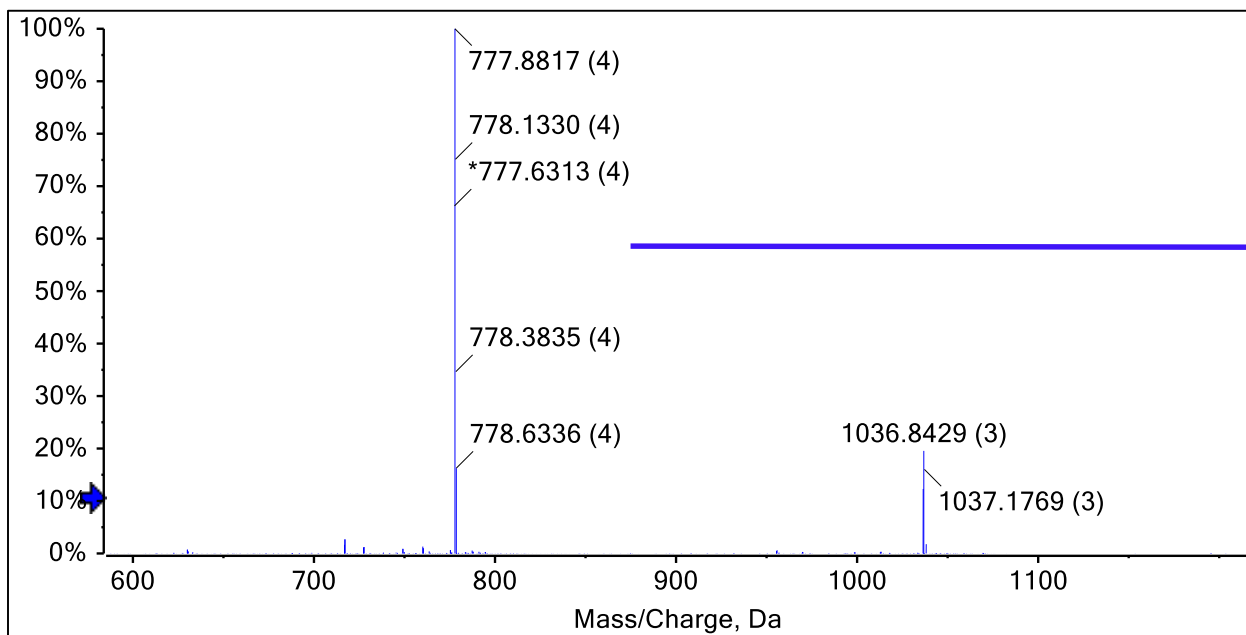
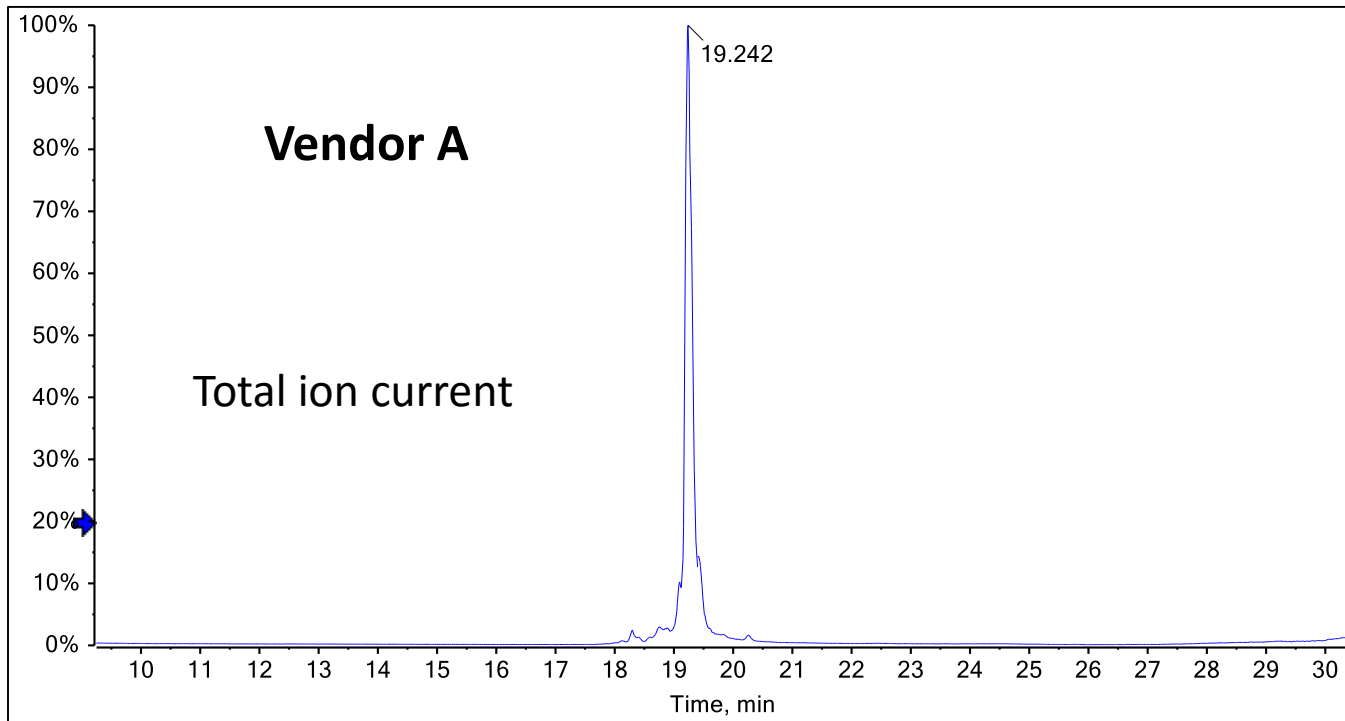
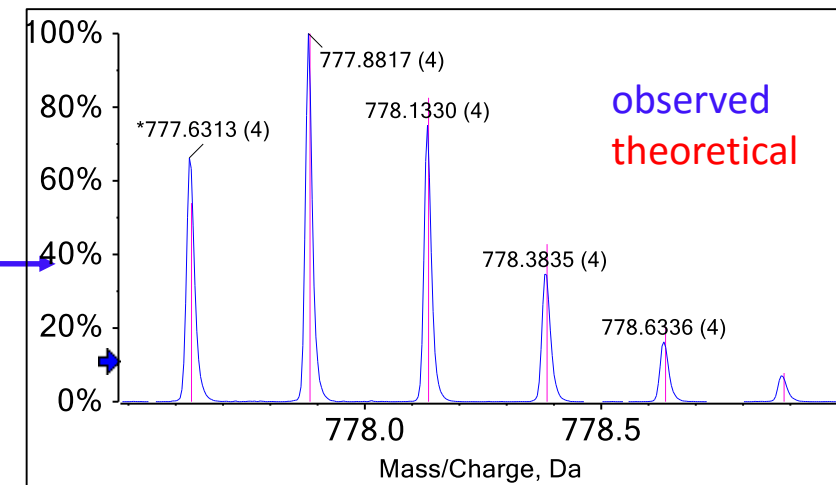


