

Questions – Chapter 03

1- Why is MS-based proteomics not inherently quantitative?

- There are losses of peptides during analysis and differences in the ionization efficiency of peptides
- The intensity of a peak in a mass spectrum is not a good indicator of the amount of the analyte in the sample, although differences in peak intensity of the same analyte between multiple samples accurately reflect relative differences in its abundance
- Detection efficiencies for ions with different m/z values are unequal
- The relationship between the amount of analyte present and measured signal intensity is complex and incompletely understood

2- What technique(s) can provide comparison of each individual peptide between experiments/samples?

- Label-free techniques
- SILAC
- Isobaric labelling
- MS gives directly the concentrations of analytes, which can be compared between the samples

3- What does the spectral counting approach compare in different analyzed samples?

- The number of all spectra associated with a specific protein
- The sum of all precursor intensities of peptides associated with a specific protein
- The precursor intensities of the 3 most intense detected peptides
- The count of observed peptides *versus* all possible peptides

4- What are some advantages of the label-based techniques?

- As the labeling occurs during sample preparation, quantitative artifacts are minimized
- The techniques are usually very cheap
- Multiplexing of samples is possible
- They can be performed at protein or peptide level

5- What does SILAC stand for?

- Selective *In-vivo* Labeling After Chemical reaction
- Nothing in particular
- Static Isoforms Labels for Affinity Capture
- Stable Isotope Labeling with Amino Acids in Cell Culture

6- At what MS level does SILAC quantification occur?

- MS¹ MS² MS³ MSⁿ

7- At what MS level does TMT or ITRAQ quantification occur?

- MS¹ MS² MS³ MSⁿ

8- How is it possible to compare the liver proteome of two mice?

- Using *in-vivo* SILAC Using SILAC Using isobaric labeling Using a label-free approach

9- In general, how many generations of animals are needed at least to completely label all organs in a SILAC mouse?

- 1 2 10 5

10- What amino acids are generally available to perform a SILAC experiment?

- Lysine and arginine Glycine and Leucine Lysine and proline Leucine and isoleucine

11- What is called a super-SILAC mix?

- A mixture of samples labelled with TMT A mixture of cell lines labelled by SILAC A mixture of non-labeled healthy tissues

12- What multiplexing capabilities are available with TMT?

- 2-plex 4-plex 6-plex 10-plex

13- What amino acid(s) is/are labeled with TMT or iTRAQ?

- Lysine Arginine N-terminus Cysteine

14- At what level is TMT or iTRAQ labeling generally performed?

- Amino acid level Peptide level Protein level Cell culture level

15- In order to decipher subtle changes in phosphorylation regulation in two cell lines, what quantitative approach(es) would you recommend?

- SILAC *In-vivo* SILAC Isobaric labeling Spectral counting

16- What mass spectrometer is commonly used for targeted protein quantification with stable isotope dilution?

- FT-ICR
- QqQ
- Ion trap
- MADLI-TOF

17- How are peptides selected for their use as heavy AQUA standards?

- They need to be proteotypic
- They need to contain more than 25 amino acids
- They need to be heavily modified post-translationally
- They need to fragment efficiently

18- What does PSAQ stand for?

- Protein Standard Absolute Quantification
- Protein for Stable Accurate Quantification
- Peptide Standard for Absolute Quantification
- None of these

19- How is abbreviated multiple selected reaction monitoring?

- SRM
- MRM
- mSRM
- MSM

20- What type of mass spectrometer is used for parallel reaction monitoring?

- QqQ
- FT-ICR
- Orbitrap
- Ion trap

21- What does PRM required for its development?

- Selection of transitions
- Optimization of collision energies
- Selection of peptides to be used as heavy standards
- An LC system