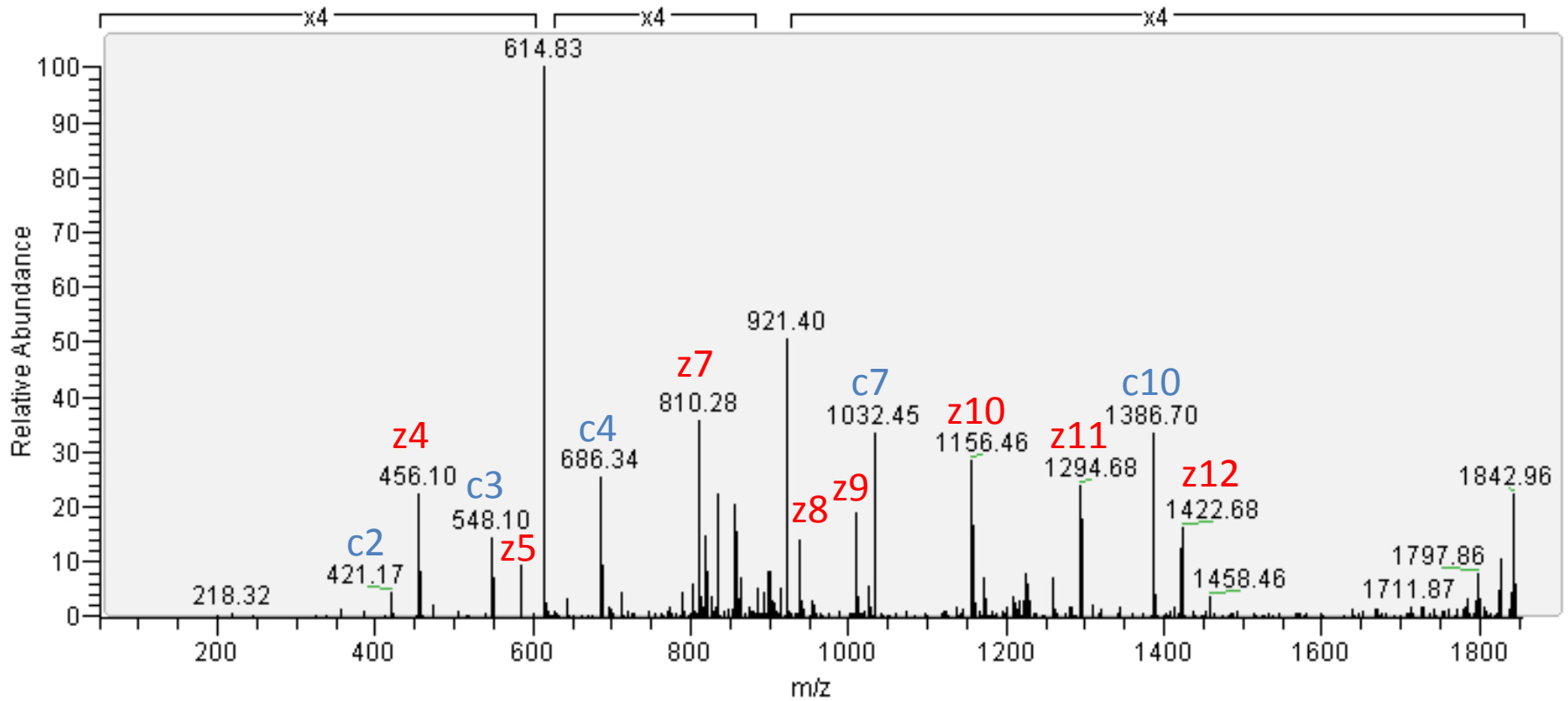


UV Chromatogram of LWAC separation of mouse PSD tryptic peptides.

The following pages contain annotated spectra supporting the O-GlcNAc site identifications determined in this study. Peptide sequence and site assignments are displayed at the top of the page, then spectra with peak assignments are below. These assignments are based on search engine results, where no attempt was made to distinguish between  $z$  and  $z+1$  ions.

m/z 614.318 3+  
VT(HexNAc)QHFAKEPQDPLK – Protein Bassoon  
Modified c2 identifies site as residue 1354

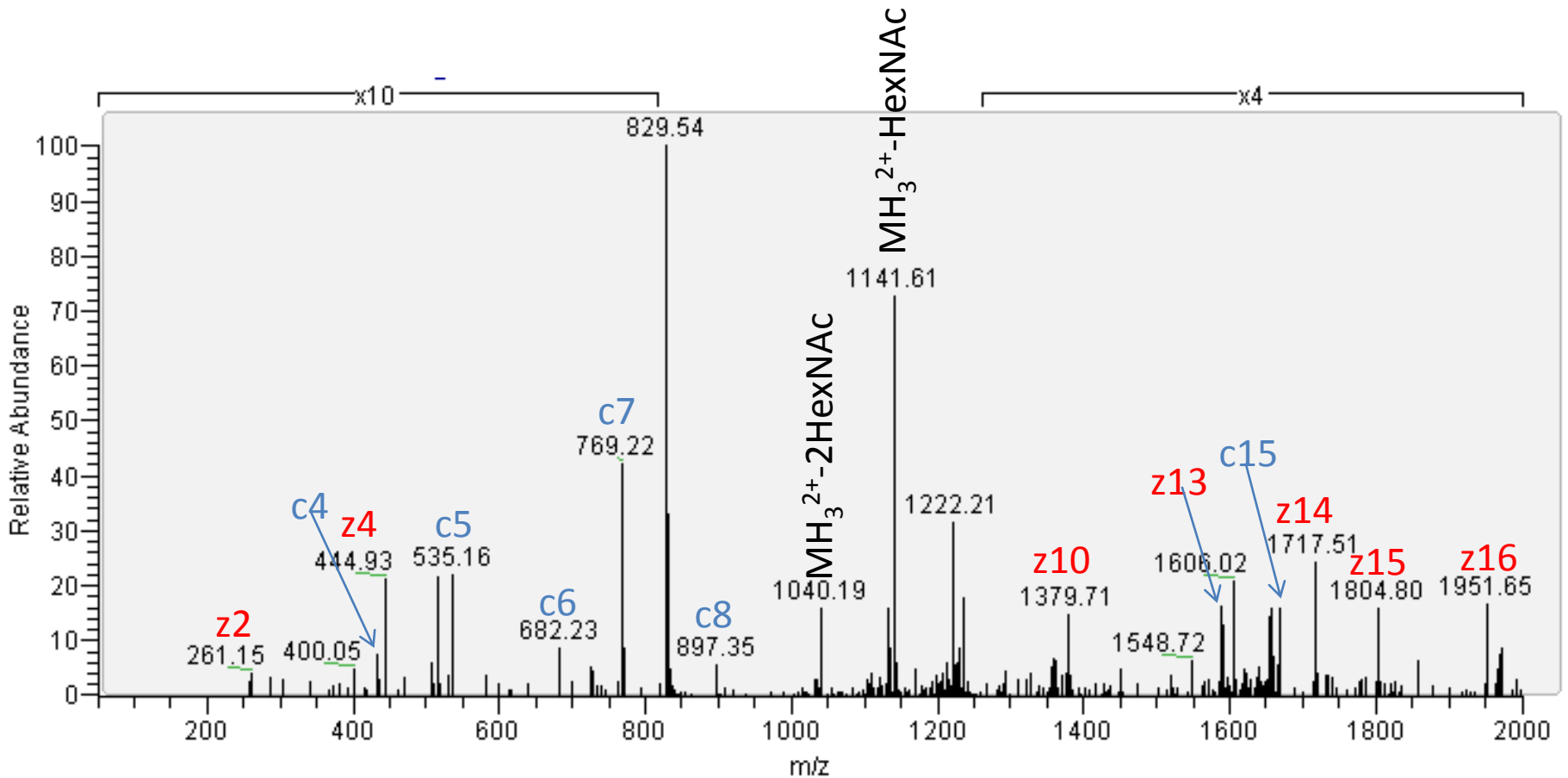


m/z 829.050 3+

EVGMTFSQGPGS(HexNAc)PATT(HexNAc)ASPTR – Protein Bassoon

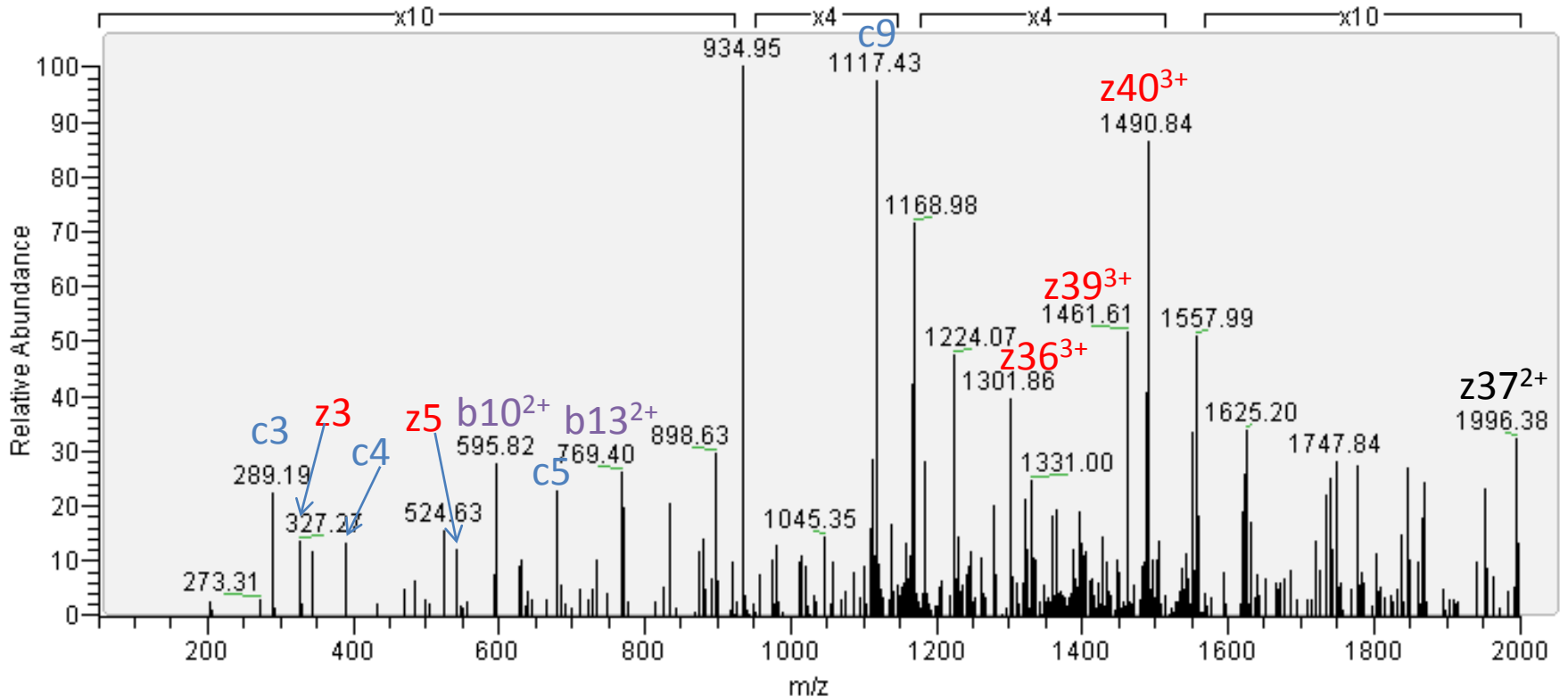
Singly modified c15 and unmodified z4 identifies site as residue1395

Other modified residue is either 1391 or 1394



m/z 934.439 5+  
SPSTS(HexNAc)STIHSYGQPPTTANYGSQTEELPHAPS(HexNAc)GPPGSGRAPR  
– Protein Bassoon

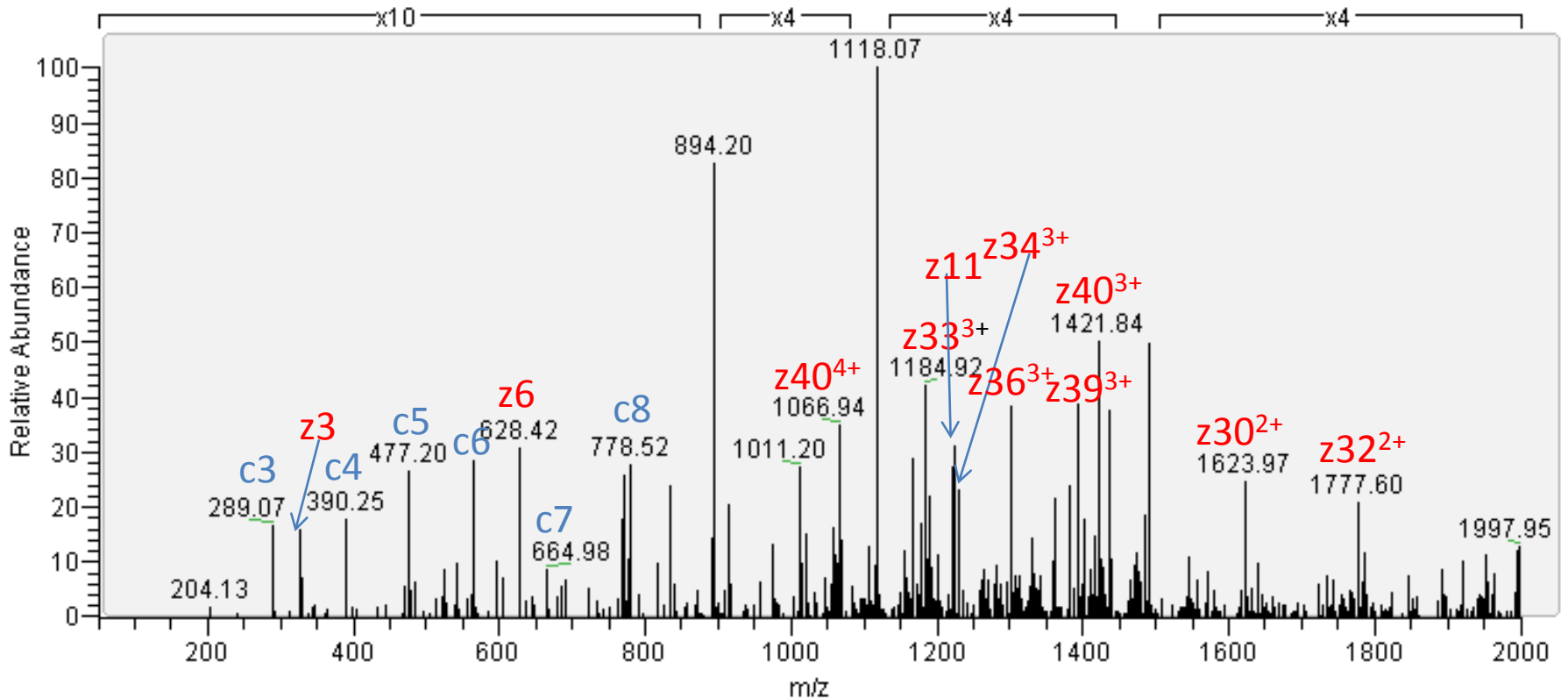
Mass difference between c4-c5 identifies site as residue1418



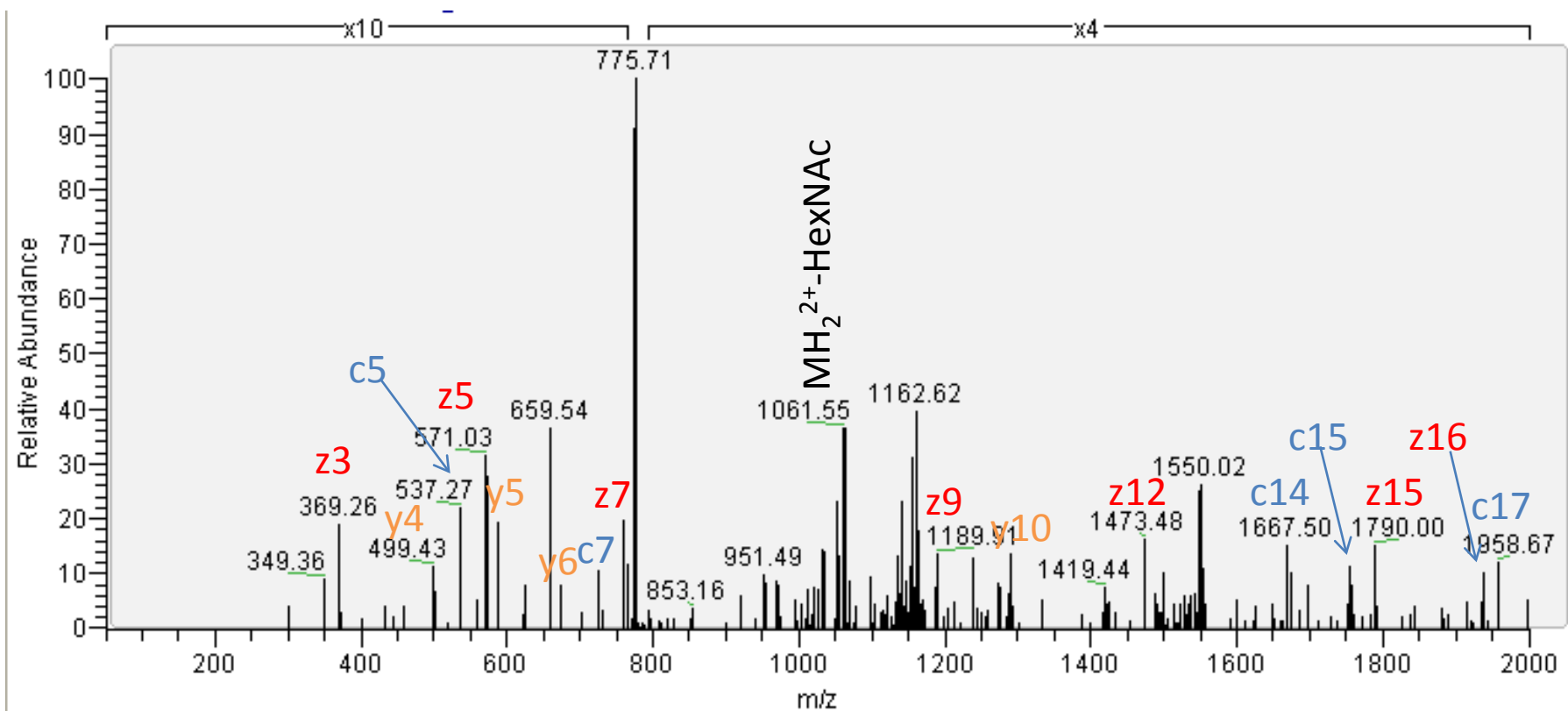
m/z 893.82 5+

SPSTSSTIHSYGQPPTTANYGSQTEELPHAPS(HexNAc)GPPGSGRAPR  
– Protein Bassoon

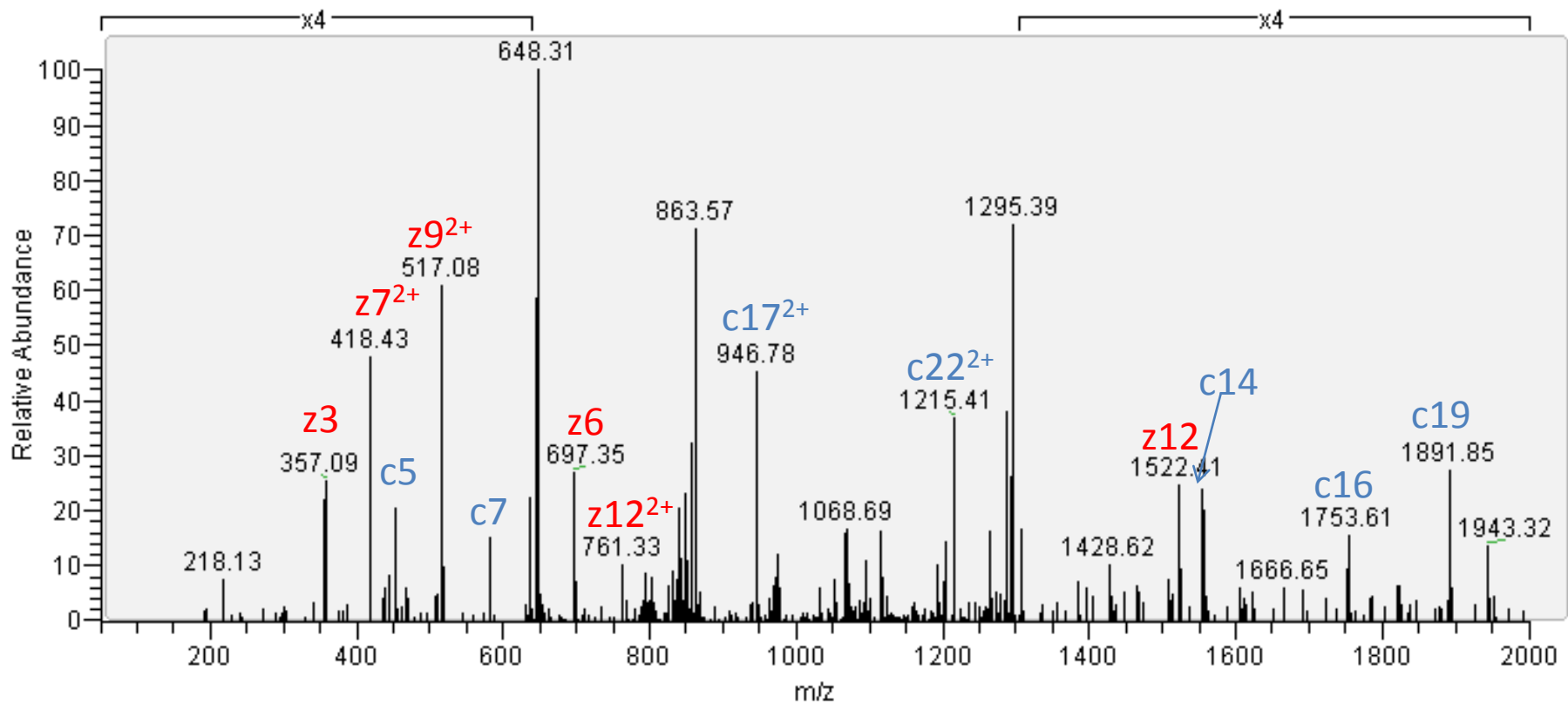
Unmodified z6 and modified z11 identifies site as residue 1445



m/z 775.718 3+  
ATAEFSTQTPSLT(HexNAc)LSSDIPR – Protein Bassoon  
Unmodified z7 and modified z9 identify site as residue 1517



m/z 647.320 4+  
SPGPPSPMVAQGT(HexNAc)QTPHRPSTPR – Protein Bassoon  
Modified c14 and z12 identifies site as residue 1537

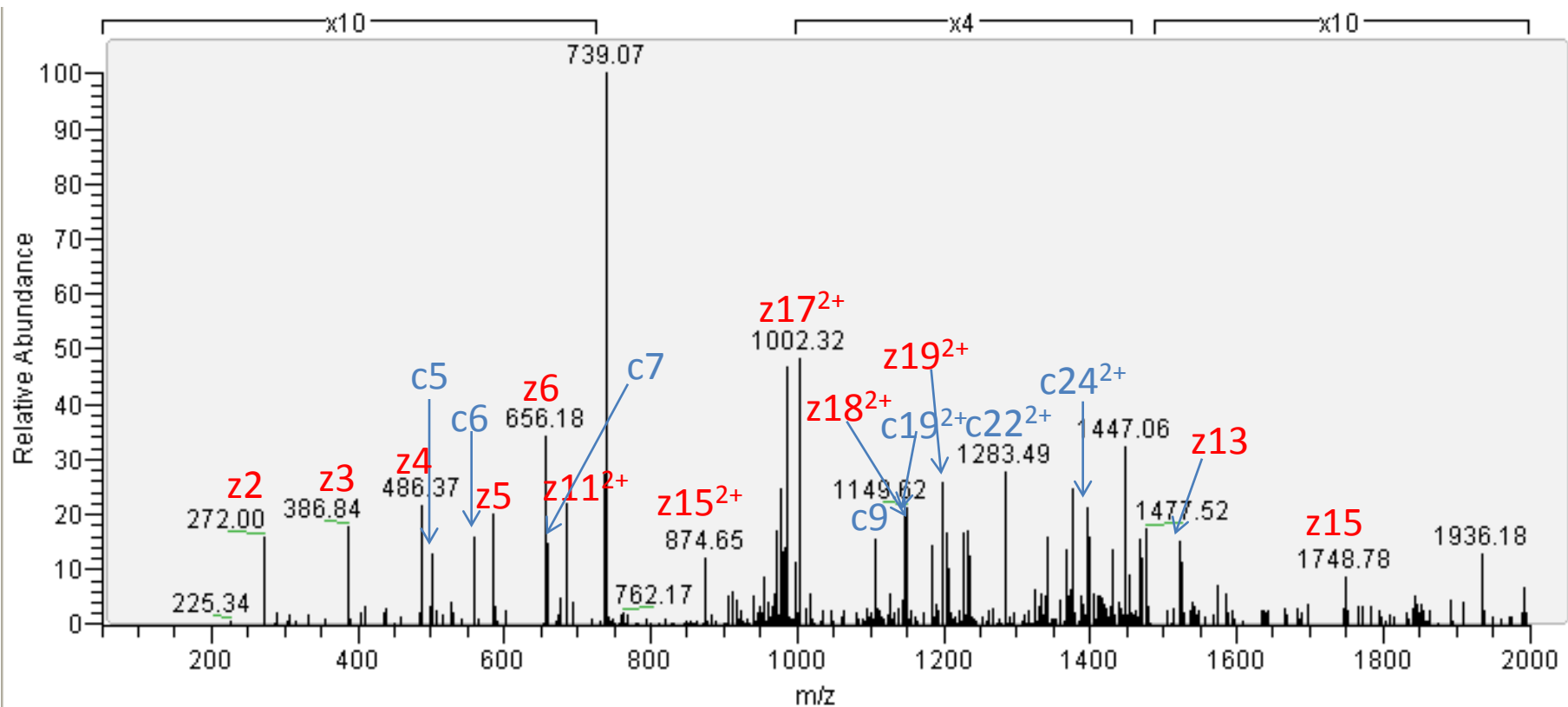


m/z 738.392 4+

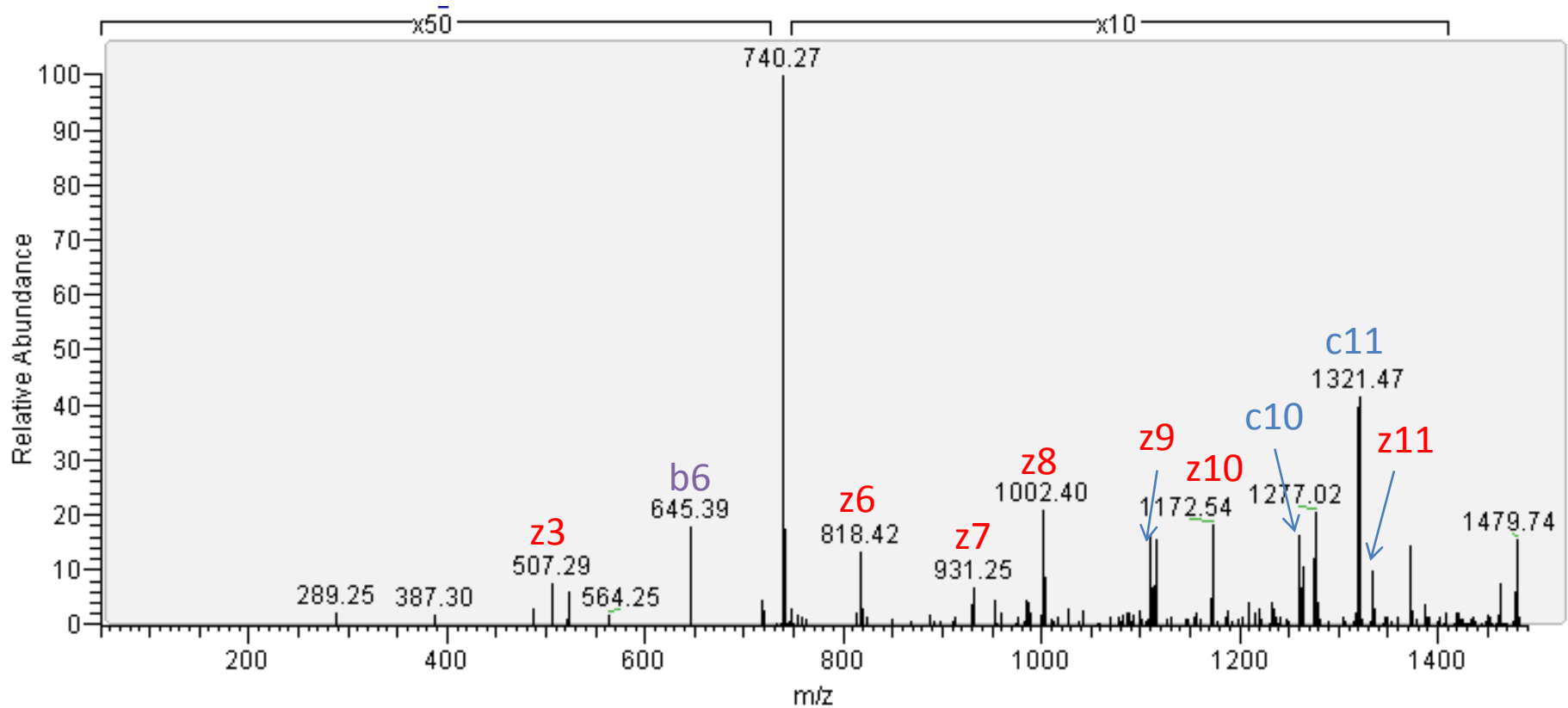
ISSVPGTS(HexNAc)RVEPGPRPPGT(HexNAc)AVVDLR – Protein Bassoon

