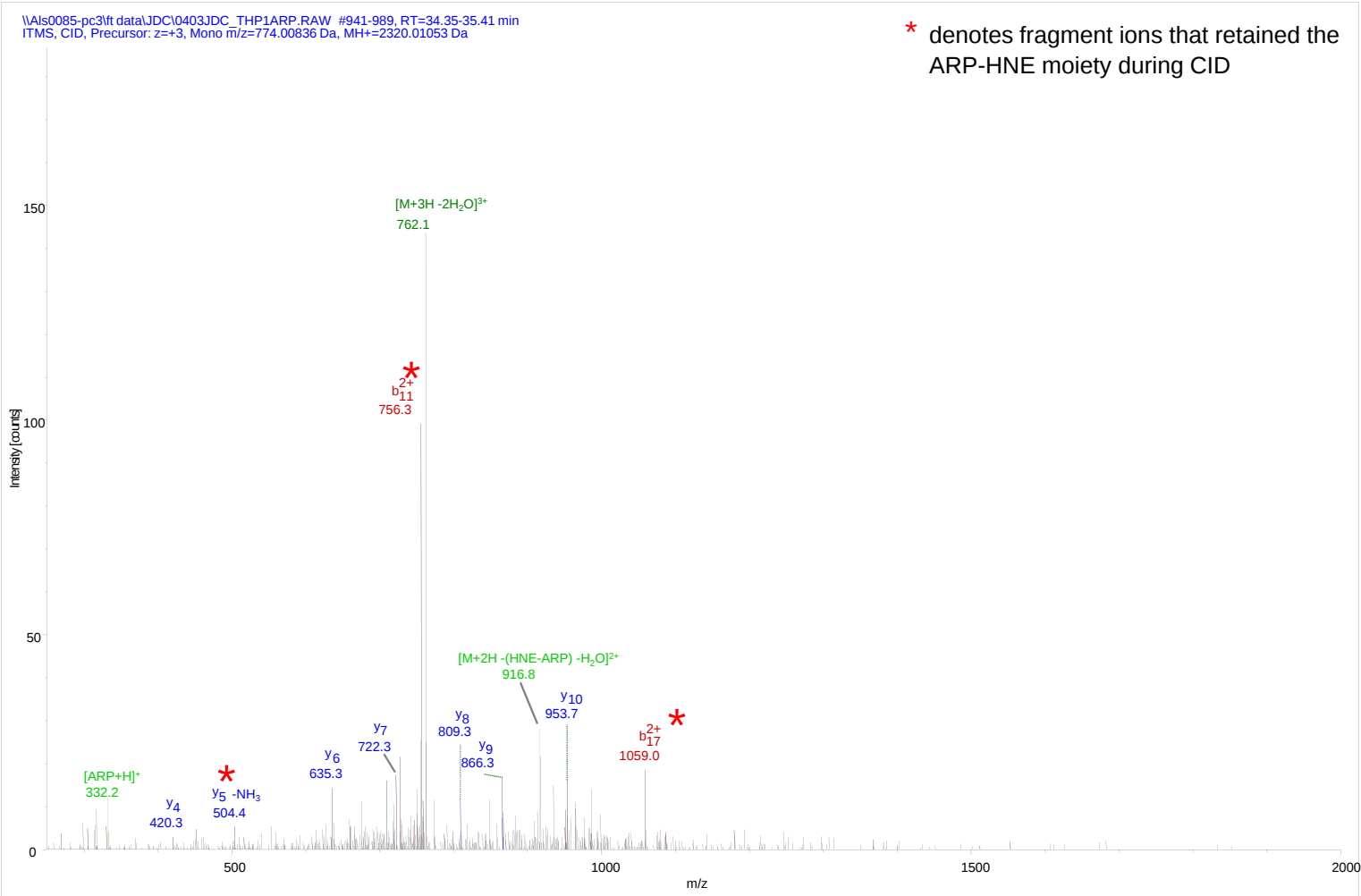
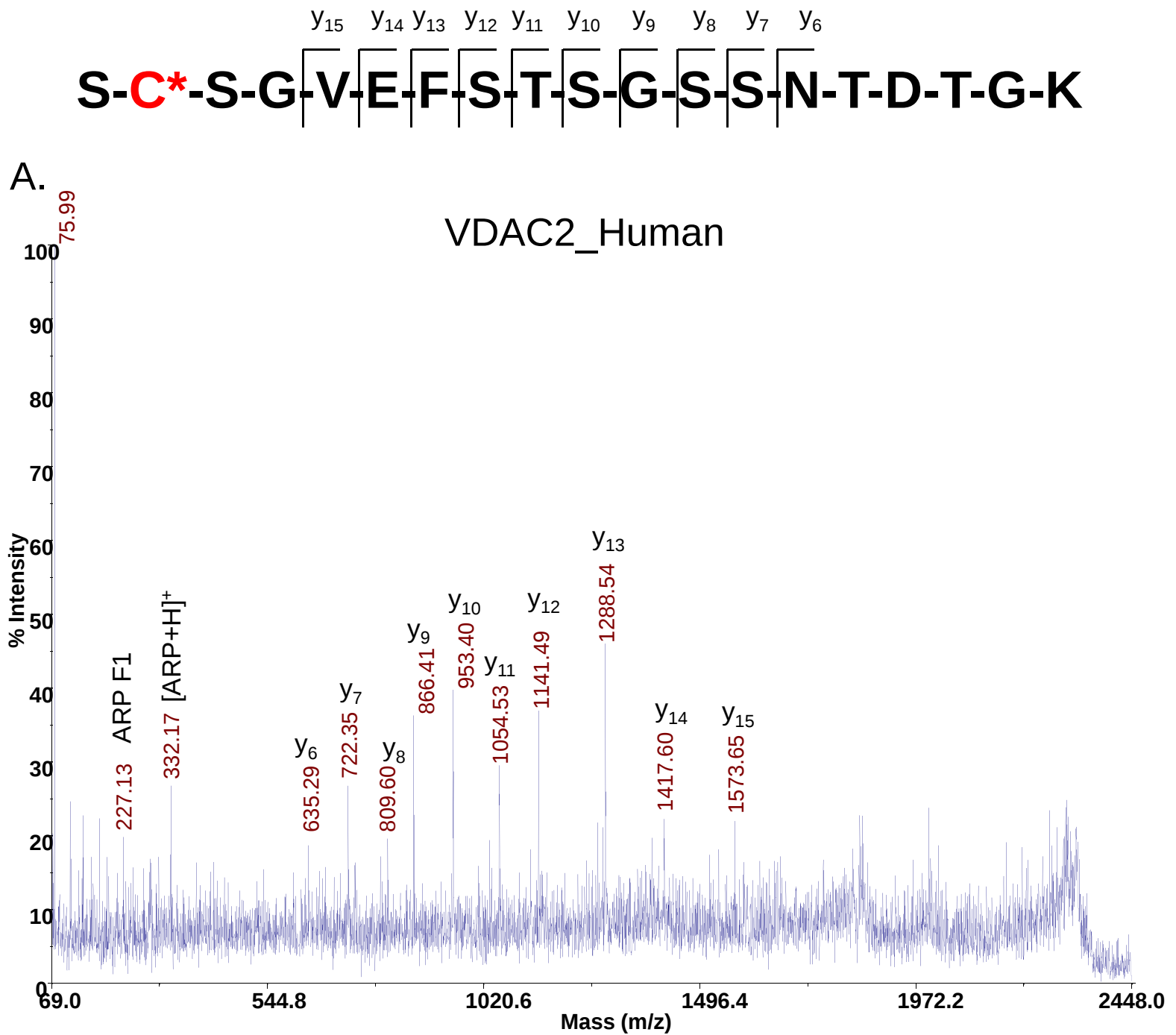
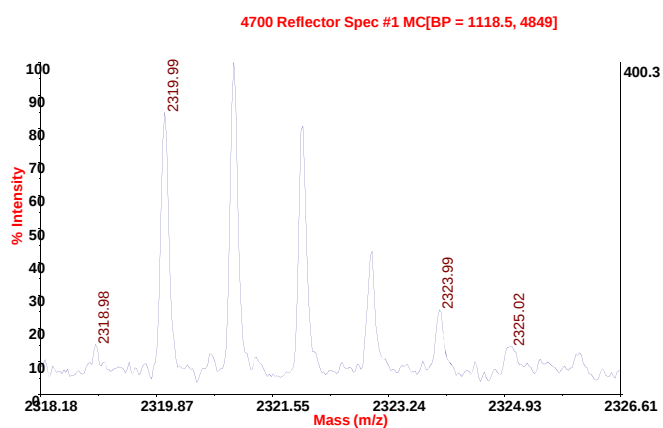


Figure S11. (A) MS/MS spectrum and (B) Full scan mass spectrum of the $[M+3H]^{3+}$ ion of the ARP-HNE modified peptide SC*SGVEFSTSGSSNTDTGK; monoisotopic m/z_{calc} 774.0071; accuracy $\Delta(m/z)$ = 1.7 ppm.

