

Questions – Chapter 02

1- In what workflow(s) are peptides analyzed?

Top-down Shotgun Bottom-up Native MS

2- What is the most common enzyme used for protein digestion in MS-based proteomics?

Lys-C Trypsin Chymotrypsin Pepsin

3- Explain the principle of peptide mass fingerprinting (PMF)?

4- What is the difference between PMF and MS/MS ion search?

5- For what does DDA stand?

Data-derived acquisition Data-dependent acquisition Data-independent acquisition None of these

6- In DDA, typically how many tandem mass spectra are recorded after an MS survey scan?

One Ten One hundred It depends

7- In shotgun proteomics, how proteins are finally identify?

By comparing experimental tandem mass spectra with theoretical *in silico*-generated tandem mass spectra By using DNA sequence databases By using mass information only By *de novo* annotating every tandem mass spectrum

8- Why top-down is relevant?

Every proteomic lab is using top-down workflows Proteoforms can be characterized Protein sequence coverage is comprehensive Entire proteins are easier to separate

9- In what application(s) is top-down particularly employed?

Characterization of biosimilars Study of post-translational modifications Study of sequence variants Protein identification

10- For what step(s) of sample preparation are DTT or TCEP used?

- Protein digestion
- Cell lysis
- Disulfide bridge reduction
- Sample fractionation

11- What reagent(s) can be used for cell lysis?

- Several detergents
- Iodoacetamide
- Sodium dodecyl sulfate
- Acetone

12- What method(s) is(are) used for protein/peptide enrichment?

- Antibody-based capture
- Depletion of abundant proteins
- Lysis
- Alkylation

13- Where does trypsin hydrolyze the peptide bonds?

- The carboxyl terminal side of arginine and glycine amino acid residues
- The carboxyl terminal side of arginine and lysine amino acid residues
- The carboxyl terminal side of arginine and tryptophan amino acid residues
- The carboxyl terminal side of cysteine and methionine amino acid residues

14- Why using different enzymes for protein digestion?

- Increase protein coverage
- Generate proteotypic peptides
- Trypsin is unspecific and additional enzymes are needed
- Digestion efficiency is not enough

15- You have received a gel piece in a tube. You want to identify the protein(s) present in the sample. What workflow(s) would you recommend?

- A top-down approach
- In-gel digestion
- A enrichment of the protein of interest
- Use of chymotrypsin for protein digestion

16- You have received human blood plasma to analyze. You want to identify the maximal number of protein present in the sample. What workflow(s) would you recommend?

- A top-down approach
- A shotgun proteomic workflow
- Not using mass spectrometry-based workflow
- Depletion of abundant proteins

17- You have received a purified protein to analyze. You want to characterize it. What workflow(s) would you recommend?

- A top-down approach
- A bottom-up approach
- Use of different enzymes for protein digestion
- An initial separation with 1D-gel electrophoresis