Mass difference between c6-c7 identifies one site as residue 1657

Modified z11 identifies other site as residue 1666



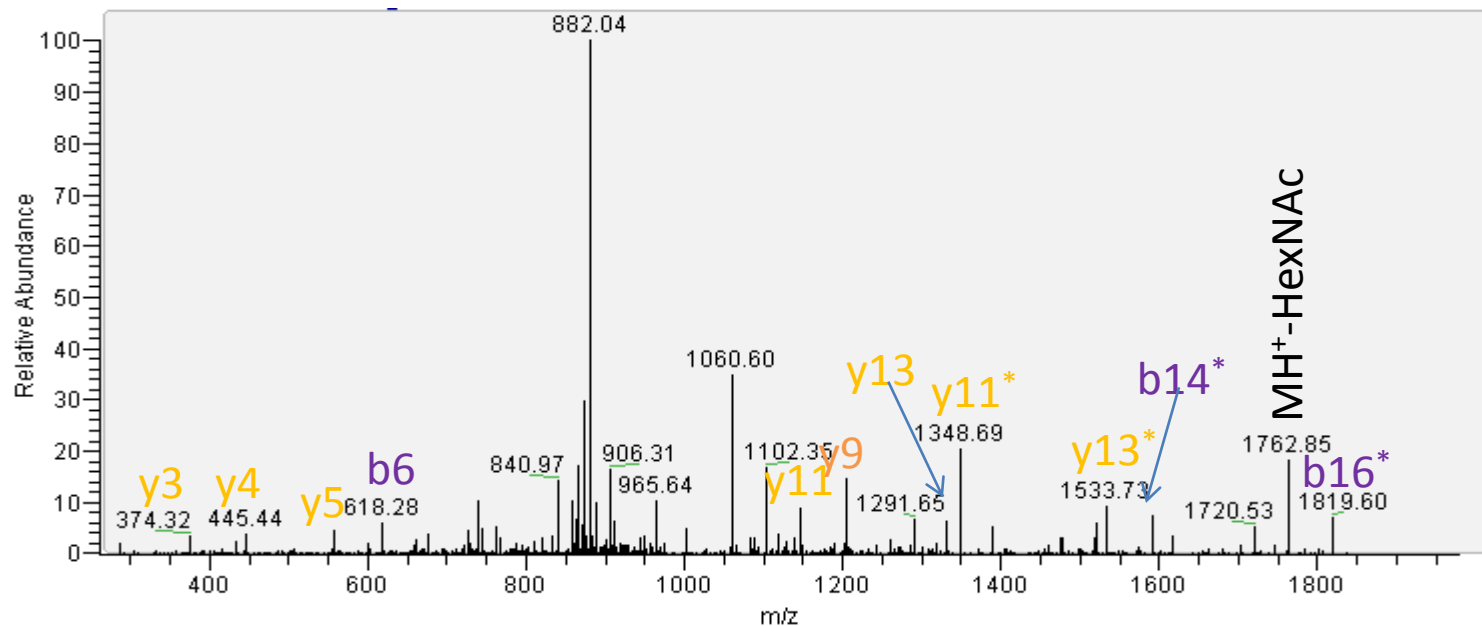
m/z 493.600 3+  
KYGLALDPVS(HexNAc)GR – Protein Bassoon  
Modified z3 identifies residue 1707



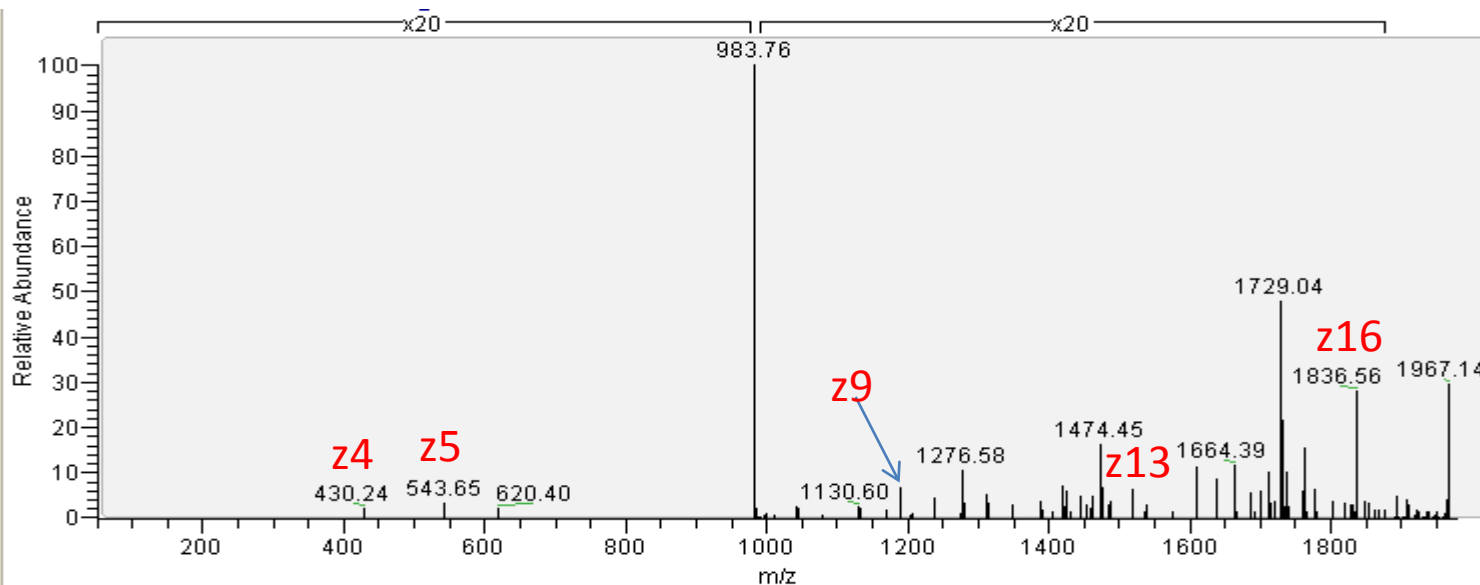
m/z 983.481 2+

LDFGQGSGS(HexNAc)PVC(Carbamidomethyl)LAQVK – Protein Bassoon  
z9 in ETD identifies site as residue 1772

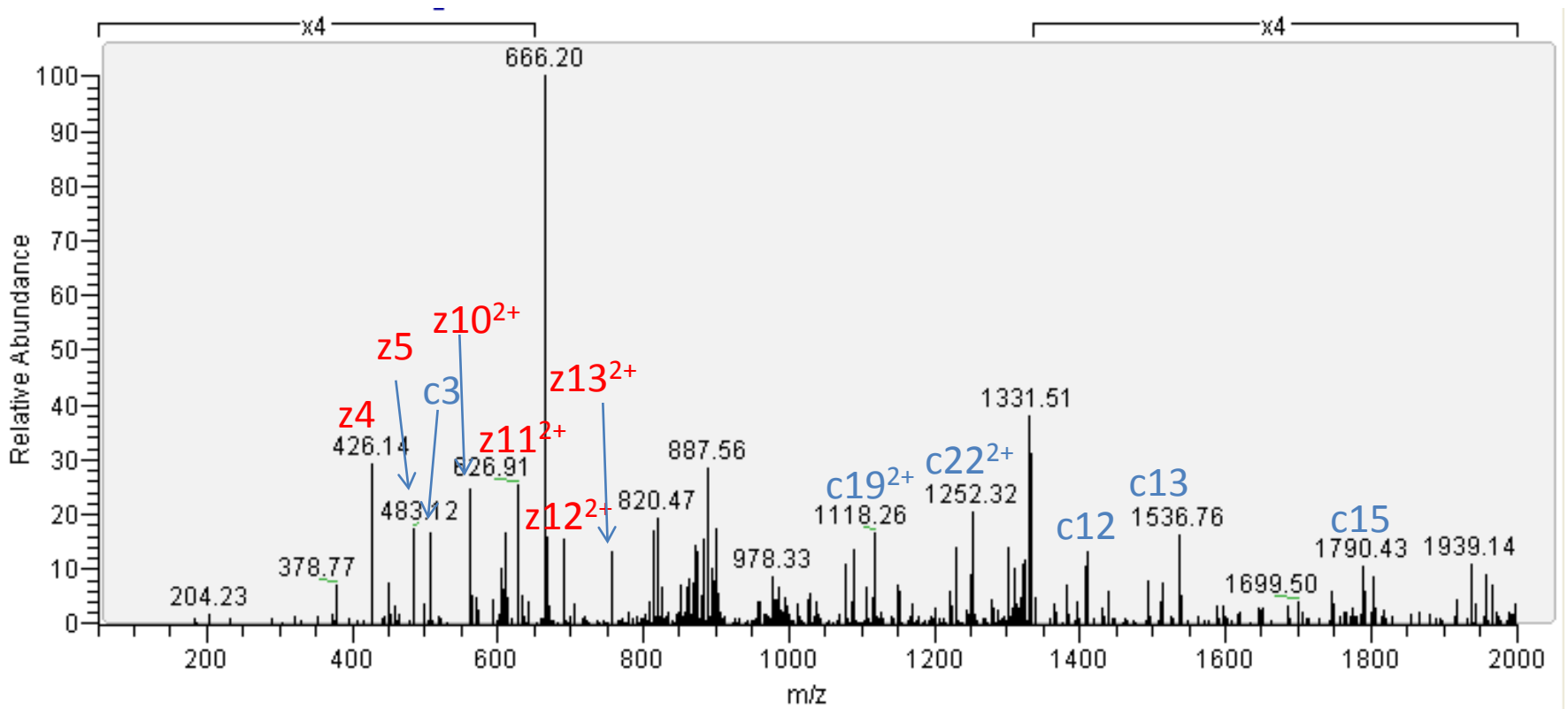
CID



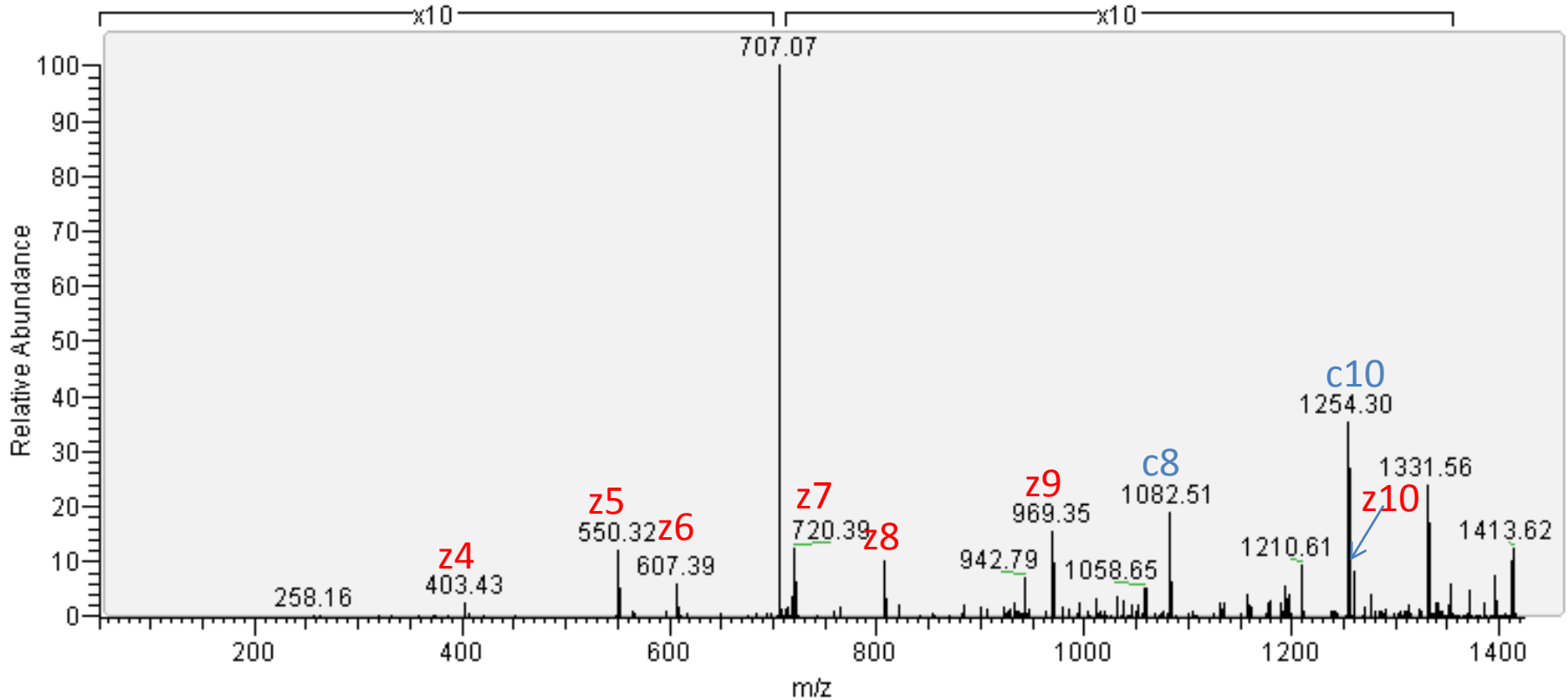
ETD



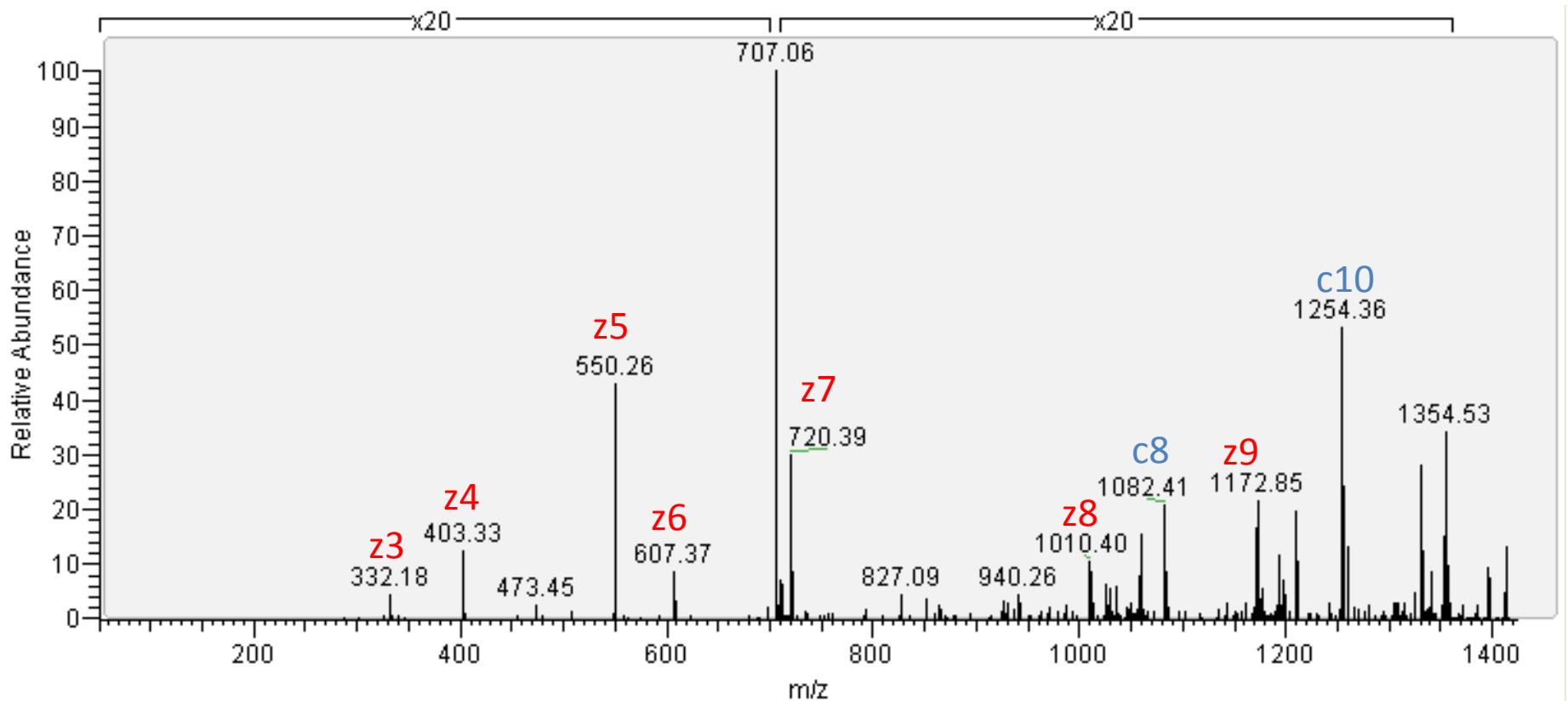
m/z 665.585 4+  
T(HexNAc)QGVVGGPHEEQRPYPQGLPGR – Protein Bassoon  
Modified c3 identifies site as residue 1962



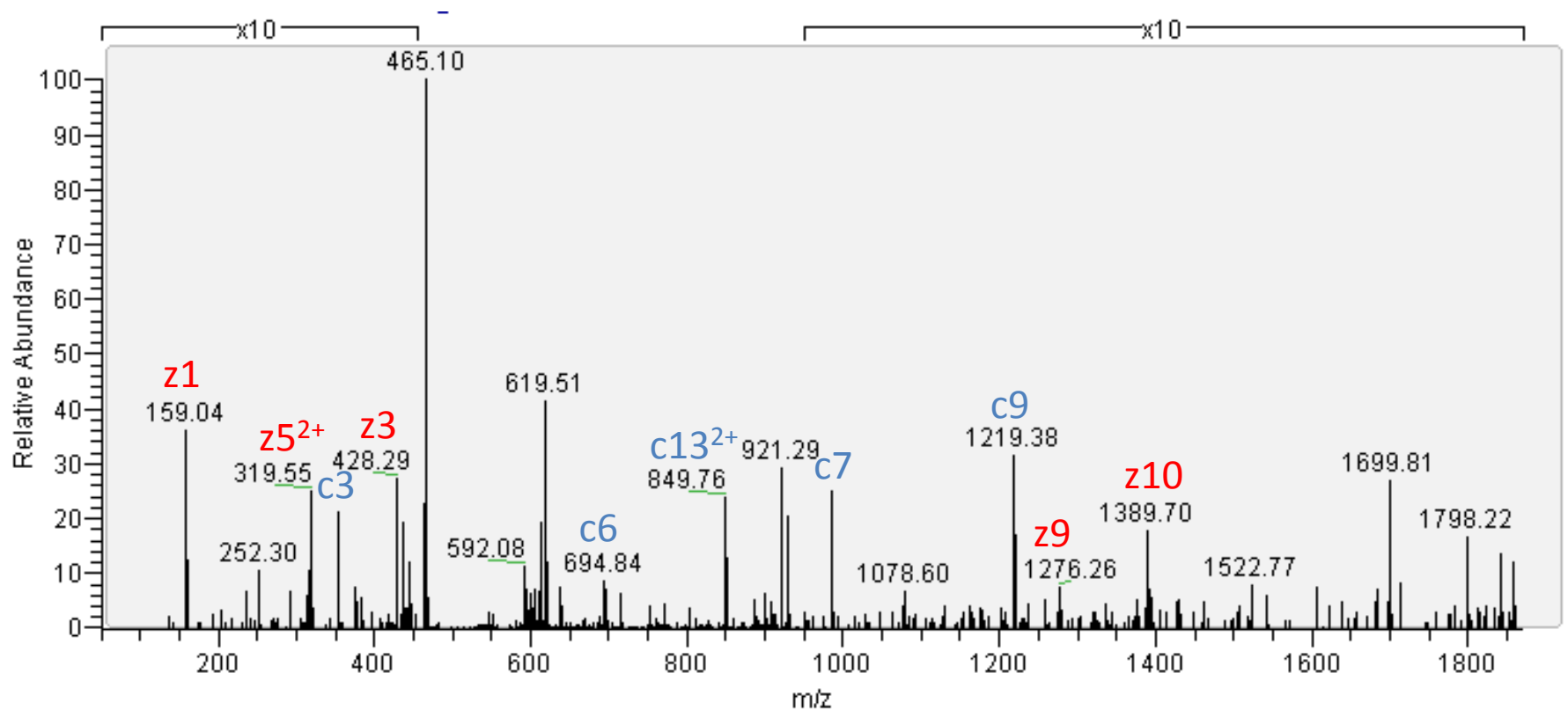
m/z 706.823 2+  
HS(HexNAc)YSLGFADGR – Protein Bassoon  
Mass difference between z9-z10 identifies site as residue 2027



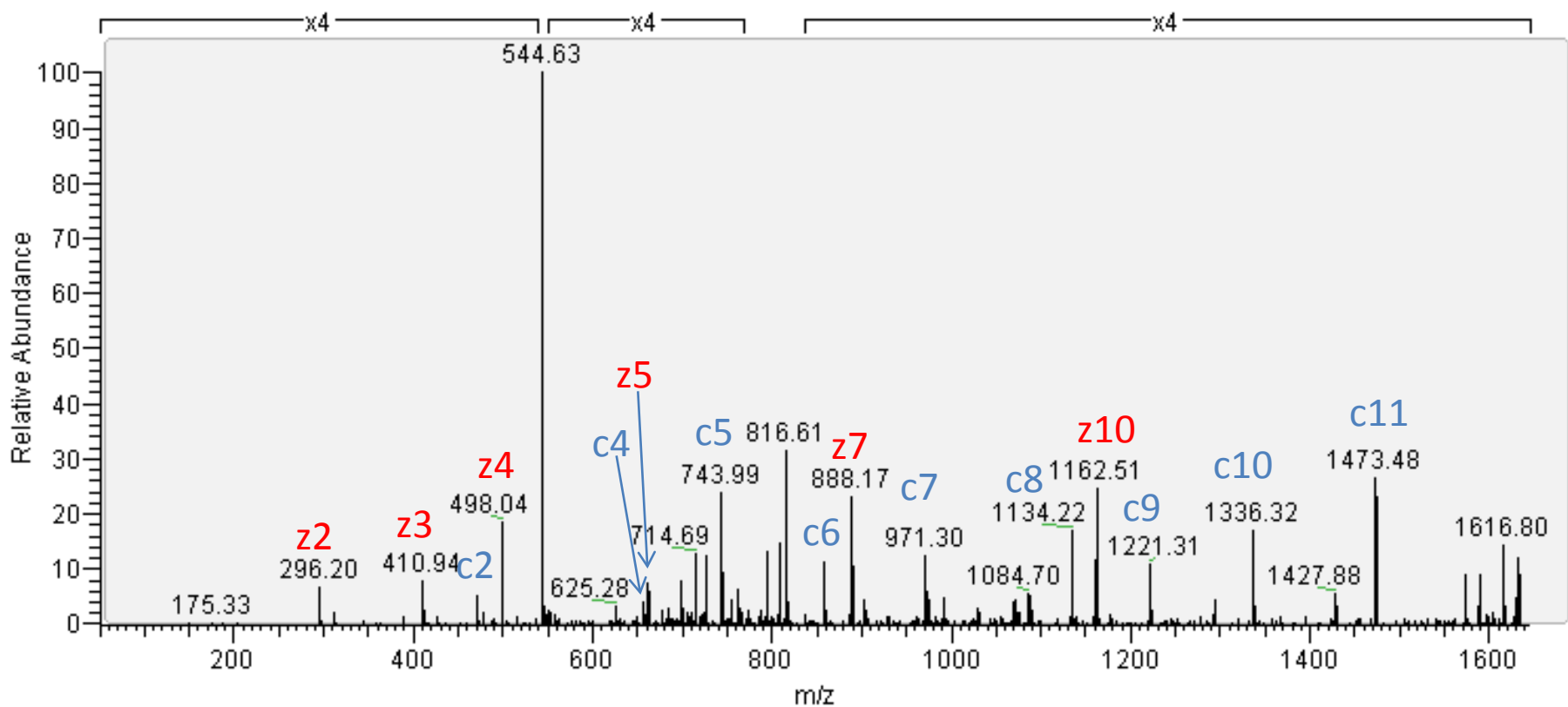
m/z 706.824 2+  
HSYS(HexNAc)LGFADGR – Protein Bassoon  
z7-z8 identifies residue 2029



