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reviewer2@nptel.iitm.ac.in ▼

Courses » Proteins and Gel-Based Proteomics

Announcements **Course** Ask a Question Progress

Unit 4 - Week-2: Gel-based proteomics and sample preparation



Course outline

How to access the portal

Introduction to Gel-Based Proteomics

Week-1: Basics of amino acids and proteins

Week-2: Gel-based proteomics and sample preparation

- L6. Systems biology and proteomics
- L7. Sample preparation and pre-analytical factors
- L8. Sample preparation: Pre-analytical factors (contd.)
- L9. Sample preparation: Protein extraction and quantification
- L10. One-dimensional electrophoresis
- Lab session-2.1: Sample preparation: Tissue sample preservation technology
- Lab session-2.2: Protein/peptide pre-fractionation

Week-2 Assignment-2

The due date for submitting this assignment has passed. **Due on 2016-04-06, 23:45 IST.** As per our records you have not submitted this assignment.

1) A protein has a molecular mass of 400 kDa when measured by gel filtration chromatography. When this protein was subjected to polyacrylamide gel electrophoresis in the presence of SDS, the protein showed 3 bands with molecular mass of 180, 160 and 60 kDa. When electrophoresis was performed in presence of SDS and dithiothreitol (DTT)/β-mercaptoethanol, again 3 bands were formed but this time with molecular masses of 160, 90 and 60 kDa. What is the subunit composition of the protein? 0.5 points

- 60, 90, 90 and 160 kDa
- 60, 80, 100 and 160 kDa
- 60, 160 and 180 kDa
- 60, 85, 95 and 180 kDa

No, the answer is incorrect.

Score: 0

Accepted Answers:

60, 90, 90 and 160 kDa

2) If your quantitative proteomics experiment contains large number of samples, which of the following would be the preferred choice of method? 0.5 points

- ICAT
- SILAC
- Label-free quantification
- Western blotting

No, the answer is incorrect.

Score: 0

Accepted Answers:

Label-free quantification

3) Serum contains a myriad of proteins and majorly consists of albumin and globulin proteins. These are high abundant proteins and make up ~80% of the protein content. You are working on a serum sample and your aim is to identify low abundant proteins. Which of the following method will you use to increase the protein coverage? 0.5 points

- TCA-acetone protein precipitation
- Ammonium sulphate precipitation
- Depletion of high abundant proteins
- Desalting of the sample

using Off-gel fractionator

- Week-2 Assignment Answer Key

- Quiz : Week-2 Assignment-2

Week-3: Two-dimensional gel electrophoresis (2-DE)

Week-4: Difference gel electrophoresis (DIGE) & Mass spectrometry

No, the answer is incorrect.

Score: 0

Accepted Answers:

Depletion of high abundant proteins

4) You want to study mitochondrial enzymes involved in β -oxidation of fatty acids. Which of the following method will you use for fractionation of sub-cellular organelles? 0.5 points

- Affinity chromatography
- Electrophoresis
- Centrifugation
- Sequential extraction

No, the answer is incorrect.

Score: 0

Accepted Answers:

Centrifugation

5) Which one of the following is NOT an element of systems biology? 0.5 points

- Biological Networks
- Modelling
- Computation
- Pre-fractionation

No, the answer is incorrect.

Score: 0

Accepted Answers:

Pre-fractionation

6) Protein precipitation can be performed using which of the following solvents? 0.5 points

- Acetone
- TCA-acetone
- Isopropanol
- All of the above

No, the answer is incorrect.

Score: 0

Accepted Answers:

All of the above

7) The cross-linking agent used in polyacrylamide gels is..? 0.5 points

- Acrylamide.
- TEMED.
- APS.
- Bisacrylamide.

No, the answer is incorrect.

Score: 0

Accepted Answers:

Bisacrylamide.

8) The following 5 proteins (A-E), which are listed with their molecular weights (MW) and isoelectric points (pI) were separated by SDS-PAGE. Give the order of their migration from the top (the point of sample loading) to the bottom of the gel. 0.5 points

Protein A (MW 35000, pI 5.4), Protein B (MW 83400, pI 10.6), Protein C (MW 47000, pI 7.0), Protein D (MW 19000, pI 4.8), Protein E (MW 97000, pI 5.9)

- E, D, C, B, A



- E, B, A, C, D
- E, B, C, A, D
- E, D, A, B, C

No, the answer is incorrect.