LC-MS/MS SEQUENCE ANALYSIS OF
SYNTHETIC THYMOSIN 1 α PURCHASED
FROM TWO DIFFERENT VENDORS

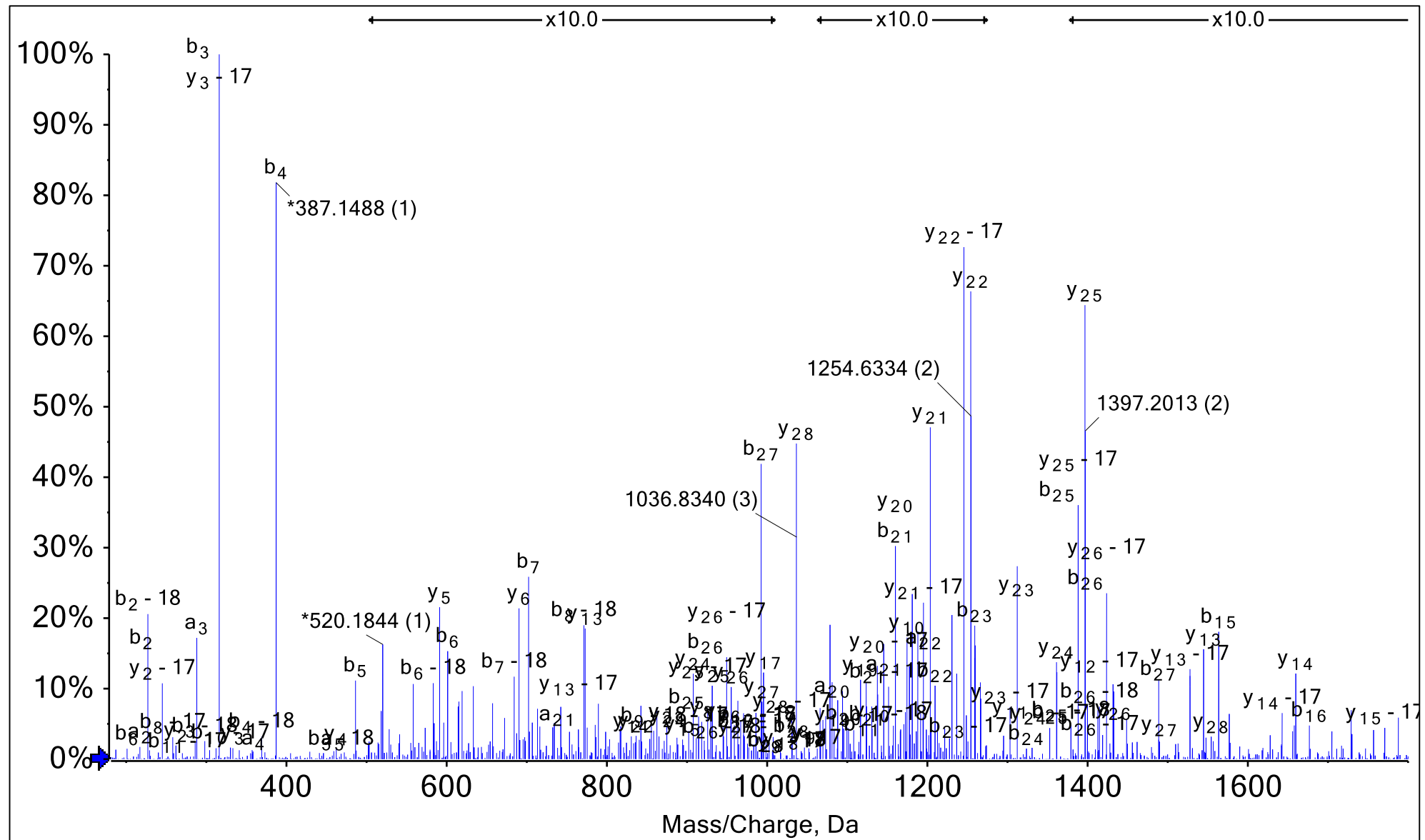


Charge state 4 isotopic profile



Calculated monoisotopic mass: **777.63331 m/z**
Measured monoisotopic mass: **777.63370 m/z**
Mass error = 0.5 ppm

MS² spectrum of 777.6330 *m/z* with fragment ions for sequence [Ac]-SDAAVDTSEITTKDLKEKKEVVEEAEN outlined (10X magnification where indicated)