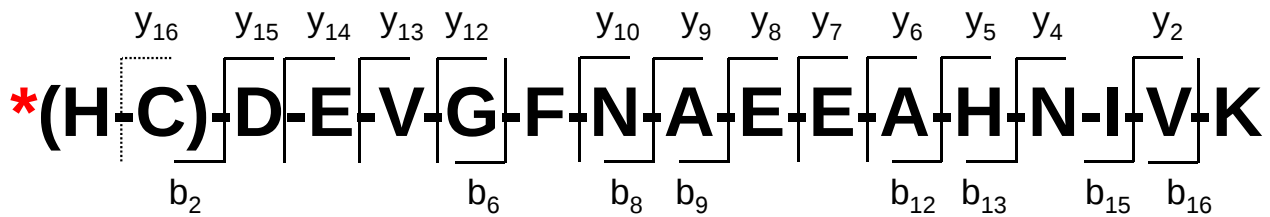


B. Mascot search result for the MS/MS spectrum depicted above:



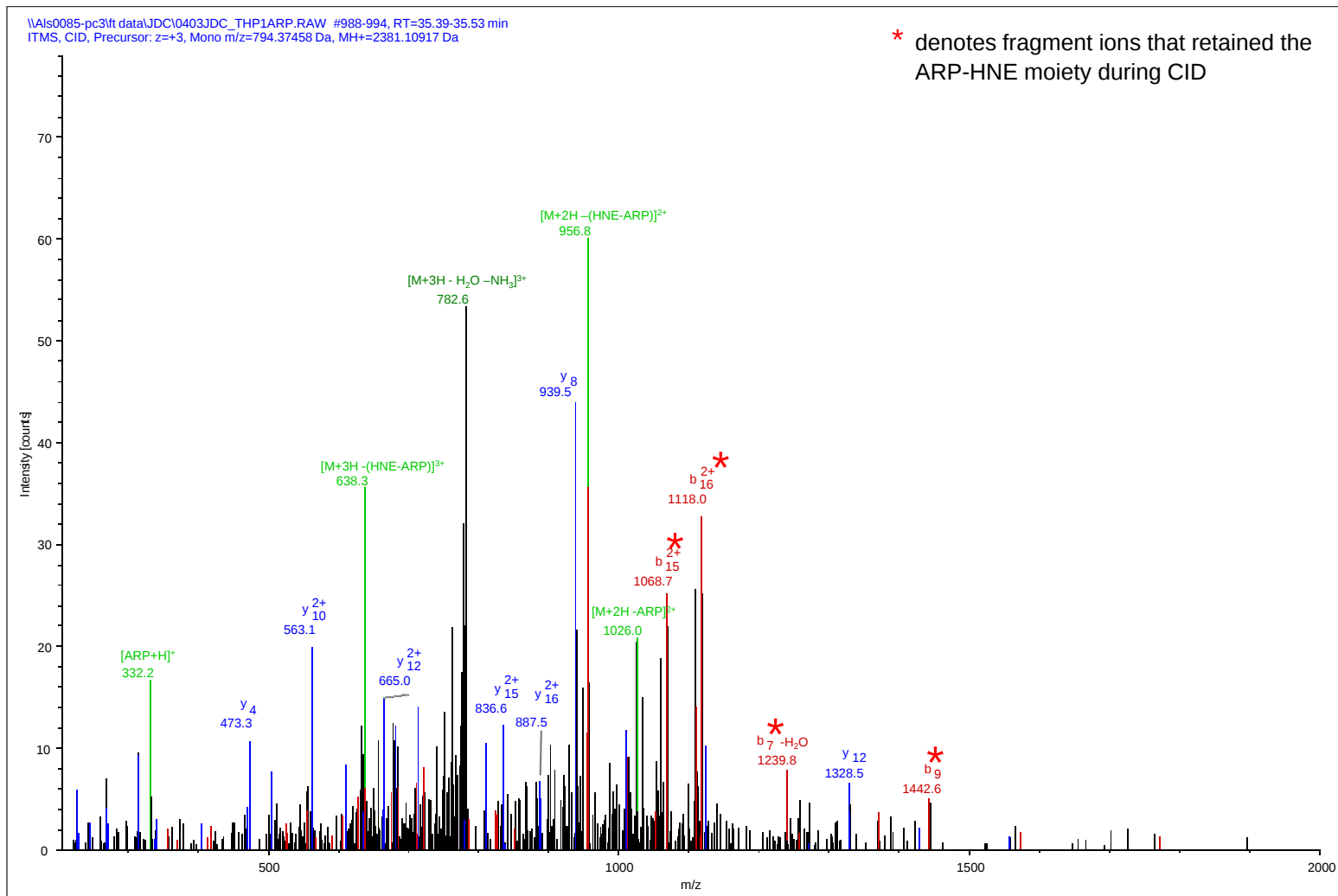
#	Ion	a	a ⁺	a ⁰	b	b ⁺	b ⁰	Seq	y	y ⁺	y ⁰	#
1	60.04	60.04		42.03	88.04		70.03	S				19
2	545.26	632.29		614.28	660.28		642.27	C	2232.97	2215.95	2214.96	18
3	60.04	719.32		701.31	747.32		729.31	S	1660.73	1643.70	1642.72	17
4	30.03	776.34		758.33	804.34		786.33	G	1573.70	1556.67	1555.69	16
5	72.08	875.41		857.40	903.41		885.40	V	1516.68	1499.65	1498.67	15
6	102.05	1004.45		986.44	1032.45		1014.44	E	1417.61	1400.58	1399.60	14
7	120.08	1151.52		1133.51	1179.52		1161.51	F	1288.57	1271.54	1270.55	13
8	60.04	1238.55		1220.54	1266.55		1248.54	S	1141.50	1124.47	1123.49	12
9	74.06	1339.60		1321.59	1367.60		1349.59	T	1054.46	1037.44	1036.45	11
10	60.04	1426.63		1408.62	1454.63		1436.62	S	953.42	936.39	935.41	10
11	30.03	1483.66		1465.65	1511.65		1493.64	G	866.39	849.36	848.37	9
12	60.04	1570.69		1552.68	1598.68		1580.67	S	809.36	792.34	791.35	8
13	60.04	1657.72		1639.71	1685.71		1667.70	S	722.33	705.30	704.32	7
14	87.06	1771.76	1754.74	1753.75	1799.76	1782.73	1781.75	N	635.30	618.27	617.29	6
15	74.06	1872.81	1855.78	1854.80	1900.81	1883.78	1882.79	T	521.26	504.23	503.25	5
16	88.04	1987.84	1970.81	1969.83	2015.83	1998.81	1997.82	D	420.21	403.18	402.20	4
17	74.06	2088.88	2071.86	2070.87	2116.88	2099.85	2098.87	T	305.18	288.16	287.17	3
18	30.03	2145.91	2128.88	2127.90	2173.90	2156.87	2155.89	G	204.13	187.11		2
19	101.11							K	147.11	130.09		1

Figure S12. (A) MS/MS and (B) MS spectrum of the MH⁺ ion of the ARP-HNE modified peptide SC*SGVEFSTSGSSNTDTGK obtained on the MALDI-TOF/TOF instrument; expected monoisotopic MH⁺ 2320.01 Da; mass accuracy $\Delta m = -0.02$ Da (-7 ppm). P, precursor ion; F1 and F2, non-peptide fragment ions related to the ARP tag (see Figure S26).



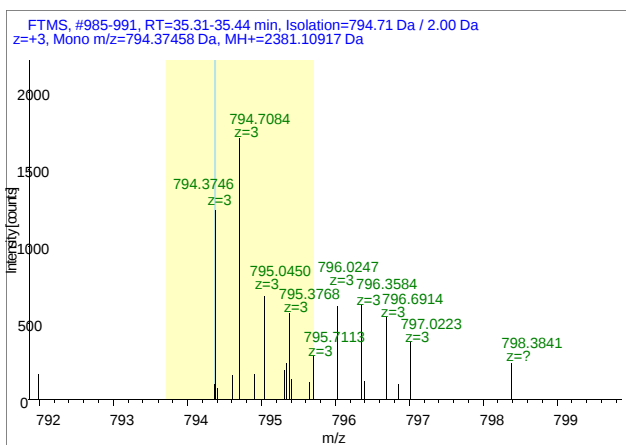
DYLT3_Human

A.



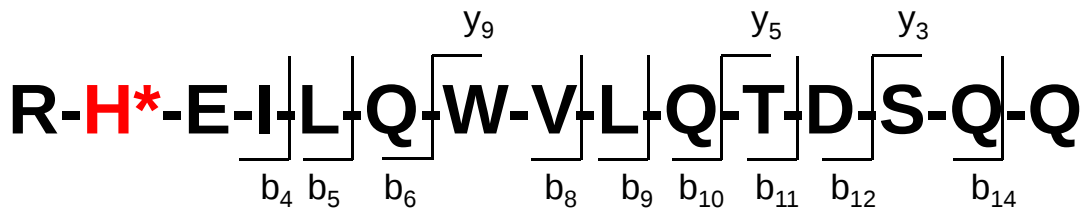
B.

Mascot search result for the MS/MS spectrum depicted above:



