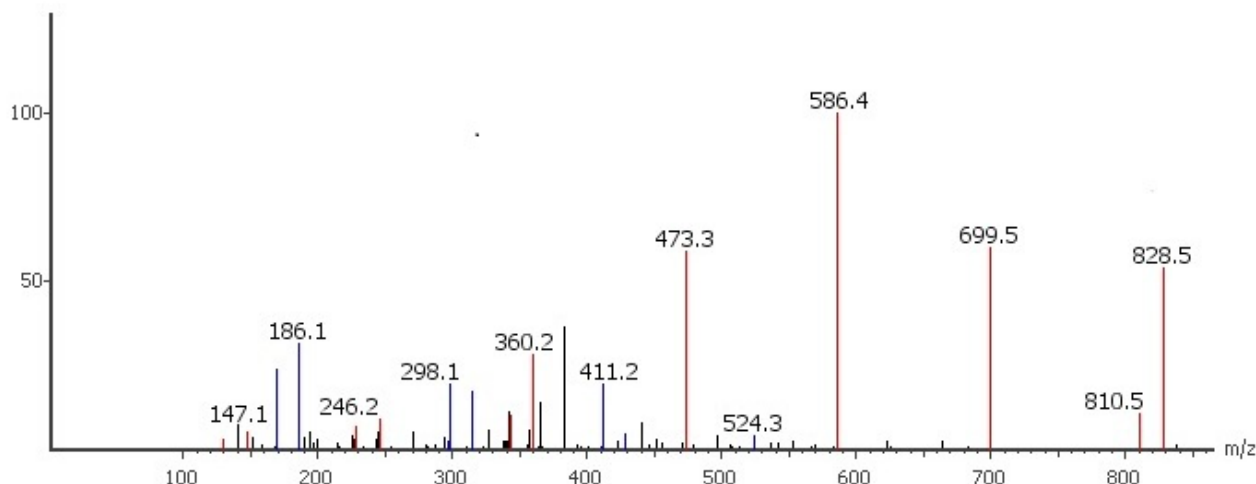
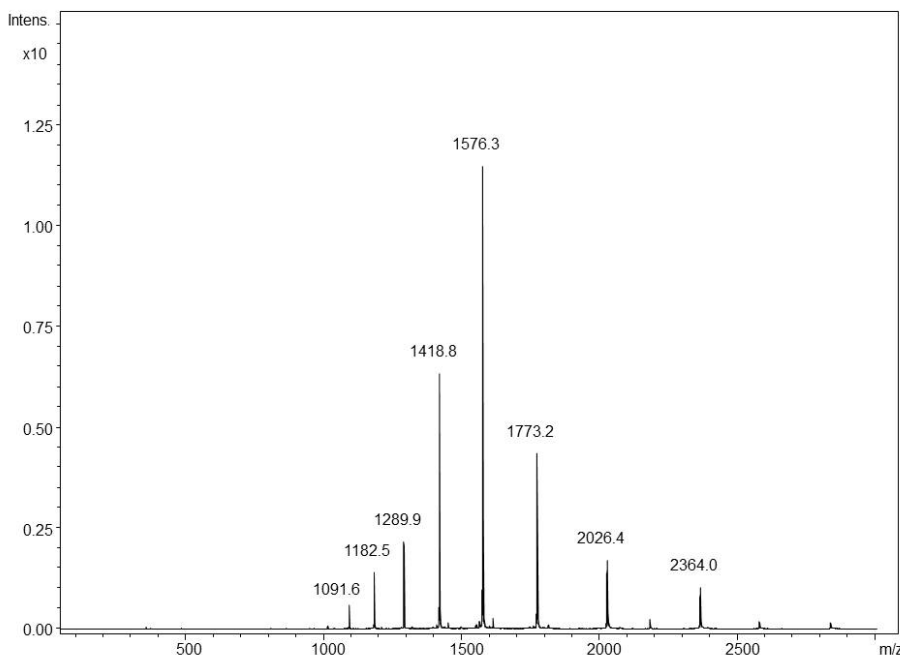


Tutorial 7 (13 Nov 2024)

1. A m/z spectrum of several peptides obtained after tandem mass spectrometry is shown below. The original peptide before fragmentation contained NINE amino acids comprising of sequence ANELLLNVK. How many b and y ions can you identify? Occasionally some y ion may appear with mass of water removed while a b ion may appear with mass of ammonia removed.