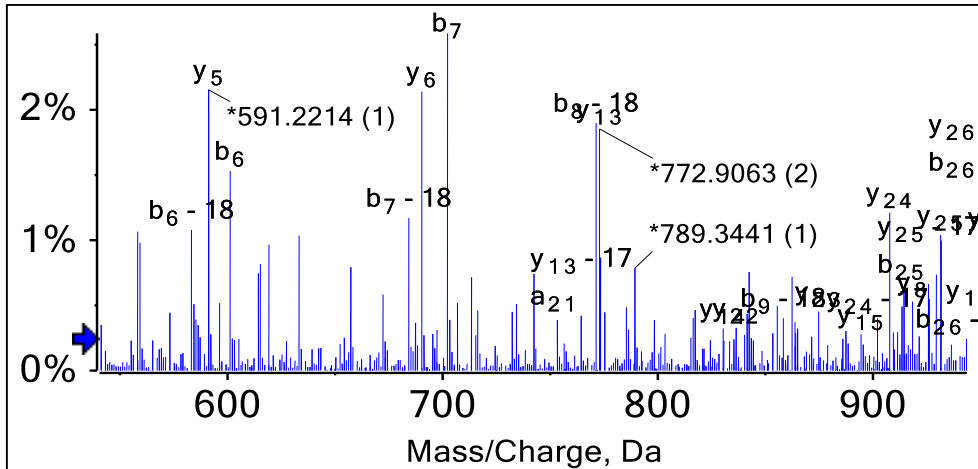
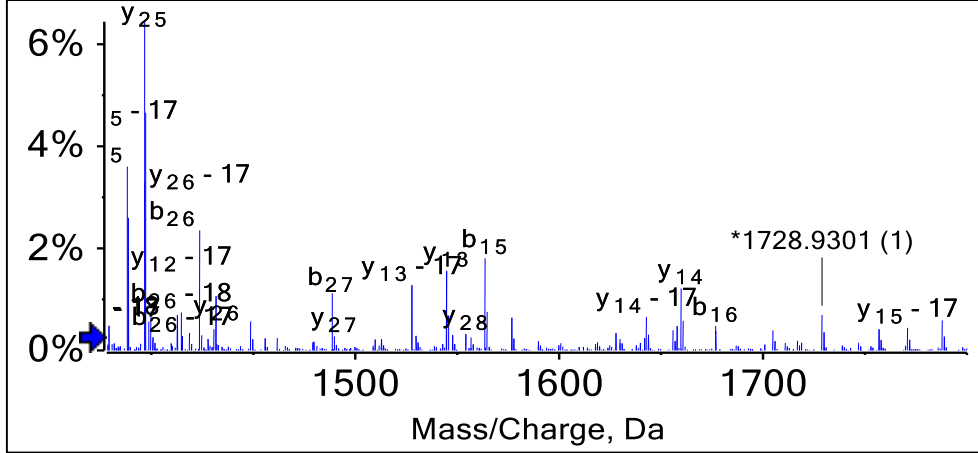
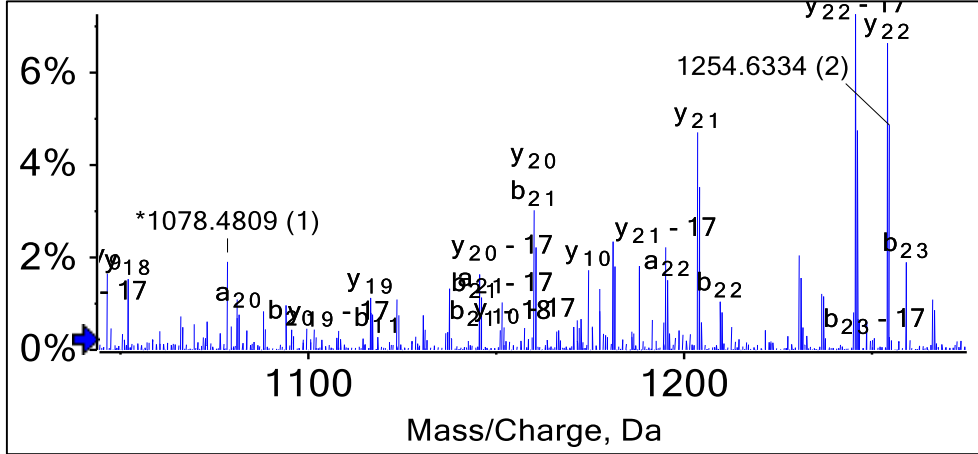
Observed fragment ions matching sequence
[Ac]-SDAAVDTSS~~ITTKDLKEKKEVVEE~~AE~~N~~
are indicated in **bold red** (max 5ppm mass accuracy)