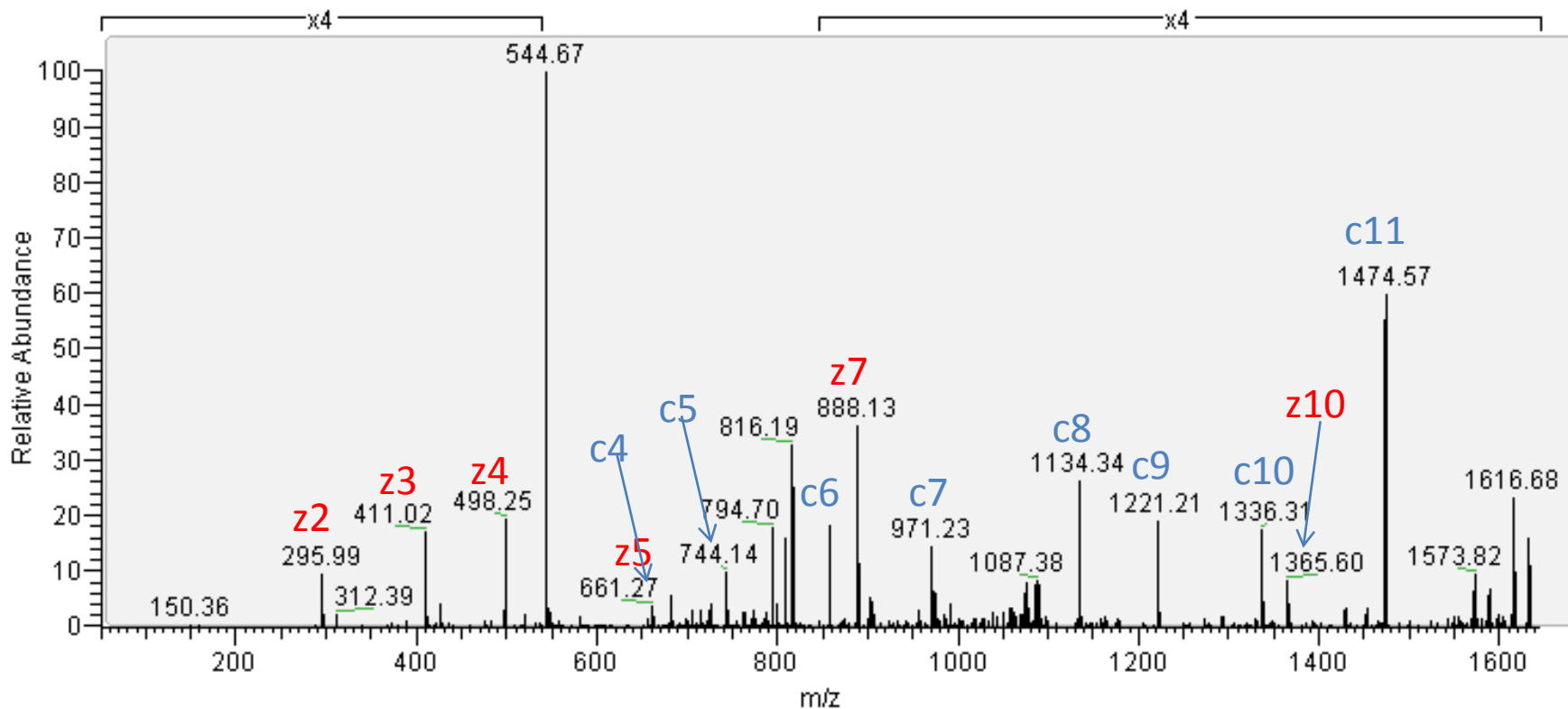
m/z 464.509 4+  
HPTDLLS(HexNAc)HPLPLRR – Protein Bassoon  
Mass difference between c6-c7 identifies site as residue 2058



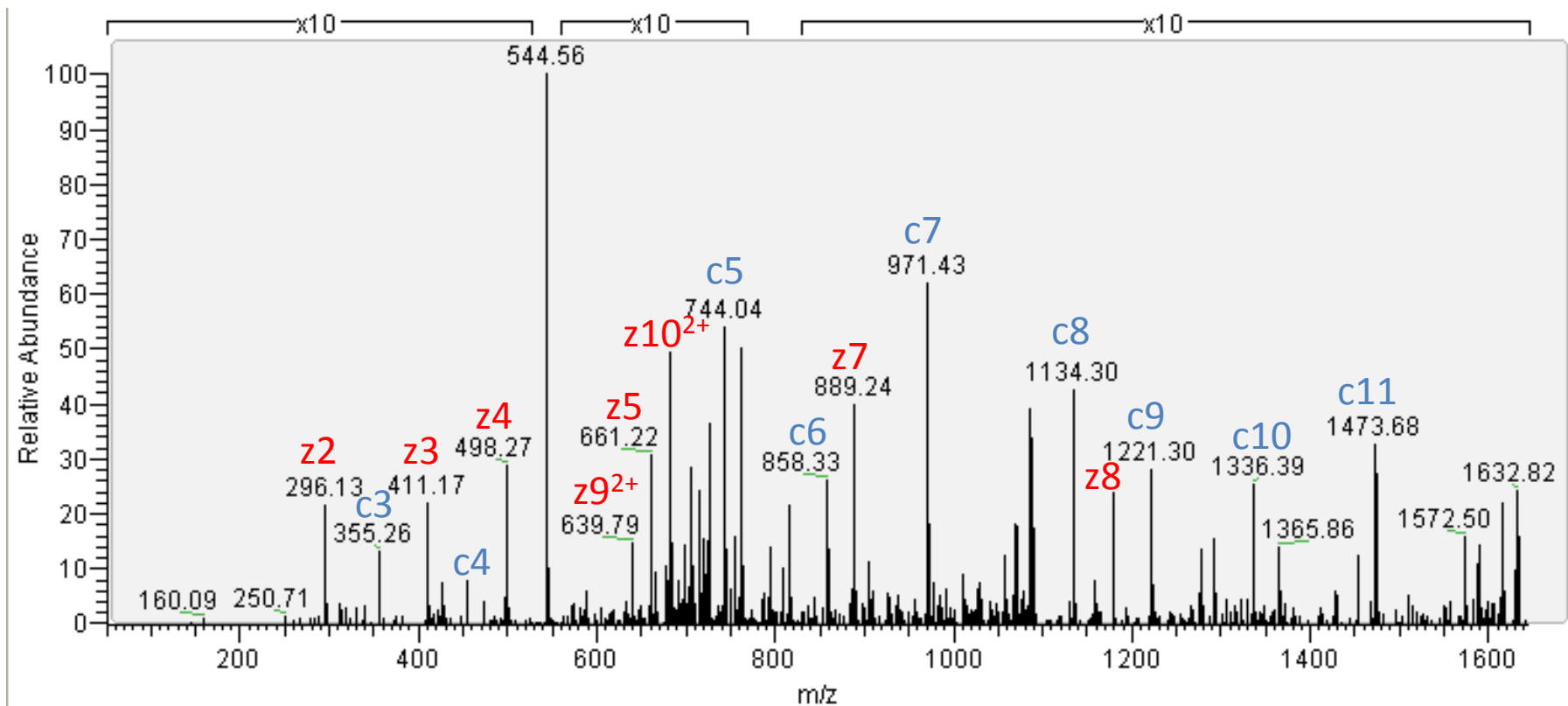
m/z 544.248 3+  
YS(HexNAc)SVSNIYSDHR – Protein Bassoon  
Unmodified z10 and modified c2 identify residue 2067



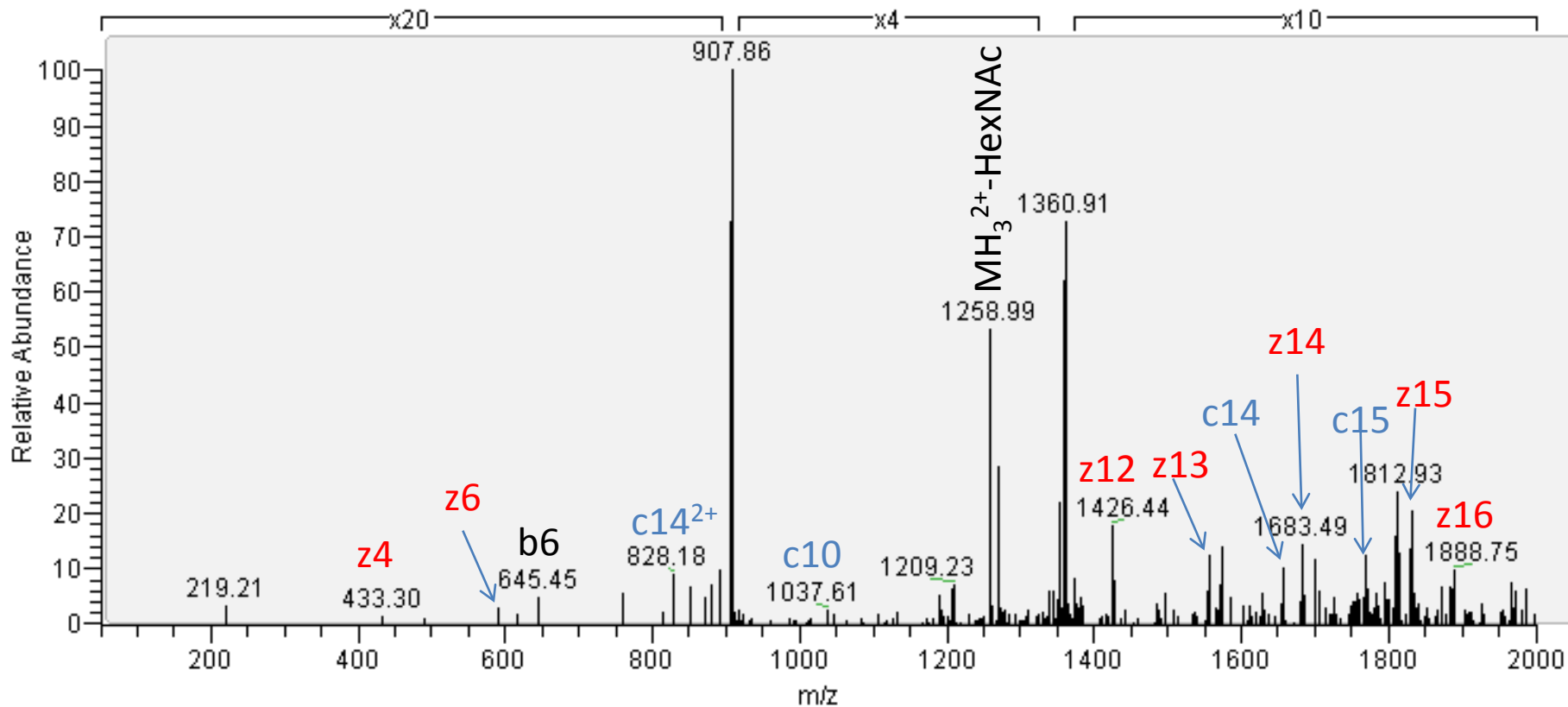
544.248 3+  
YSS(HexNAc)VSNIYSDHR – Protein Bassoon  
Modified c4 and z10 identify site as residue 2068



m/z 544.249 3+  
YSSVS(HexNAc)NIYSDHR – Protein Bassoon  
Mass difference between z7-z8 identifies site as residue 2070

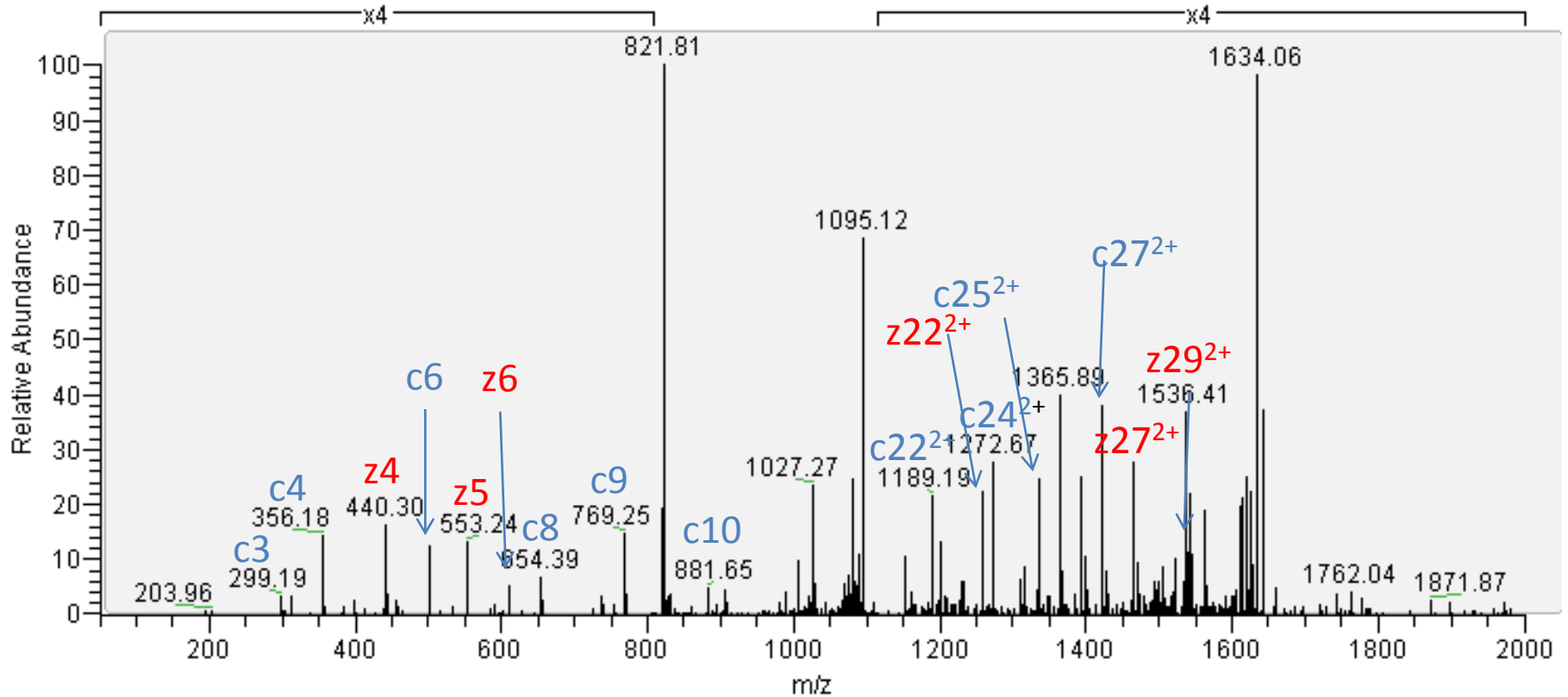


m/z 907.104 3+  
YGPRGDAVGFQEAS(HexNAc)LAQYSATTAR – Protein Bassoon  
Modified c14 identifies site as residue 2091



m/z 821.163 4+

HGSGSGGPDLVQYQPQHGPGLS(HexNAc)APQGLAPLR – Protein Bassoon  
Unmodified c10 identifies site as residue 2141

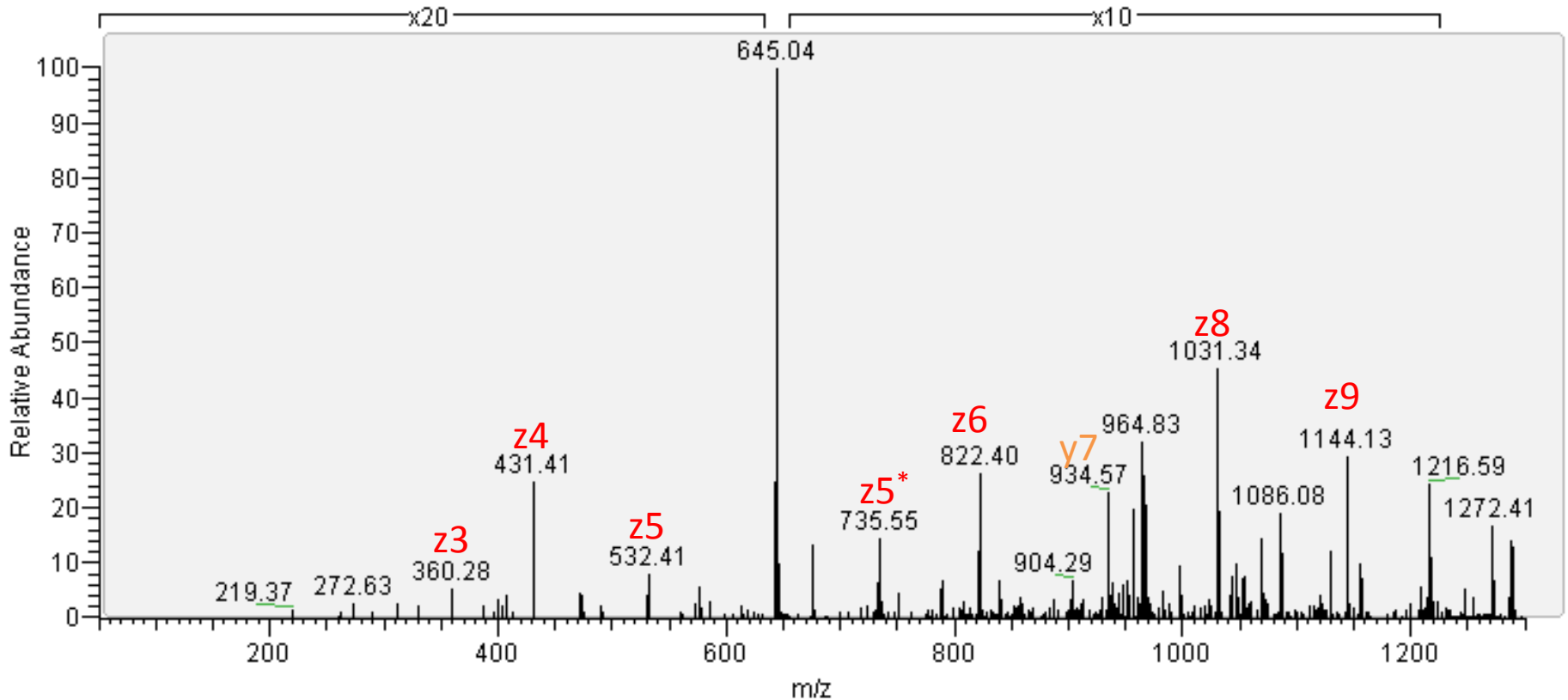


m/z 644.857 2+

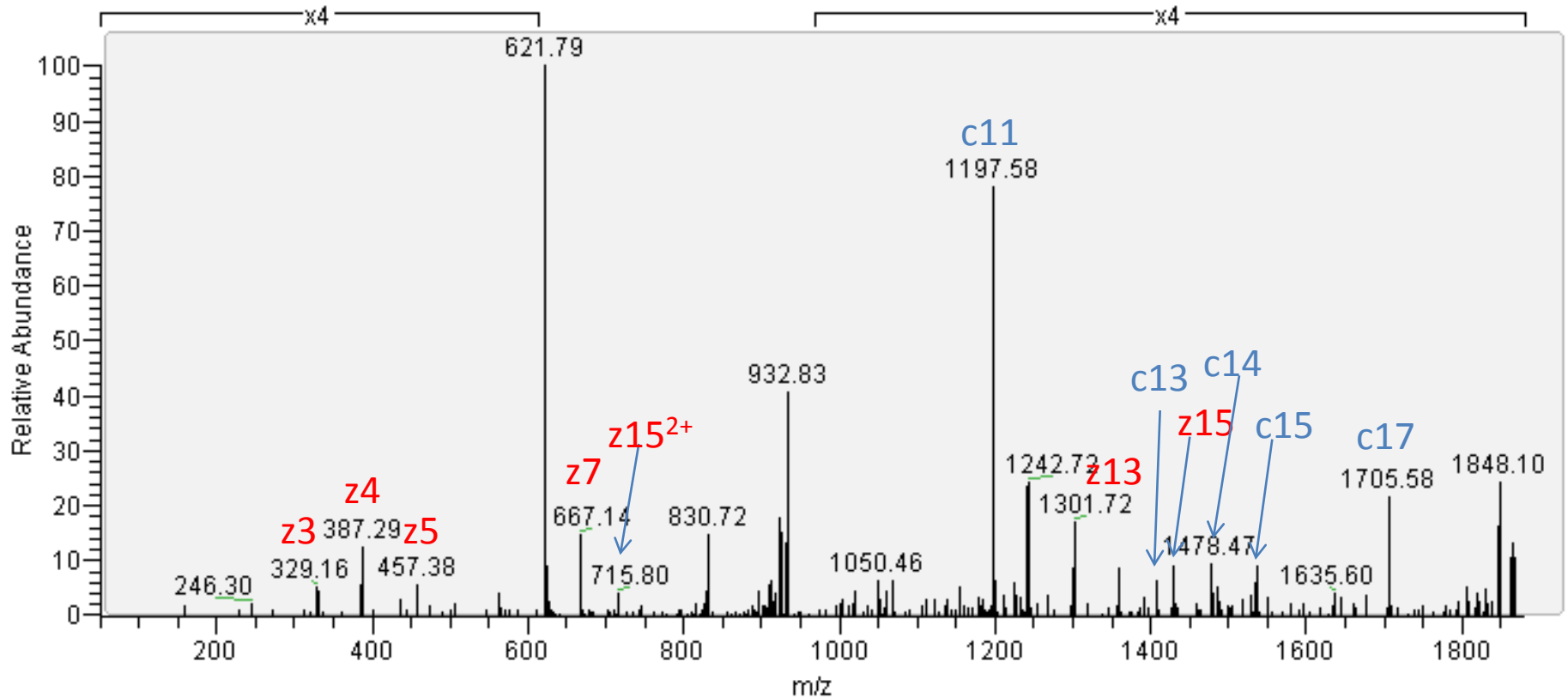
QLLPS(HexNAc)TATVR – Protein Bassoon

Mixture of peptides modified on two different residues:

Unmodified z5 identifies one site as residue 2188



m/z 621.661 3+  
AS(HexNAc)GAGGPPRPELPAGVAR – Protein Bassoon  
Residue 2295 is the only potential modification site

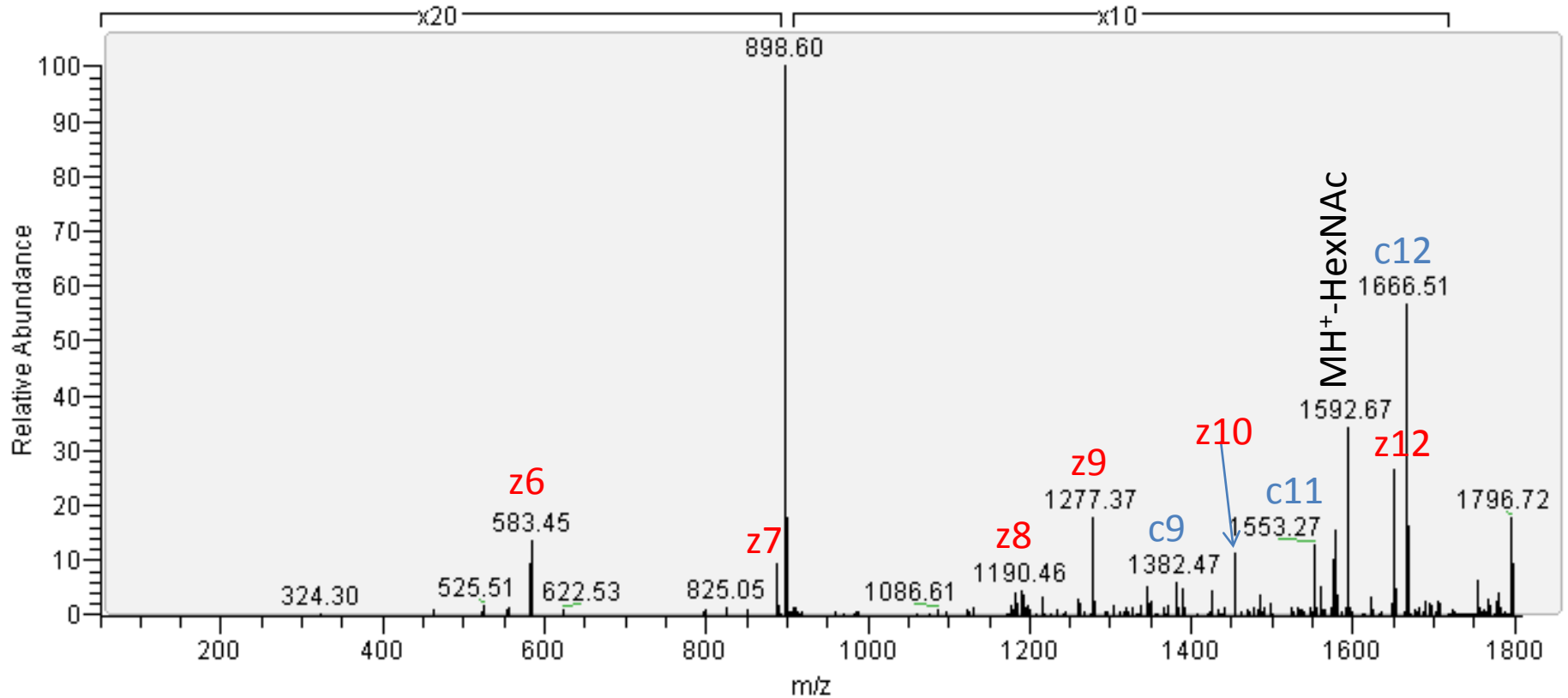


m/z 898.445 2+

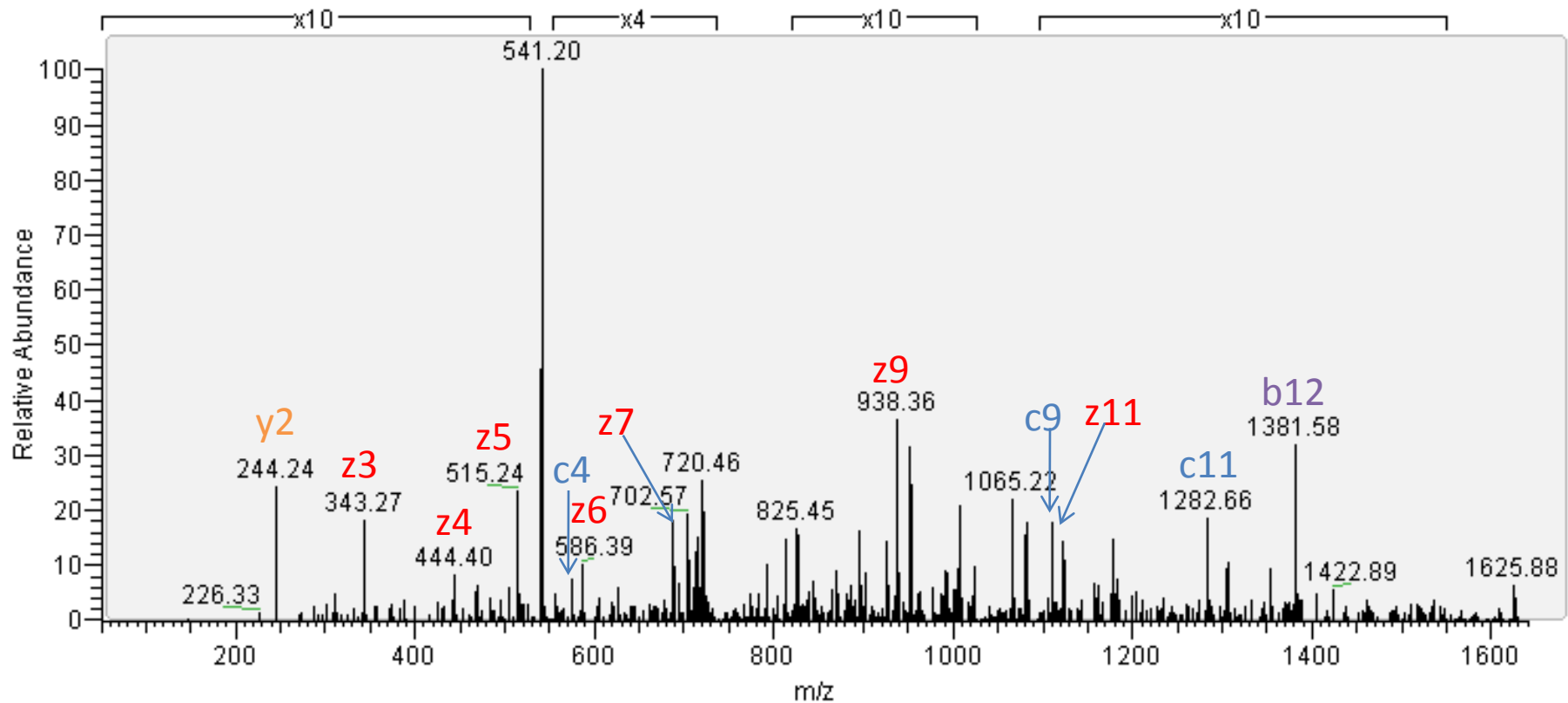
EEPFST(HexNAc)T(HexNAc)APAVIK – Protein Bassoon

