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NPTEL

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Courses » Mass spectrometry based proteomics

Announcements Course Ask a Question Progress

# Unit 2 - Week 1: Proteomics introduction and sample preparation



## Course outline

### How to access the portal

### Week 1: Proteomics introduction and sample preparation

- Introductory lecture
- Lecture 1: Introduction to proteomics
- Lecture 2: Proteomics and sample preparation
- Lecture 3: Bacterial protein extraction
- Lecture 4: In-gel digestion
- Lecture 5: Fundamentals of mass spectrometry
- Lab Session- 1.1: Protein/peptide pre-fractionation using OFFGEL FRACTIONATOR
- Lab session 1.2: Demonstration of Q-TOF MS technology
- Assignment-I Answer key
- Quiz : Assignment-1

## Assignment-1

The due date for submitting this assignment has passed. **Due on 2016-03-29, 05:15 IST.** As per our records you have not submitted this assignment.

1) Find the best suitable technique and the application it is best suited for 0.5 points

- |                      |                               |
|----------------------|-------------------------------|
| a) Mass spectrometry | i) Protein sequencing         |
| b) Realtime PCR      | ii) DNA sequencing            |
| c) Edman degradation | iii) Gene Expression Analysis |
| d) ELISA             | iv) Protein identification    |
| e) Sanger's method   | v) Quantitation of an analyte |

- a-i; b-iii; c-ii; d-v; e-iv
- a-iv; b-ii; c-v; d-i; e-iii
- a-iv; b-ii; c-i; d-v; e-iii
- a-iv; b-iii; c-i; d-v; e-ii

**No, the answer is incorrect.**  
**Score: 0**

**Accepted Answers:**  
*a-iv; b-iii; c-i; d-v; e-ii*

2) The central dogma of life which states that the information coded by the genome is transcribed and translated is best described by which of the following 0.5 points

- DNA to RNA to DNA
- RNA to DNA to Protein
- Protein to RNA to DNA
- DNA to RNA to Protein

**No, the answer is incorrect.**  
**Score: 0**

**Accepted Answers:**  
*DNA to RNA to Protein*

3) Cell or tissue lysis is an essential technique which enables extraction of several cellular components including protein; which of the following lysis methods is gentle lysis method? 0.5 points

- Sonication
- Freeze-thaw
- Bead Beater homogenization
- French-press

**No, the answer is incorrect.**  
**Score: 0**

**Accepted Answers:**

Week 2: Basics of mass spectrometry

Week 3: Quantitative proteomics

Week-4: Proteomics and systems biology

### Freeze-thaw

- 4) Pair the chemical reagents with its respective role
- |                   |                            |
|-------------------|----------------------------|
| a) Urea           | i) Reducing agent          |
| b) Iodoacetamide  | ii) Cysteine blocking      |
| c) Dithiothreitol | iii) Protein precipitation |
| d) Acetone        | iv) DNA precipitation      |
| e) Ethanol        | v) Protein denaturation    |
| f) Isopropanol    | vi) RNA precipitation      |

0.5 points

- a-iv; b-i; c-ii; d-v; e-iii; f-vi  
 a-v; b-ii; c-i; d-iii; e-iv; f-vi  
 a-v; b-ii; c-iv; d-i; e-iii; f-vi  
 a-iv; b-i; c-iii; d-ii; e-v; f-vi

**No, the answer is incorrect.**

**Score: 0**

**Accepted Answers:**

*a-v; b-ii; c-i; d-iii; e-iv; f-vi*

- 5) Protein mixture having five different proteins namely P, Q, R, S and T. If they are separated using SDS-PAGE, find the separation of proteins from high to low mobility

0.5 points

Protein Name	Mol. wt.
P	25 kDa
Q	17 kDa
R	36 kDa
S	64 kDa
T	45 kDa

- T, Q, S, R, P  
 Q, S, R, T, P  
 S, T, R, P, Q  
 Q, P, R, T, S

**No, the answer is incorrect.**

**Score: 0**

**Accepted Answers:**

*Q, P, R, T, S*

- 6) What is the advantage of TRIzol based protein extraction protocol?

0.5 points

- Add salts to protein essential for ionization  
 TRIzol digest the protein used for mass spectrometry analysis  
 Protein extraction is free from nucleic acid contamination  
 All of the above

**No, the answer is incorrect.**

**Score: 0**

**Accepted Answers:**

*Protein extraction is free from nucleic acid contamination*

- 7) In the process of in-gel digestion you forgot to add cysteine blocking agent to the protein sample. The data analysis of MALDI-TOF/TOF data has showed poor results. What might be the reason? **0.5 points**

- The protein sample might not be denatured  
 The protein sample might not be having disulphide bonds, which resulted in improper protein digestion followed by its identification in MS/MS analysis with low protein score  
 The protein sample might be having disulphide bonds, which resulted in improper protein digestion followed by its identification in MS/MS analysis with low protein score  
 All of the above



**No, the answer is incorrect.**

**Score: 0**

**Accepted Answers:**

*The protein sample might be having disulphide bonds, which resulted in improper protein digestion followed by its identification in MS/MS analysis with low protein score*

8) In-the process of in-gel digestion, which of the following reagent act as dehydrating agent **0.5 points** to remove the CBB stain?