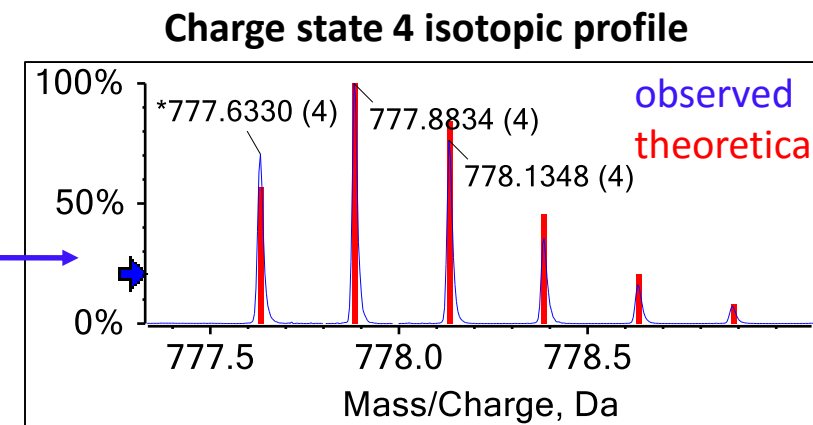
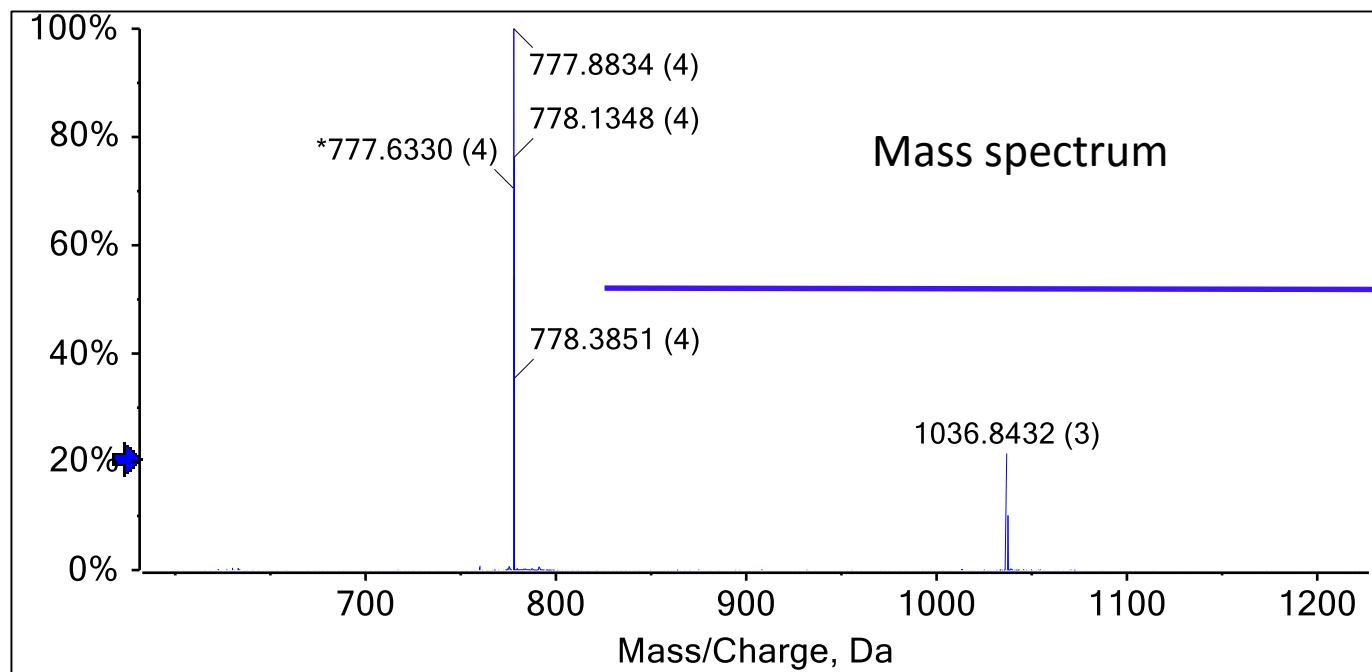