Mass difference between z6-z7 identifies one site as residue 2318

Mass difference between z7-z8 identifies other site as residue 2317



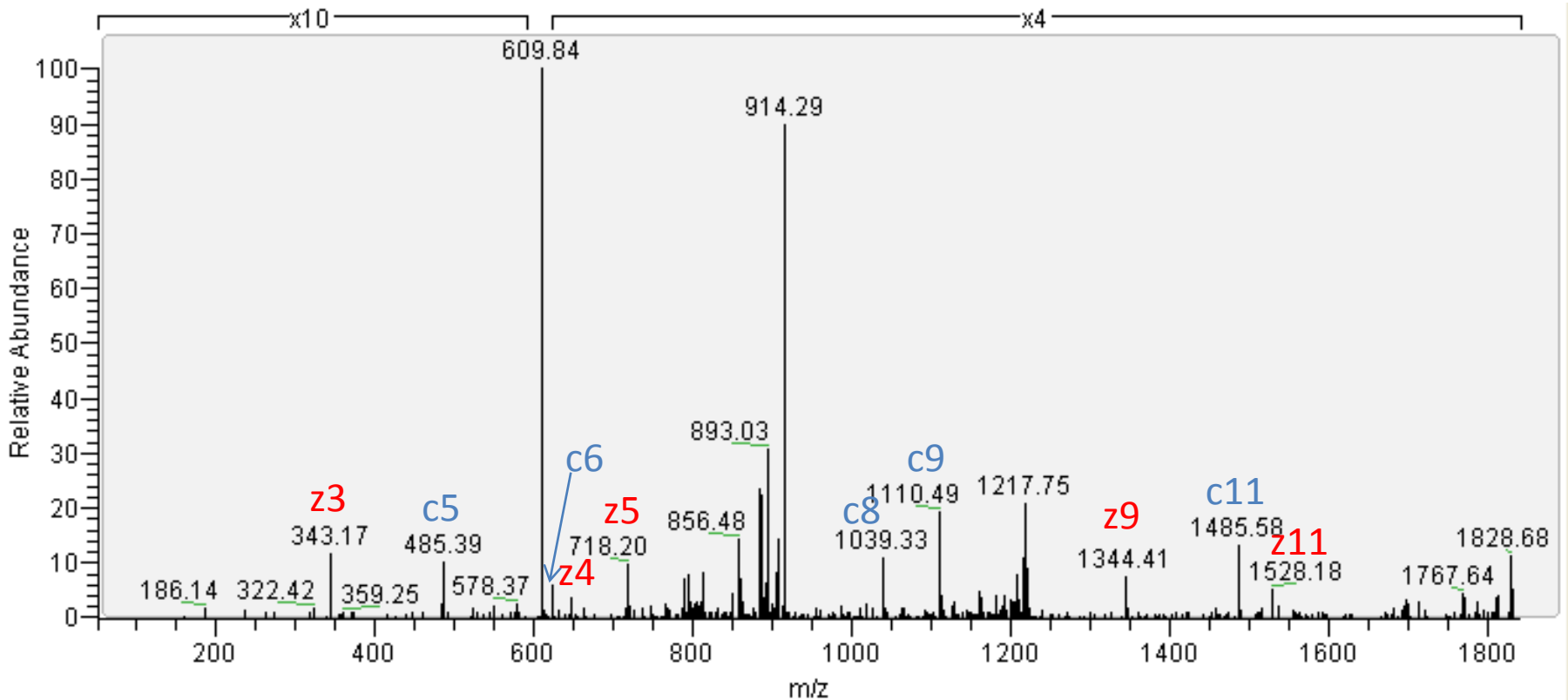
m/z 541.957 3+  
VS(HexNAc)PAIHITAATDPK – Protein Bassoon  
Modified c4 identifies residue 2694



m/z 606.580 3+

VSPAIIIT(HexNAc)AAT(HexNAc)DPK – Protein Bassoon

Modified z5 and doubly modified z9 identify sites as residues 2700 and 2703

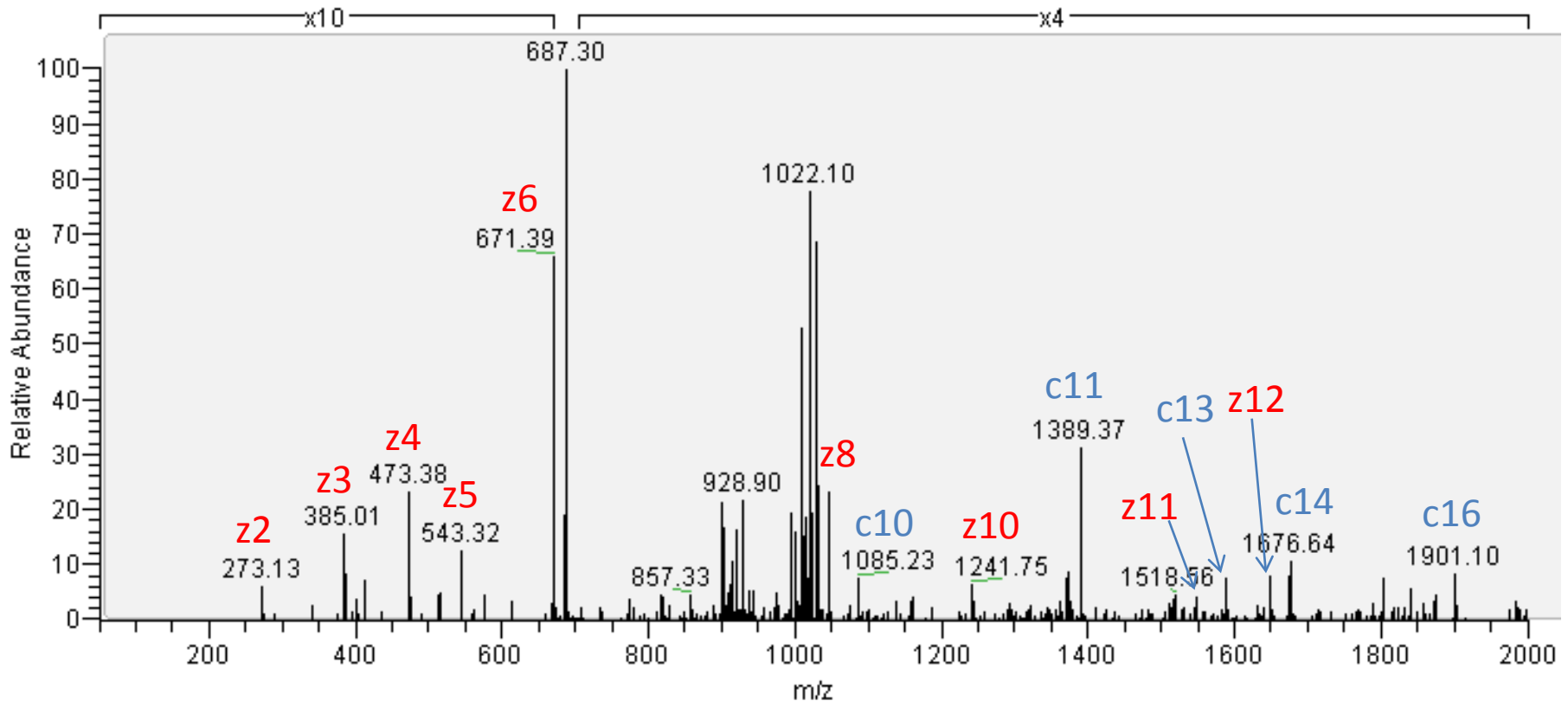


m/z 687.046 3+

GLAGPTT(HexNAc)VPAT(HexNAc)KASLLR - Protein bassoon

Mass difference between z10-z11 identifies one site as residue 2941.

Mass difference between c10-c11 identifies other site as residue 2945.

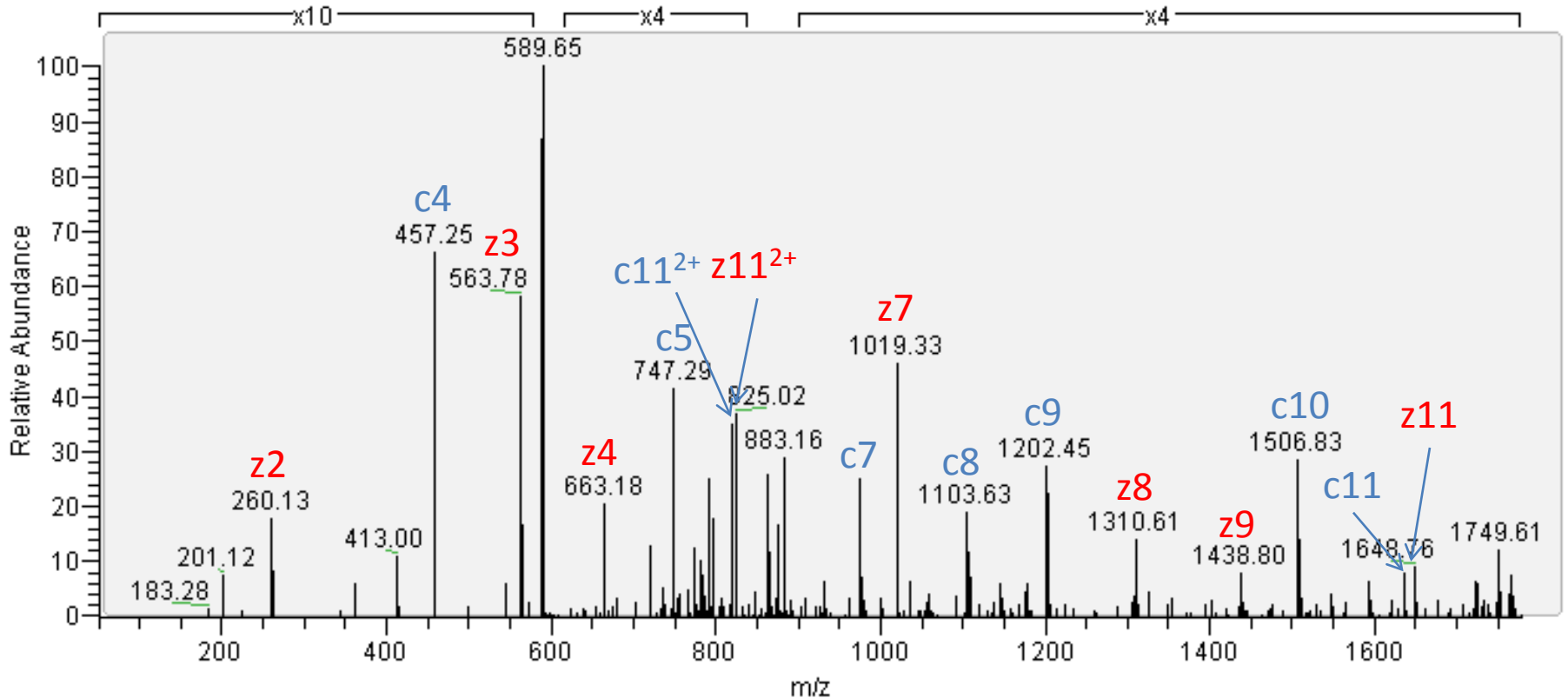


m/z 588.986 3+

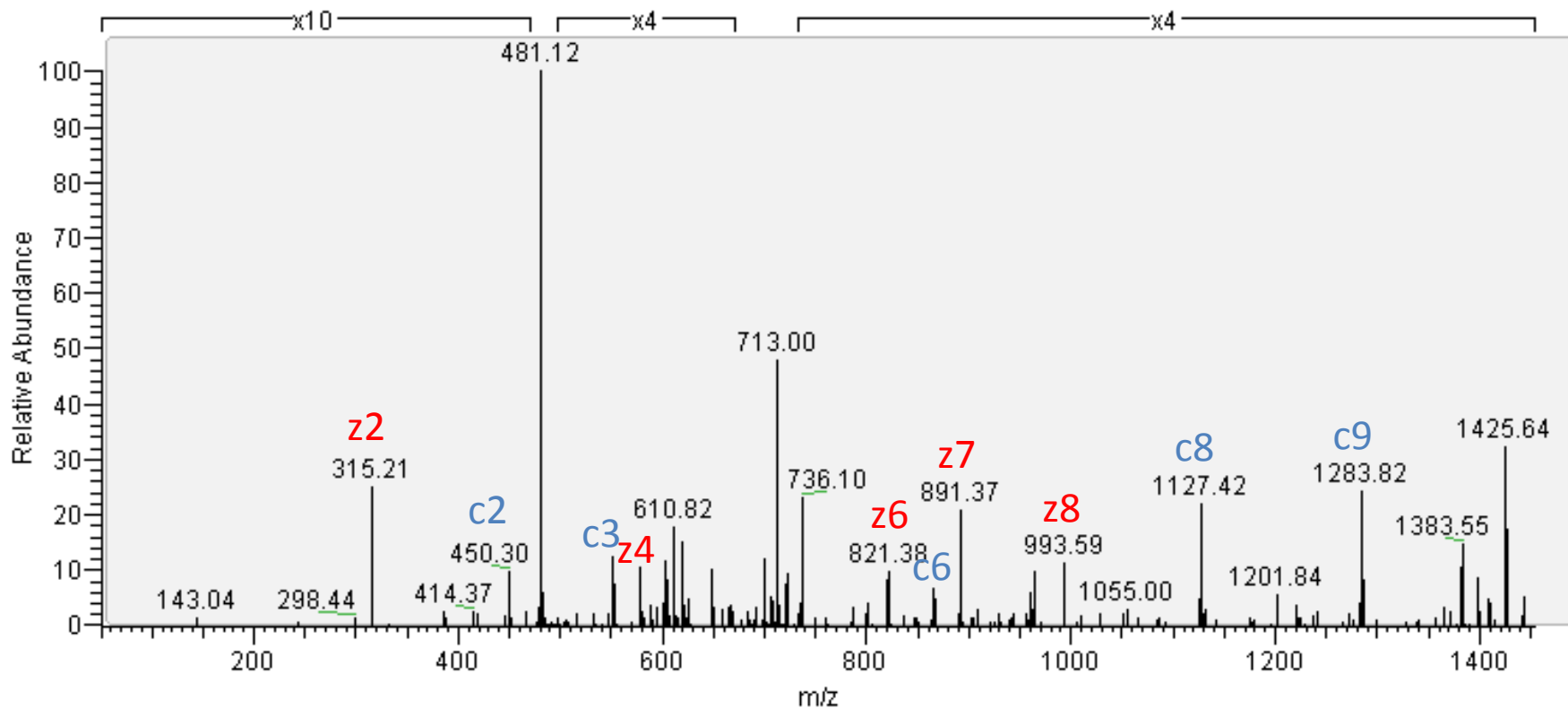
TIPKS(HexNAc)EVKVT(HexNAc)EK – Protein Piccolo

Mass difference between c4-c5 identifies one site as residue 2634

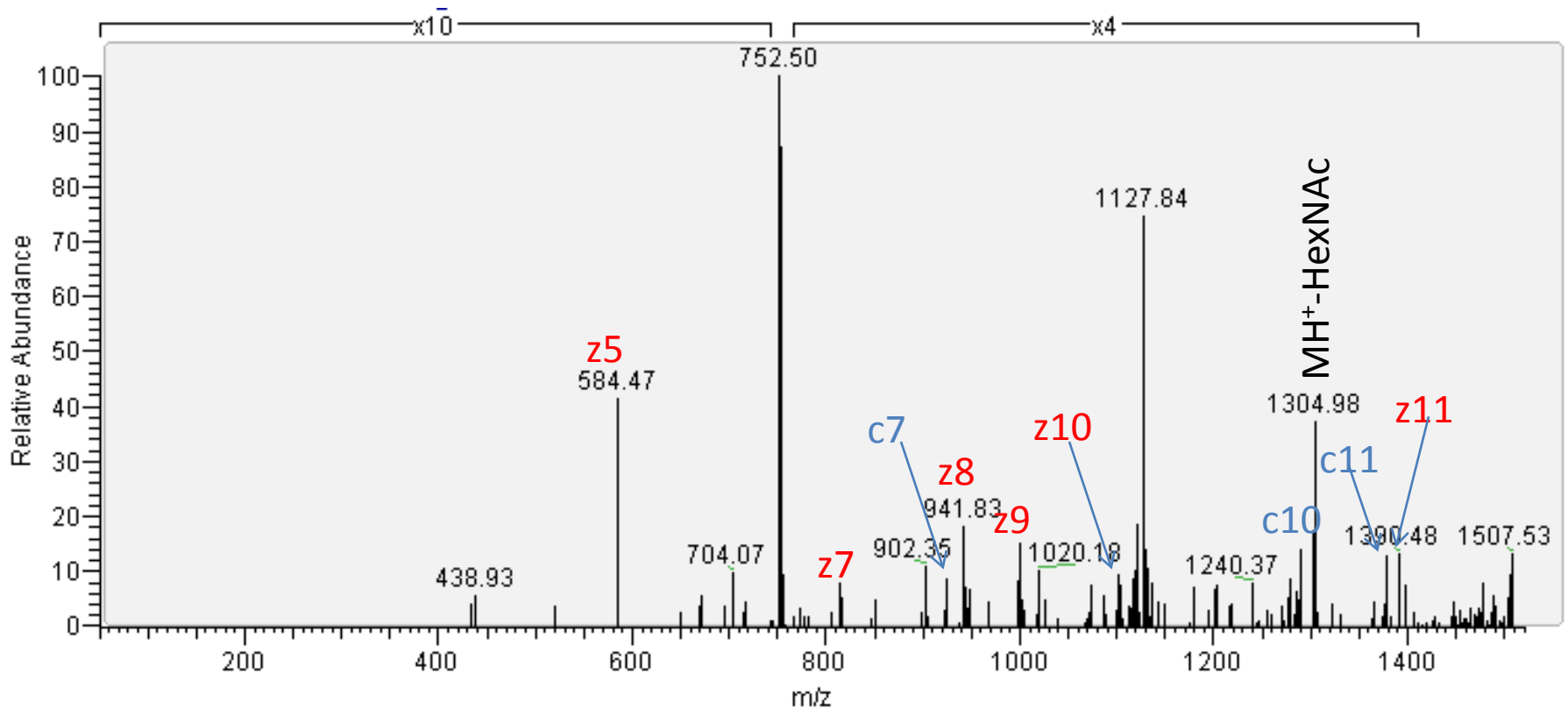
Mass difference between z2-z3 identifies other site as residue 2639



m/z 480.905 3+  
QT(HexNAc)TANEVYRR – Protein Piccolo  
Modified c2 identifies site as residue 2656

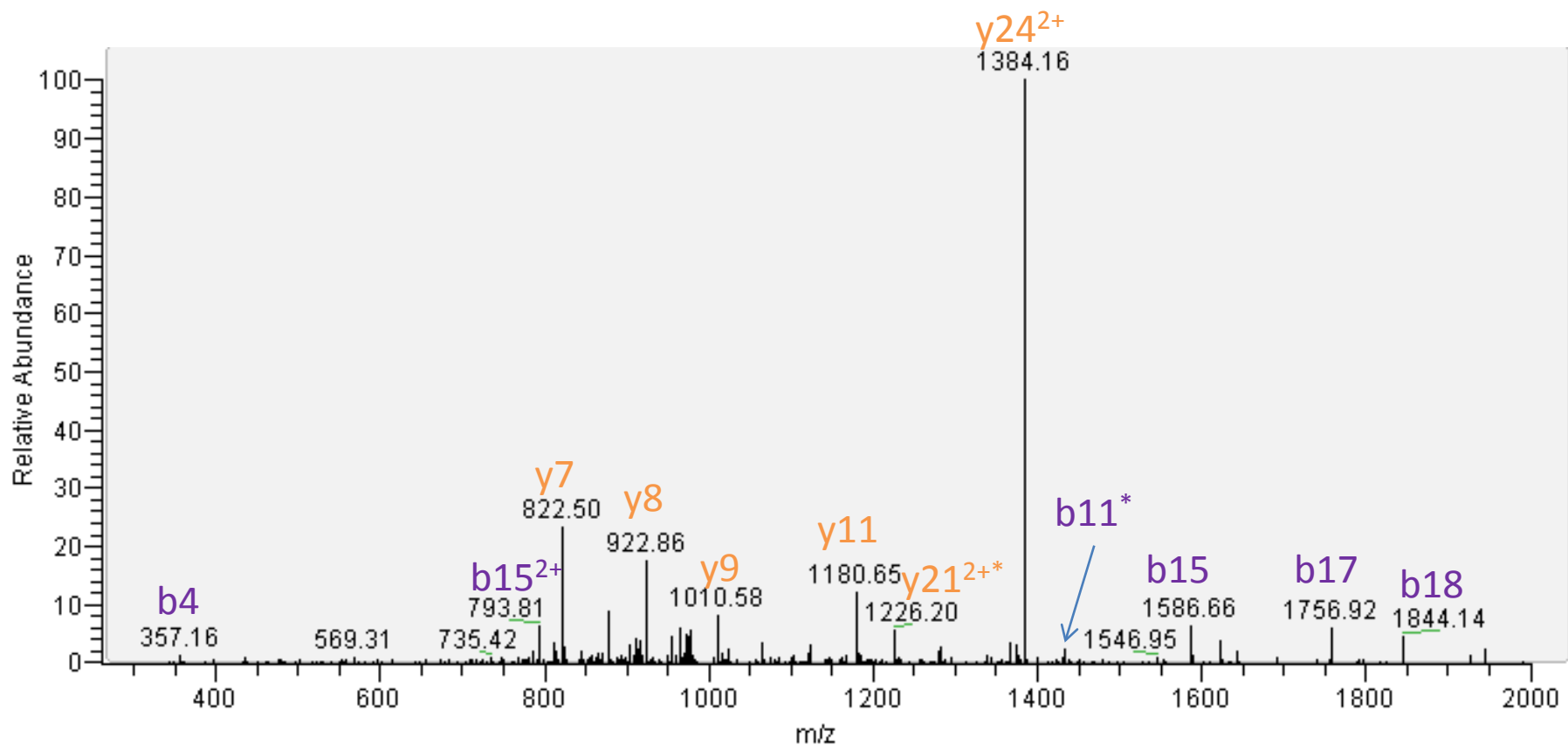


m/z 753.337 2+  
VS(HexNAc)TGEVMDYSSK - Protein piccolo  
Mass difference between z10-z11 identifies site as residue 2930

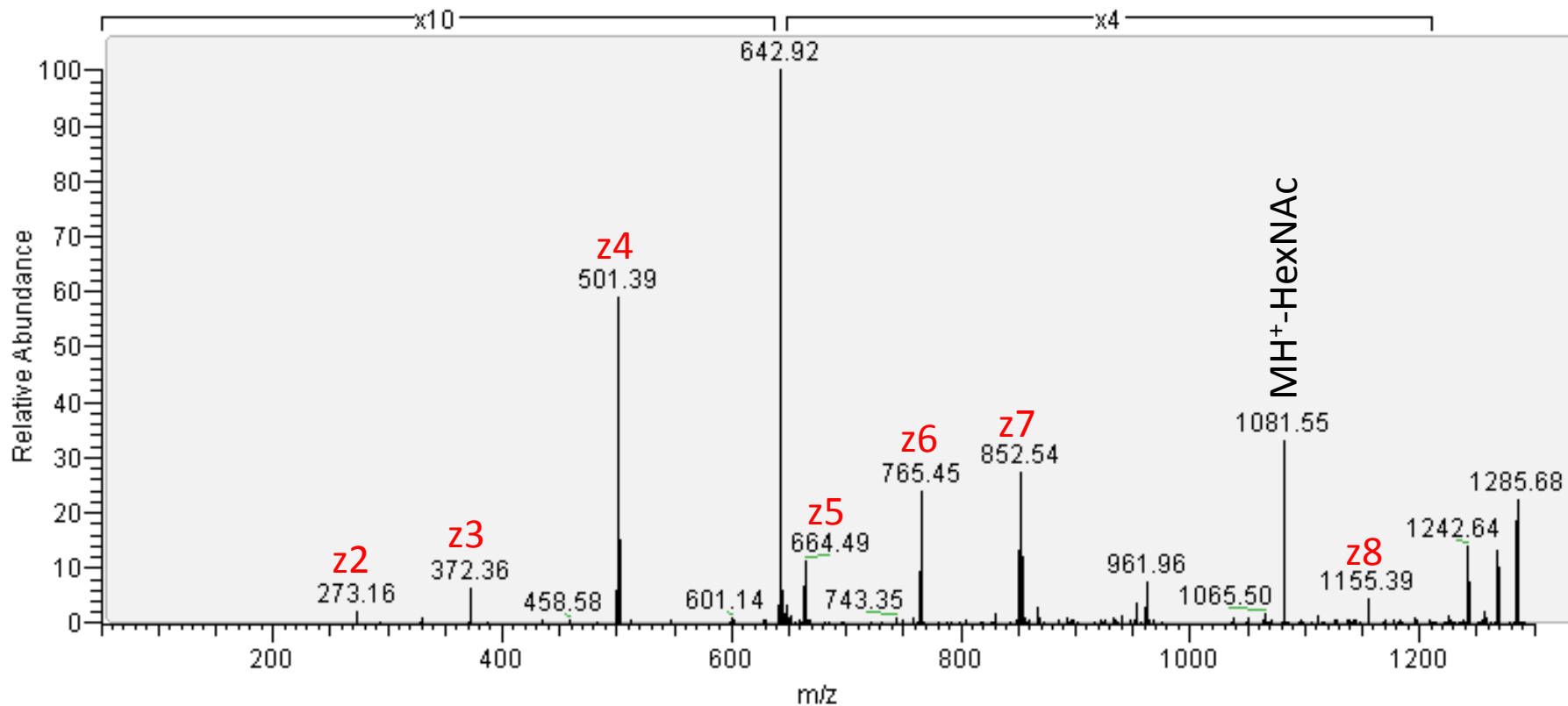


m/z 990.500 3+  
TTGPYPET(HexNAc)RQVISGVGISTPQYSTAR – Protein Piccolo  
Modified b11 and y21 identify site as residue 2948