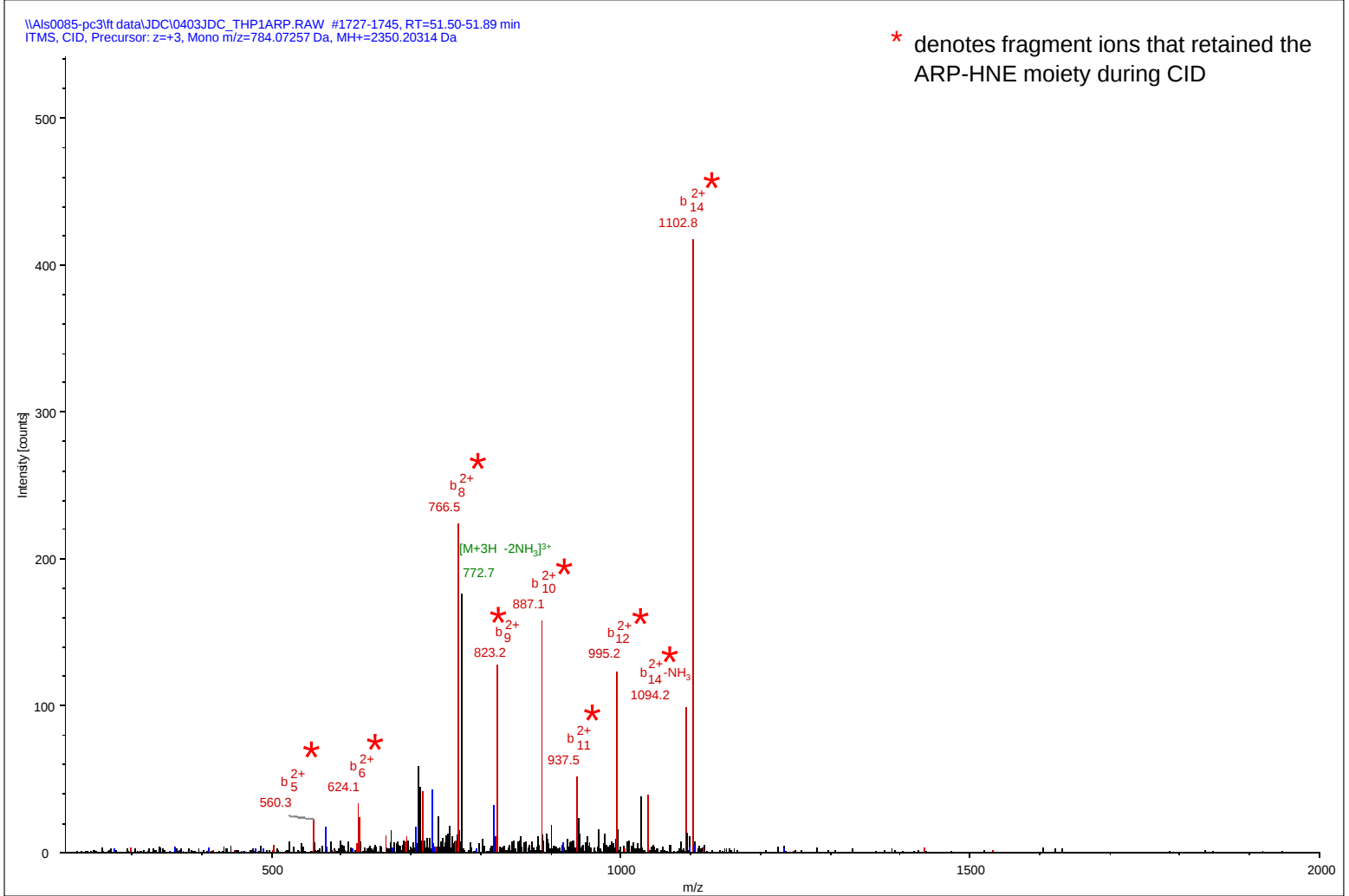
#	b	b ⁺⁺	b [*]	b ⁺⁺	b ⁰	b ⁰⁺⁺	Seq.	y	y ⁺⁺	y [*]	y ⁺⁺	y ⁰	y ⁰⁺⁺	#
1	607.30	304.15					H							17
2	710.31	355.66					C	1774.81	887.91	1757.78	879.39	1756.80	878.90	16
3	825.34	413.17			807.33	404.17	D	1671.80	836.40	1654.77	827.89	1653.79	827.40	15
4	954.38	477.69			936.37	468.69	E	1556.77	778.89	1539.74	770.38	1538.76	769.88	14
5	1053.45	527.23			1035.44	518.22	V	1427.73	714.37	1410.70	705.85	1409.72	705.36	13
6	1110.47	555.74			1092.46	546.73	G	1328.66	664.83	1311.63	656.32	1310.65	655.83	12
7	1257.54	629.27			1239.53	620.27	F	1271.64	636.32	1254.61	627.81	1253.63	627.32	11
8	1371.58	686.29	1354.56	677.78	1353.57	677.29	N	1124.57	562.79	1107.54	554.28	1106.56	553.78	10
9	1442.62	721.81	1425.59	713.30	1424.61	712.81	A	1010.53	505.77	993.50	497.25	992.52	496.76	9
10	1571.66	786.33	1554.64	777.82	1553.65	777.33	E	939.49	470.25	922.46	461.74	921.48	461.24	8
11	1700.70	850.86	1683.68	842.34	1682.69	841.85	E	810.45	405.73	793.42	397.21	792.44	396.72	7
12	1771.74	886.37	1754.71	877.86	1753.73	877.37	A	681.40	341.21	664.38	332.69			6
13	1908.80	954.90	1891.77	946.39	1890.79	945.90	H	610.37	305.69	593.34	297.17			5
14	2022.84	1011.93	2005.82	1003.41	2004.83	1002.92	N	473.31	237.16	456.28	228.64			4
15	2135.93	1068.47	2118.90	1059.95	2117.92	1059.46	I	359.27	180.14	342.24	171.62			3
16	2235.00	1118.00	2217.97	1109.49	2216.99	1109.00	V	246.18	123.59	229.15	115.08			2
17							K	147.11	74.06	130.09	65.55			1

Figure S13. (A) MS/MS and (B) Full scan mass spectrum of the $[M+3H]^{3+}$ ion of the ARP-HNE modified peptide (HC)*DEVGFNAEEAHNIVK; monoisotopic m/z_{calc} 794.3719; accuracy $\Delta(m/z) = 3.4$ ppm. Note, the low abundant y_{16} (2+) fragment ion at m/z 877.5 may indicate adduction of the N-terminal His residue.



LRC59_Human

A.



B.

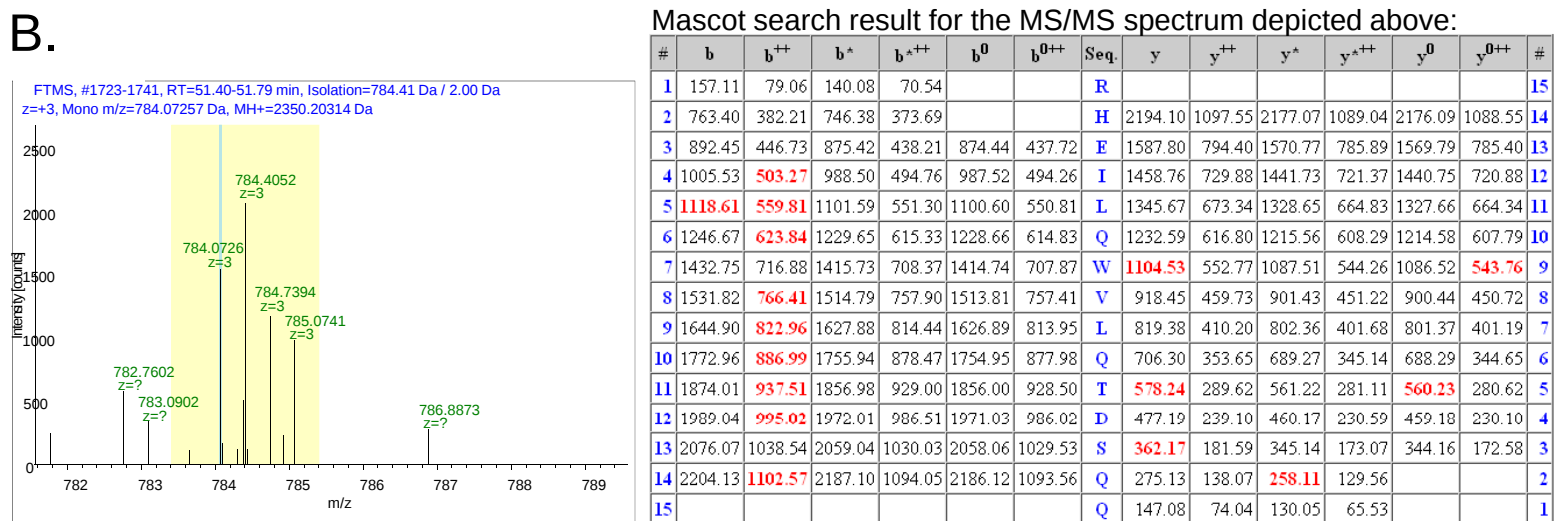
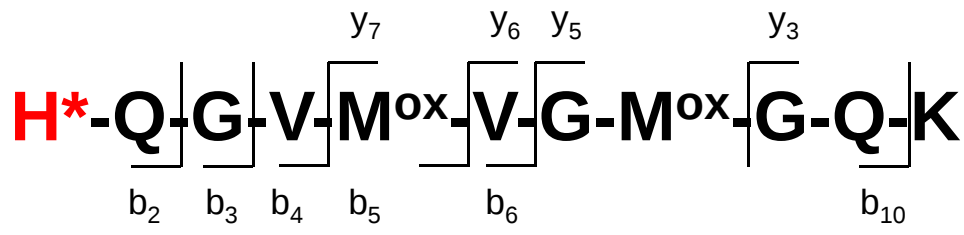
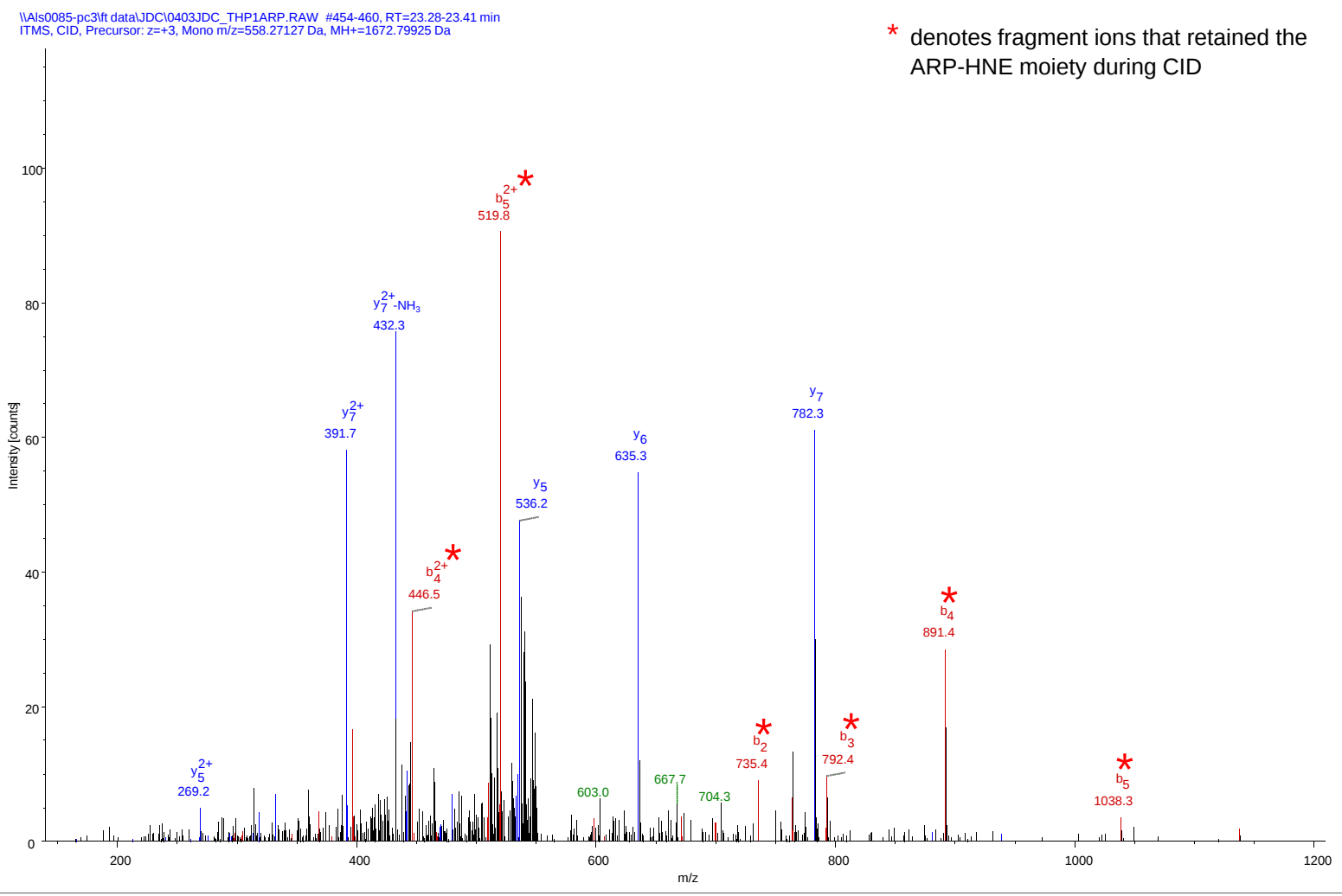


Figure S14. (A) MS/MS spectrum and (B) Full scan mass spectrum of the $[M+3H]^{3+}$ ion of the ARP-HNE modified peptide RH*EILQWVLQTD SQQ; monoisotopic m/z_{calc} 784.0706; accuracy $\Delta(m/z)$ = 2.6 ppm.