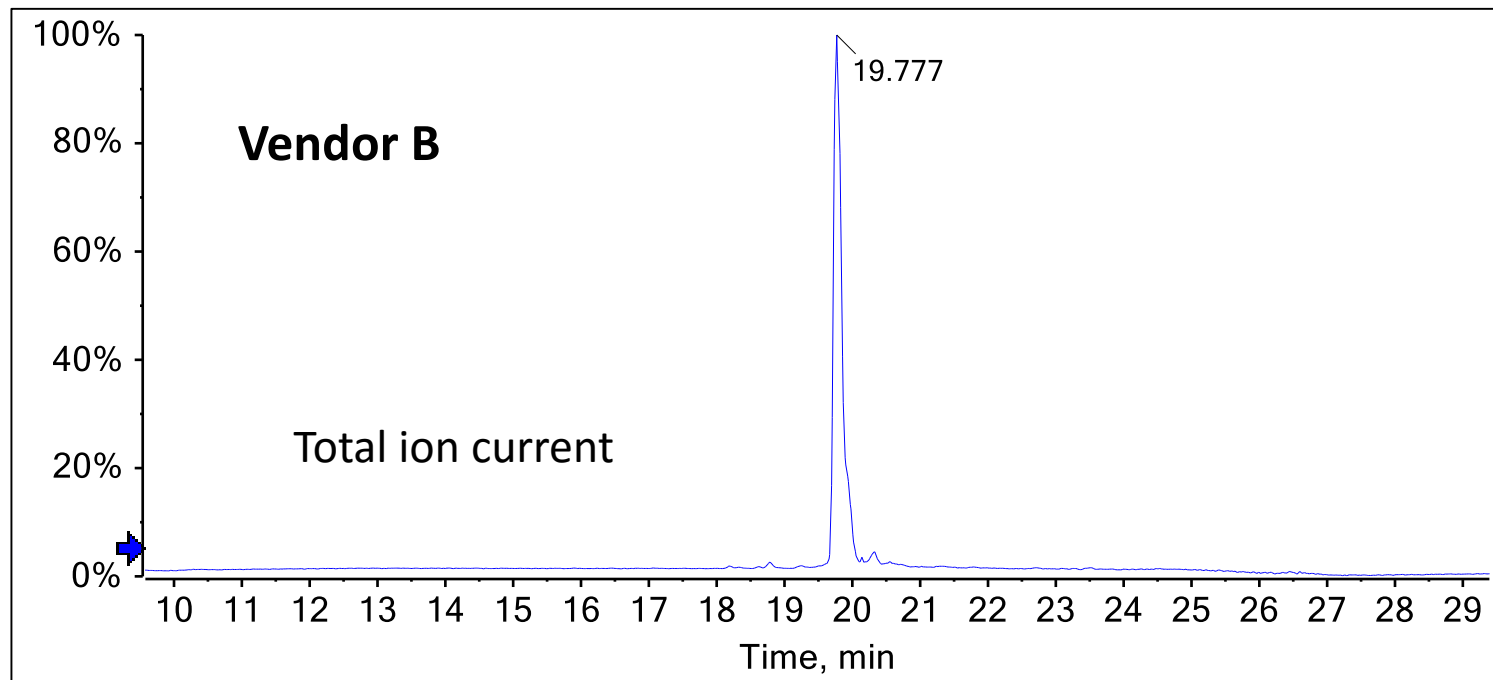
Sequence: [1Ac]-SDAAVDTSS~~ITTKDLKEKKEVVEE~~AE~~N~~