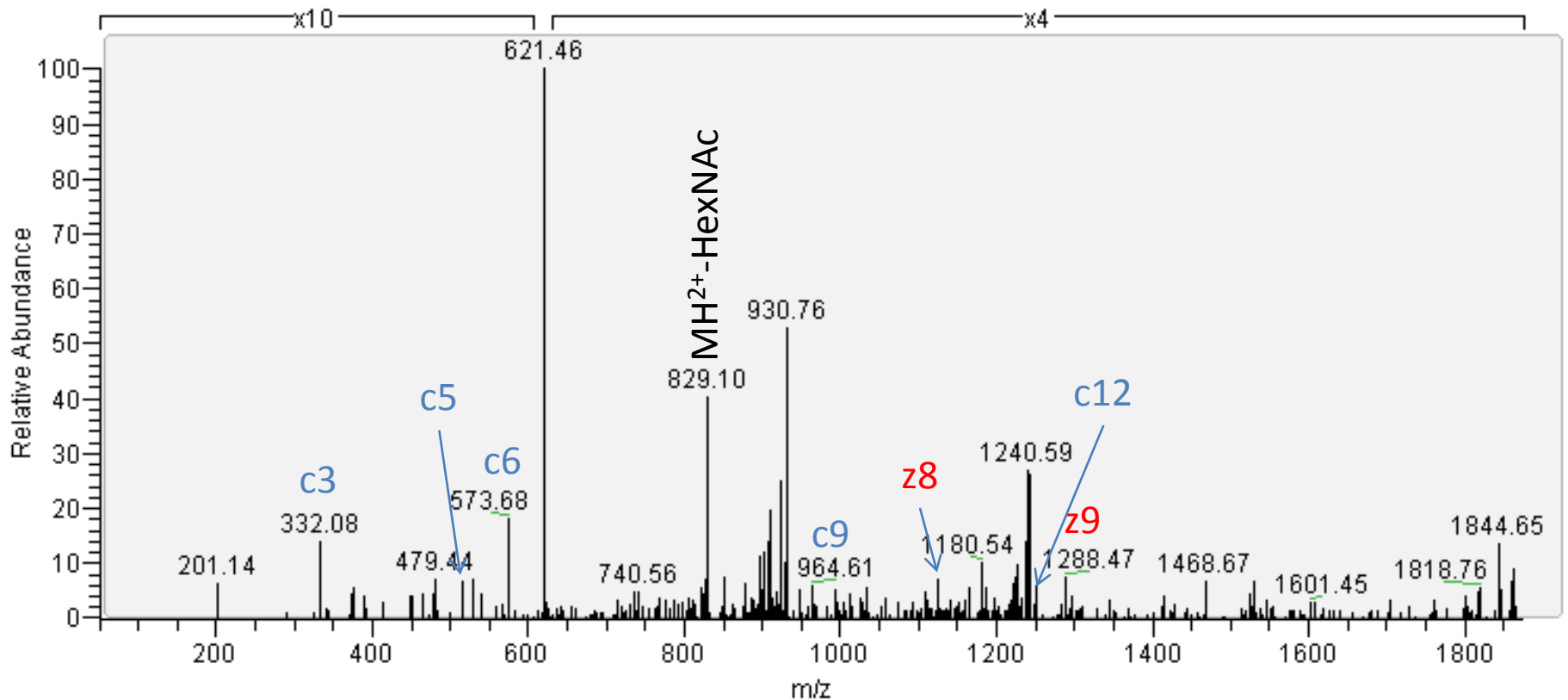
CID



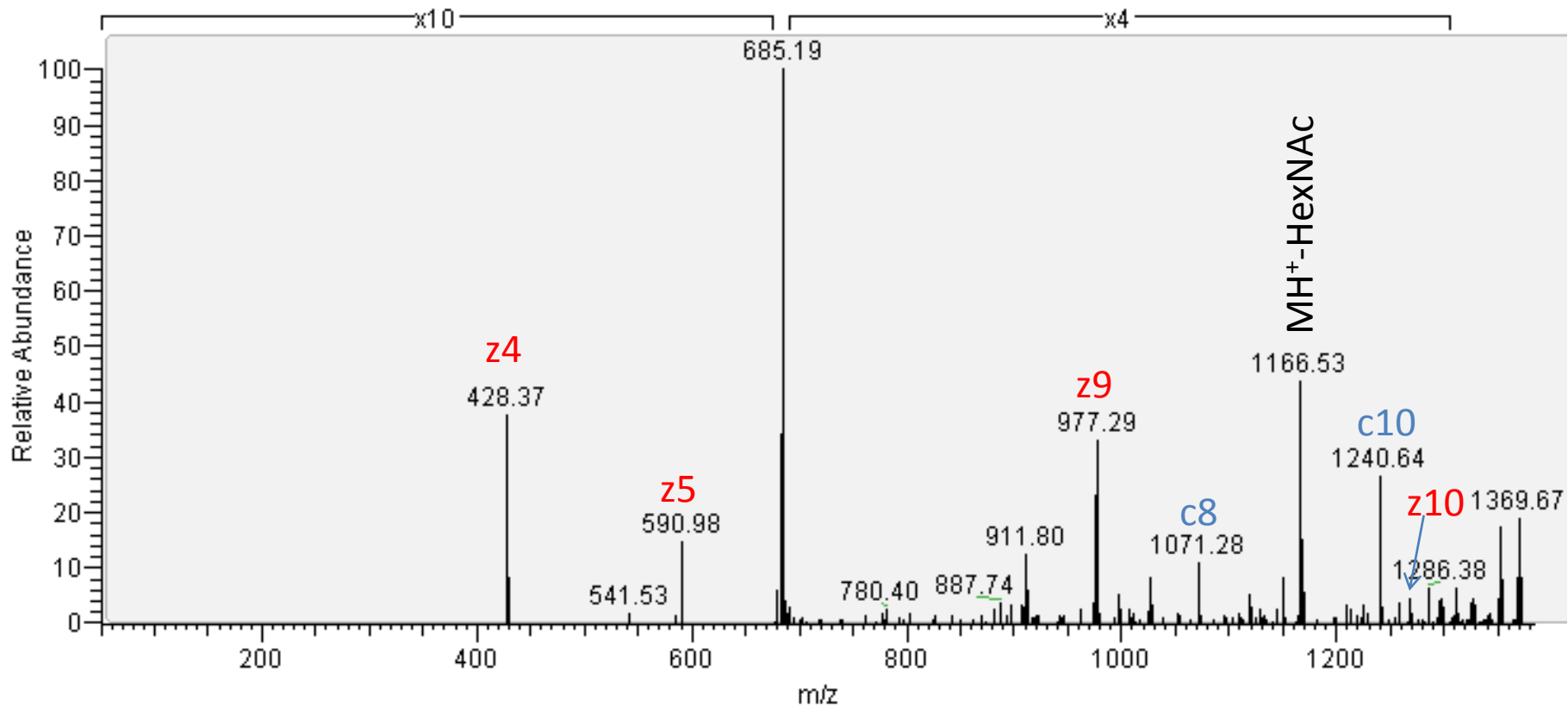
m/z 642.836 2+  
IT(HexNAc)STYEVIR - Protein piccolo  
Mass difference between z7-z8 identifies site as residue 3873



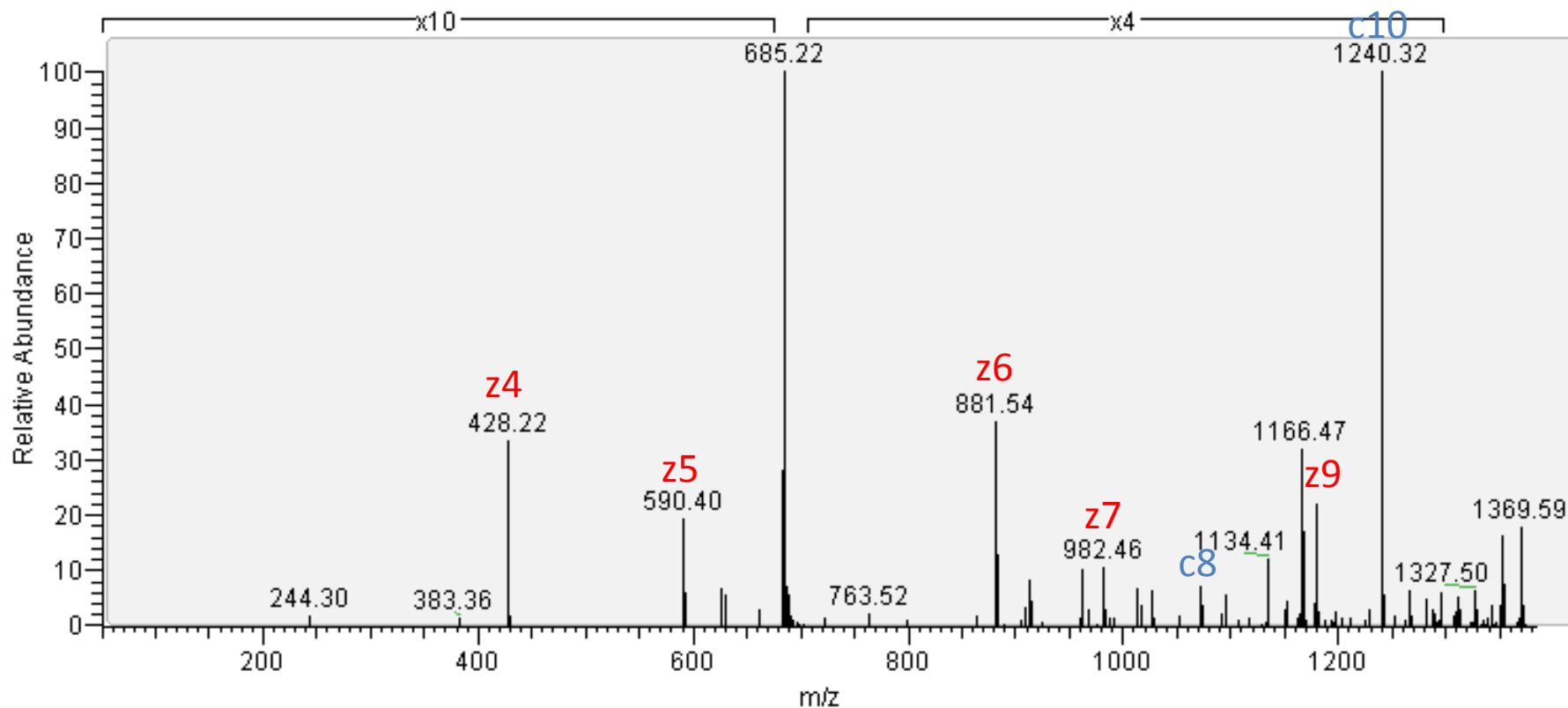
m/z 620.629 3+  
RASSPGYIDSPTYS(HexNAC)R – Actin-binding LIM protein 3  
Unmodified c12 identifies site as residue 383



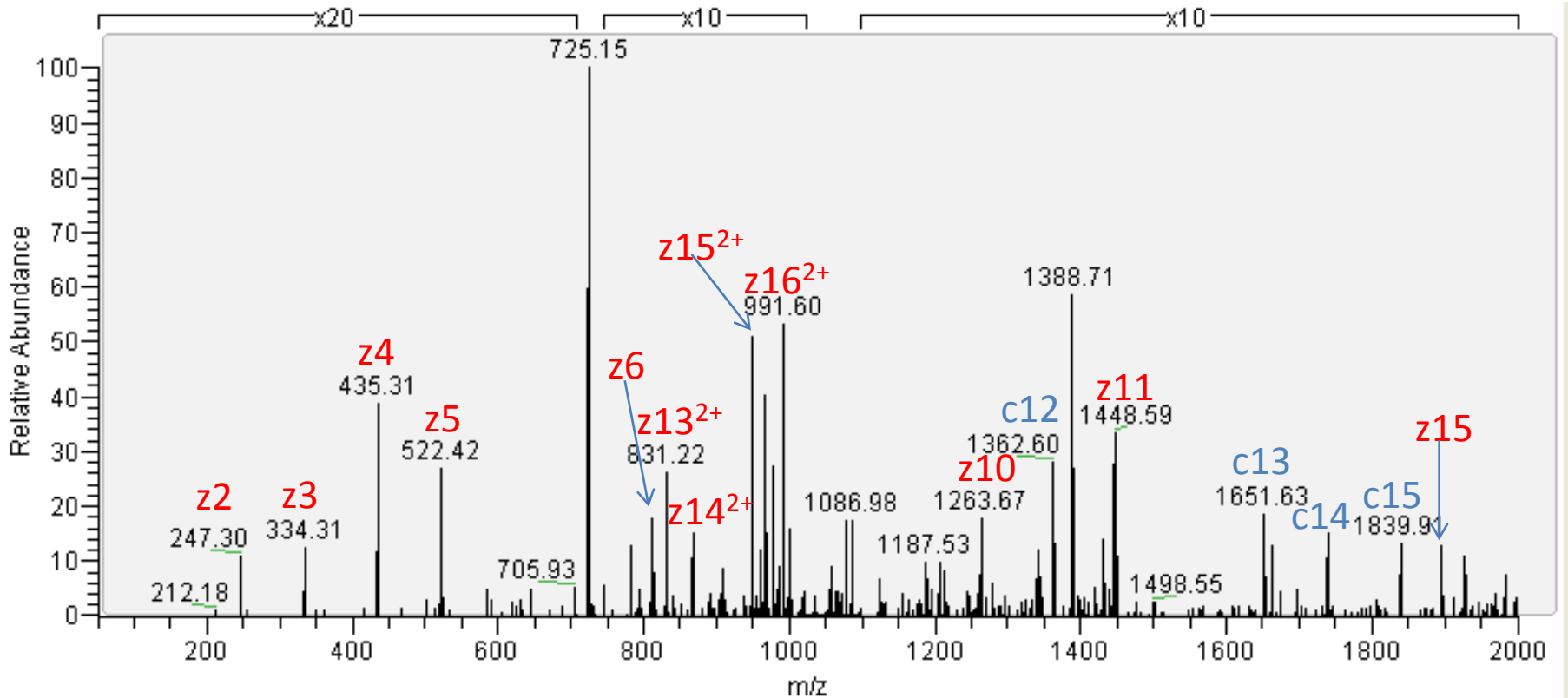
m/z 685.329 2+  
SS(HexNAc)TPTS<sup>Y</sup>QAPK – Actin-binding LIM protein 3  
Mass difference between z9-z10 identifies site as residue 419



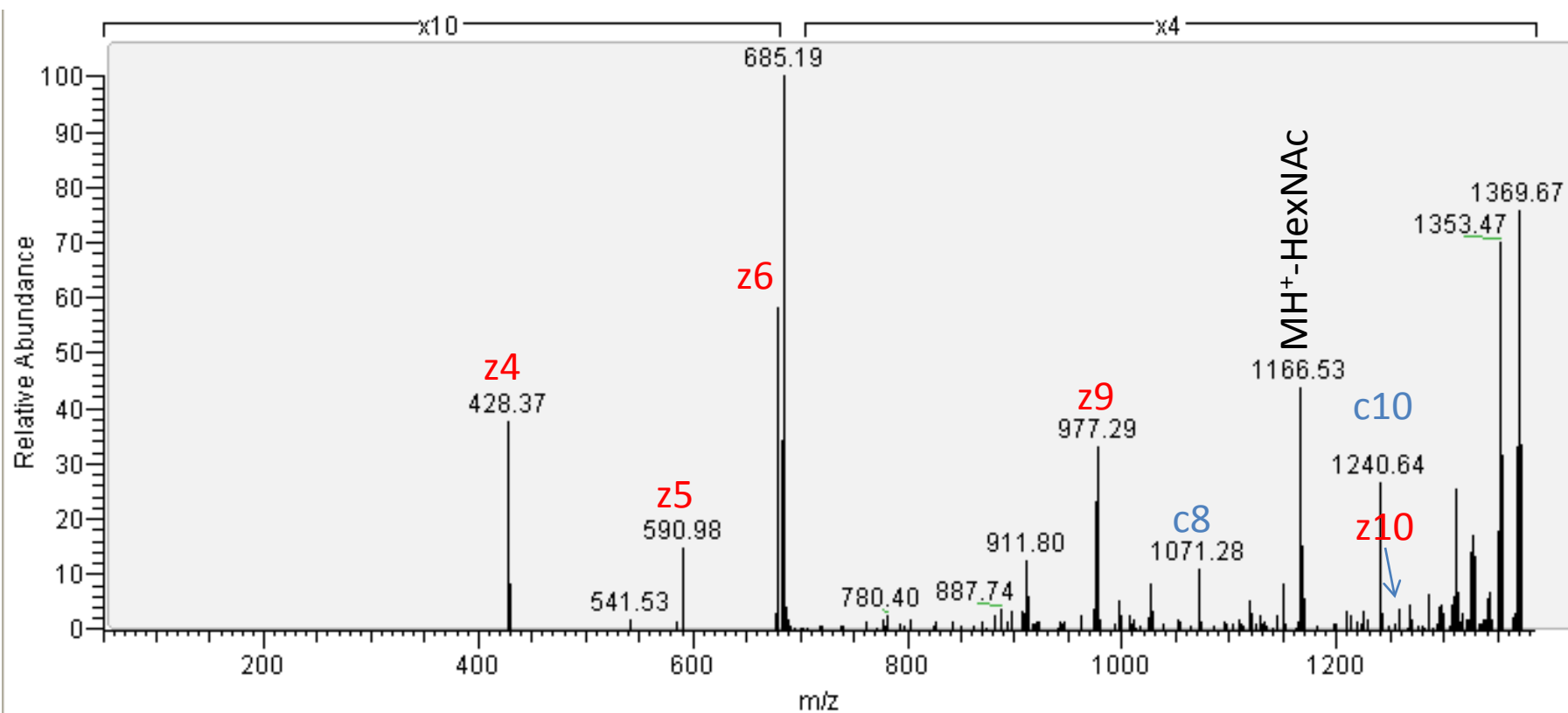
m/z 685.329 2+  
SSTPTS(HexNAc)YQAPK – Actin-binding LIM protein 3  
Mass difference between z5-z6 identifies residue 423



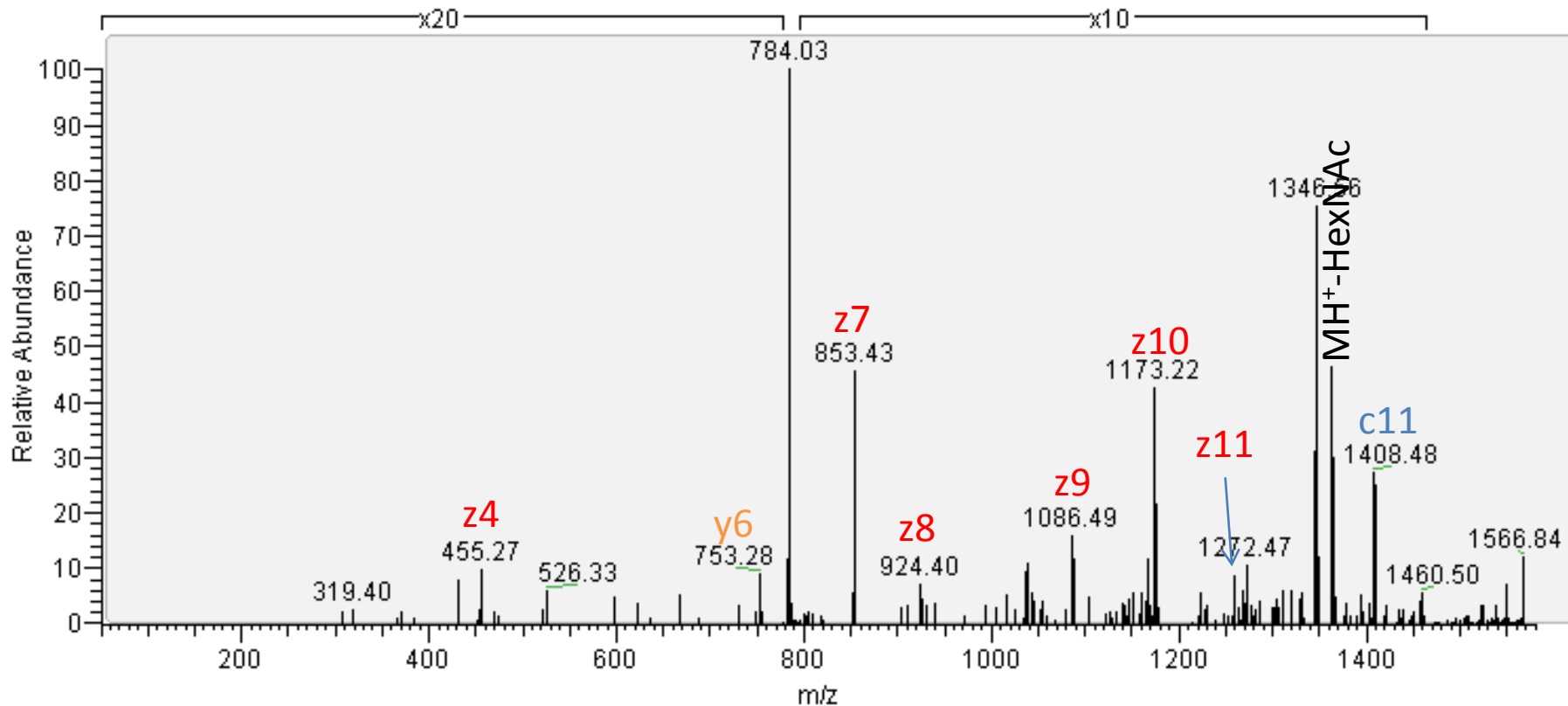
m/z 724.666 3+  
SSSYADPWTPPRS(HexNAc)STSSR - Actin-binding LIM protein 3  
Mass difference between c12-c13 identifies residue 546



m/z 685.329 2+  
SS(HexNAc)TPTSYPQAPK - Actin-binding LIM protein 3  
Mass difference between z9-z10 identifies site as residue 547

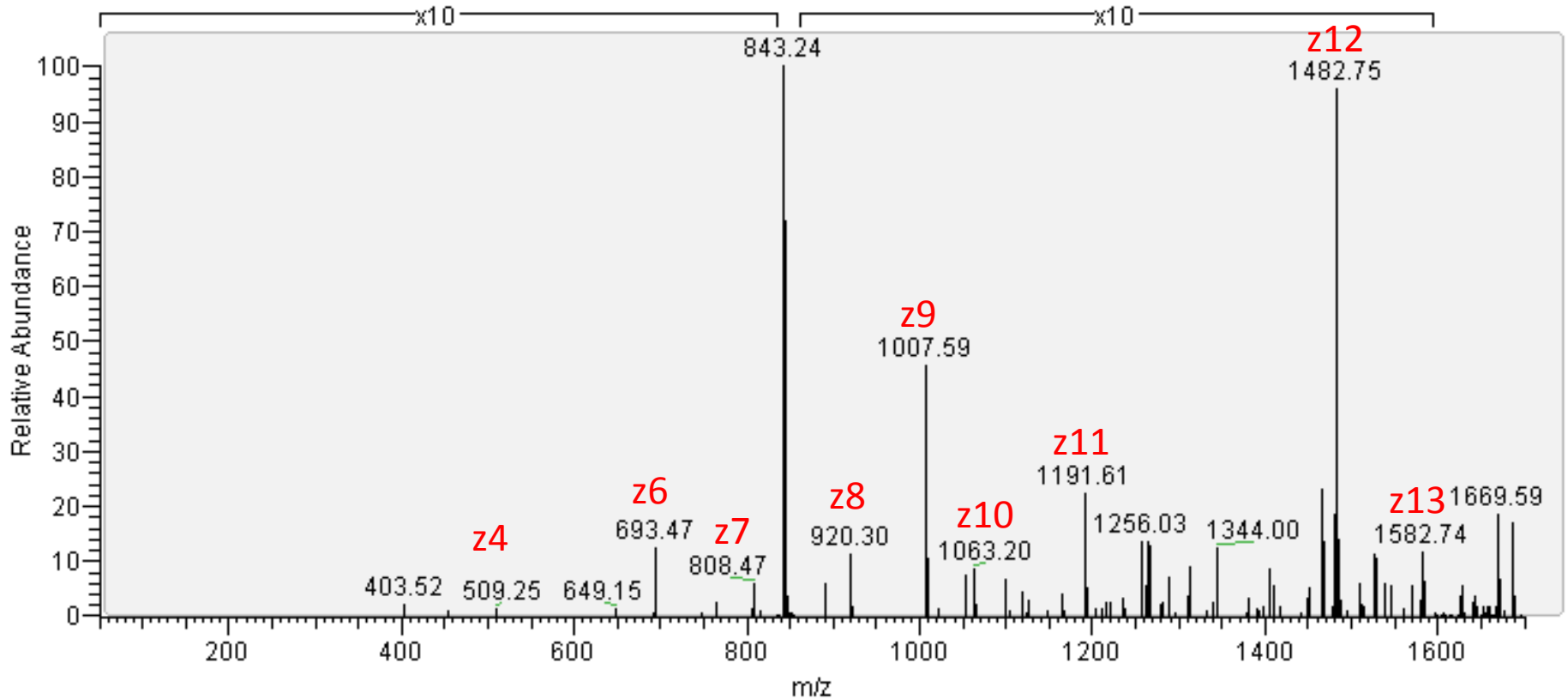


m/z 783.854 2+  
S(HexNAc)SSYADPWTPPR - Actin-binding LIM protein 3  
Unmodified z11 identifies site as residue 534

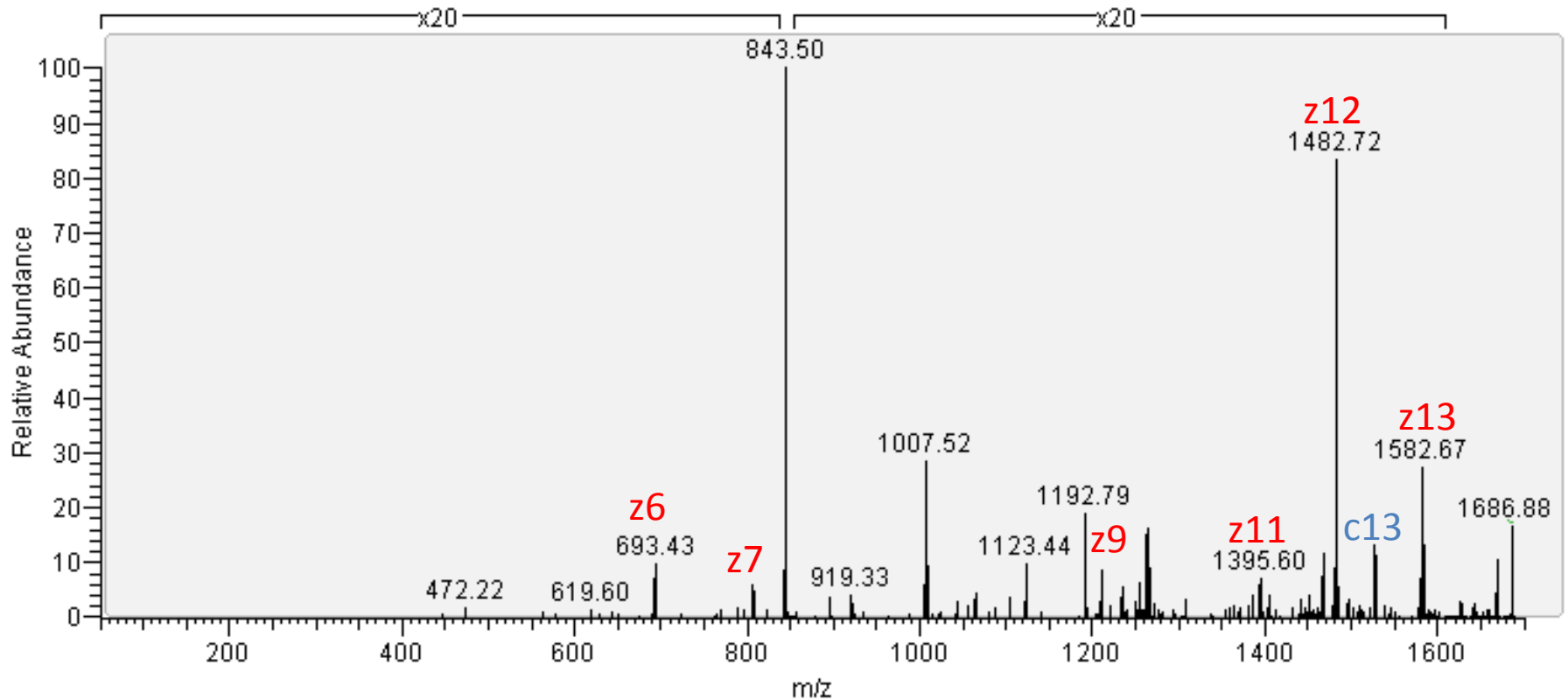


m/z 843.402 2+

STS(HexNAc)QGSINSPVYSR - Actin-binding LIM protein 1  
Mass difference between z11-z12 identifies site as residue 496