Score: 0

Accepted Answers:

E, B, C, A, D

9) Membrane proteins are highly hydrophobic in nature and thus are difficult to extract from the membrane. In light of this, which of the following reagent will be used for solubilisation of membrane proteins during protein extraction? 0.5 points

- Thiourea
- β -mercaptoethanol
- Dithiothreitol
- Urea

No, the answer is incorrect.

Score: 0

Accepted Answers:

Thiourea

10) Your protein sample has a lot of phosphorylated proteins. In this light, which of the following stain would you prefer to use for the detection of phosphorylated proteins specifically? 0.5 points

- Pro-Q Diamond
- Coomassie Brilliant Blue
- Silver stain
- SYPRO Ruby

No, the answer is incorrect.

Score: 0

Accepted Answers:

Pro-Q Diamond

11) Which of the following reagent is added to the protein sample to provide charge in Blue native PAGE? 0.5 points

- Dithiothreitol
- Sodium dodecyl sulfate
- Coomassie brilliant blue
- Iodoacetamide

No, the answer is incorrect.

Score: 0

Accepted Answers:

Coomassie brilliant blue

12) Lipids often interfere in proteomics experiments and their removal is essential for enhanced proteome coverage. Which of these organic solvents is used to remove lipid contamination from protein sample? 0.5 points

- Acetic acid
- Acetonitrile
- Methanol
- Formic acid

No, the answer is incorrect.

Score: 0

Accepted Answers:



Methanol

13) Which of the following statement(s) is true for SDS-PAGE? 0.5 points

- Proteins are denatured by the detergent SDS and boiling of sample
- Upon SDS treatment, all proteins possess the same charge-to-mass ratio
- Smaller proteins migrate more rapidly through the gel
- All of the above

No, the answer is incorrect.

Score: 0

Accepted Answers:

All of the above



14) Salts often interfere in 2-DE experiments and should be removed to avoid clogging of IPG strip. Which of the following technique is NOT employed for removing salts from protein sample? 0.5 points

- Equilibration
- Gel permeation chromatography
- Precipitation
- Dialysis

No, the answer is incorrect.

Score: 0

Accepted Answers:

Equilibration

15) Which of the following is an aim of sample pre-fractionation? 0.5 points

- Increasing the proteome coverage
- Reducing sample complexity
- Removal of high abundant proteins
- All of the above

No, the answer is incorrect.

Score: 0

Accepted Answers:

All of the above

16) Which of the following statement(s) is TRUE for off-gel fractionation? 0.5 points
(Note: Question 16-20 are lab session based questions)

- The instrument can fractionate only one sample at a time
- Gel side of the IPG strip should be kept upwards
- It results in decreased proteome coverage
- All of the above

No, the answer is incorrect.

Score: 0

Accepted Answers:

Gel side of the IPG strip should be kept upwards

17) Heat stabilization techniques have significant advantages over the conventional approaches like snap-freeze because..? 0.5 points

- they include addition of an inhibitor.
- they are free of additives.
- they include addition of organic solvents.
- they involve maintenance of pH.

No, the answer is incorrect.

Score: 0

Accepted Answers:

they are free of additives.

18 On which of the following basis, does protein/peptide separation occur in off-gel fractionation? 0.5 points

- Mass
- Molecular weight
- Isoelectric point
- None of the above

No, the answer is incorrect.

Score: 0

Accepted Answers:

Isoelectric point

19 Proteomics studies using brain tissue samples are often used to understand the progression of brain tumor. Tissue samples should be handled with care and different strategies are often exploited to get a better coverage. Heat stabilization of tissue samples using “Denator” involves..? 0.5 points

- denaturation of proteins.
- reduction in enzyme activity.
- unfolding of proteins.
- permanent inactivation of enzymes.

No, the answer is incorrect.

Score: 0

Accepted Answers:

permanent inactivation of enzymes.

20 Biological samples are extremely complex and contain different biomolecules and small metabolites. The main role of off-gel fractionation in case of biological samples is to..? 0.5 points

- reduce their complexity.
- resolve proteins/peptides on the basis of their pI.
- separate proteins/peptides on the basis of their hydrophobicity.
- Both A and B.

No, the answer is incorrect.

Score: 0

Accepted Answers:

Both A and B.

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