Table List

Symbol	Res. Mass	# (N)	b	y	# (C)
S	87.03203	1	65.52857	1554.25934	28
D	115.02694	2	123.04204	1489.73805	27
A	71.03711	3	158.56060	1432.22457	26
A	71.03711	4	194.07916	1396.70602	25
V	99.06841	5	243.61337	1361.18746	24
D	115.02694	6	301.12684	1311.65325	23
T	101.04768	7	351.65068	1254.13978	22
S	87.03203	8	395.16669	1203.61594	21
S	87.03203	9	438.68270	1160.09993	20
E	129.04259	10	503.20400	1116.58391	19
I	113.08406	11	559.74603	1052.06262	18
T	101.04768	12	610.26987	995.52058	17
T	101.04768	13	660.79371	944.99675	16
K	128.09496	14	724.84119	894.47291	15
D	115.02694	15	782.35467	830.42542	14
L	113.08406	16	838.89670	772.91195	13
K	128.09496	17	902.94418	716.36992	12
E	129.04259	18	967.46548	652.32244	11
K	128.09496	19	1031.51296	587.80114	10
K	128.09496	20	1095.56044	523.75366	9
E	129.04259	21	1160.08174	459.70618	8
V	99.06841	22	1209.61594	395.18488	7
V	99.06841	23	1259.15015	345.65068	6
E	129.04259	24	1323.67145	296.11647	5
E	129.04259	25	1388.19274	231.59517	4
A	71.03711	26	1423.71130	167.07388	3
E	129.04259	27	1488.23260	131.55532	2
N	114.04293	28	1545.25406	67.03402	1