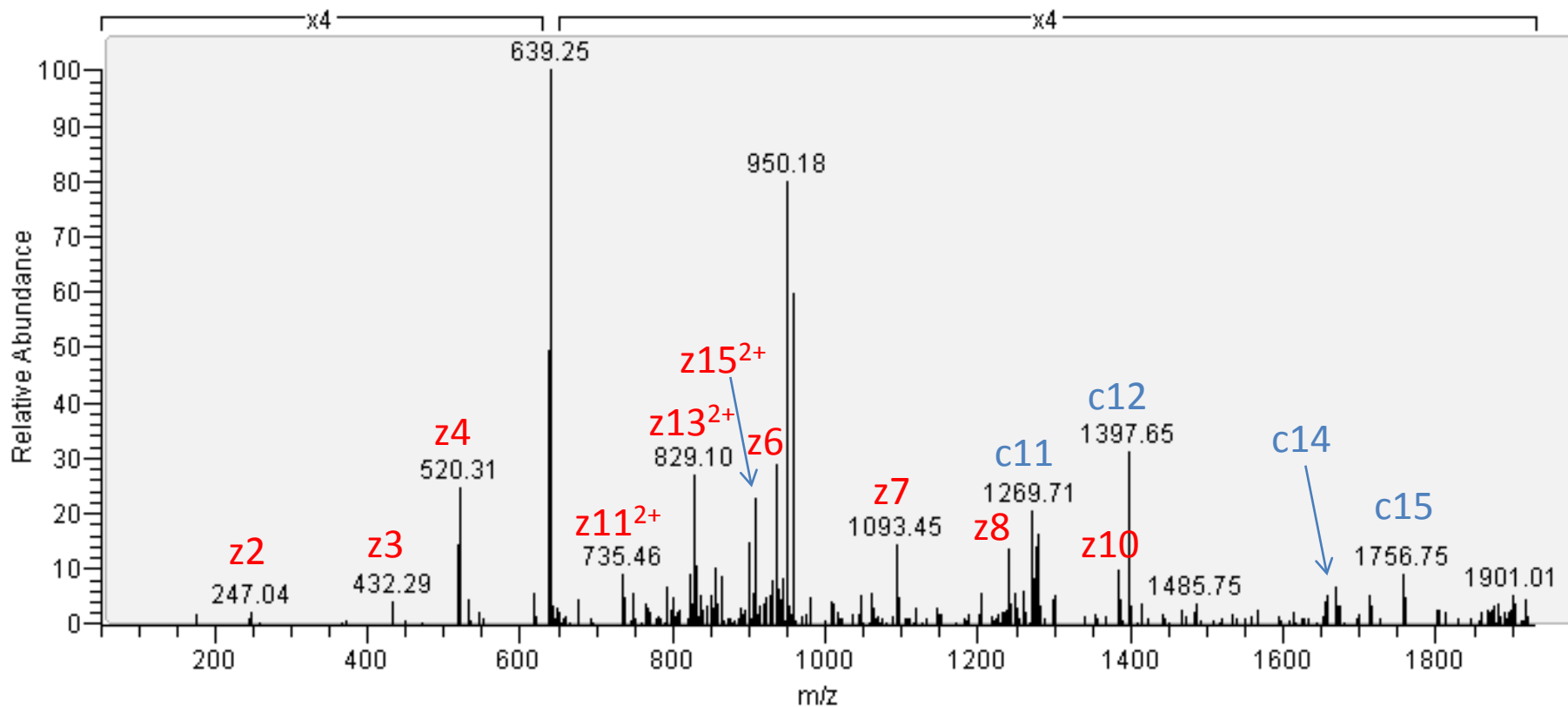
m/z 843.402 2+  
STSQGS(HexNAc)INSPVYSR - Actin-binding LIM protein 1  
Unmodified z7 and modified z9 identify site as residue 499



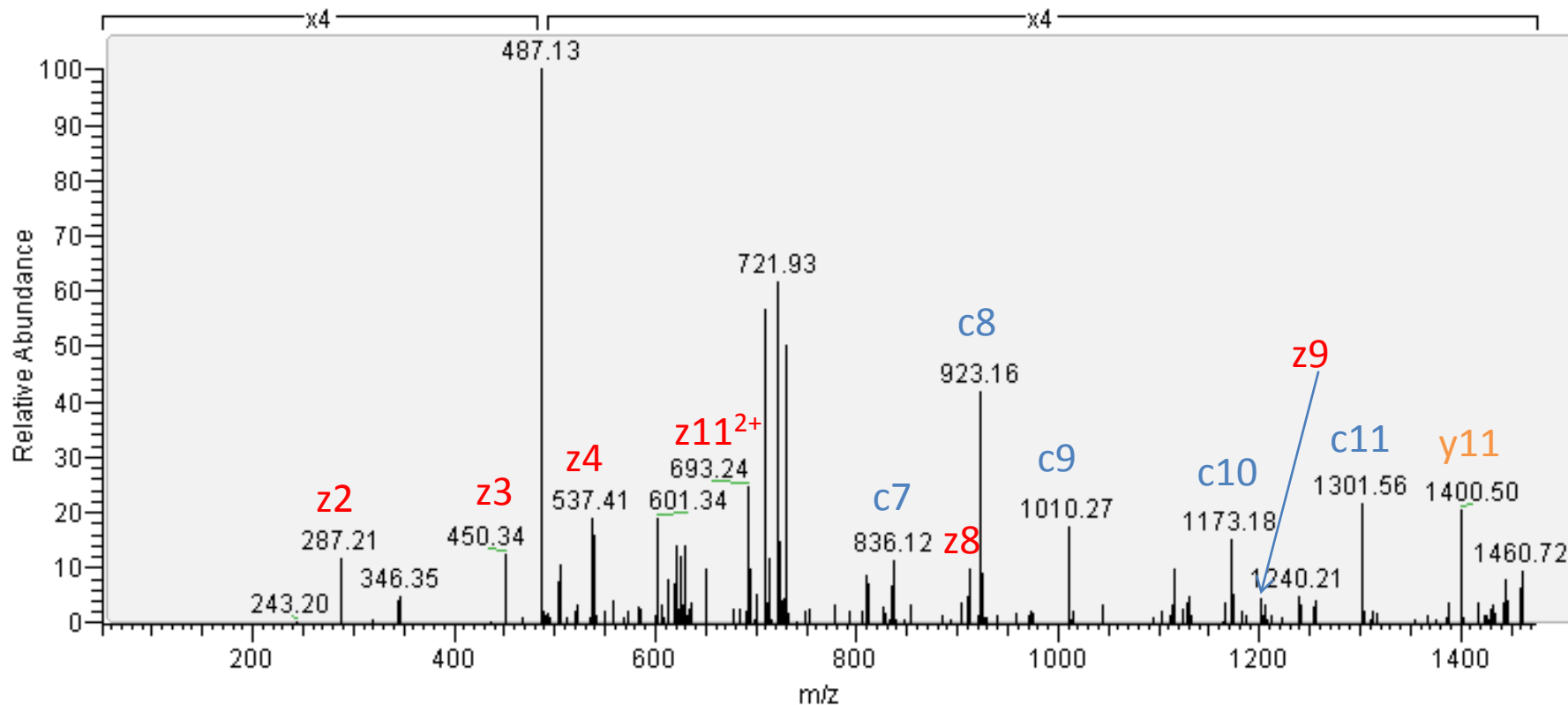
m/z 638.965 3+

VSGSPSSGFRS(HexNAc)QSWSR - Neurofilament medium polypeptide

Unmodified z4, but modified z6 identifies site as residue 37



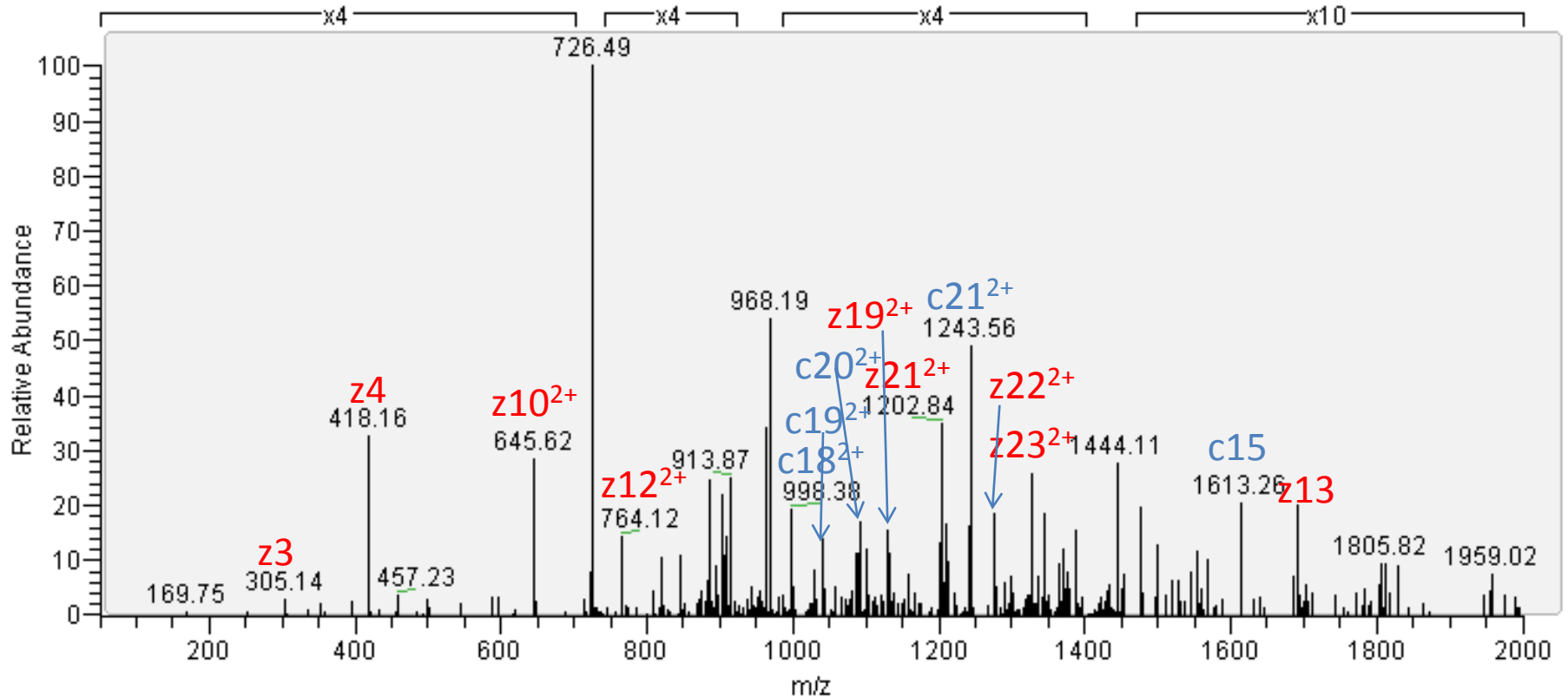
m/z 486.903 3+  
GSPS(HexNAc)TVSSSYKR - Neurofilament medium polypeptide  
Mass difference between z8-z9 identifies site as residue 46



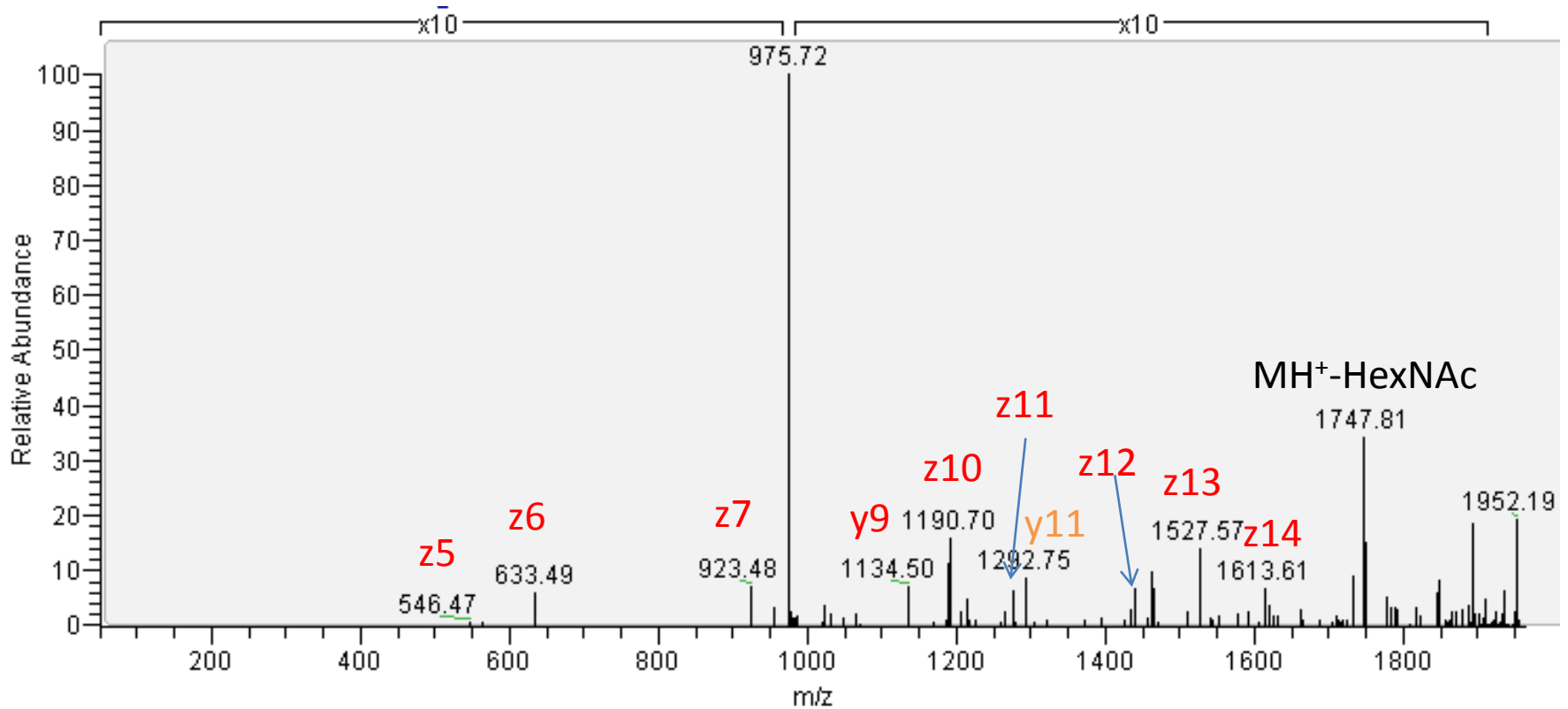
m/z 726.121 4+

FSTFSGSITGPLYTHRQPSVT(HexNAc)ISSK - Neurofilament medium polypeptide

Mass difference between c20-c21 identifies site as residue 430



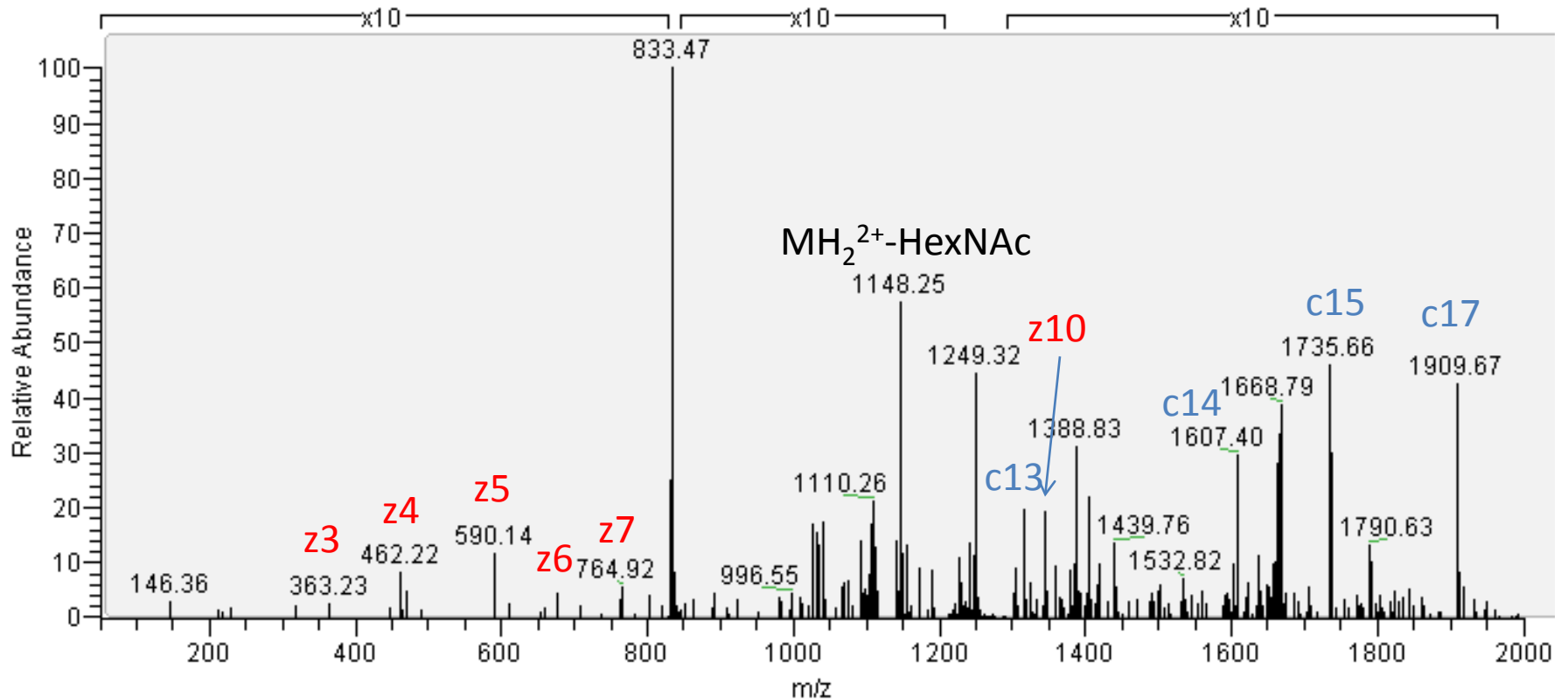
m/z 975.965 2+  
SAYSSYSAPVS(HexNAc)SSLSVR - Neurofilament light polypeptide  
Mass difference between z6-z7 identifies site as residue 48



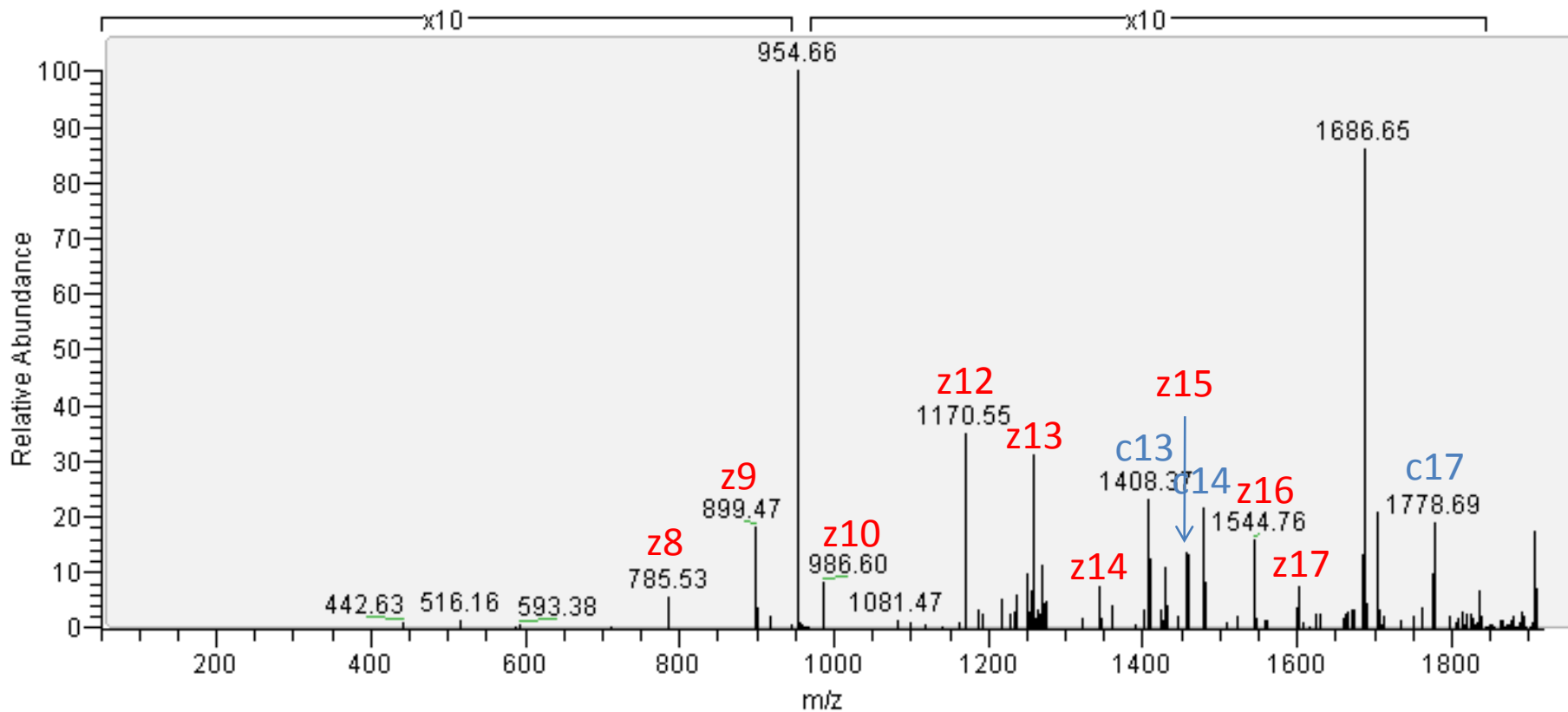
m/z 833.406 3+

LSFTSVGSITSGYS(HexNAc)QSSQVFGR - Neurofilament light polypeptide

Mass difference between c13-c14 identifies site as residue 414



m/z 953.966 2+  
S(HexNAc)GSLSSSPNTPSASPLK - 270 kDa ankyrin G isoform  
Unmodified z17 identifies site as residue 1520



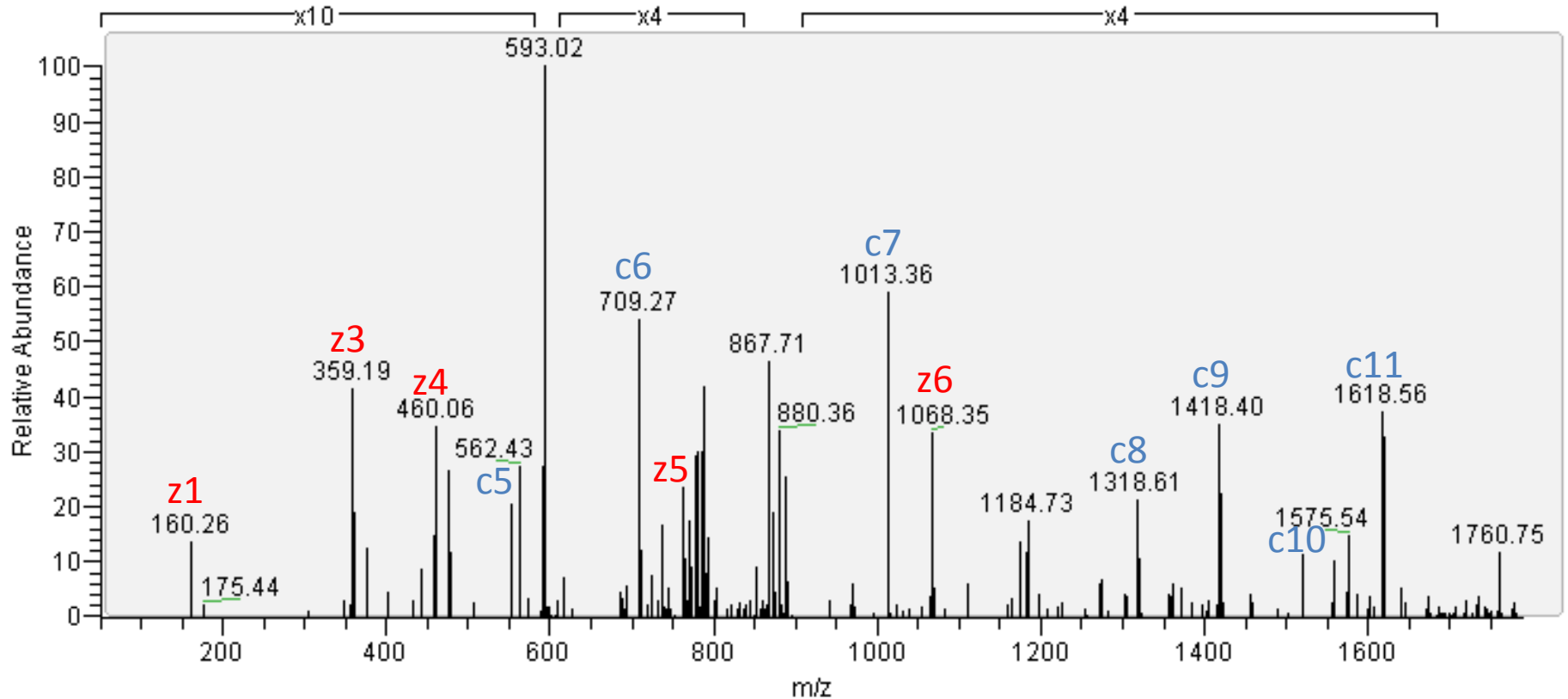
m/z 592.605 3+

S(Phospho)SSPPRT(HexNAc)T(HexNAc)TTVR - Disks large-associated protein 1

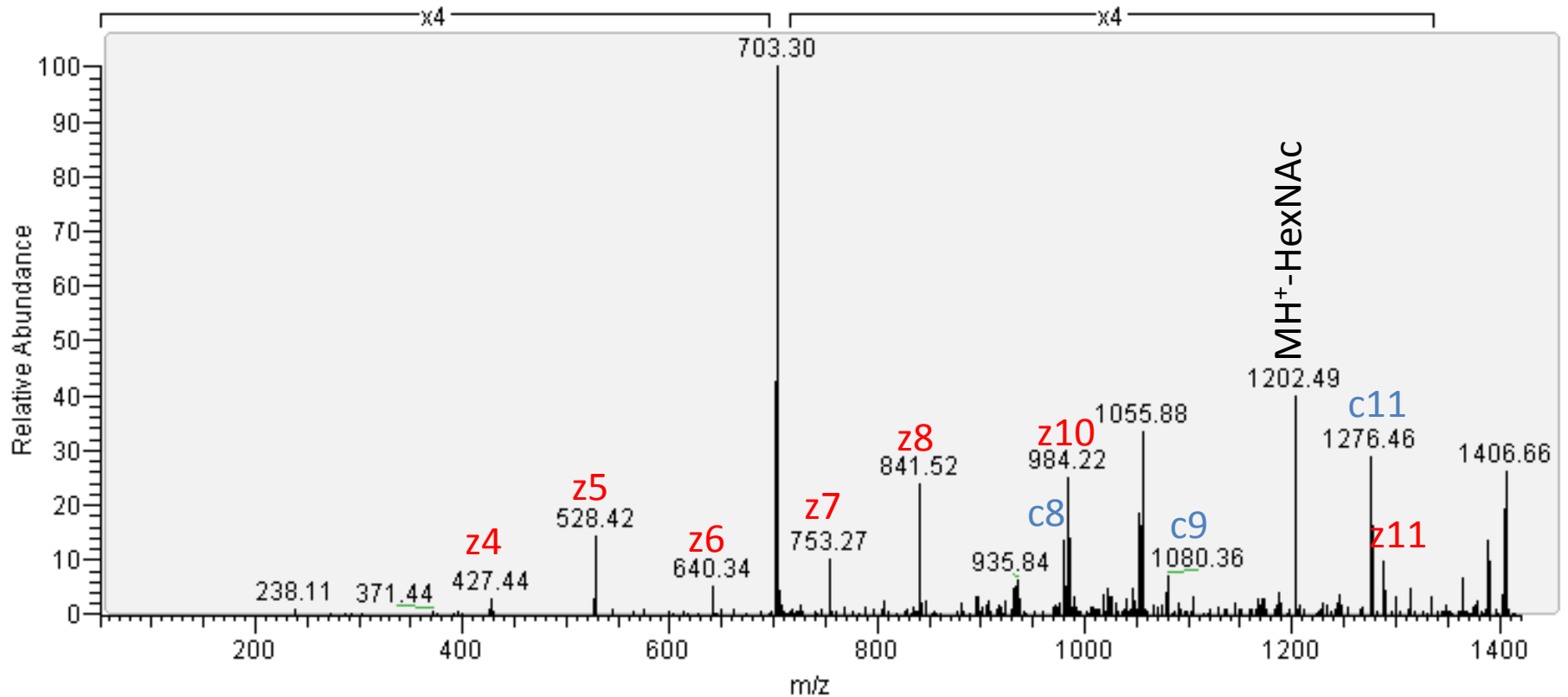
Mass difference between z4-z5 identifies site as residue 525

