



7th Assignment - Structure and Systems Bioinformatics

Hand in: 2023-06-29 20:00 CEST (source code and pdf in a single archive, uploaded via ILIAS)

Task 1 – Proteomics Basics (100 P)

1. In order to perform shotgun (bottom-up) proteomics, proteins are digested into shorter peptides before a mass spectrometric measurement is performed. Trypsin is the most commonly used protease. Give two additional examples for proteases used in proteomics and indicate their cleavage specificity, including references (5 P).
2. Write a Python program that performs *in silico* digestion with trypsin for any given amino acid sequence.
 - Look up and briefly summarize the digestive specificity of trypsin, including references (3 P).
 - The only input of your program should be a **fasta file containing a single sequence entry** (2 P).
 - With your program calculate the **ideal** tryptic digestion of *Alcohol dehydrogenase 1A* (Uniprot: P07327, sequence at <http://www.uniprot.org/uniprot/P07327.fasta>). Therefore, calculate the peptides, their **monoisotopic** masses and **average** masses (with respect to the isotope occurrences) (17 P).
 - List the sequences, **monoisotopic** masses and **average** masses of the resulting peptides in **standard console output** (5 P).
 - In your report, include a table containing the sequences and both **monoisotopic** and **average** masses of the **6 longest** tryptic peptides of this protein (3 P).
3. Assume you performed an MS experiment with no fragmentation using a TOF instrument.

The complete task refers to the singly positively charged peptide HFEEDMGRK.

- At what mass-to-charge ratio would you expect to find the mono-isotopic peak of the singly positively charged peptide HFEEDMGRK? Provide your calculation/thoughts (5 P).
- Calculate and plot the three most intense peaks on the m/z axis (take care of the correct expected distance of the peaks and the correct intensity proportions, a sketch is not sufficient) (15 P).
- The kinetic energy of the peptide ion at the entrance of the TOF is estimated to be 3,000 eV. The length of the drift tube is 1.2 m (no reflectron). Calculate the time of flight of this peptide ion (10 P).

4. In order to get richer information on the peptide you follow up with a CID experiment.
 - Implement a program that calculates the masses of the HFEEDMGRK peptide's *b* and *y* fragment ions. Your program should report the mass-to-charge ratio of all fragment ions' monoisotopic, singly charged versions (MH+) (**25 P**).
 - Please provide the mass-to-charge ratio of all fragment ions' monoisotopic, singly charged versions (MH+) also in a table in your pdf (**10 P**).

Questions can be directed to ssbi-ss23@informatik.uni-tuebingen.de or the ILIAS course forum. We highly encourage you to use ILIAS for communication.