ACTB_Human

A.



B.

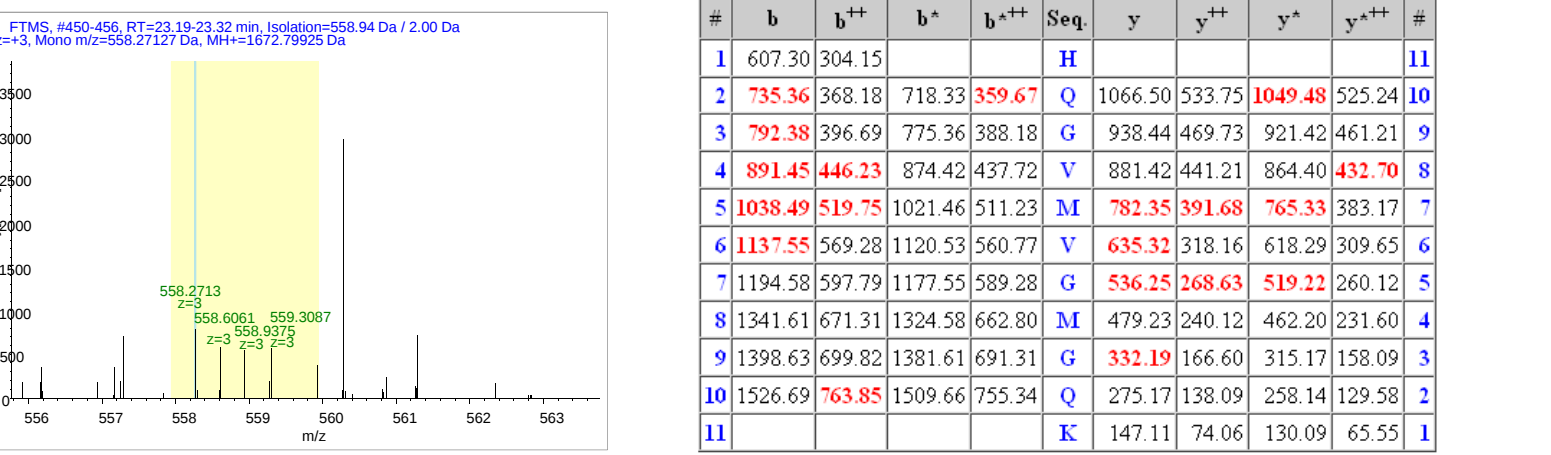
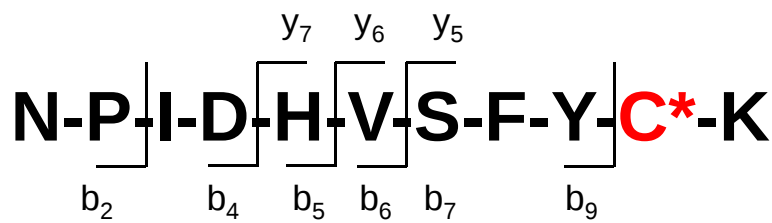
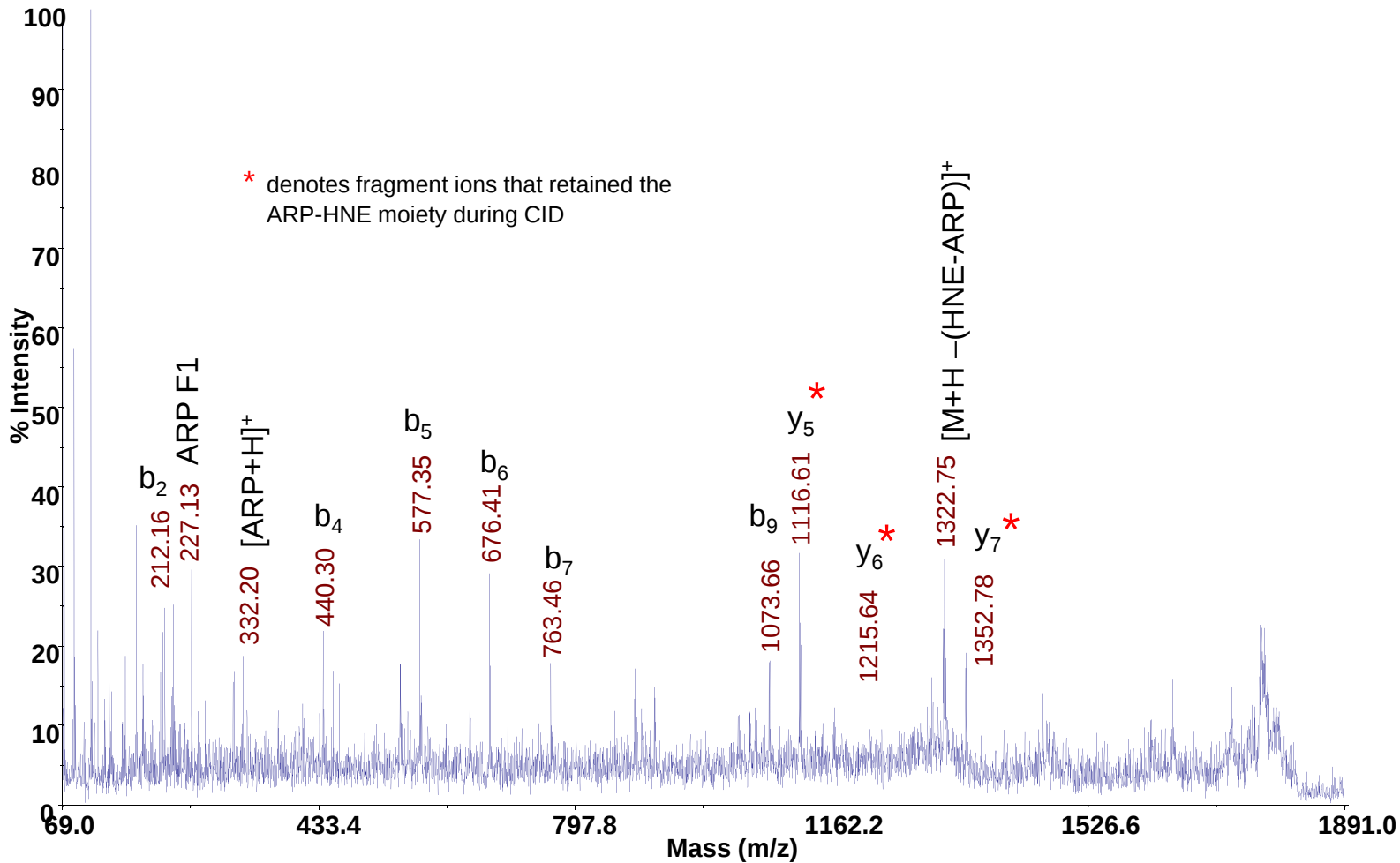


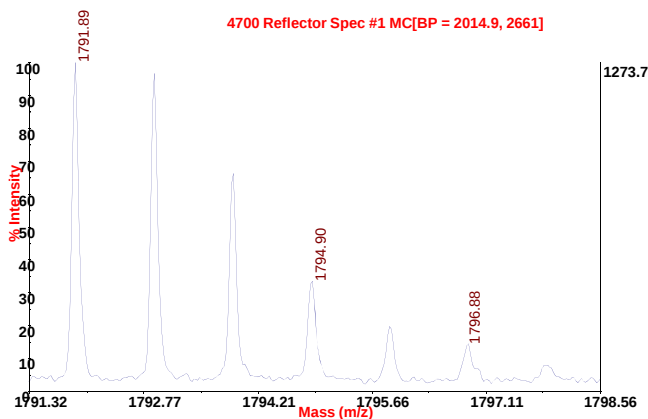
Figure S16. (A) MS/MS spectrum and (B) Full scan mass spectrum of the $[M+3H]^{3+}$ ion of the ARP-HNE modified peptide H*QGVMVGMGQK; monoisotopic m/z_{calc} 558.2704; accuracy $\Delta(m/z)$ = 1.5 ppm.



A. SAMH1_Human



B.



Mascot search result for the MS/MS spectrum depicted above:

#	Ion	a	a*	a ⁰	b	b*	b ⁰	Seq.	y	y*	y ⁰	#
1	87.06	87.06	70.03		115.05	98.02		N				11
2	70.07	184.11	167.08		212.10	195.08		P	1677.81	1660.79	1659.80	10
3	86.10	297.19	280.17		325.19	308.16		I	1580.76	1563.73	1562.75	9
4	88.04	412.22	395.19	394.21	440.21	423.19	422.20	D	1467.68	1450.65	1449.67	8
5	110.07	549.28	532.25	531.27	577.27	560.25	559.26	H	1352.65	1335.62	1334.64	7
6	72.08	648.35	631.32	630.34	676.34	659.31	658.33	V	1215.59	1198.56	1197.58	6
7	60.04	735.38	718.35	717.37	763.37	746.35	745.36	S	1116.52	1099.50	1098.51	5
8	120.08	882.45	865.42	864.44	910.44	893.42	892.43	F	1029.49	1012.46		4
9	136.08	1045.51	1028.48	1027.50	1073.51	1056.48	1055.49	Y	882.42	865.39		3
10	76.02	1148.52	1131.49	1130.51	1176.51	1159.49	1158.50	C	719.36	702.33		2
11	570.34							K	616.35	599.32		1

Figure S17. (A) MS/MS and (B) mass spectrum of the MH⁺ ion of the ARP-HNE modified peptide NPIDHVSFYC*K obtained on the MALDI-TOF/TOF instrument; expected monoisotopic MH⁺ 1791.86 Da; $\Delta m = 0.03$ Da (19 ppm). P, precursor ion; F1 and F2, non-peptide fragment ions related to the ARP tag (see Figure S26).