Mass difference between z5-z6 identifies site as residue 526

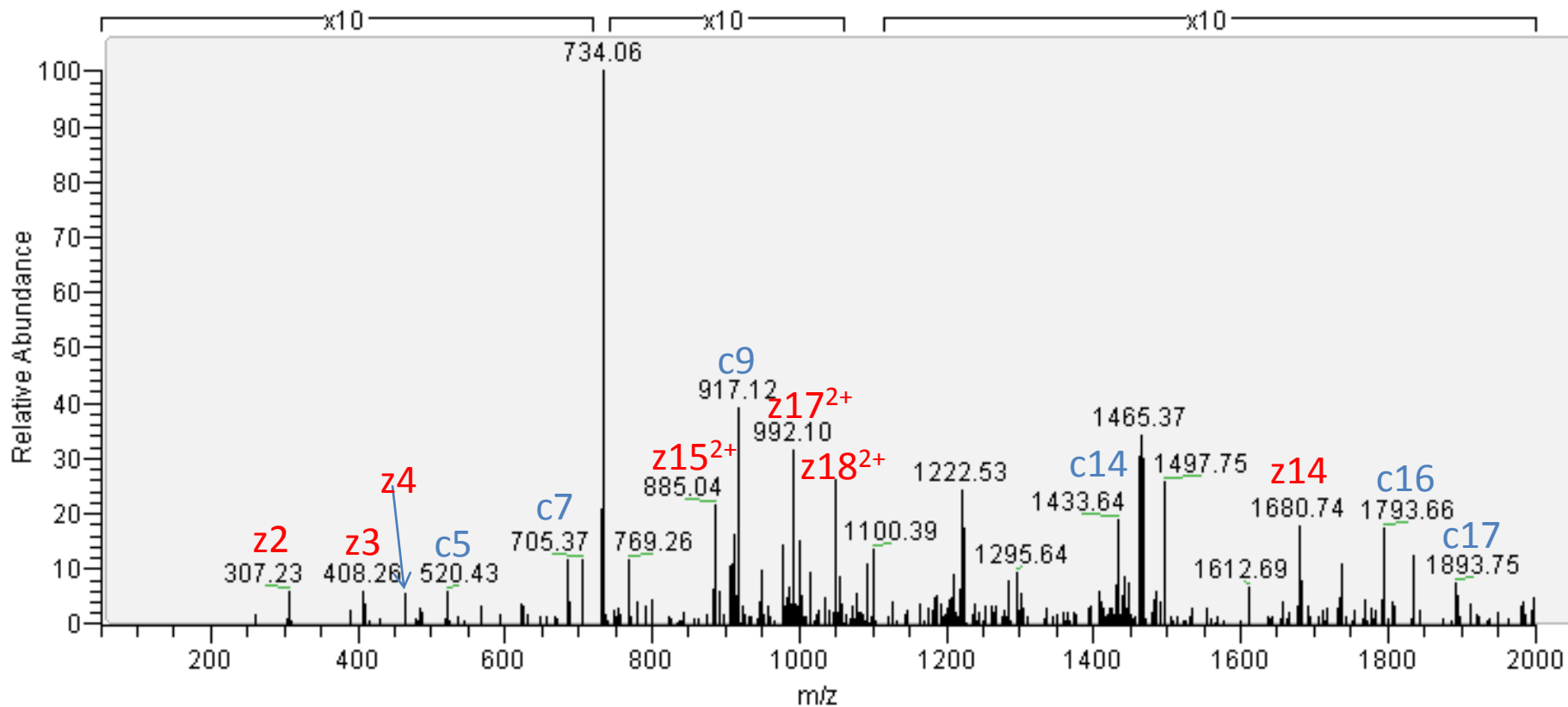
One of the serines is phosphorylated.



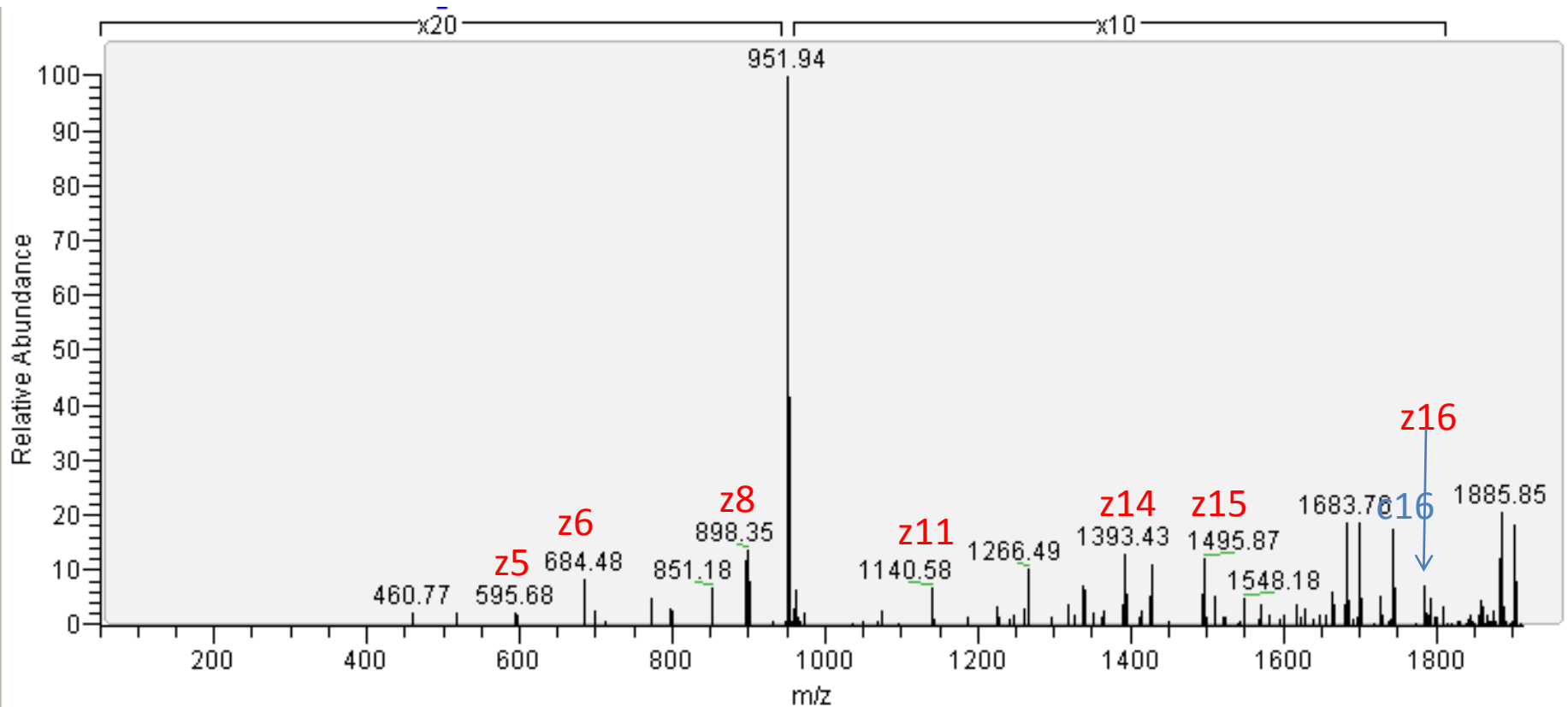
m/z 703.392 2+  
TT(HexNAc)SGSIITVVPK – Protein EMSY  
Mass difference between z10-z11 identifies site as residue 499



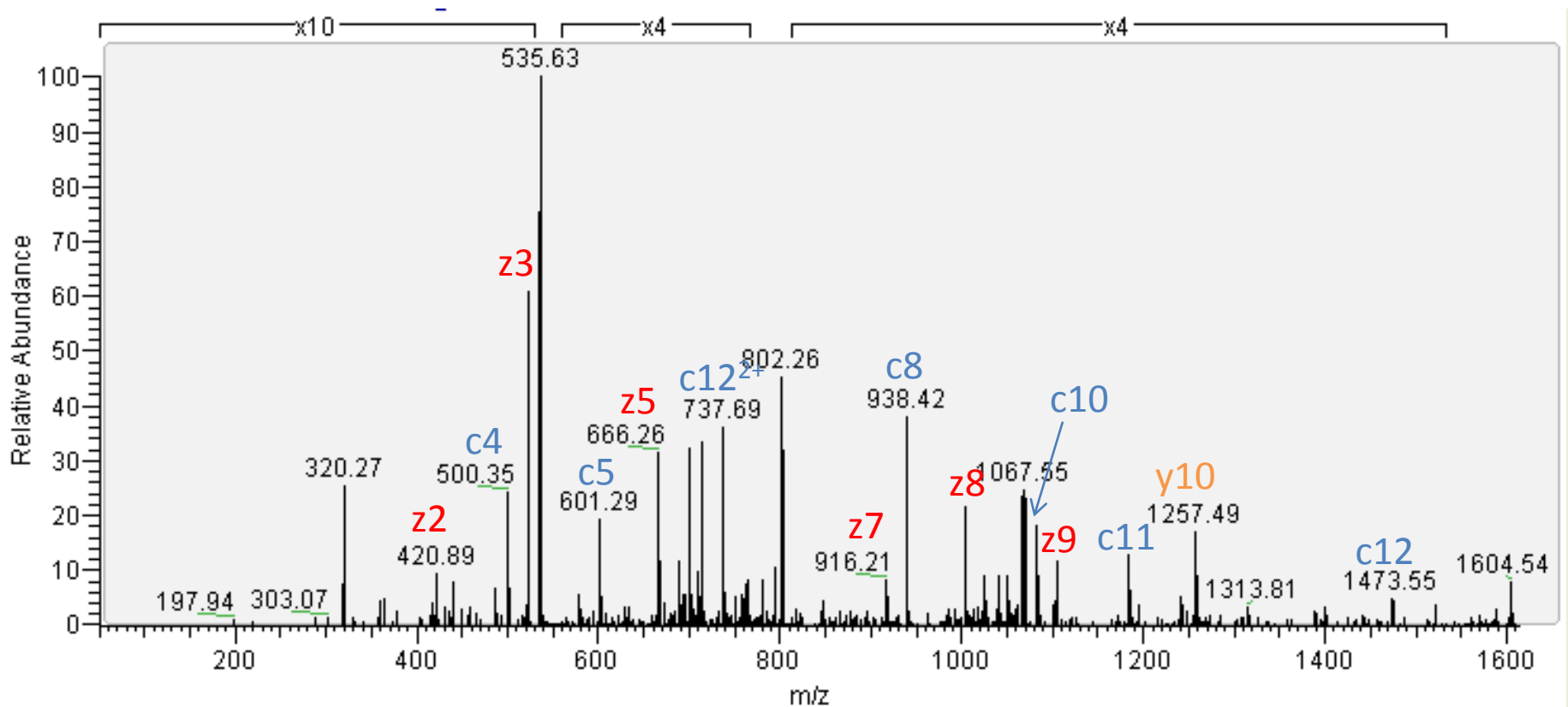
m/z 733.694 3+  
SLSQSQGDPLPPAHT(HexNAc)GTFR - Catenin delta-2  
Unmodified c14 and modified c16 identify site as residue 447



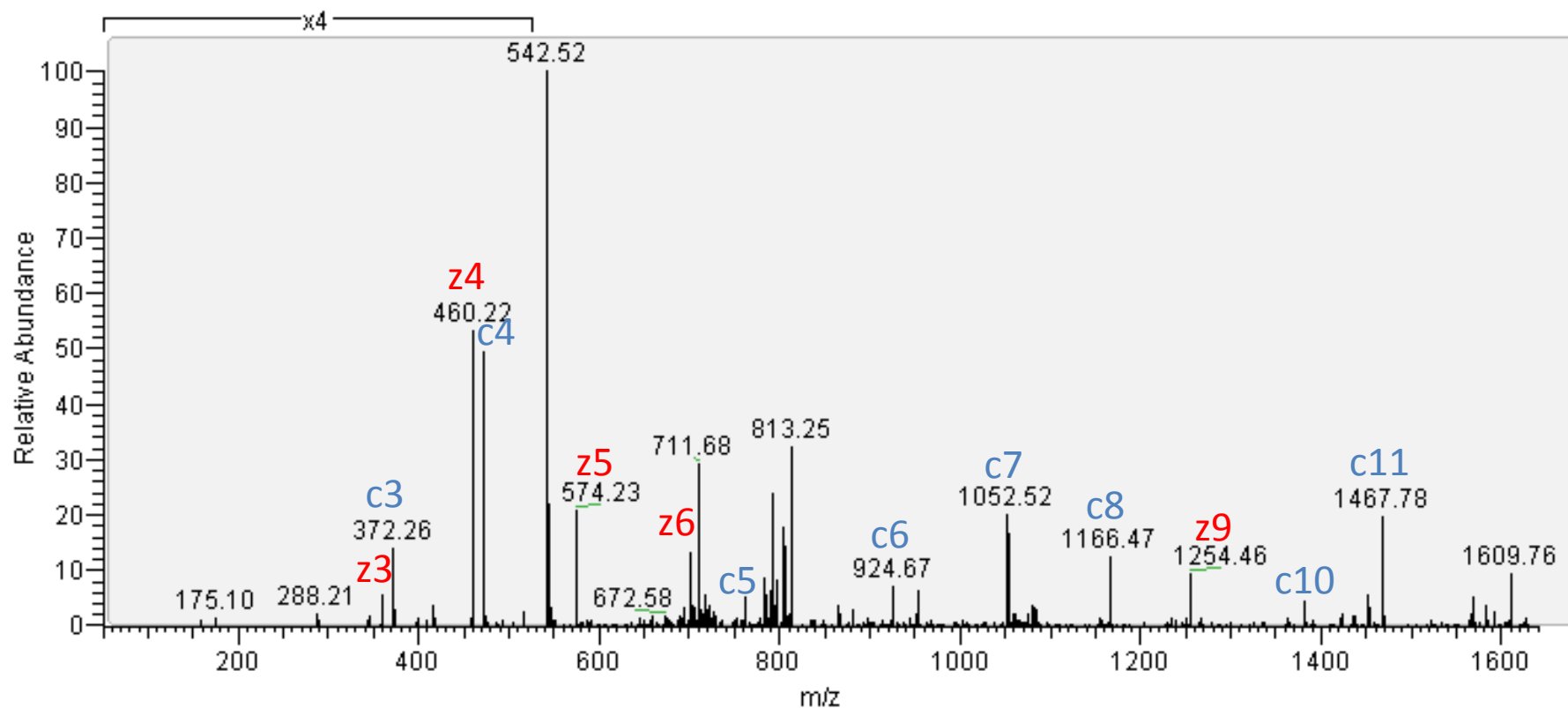
m/z 951.475 2+  
TS(HexNAc)TAPSSPGVDSVPLQR - Catenin delta-2  
Mass difference between z15-z16 identifies site as residue 453



m/z 534.930 3+  
TPFHTSLHSGTS(HexNAc)K – Dematin  
Modified z2 identifies site as residue 285



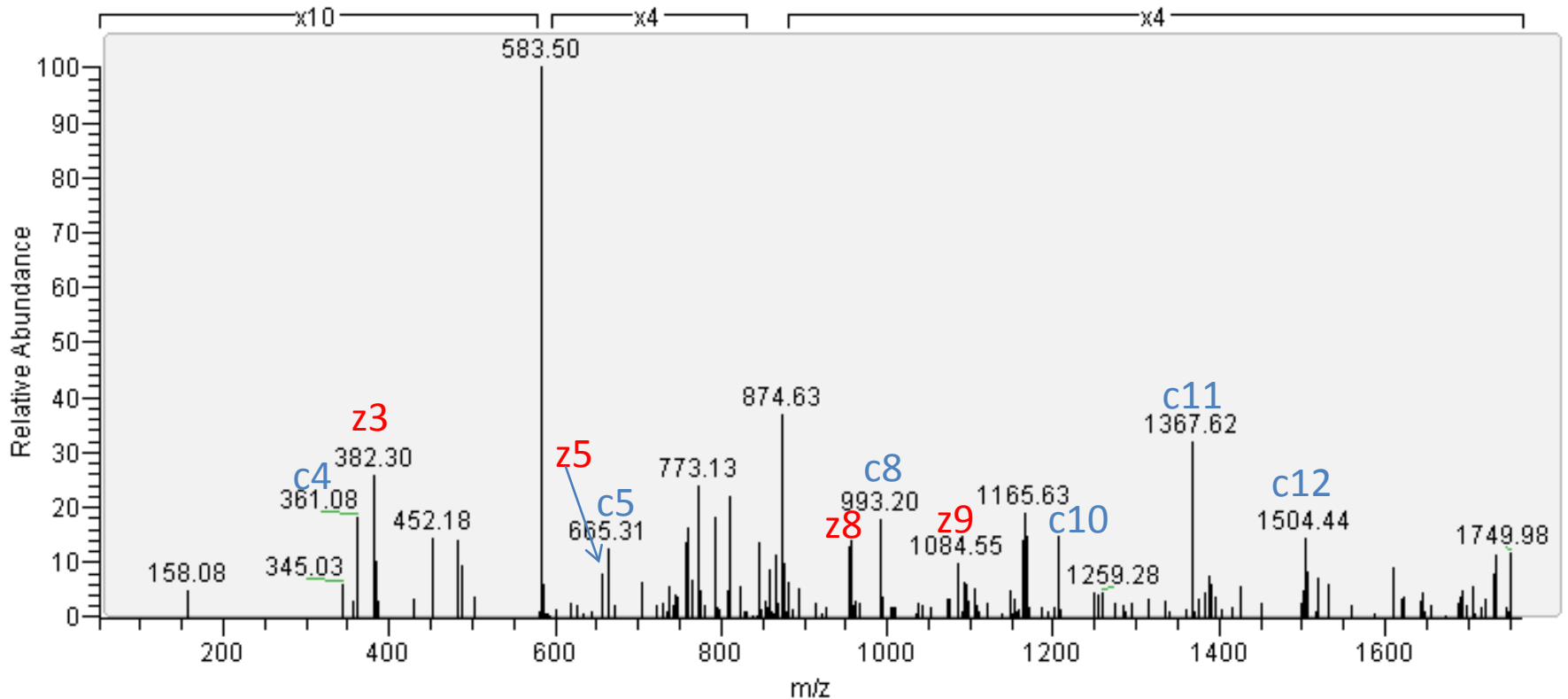
m/z 542.279 3+  
RTPVS(HexNAc)YQNTISR - Nuclear receptor corepressor 1  
Mass difference between c4-c5 identifies site as residue 1496



m/z 583.280 3+

SAGQT(HexNAc)QSLTIC(Carbamidomethyl)HNK - Polyhomeotic-like protein 3

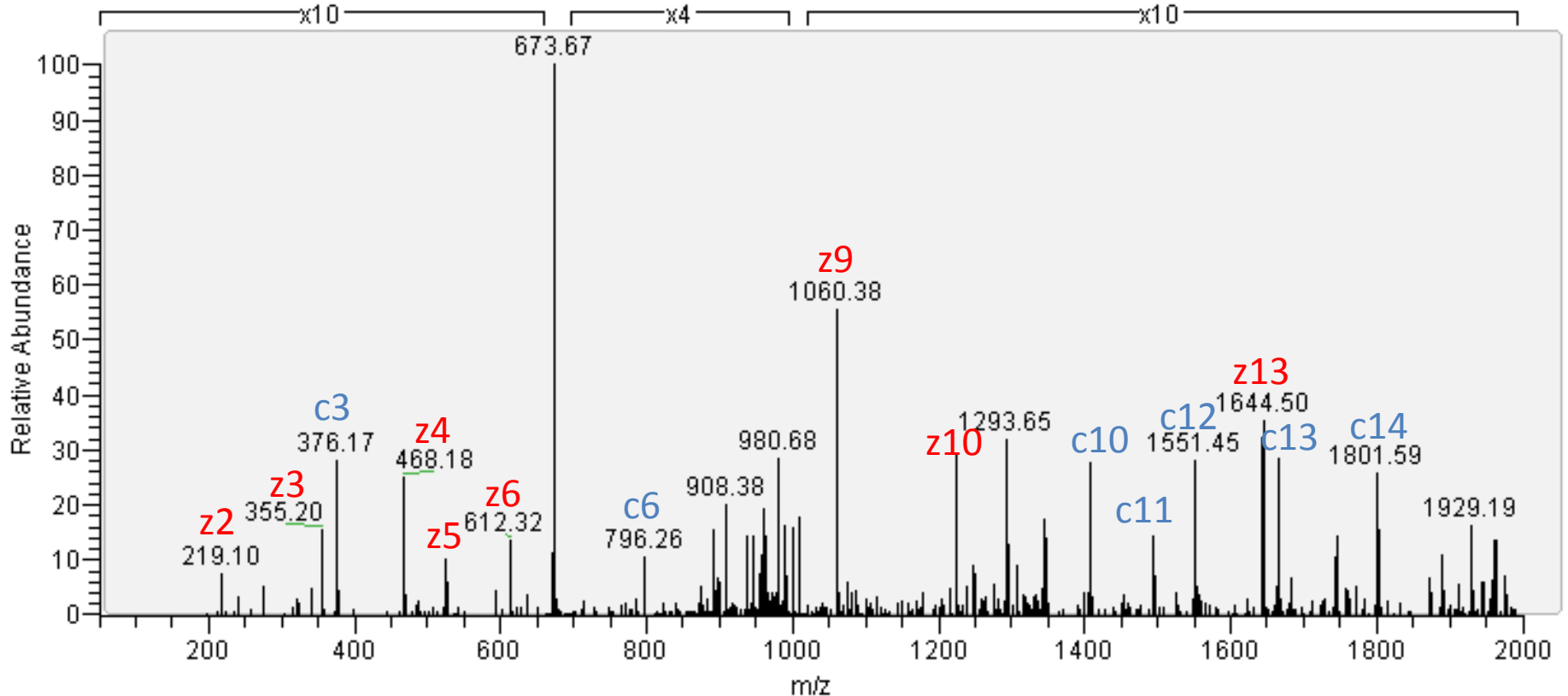
Mass difference between c4-c5 identifies site as residue 238



m/z 673.296 3+

EQTYPC(Carbamidomethyl)YS(HexNAc)GTSGLHSK - Human immunodeficiency virus type I enhancer-binding protein 2