- 50 mM Iodoacetamide solution
- Ammonium bicarbonate solution
- 10 mM DTT solution
- Acetonitrile solution

**No, the answer is incorrect.**

**Score: 0**

**Accepted Answers:**

*Acetonitrile solution*

9) Match the following

- |                            |                                |
|----------------------------|--------------------------------|
| a) Electron ionization     | i) Solid phase                 |
| b) Fast atom bombardment   | ii) Liquid phase               |
| c) Electrospray ionization | iii) Gas phase                 |
| d) MALDI                   | iv) Singly charged ion species |

- a-iii; b-i; c-ii; d-iv
- a-iv; b-ii; c-i; d-iii
- a-iii; b-ii; c-i; d-iv
- a-iv; b-i; c-ii; d-iii

**No, the answer is incorrect.**

**Score: 0**

**Accepted Answers:**

*a-iii; b-i; c-ii; d-iv*

10) A protein mixture contains 5 different proteins namely P, Q, R, S and T. They were separated using gel filtration chromatography. Find the order of mobility (High to low) of these proteins in gel filtration chromatography. **0.5 points**

Protein Name	Mol. wt.
P	13 kDa
Q	17 kDa
R	86 kDa
S	44 kDa
T	10 kDa

- R, S, Q, P, T
- T, P, Q, S, R
- P, T, S, R, Q
- Q, R, S, T, P

**No, the answer is incorrect.**

**Score: 0**

**Accepted Answers:**

*R, S, Q, P, T*

11) You have been given a solution having glutamic acid, aspartic acid and lysine amino acids. If you want to purify lysine amino acid using strong cation exchange chromatography. What is the pH of the buffer that you will use for the experiment? **0.5 points**

- pH 3
- pH 7
- pH 8
- pH 10



**No, the answer is incorrect.**

**Score: 0**

**Accepted Answers:**

*pH 7*

12) If you are purifying the protein on Cation Exchange Chromatography, What will be the salt used in elution buffer? **0.5 points**

- High concentration of phosphate buffer
- Low concentration phosphate buffer
- A buffer made with KCl
- Very low concentration imidazole solution

**No, the answer is incorrect.**

**Score: 0**

**Accepted Answers:**

*A buffer made with KCl*

13) Match the following techniques with their respective principles/ advantages

- |                         |   |
|-------------------------|---|
| a) 2DE                  | i) Separation based on mol. Wt.             |
| b) SDS-PAGE             | ii) Separation based on pI                  |
| c) 2D-DIGE              | iii) Less gel to gel variations             |
| d) Offgel fractionation | iv) Separation based on pI & mol. wt.       |
| e) Native PAGE          | v) Separation based on charge to mass ratio |

- a-iv; b-i; c-ii; d-v; e-iii
- a-v; b-ii; c-i; d-iii; e-iv
- a-v; b-ii; c-iv; d-i; e-iii
- a-iv; b-i; c-iii; d-ii; e-v

**No, the answer is incorrect.**

**Score: 0**

**Accepted Answers:**

*a-iv; b-i; c-iii; d-ii; e-v*

14) Which one of the following method is used for ionization of volatile compounds? **0.5 points**

- Electrospray ionization
- MALDI
- Electron ionization
- FAB

**No, the answer is incorrect.**

**Score: 0**

**Accepted Answers:**

*Electron ionization*

15) There are many ionization sources that has been tried and tested for various applications. The ionizations sources are categorized into gas phase, solid phase and solution phases. Which of the falls under the gas phase ionization source? **0.5 points**

- ESI
- MALDI
- Electron ionization
- All of the above

**No, the answer is incorrect.**

**Score: 0**

**Accepted Answers:**

*Electron ionization*

16) What is the basic principle of OFFGEL fractionator for fractionation of the proteins? **0.5 points**

- Based on Molecular weight
- Based on isoelectric pH



**0.5 points**

**0.5 points**

**0.5 points**

- Both Molecular weight and isoelectric pH
- Based on charge

**No, the answer is incorrect.**

**Score: 0**

**Accepted Answers:**

*Based on isoelectric pH*

17) DFFGEL fractionator is used for fractionation of

0.5 points

- Proteins
- Peptides
- Both
- DNA

**No, the answer is incorrect.**

**Score: 0**

**Accepted Answers:**

*Both*

18) Which of the following is tandem mass spectrometry

0.5 points

- Q-TOF
- TOF-TOF
- Q-TRAP
- All of the above

**No, the answer is incorrect.**

**Score: 0**

**Accepted Answers:**

*All of the above*

19) What is advantage of Chip cube integrated with QTOF mass spectrometry?

0.5 points

- Superior sensitivity
- Integrated RP column
- Low sample injection
- All of the above

**No, the answer is incorrect.**

**Score: 0**

**Accepted Answers:**

*All of the above*

20) What are the major application of chip cube QTOF instrument?

0.5 points

- Metabolomic analysis
- iTRAQ based quantitation
- Label-free quantization
- All of the above

**No, the answer is incorrect.**

**Score: 0**

**Accepted Answers:**

*All of the above*

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