#	Immon.	a	a ⁰	b	b ⁰	Seq.	y	y [*]	y ⁰	#
1	72.08	72.08		100.08		V				11
2	74.06	173.13	155.12	201.12	183.11	T	1637.83	1620.80	1619.82	10
3	88.04	288.16	270.14	316.15	298.14	D	1536.78	1519.75	1518.77	9
4	88.04	403.18	385.17	431.18	413.17	D	1421.75	1404.73	1403.74	8
5	86.10	516.27	498.26	544.26	526.25	L	1306.73	1289.70		7
6	72.08	615.33	597.32	643.33	625.32	V	1193.64	1176.62		6
7	76.02	718.34	700.33	746.34	728.33	C	1094.57	1077.55		5
8	86.10	831.43	813.42	859.42	841.41	L	991.56	974.54		4
9	72.08	930.50	912.49	958.49	940.48	V	878.48	861.45		3
10	136.08	1093.56	1075.55	1121.55	1103.54	Y	779.41	762.39		2
11	570.34					K	616.35	599.32		1

Figure S18. (A) MS/MS and (B) MS spectrum of the MH^+ ion of the ARP-HNE modified peptide VTDDLVC*LVYK obtained on the MALDI-TOF/TOF instrument; expected monoisotopic MH^+ 1736.90 Da; mass accuracy $\Delta m = 0.17$ Da (94 ppm). F1 and F2, non-peptide fragment ions related to the ARP tag (see Figure S26).

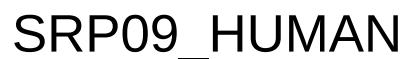
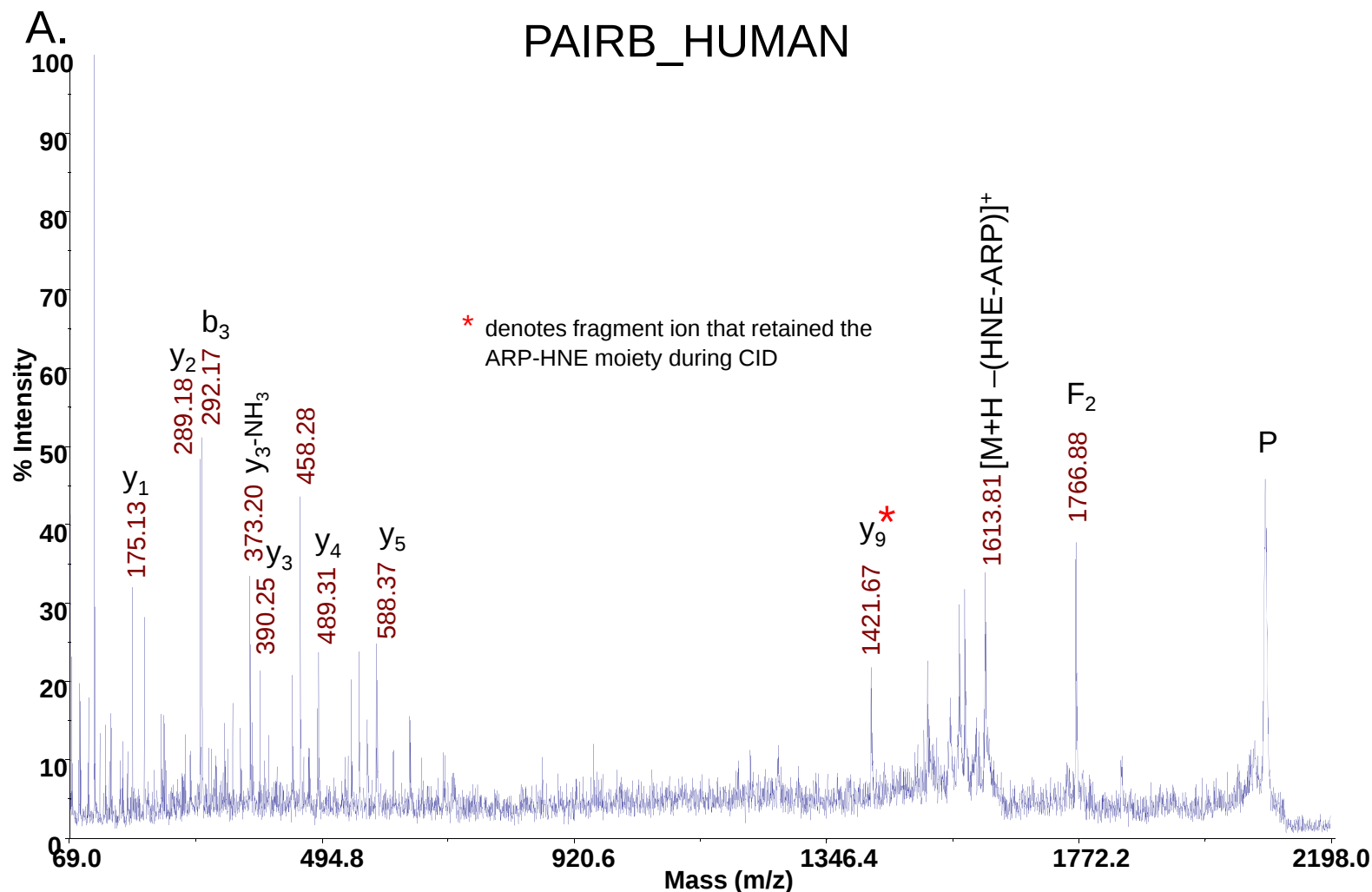
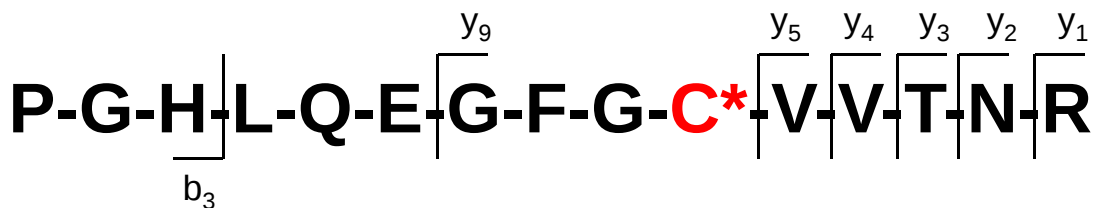
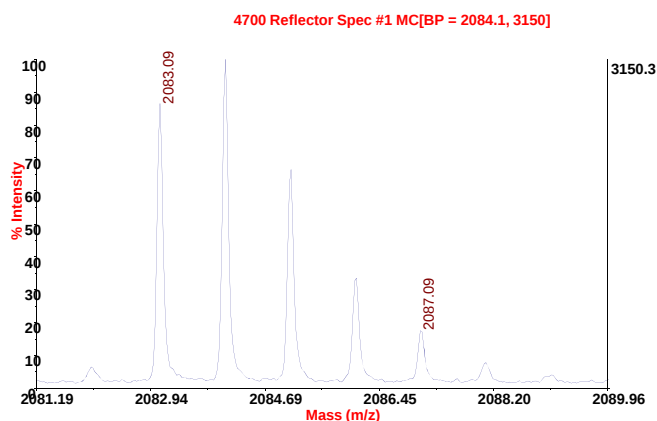


Figure S19. (A) MS/MS and (B) Full scan mass spectrum of the $[M+2H]^{2+}$ ion of the ARP-HNE modified peptide VTDDLVC*LVYK; monoisotopic m/z_{calc} 868.9517, accuracy $\Delta(m/z) = 1.8$ ppm.



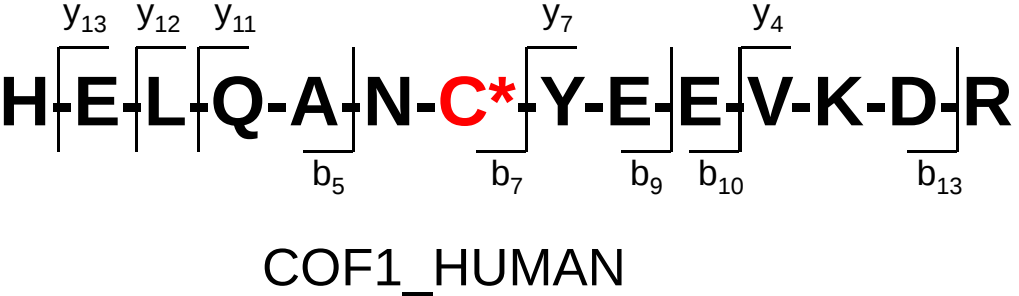
B.