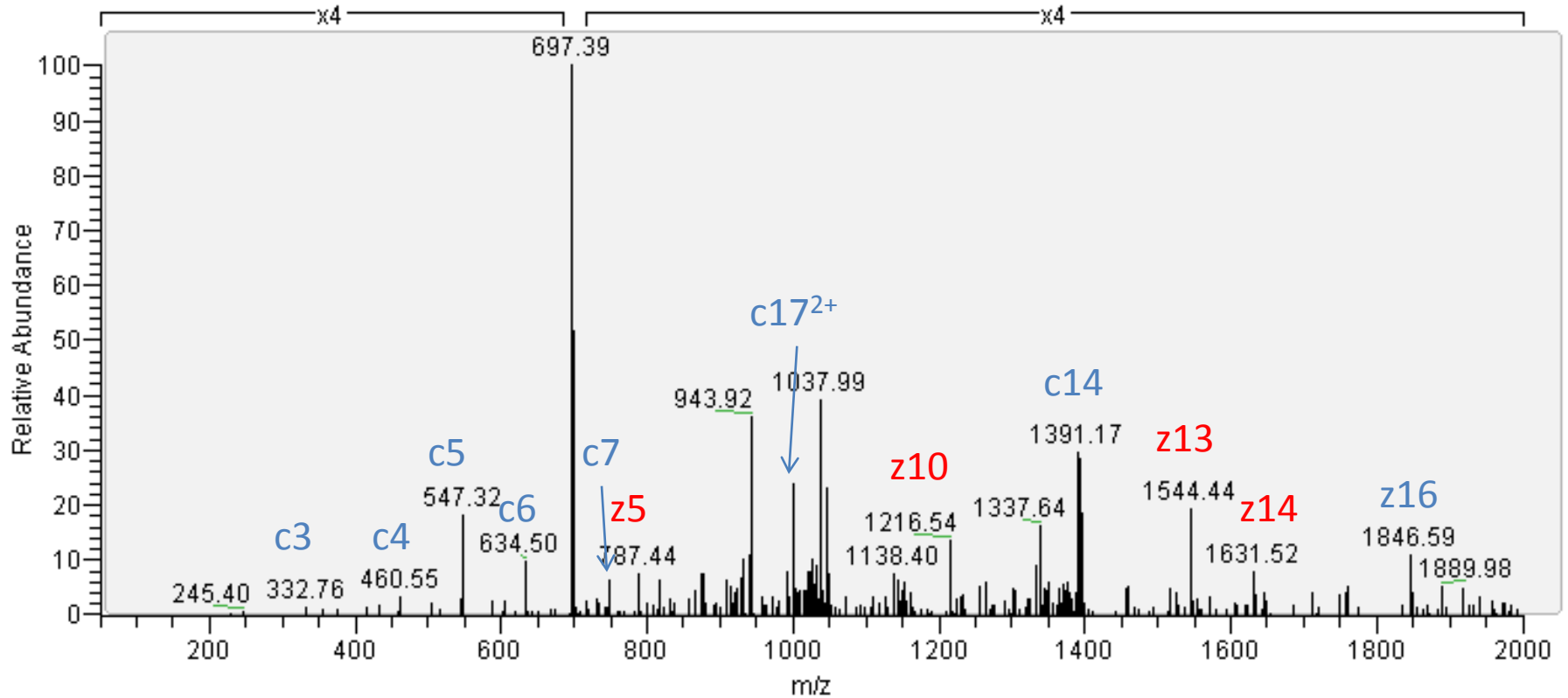
Unmodified z6 and modified z9 identify site as residue 1271



m/z 697.325 3+

RASQSSLESSTGPSYS(HexNAc)RS - Regulating synaptic membrane exocytosis protein 2

Unmodified c14 and modified c17 identify site as residue 1528

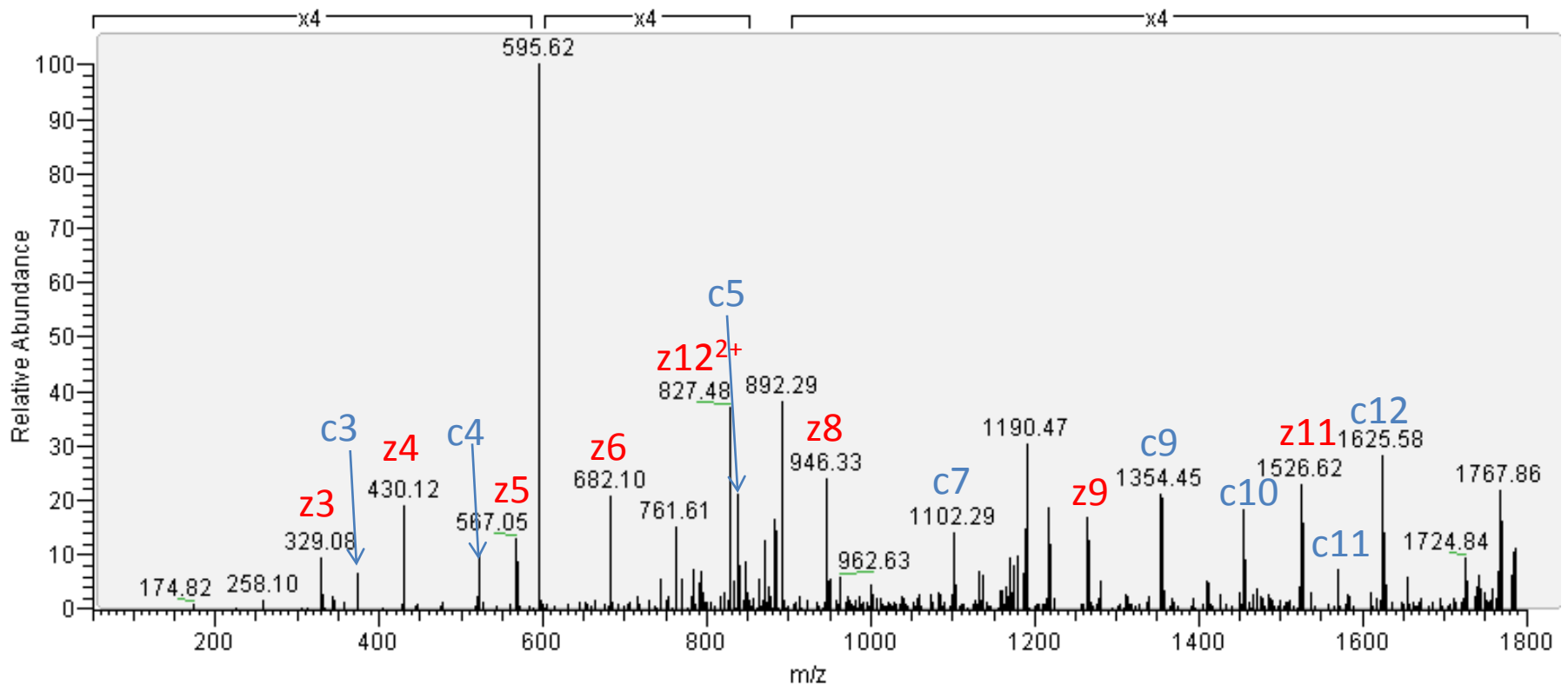


The following pages contain annotated spectra supporting the N-GlcNAc site identifications determined in this study. Peptide sequence and site assignments are displayed at the top of the page, then spectra with peak assignments are below. These assignments are based on search engine results, where no attempt was made to distinguish between  $z$  and  $z+1$  ions.

m/z 594.946 3+

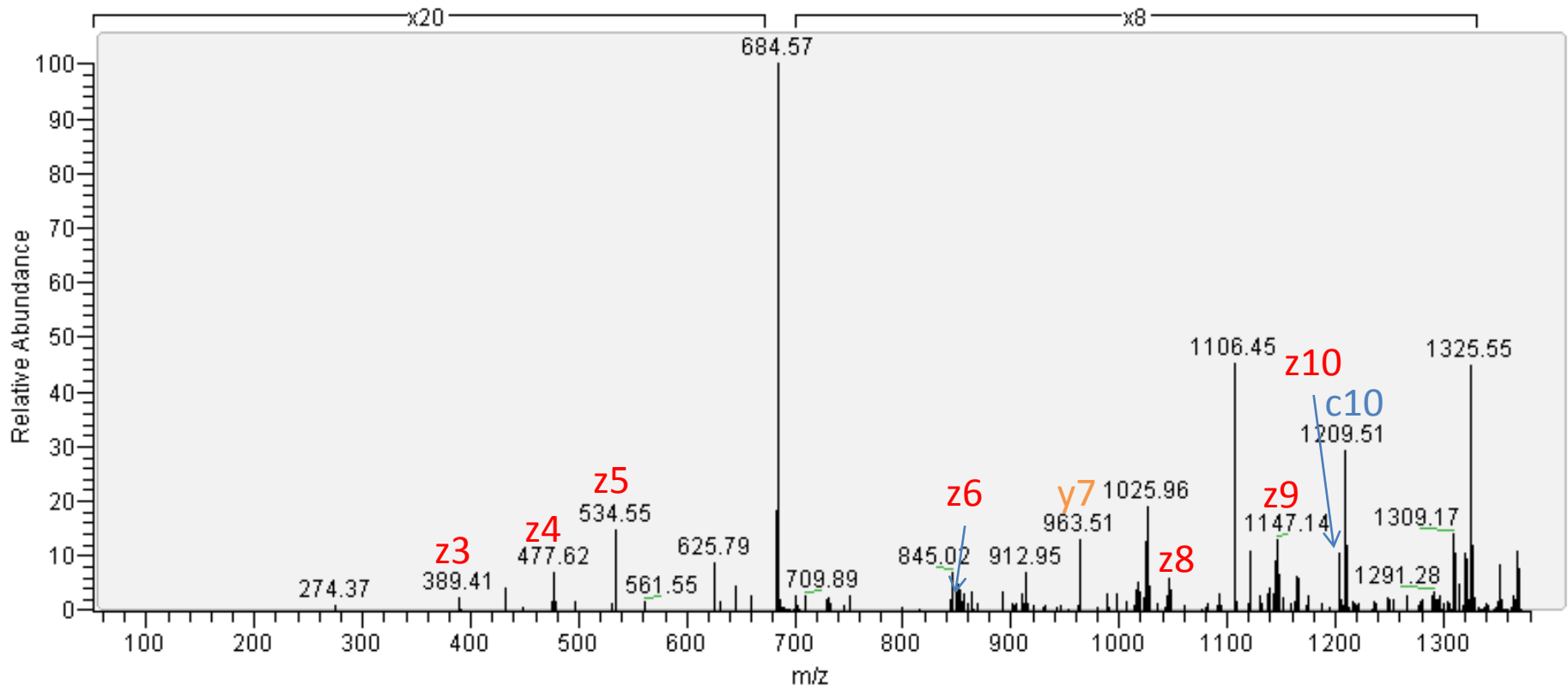
IQDFN(HexNAc)YTDHTLGR – Gamma-aminobutyric acid type B receptor subunit 2

Mass difference between c4 to c5 identifies asparagine 388 as GlcNAc modification site.



m/z 684.322 2+

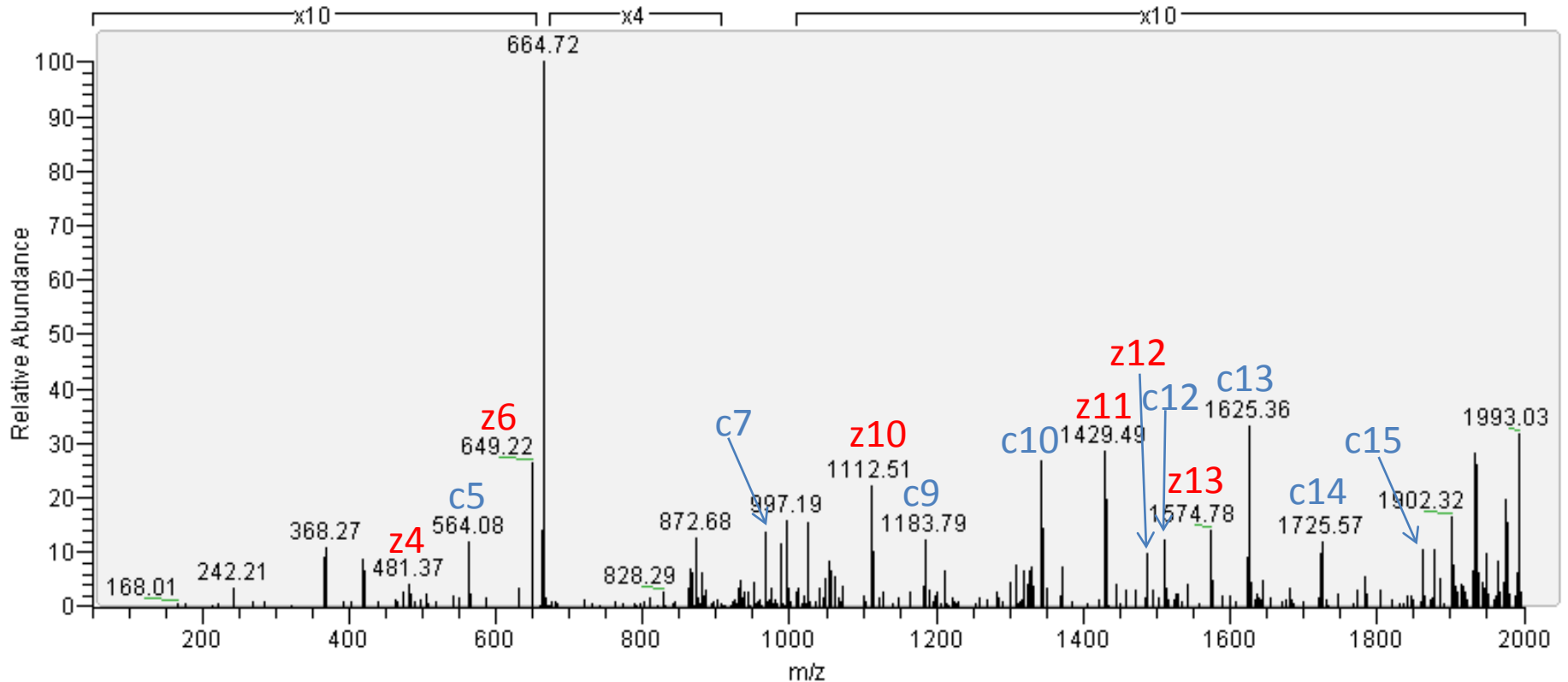
FGTVPN(HexNAc)GSTER - Glutamate [NMDA] receptor subunit epsilon-2 precursor  
Mass difference between z5 and z6 identifies asparagine 688 as GlcNAc modification site.



664.289 2+

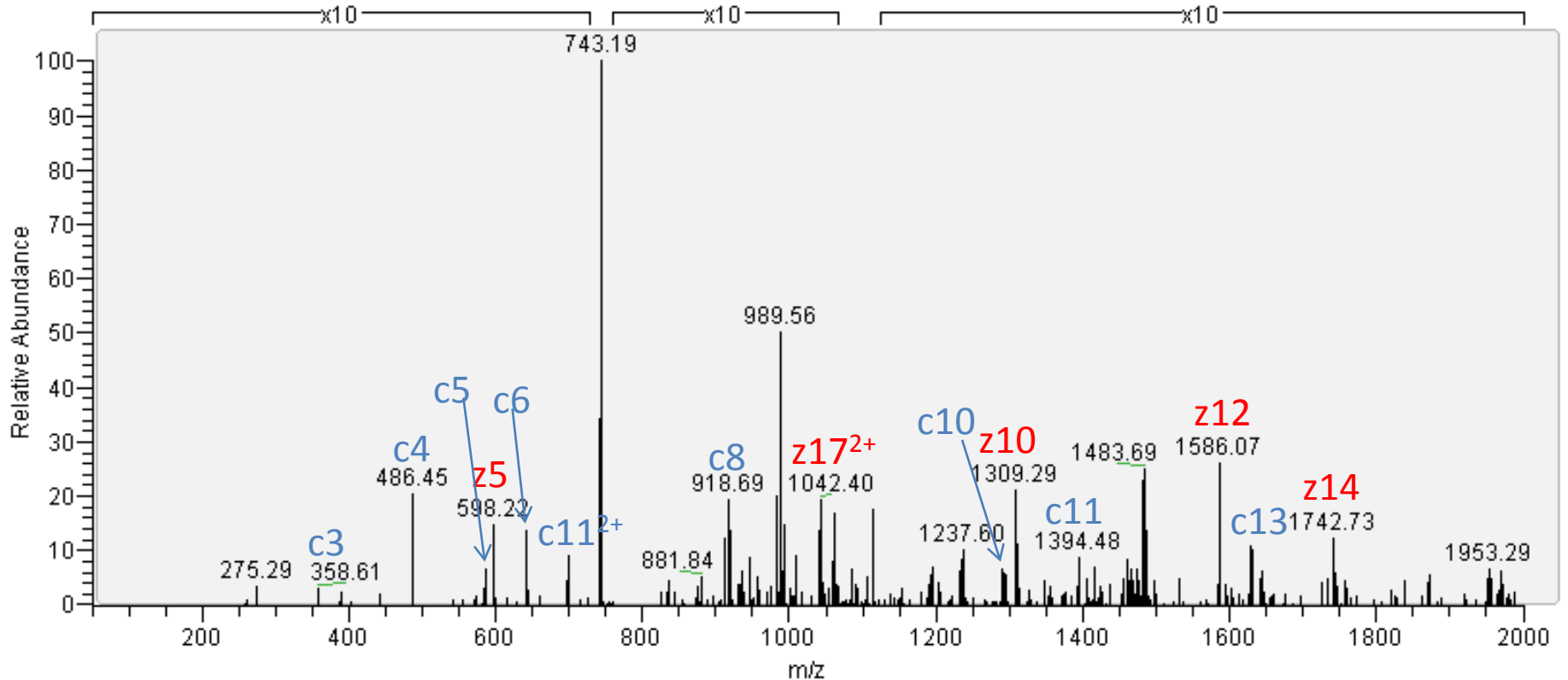
EIC(Carbamidomethyl)SGN(HexNAc)SSQC(Carbamidomethyl)APNVHK – ADAM 22

Mass difference between z10-z11 identifies asparagine 517 as GlcNAc modification site.



m/z 742.733 3+

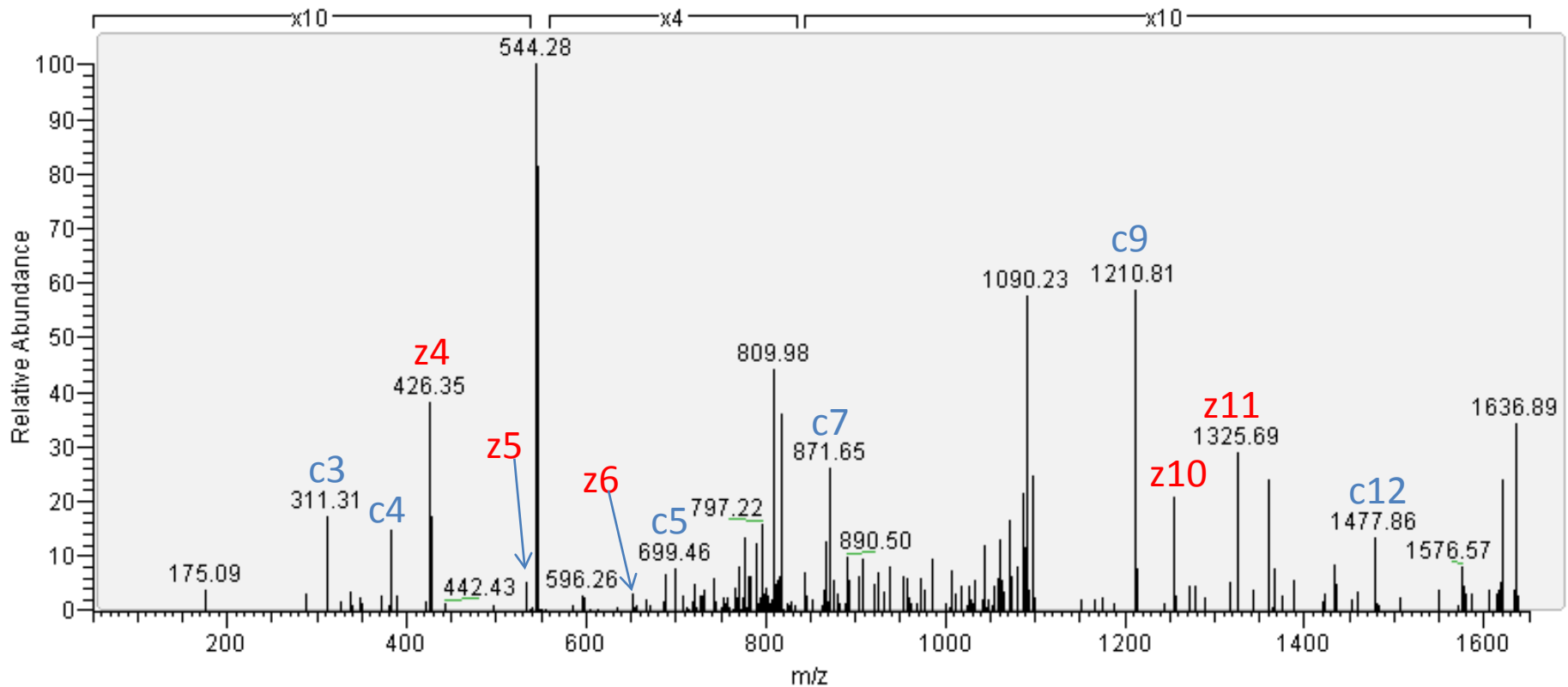
KLVQVGIYN(HexNAc)GTHVIPNDR – Glutamate [NMDA] receptor subunit zeta-1 precursor  
Unmodified c8 but modified c10 identify asparagine 368 as GlcNAc modification site.



m/z 545.649 3+

HRAN(HexNAc)ATLLGPLR - Neurocan core protein precursor

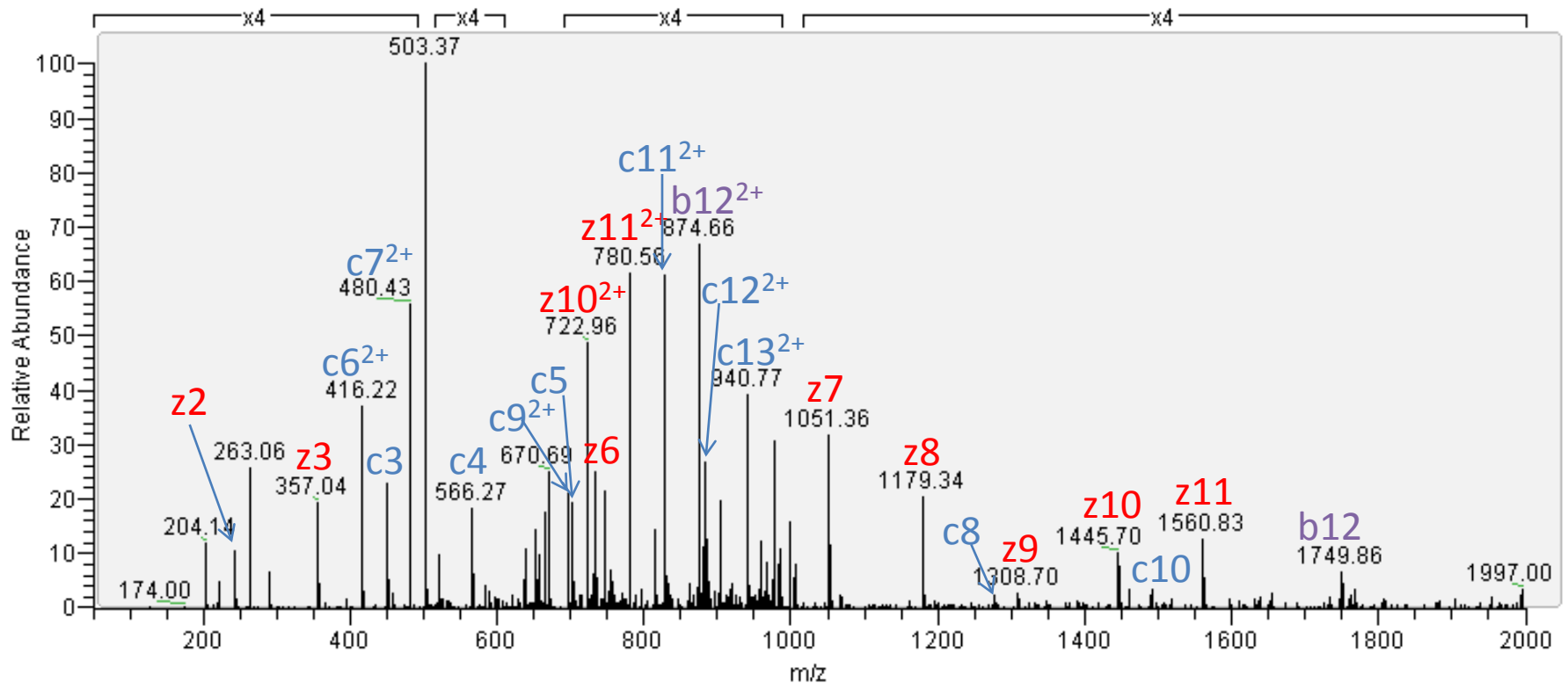
Mass difference between c4-c5 identifies asparagine 121 as GlcNAc modification site



m/z 503.000 4+

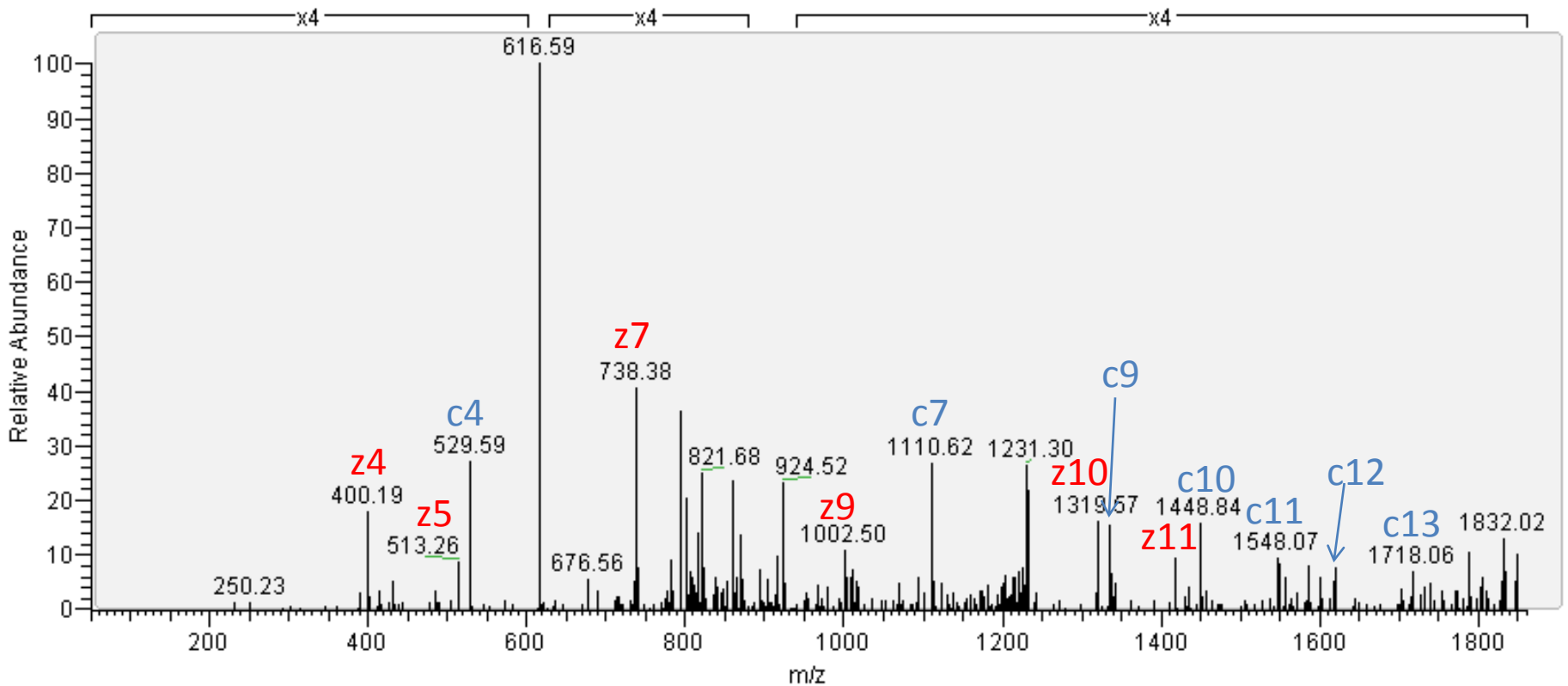
WSC(Carbamidomethyl)DHKQN(HexNAc)ITYLLK – Oligodendrocyte-myelin glycoprotein precursor

Mass difference between z6-z7 identifies asparagine 234 as GlcNAc modification site.



m/z 616.340 3+

KFHVN(HexNAc)YTQPLVAVK - Sodium/potassium-transporting ATPase subunit beta-2  
Mass difference between z9-z10 identifies asparagine 238 as GlcNAc modification site.



m/z 594.031 4+

HEN(HexNAc)NTKDNSIQHEFSLTR - Thy-1 membrane glycoprotein precursor

Unmodified c2 and z15 identify asparagine 42 as GlcNAc modification site.