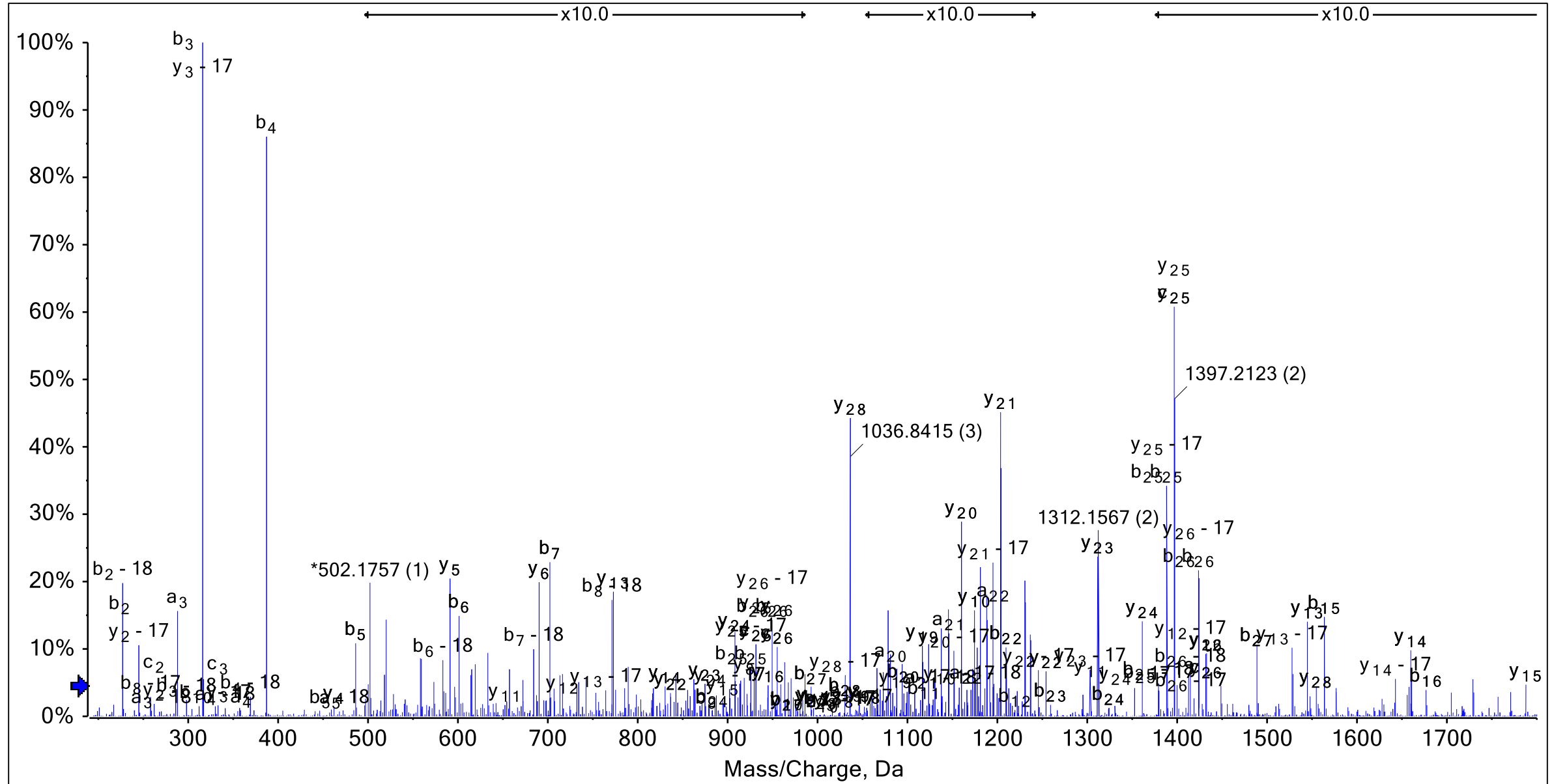
Magnified regions
of MS² spectrum
with labeled
fragment ions





Measured monoisotopic mass: 777.63303 m/z
Calculated monoisotopic mass: 777.63331 m/z
Mass error: -0.4 ppm

MS² spectrum of 777.6330 *m/z* with fragment ions for sequence [Ac]-SDAAVDTSEITTKDLKEKKEVVEEAEN outlined (10X magnification where indicated)