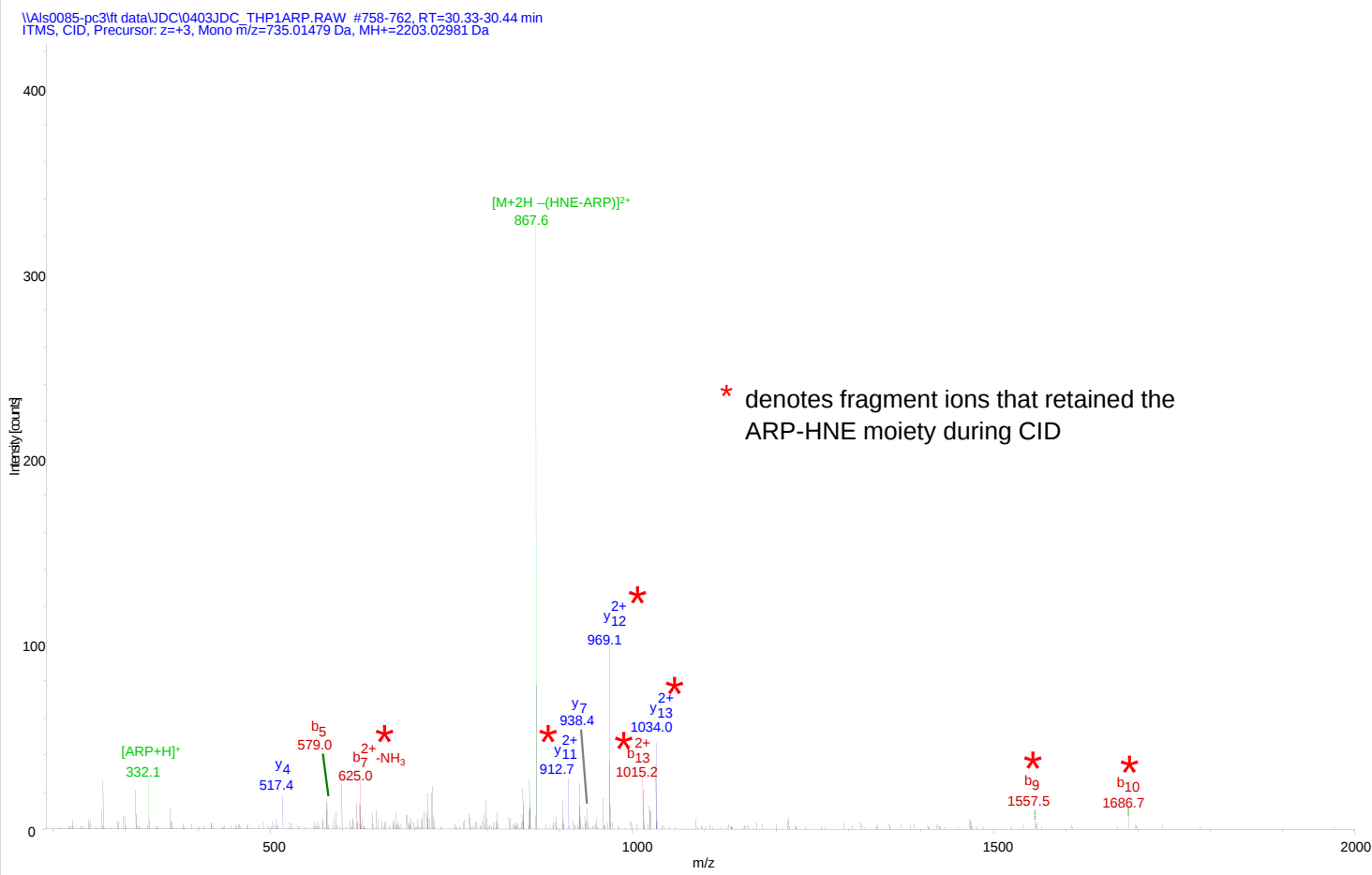
Mascot search result for the MS/MS spectrum depicted above:

#	Immon.	a	a ⁺	a ⁰	b	b ⁺	b ⁰	Seq.	v	w	w ⁺	y	y ⁺	y ⁰	#
1	70.07	70.07			98.06			P							15
2	30.03	127.09			155.08			G				1985.97	1968.94	1967.96	14
3	110.07	264.15			292.14			H	1846.89			1928.95	1911.92	1910.94	13
4	86.10	377.23			405.22			L	1733.81	1732.81		1791.89	1774.86	1773.88	12
5	101.07	505.29	488.26		533.28	516.26		Q	1605.75	1604.76		1678.80	1661.78	1660.79	11
6	102.05	634.33	617.30	616.32	662.33	645.30	644.32	E	1476.71	1475.71		1550.75	1533.72	1532.73	10
7	30.03	691.35	674.33	673.34	719.35	702.32	701.34	G				1421.70	1404.68	1403.69	9
8	120.08	838.42	821.39	820.41	866.42	849.39	848.40	F	1272.62			1364.68	1347.65	1346.67	8
9	30.03	895.44	878.42	877.43	923.44	906.41	905.43	G				1217.61	1200.59	1199.60	7
10	545.26	1467.69	1450.66	1449.68	1495.68	1478.66	1477.67	C	643.35	642.36		1160.59	1143.56	1142.58	6
11	72.08	1566.76	1549.73	1548.74	1594.75	1577.72	1576.74	V	544.28	557.30		588.35	571.32	570.34	5
12	72.08	1665.82	1648.80	1647.81	1693.82	1676.79	1675.81	V	445.22	458.24		489.28	472.25	471.27	4
13	74.06	1766.87	1749.85	1748.86	1794.87	1777.84	1776.86	T	344.17	357.19	359.17	390.21	373.18	372.20	3
14	87.06	1880.91	1863.89	1862.90	1908.91	1891.88	1890.90	N	230.12	229.13		289.16	272.14		2
15	129.11							R	74.02	73.03		175.12	158.09		1

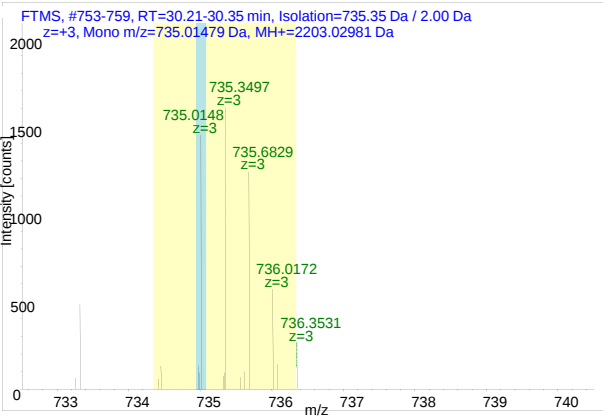
Figure S20. (A) MS/MS and (B) MS spectrum of the MH⁺ of the ARP-HNE modified peptide PGHLQEGFGC*VVTNR obtained on the MALDI-TOF/TOF instrument; expected monoisotopic MH⁺ 2083.02 Da; mass accuracy Δm = 0.07 Da (33 ppm). P, precursor ion; F1 and F2, non-peptide fragment ions related to the ARP tag (see Figure S26).



A.



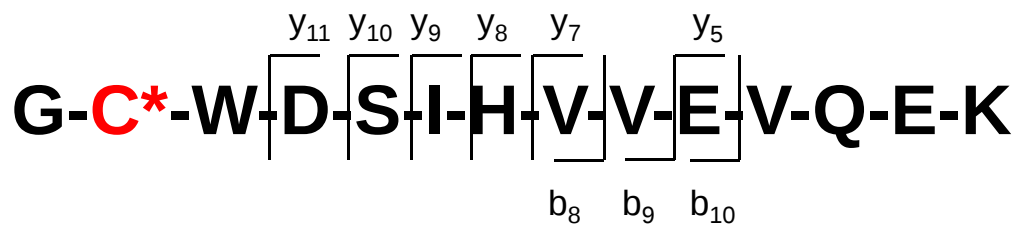
B.



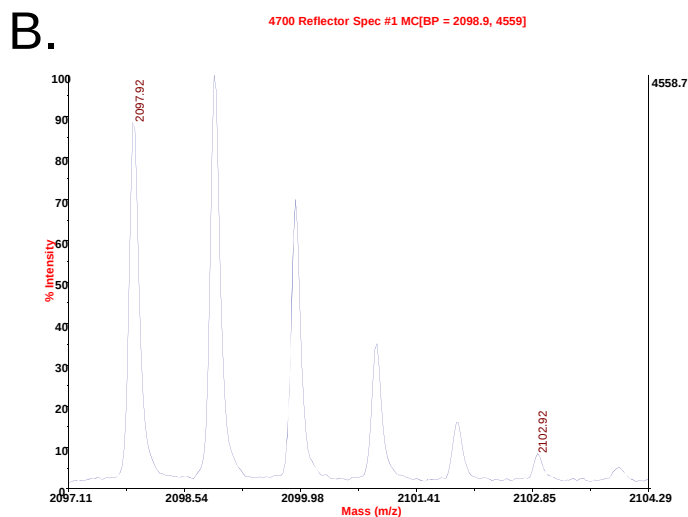
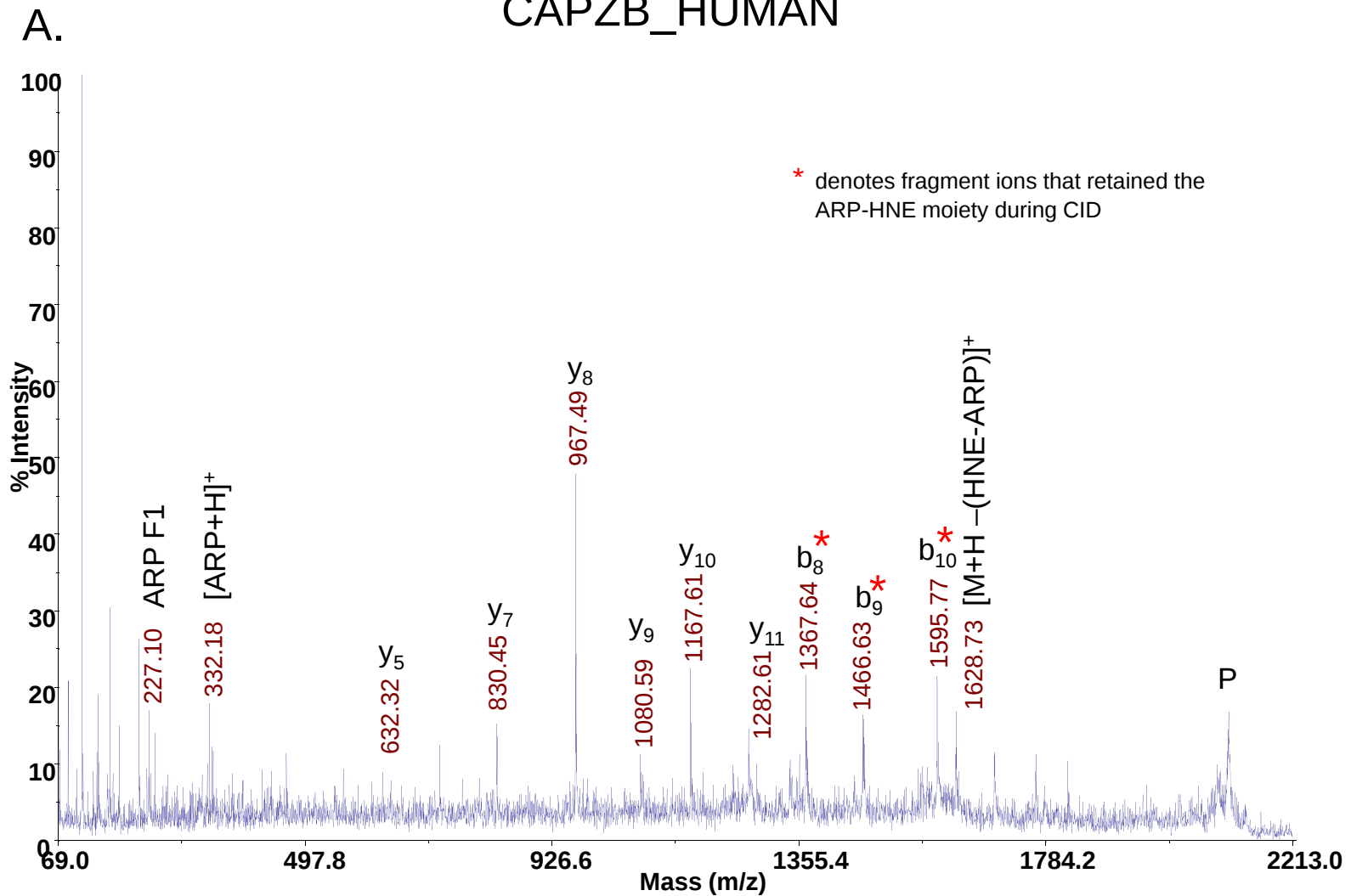
Mascot search result for the MS/MS spectrum depicted above:

#	b	b ⁺⁺	b ⁺	b ⁺⁺	b ⁰	b ⁰⁺⁺	Seq.	y	y ⁺⁺	y ⁺	y ⁺⁺	y ⁰	y ⁰⁺⁺	#
1	138.07	69.54					H							14
2	267.11	134.06			249.10	125.05	E	2065.97	1033.49	2048.94	1024.97	2047.96	1024.48	13
3	380.19	190.60			362.18	181.59	L	1936.93	968.97	1919.90	960.45	1918.91	959.96	12
4	508.25	254.63	491.22	246.12	490.24	245.62	Q	1823.84	912.42	1806.81	903.91	1805.83	903.42	11
5	579.29	290.15	562.26	281.63	561.28	281.14	A	1695.78	848.40	1678.76	839.88	1677.77	839.39	10
6	693.33	347.17	676.30	338.66	675.32	338.16	N	1624.75	812.88	1607.72	804.36	1606.74	803.87	9
7	1265.58	633.29	1248.55	624.78	1247.57	624.29	C	1510.70	755.86	1493.68	747.34	1492.69	746.85	8
8	1428.64	714.82	1411.61	706.31	1410.63	705.82	Y	938.46	469.73	921.43	461.22	920.45	460.73	7
9	1557.68	779.34	1540.66	770.83	1539.67	770.34	E	775.39	388.20	758.37	379.69	757.38	379.20	6
10	1686.73	843.87	1669.70	835.35	1668.71	834.86	E	646.35	323.68	629.33	315.17	628.34	314.67	5
11	1785.79	893.40	1768.77	884.89	1767.78	884.40	V	517.31	259.16	500.28	250.64	499.30	250.15	4
12	1913.89	957.45	1896.86	948.93	1895.88	948.44	K	418.24	209.62	401.21	201.11	400.23	200.62	3
13	2028.92	1014.96	2011.89	1006.45	2010.90	1005.96	D	290.15	145.58	273.12	137.06	272.14	136.57	2
14							R	175.12	88.06	158.09	79.55			1

Figure S23. (A) MS/MS spectrum and (B) Full scan mass spectrum of the [M+3H]₃⁺ ion of the ARP-HNE modified peptide HELQANC*YEEVKDR; monoisotopic m/z_{calc} 735.0139; accuracy Δ(m/z) = 1.3 ppm.



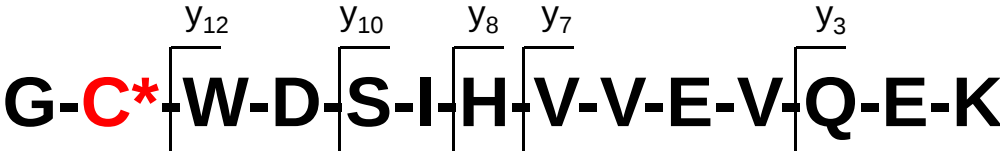
CAPZB_HUMAN



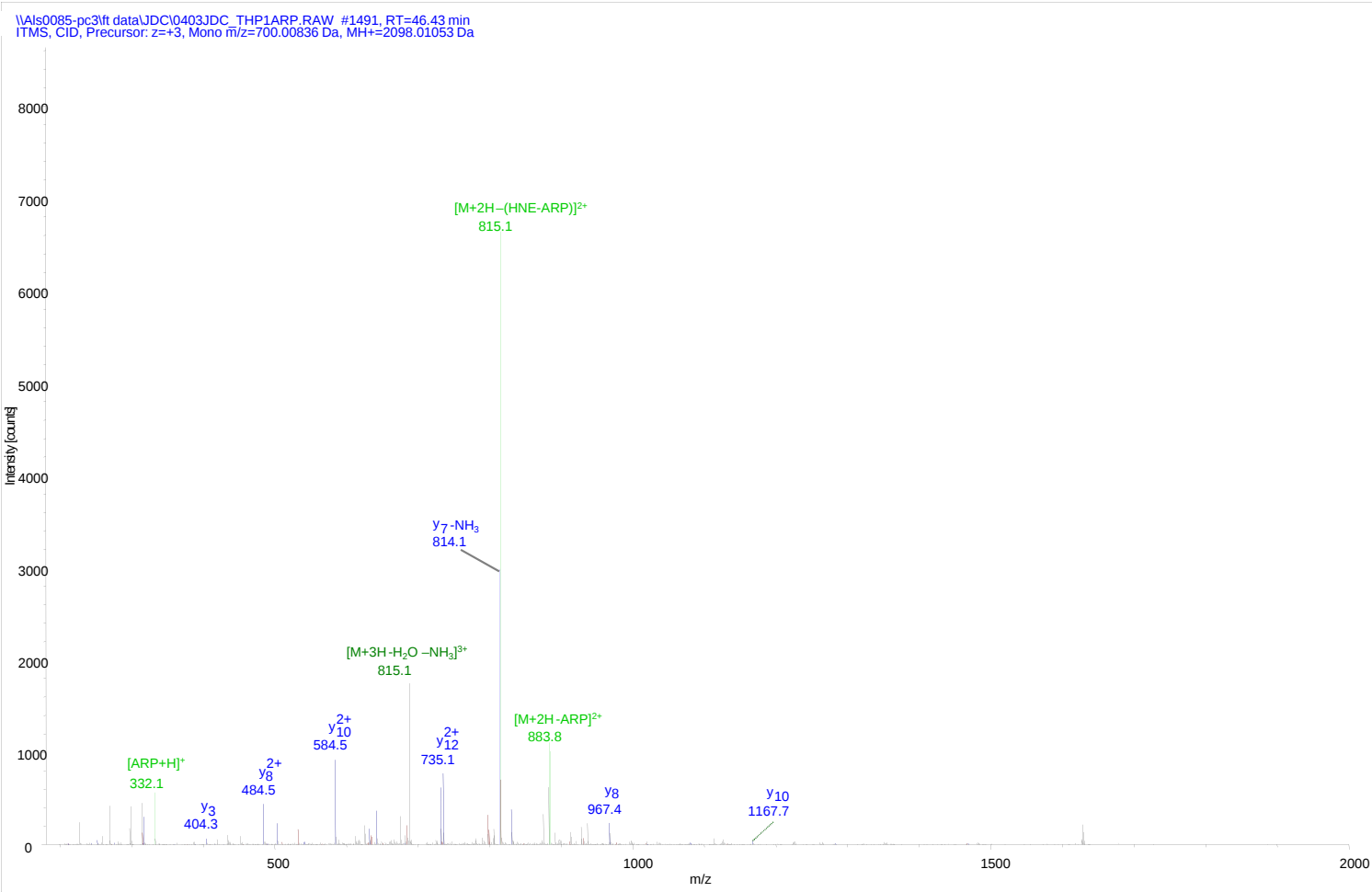
Mascot search result for the MS/MS spectrum depicted above:

#	Immon.	a	a*	a ⁰	b	b*	b ⁰	Seq.	y	y*	y ⁰	#
1	30.03	30.03			58.03			G				14
2	545.26	602.28			630.27			C	2040.99	2023.96	2022.98	13
3	159.09	788.36			816.35			W	1468.74	1451.72	1450.73	12
4	88.04	903.39		885.37	931.38		913.37	D	1282.66	1265.64	1264.65	11
5	60.04	990.42		972.41	1018.41		1000.40	S	1167.64	1150.61	1149.63	10
6	86.10	1103.50		1085.49	1131.50		1113.49	I	1080.60	1063.58	1062.59	9
7	110.07	1240.56		1222.55	1268.56		1250.54	H	967.52	950.49	949.51	8
8	72.08	1339.63		1321.62	1367.62		1349.61	V	830.46	813.44	812.45	7
9	72.08	1438.70		1420.69	1466.69		1448.68	V	731.39	714.37	713.38	6
10	102.05	1567.74		1549.73	1595.73		1577.72	E	632.32	615.30	614.31	5
11	72.08	1666.81		1648.80	1694.80		1676.79	V	503.28	486.26	485.27	4
12	101.07	1794.87	1777.84	1776.86	1822.86	1805.83	1804.85	Q	404.21	387.19	386.20	3
13	102.05	1923.91	1906.88	1905.90	1951.90	1934.88	1933.89	E	276.16	259.13	258.14	2
14	101.11							K	147.11	130.09		1

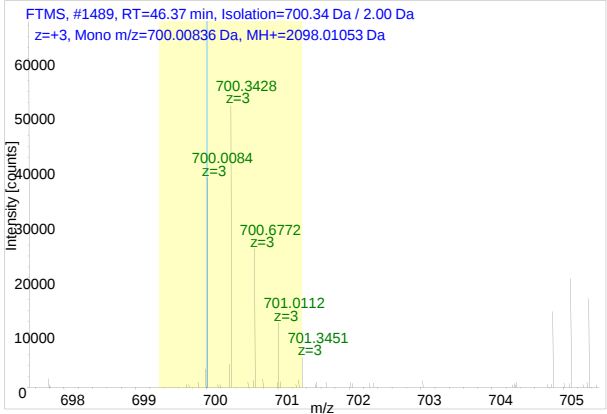
Figure S24. (A) MS/MS and (B) MS spectrum of the MH^+ ion of the ARP-HNE modified peptide $\text{GC}^*\text{WDSIHVVEVQEK}$ obtained on the MALDI-TOF/TOF instrument; expected monoisotopic MH^+ 2098.01 Da; mass accuracy $\Delta m = -0.09$ Da (-43 ppm). P, precursor ion; F1 and F2, non-peptide fragment ions related to the ARP tag (see Figure S26).