2. The m/z spectrum below represents the data obtained by LC-MS of Alpha Bovine Lactalbumin protein (BLA). The ions were created using ESI by applying a positive potential. Calculate the average mass of BLA using the data.



3. Calculate the Time of Flight for a protein of mass 10 kDa travelling a distance 0.1 m in TOF mass spectrometer under an accelerating potential difference of 100 V.
[$z = 1$; $e = 1.6 \times 10^{-19} \text{ C}$]

4. Name two amino acids which cannot be unequivocally identified from each other when trying to sequence using TANDEM MASS SPECTROMETRY.
5. The m/z spectrum of a sample from a MALDI-TOF spectrometer often contains several peaks of mass <500 Da. Explain the origin of these peaks.
6. A peptide was subjected to degradation from the N terminus (Edman degradation). The resultant mixture was subjected to MALDI/MS analysis. The following ladder of m/z was observed: 977.3, 1064.3, 1151.4, 1222.6, 1378.9 and 1465.9. Assume that z = 1 in all cases and derive the sequence of the N-terminal region of the peptide. The identical peptide was enzymatically phosphorylated with protein kinase C. For this peptide the ladder was as follows: 977.3, 1064.3, 1231.6, 1302.6, 1458.9 and 1545.9. Explain the results.
7. The peptide SPSWEPFR displayed two peaks in MS spectrum with masses 1005.77 and 1085.76 Da. Explain the origin of two peaks. How can you confirm if your explanation is right?
8. PEPTIDE MASS FINGERPRINTING produced the following peptides with mass values given below:

748.404	920.415	1271.61	1378.78
1484.97	1502.62	1562.8	1606.82
1815.83	1853.89	1884.95	1981.97

Database search revealed the following protein (UniProtKB - P68083 (MYG_EQUBU): Myoglobin from Plains ZEBRA) as a LIKELY IDENTIFICATION. Try and match at least 5 peptides from the protein below to the masses above and identify the peptides in the protein to corresponding masses.

MGLSDGEWQQ	VLNVWGKVEA	DIAGHGQEV	IRLFTGHPET
LEKFDKFKHL	KTEAEMKASE	DLKKHGTVVL	TALGGILKKK
GHHEAELKPL	AQSHATKHKI	PIKYLEFISD	AIHVLHLSKH
PGDFGADAQG	AMTK ALELFR	NDIAAKYKEL	GFQG

For example: ALELFR corresponds to a mass of 748.404 Da.

This table shows the Monoisotopic and Average mass in Daltons of each one of the amino acids.

1-letter code	3-letter code	Chemical formula	Monoisotopic	Average
A	Ala	C ₃ H ₅ ON	71.03711	71.0788
R	Arg	C ₆ H ₁₂ ON ₄	156.10111	156.1875
N	Asn	C ₄ H ₆ O ₂ N ₂	114.04293	114.1038
D	Asp	C ₄ H ₅ O ₃ N	115.02694	115.0886
C	Cys	C ₃ H ₅ ONS	103.00919	103.1388
E	Glu	C ₅ H ₇ O ₃ N	129.04259	129.1155
Q	Gln	C ₅ H ₈ O ₂ N ₂	128.05858	128.1307
G	Gly	C ₂ H ₃ ON	57.02146	57.0519
H	His	C ₆ H ₇ ON ₃	137.05891	137.1411
I	Ile	C ₆ H ₁₁ ON	113.08406	113.1594
L	Leu	C ₆ H ₁₁ ON	113.08406	113.1594
K	Lys	C ₆ H ₁₂ ON ₂	128.09496	128.1741
M	Met	C ₅ H ₉ ONS	131.04049	131.1926

F	Phe	C ₉ H ₉ ON	147.06841	147.1766
P	Pro	C ₅ H ₇ ON	97.05276	97.1167
S	Ser	C ₃ H ₅ O ₂ N	87.03203	87.0782
T	Thr	C ₄ H ₇ O ₂ N	101.04768	101.1051
W	Trp	C ₁₁ H ₁₀ ON ₂	186.07931	186.2132
Y	Tyr	C ₉ H ₉ O ₂ N	163.06333	163.1760
V	Val	C ₅ H ₉ ON	99.06841	99.1326