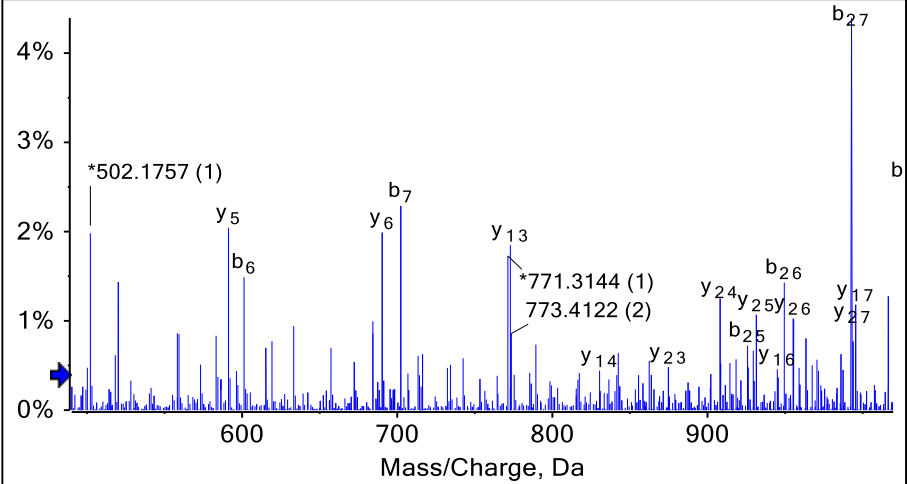
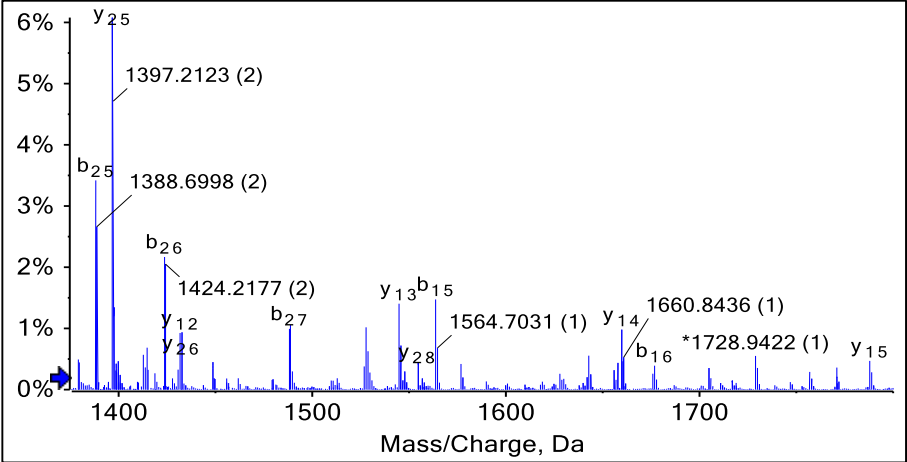
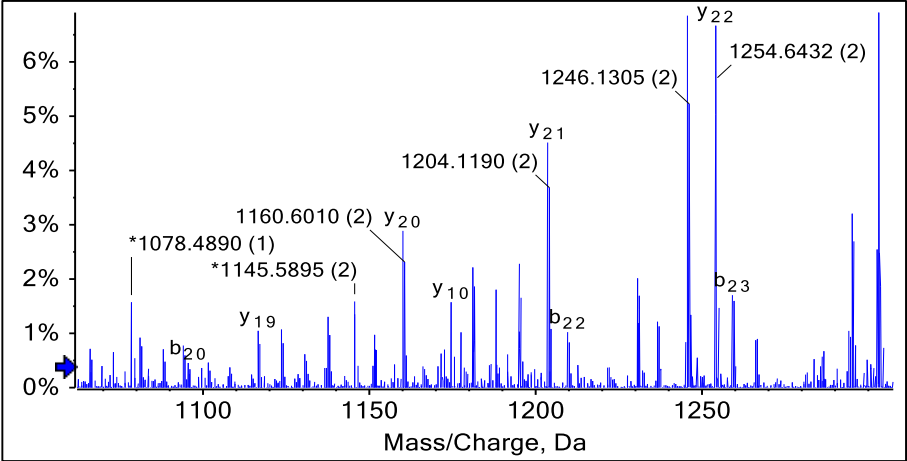
Observed fragment ions matching sequence
[Ac]-SDAAVDTSS~~ITTKDLKEKKEVVEEA~~EN
are indicated in **bold red** (max 5ppm mass accuracy)

Sequence: [1Ac]-SDAAVDTSS~~ITTKDLKEKKEVVEEA~~EN

Table List

	Symbol	Res. Mass	# (N)	b	y	# (C)
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	K	128.09496	19	1031.51296	587.80114	10
▶	K	128.09496	20	1095.56044	523.75366	9
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	E	129.04259	25	1388.19274	231.59517	4
	A	71.03711	26	1423.71130	167.07388	3
	E	129.04259	27	1488.23260	131.55532	2
	N	114.04293	28	1545.25406	67.03402	1

Magnified regions
of MS² spectrum
with labeled
fragment ions



CONCLUSIONS

- The observed peptide mass for Thymosin 1a from both Vendors matches the calculated one
- Thymosin 1 α sequence [Ac]-SDAAVDTSSEITTKDLKEKKEVVEEAEN is fully confirmed by MS/MS data for both Vendors