A. CAPZB_HUMAN



B. Mascot search result for the MS/MS spectrum depicted above:



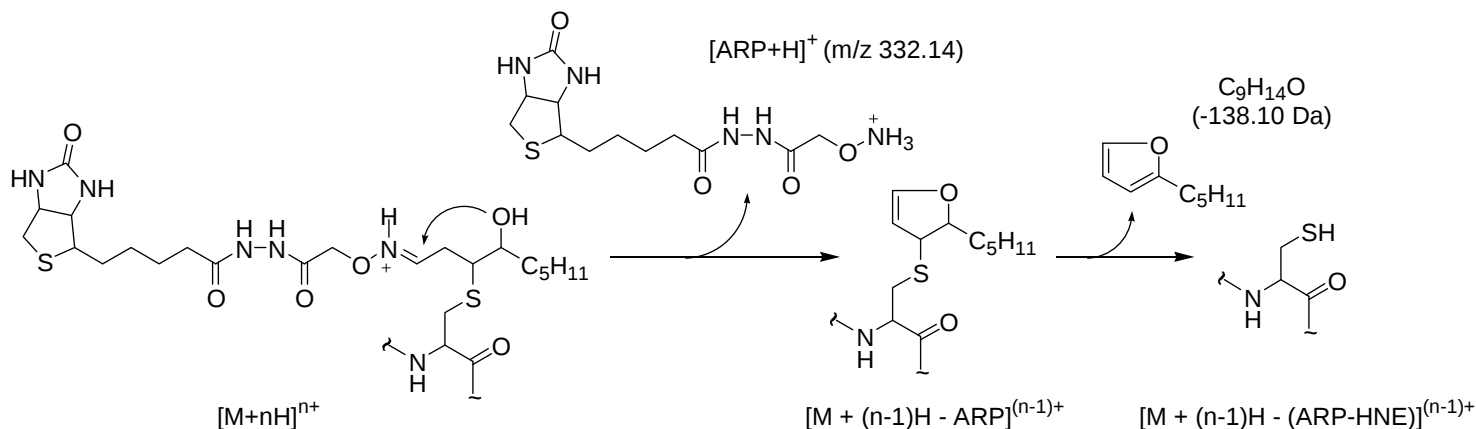
#	b	b ⁺⁺	b ⁺	b ⁺⁺⁺	b ⁰	b ⁰⁺⁺	Seq.	y	y ⁺⁺	y ⁺	y ⁺⁺⁺	y ⁰	y ⁰⁺⁺	#
1	58.03	29.52					G							14
2	630.27	315.64					C	2040.99	1021.00	2023.96	1012.48	2022.98	1011.99	13
3	816.35	408.68					W	1468.74	734.88	1451.72	726.36	1450.73	725.87	12
4	931.38	466.19			913.37	457.19	D	1282.66	641.84	1265.64	633.32	1264.65	632.83	11
5	1018.41	509.71			1000.40	500.70	S	1167.64	584.32	1150.61	575.81	1149.63	575.32	10
6	1131.50	566.25			1113.49	557.25	I	1080.60	540.81	1063.58	532.29	1062.59	531.80	9
7	1268.56	634.78			1250.54	625.78	H	967.52	484.26	950.49	475.75	949.51	475.26	8
8	1367.62	684.32			1349.61	675.31	V	830.46	415.73	813.44	407.22	812.45	406.73	7
9	1466.69	733.85			1448.68	724.84	V	731.39	366.20	714.37	357.69	713.38	357.20	6
10	1595.73	798.37			1577.72	789.37	E	632.32	316.67	615.30	308.15	614.31	307.66	5
11	1694.80	847.91			1676.79	838.90	V	503.28	252.14	486.26	243.63	485.27	243.14	4
12	1822.86	911.93	1805.83	903.42	1804.85	902.93	Q	404.21	202.61	387.19	194.10	386.20	193.61	3
13	1951.90	976.46	1934.88	967.94	1933.89	967.45	E	276.16	138.58	259.13	130.07	258.14	129.58	2
14							K	147.11	74.06	130.09	65.55			1

Figure S25. (A) MS/MS spectrum and (B) Full scan mass spectrum of the [M+3H]³⁺ ion of the ARP-HNE modified peptide GC*WDSIHVVVEVQEK; monoisotopic m/z_{calc} 700.0080; accuracy Δ(m/z) = 0.5 ppm.

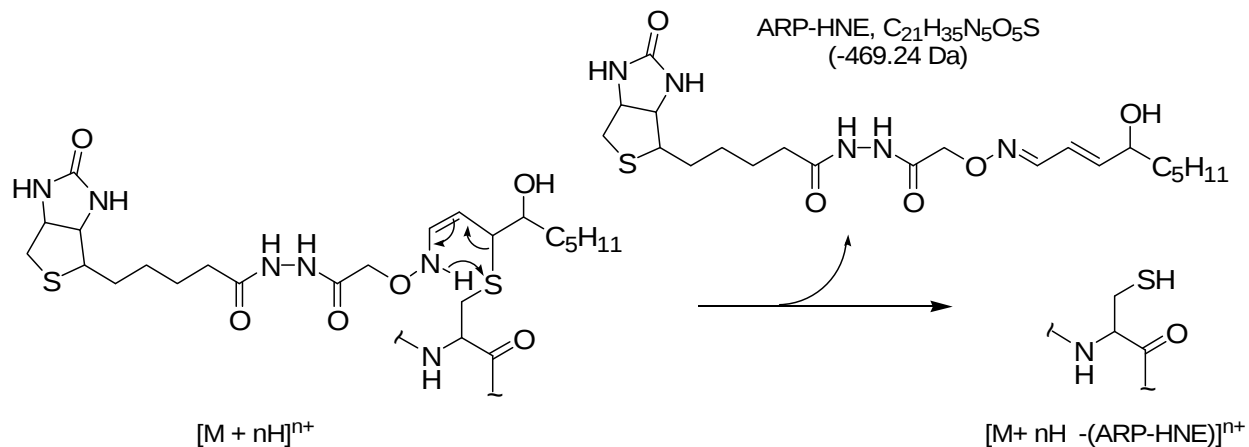
Figure S26.

(A) Proposed CID fragmentation mechanisms specific for ARP-labeled peptide-HNE adducts

Loss of $[\text{ARP}+\text{H}]^+$ followed by neutral loss of 'dehydrated HNE'



Neutral loss of ARP-HNE via retro Michael addition reaction



Neutral loss of ARP (observed in MALDI-MS/MS spectra, i.e. $\text{n}=1$)

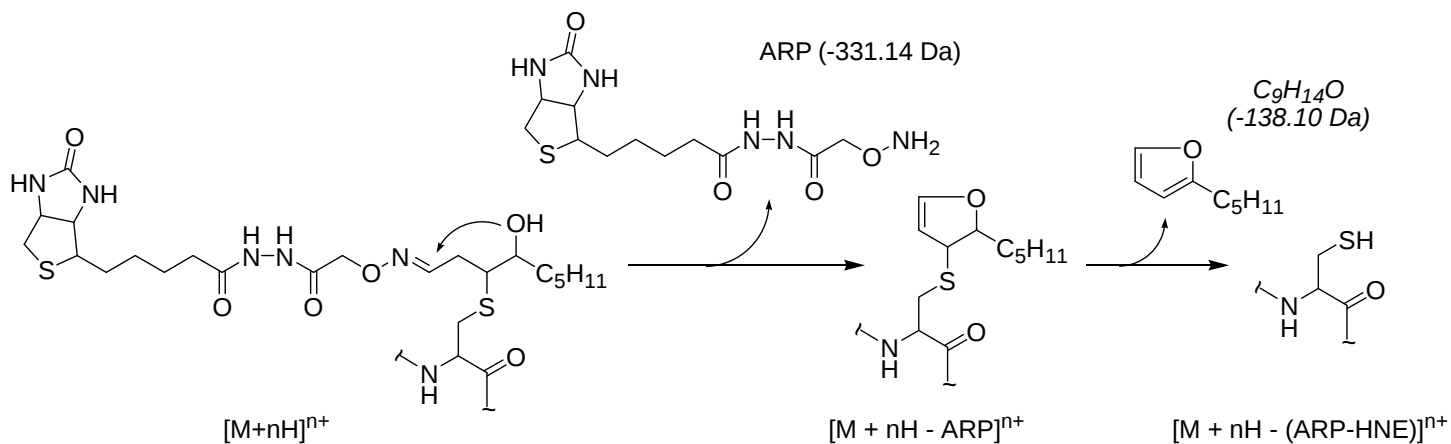
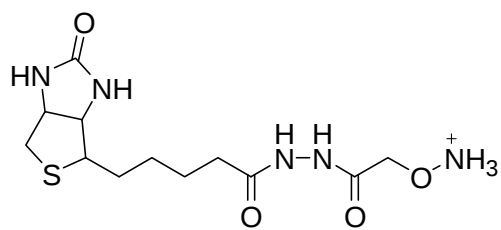
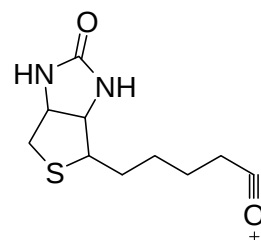


Figure S26. continued

B. Observed ARP-related fragment ions



[ARP+H]⁺ (C₁₂H₂₂N₅O₄S)⁺
(m/z 332.14)



ARP F1 (C₁₀H₁₅N₂O₂S)⁺
(m/z 227.09)