

Unit 3 - Week-2: Gel-based proteomics

Course outline

How to access the portal ?

Week-1: Basics of proteins and proteomics

Week-2: Gel-based proteomics

- L6. Sample preparation and pre-analytical factors
- L7. Sample preparation: Pre-analytical factors (contd.)
- L8. Sample preparation: Protein extraction and quantification
- L9. One-dimensional electrophoresis
- L10. Introduction to 2-DE

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

Week-4: Difference in gel electrophoresis (DIGE) & Systems Biology

Week-5: Basics of mass spectrometry

Week-6: Basics of mass spectrometry and sample preparation

Week-7: Quantitative Proteomics

Week-8: Advancement in Proteomics

Text Transcripts

Week-2 Assignment

The due date for submitting this assignment has passed.
As per our records you have not submitted this assignment.

Due on 2019-09-11, 23:59 IST.

Week-2 Assignment

- 1) In case of discontinuous SDS-PAGE, which of the following is the composition of resolving gel solution? **1 point**
- 30% Acrylamide, 10% APS, pH 8.8
 - 30% Acrylamide, 10% APS, pH 6.8
 - 10% Acrylamide, 30% APS, pH 6.8
 - 10% Acrylamide, 30% APS, pH 8.8

No, the answer is incorrect.

Score: 0

Accepted Answers:
30% Acrylamide, 10% APS, pH 8.8

- 2) During sample preparation for a proteomic experiment, protease inhibitor cocktail (PIC) is generally added. What is the purpose of adding PIC? **1 point**
- It ensures the protection of proteins against proteolytic enzymes
 - It helps to reduce the proteins
 - It helps to denature the intact proteins
 - It increases the solubility of proteins in buffer

No, the answer is incorrect.

Score: 0

Accepted Answers:
It ensures the protection of proteins against proteolytic enzymes

- 3) During sample preparation for a general proteomics experiment, various steps are performed such as protein extraction and cell lysis etc. Why cell lysis is important during the process? **1 point**
- It helps to effectively disrupt the cells
 - It helps to maximize the sample recovery
 - It helps to break the disulphide bridges in proteins
 - None of the above

No, the answer is incorrect.

Score: 0

Accepted Answers:
It helps to effectively disrupt the cells
It helps to maximize the sample recovery

- 4) Chaotropic agents such as urea and thiourea are used in solubilising buffer for solubilisation of proteins. What does urea actually do during the process? **1 point**
- Disrupts the hydrogen bonds and hydrophobic interactions between and within the proteins
 - Sample containing urea buffer when heated at 95 degrees breaks the disulphide bonds and hence protein become soluble
 - It disrupts all the covalent and non-covalent interactions between the proteins
 - For disrupting non covalent interactions urea requires the presence of SDS in the solution

No, the answer is incorrect.

Score: 0

Accepted Answers:
Disrupts the hydrogen bonds and hydrophobic interactions between and within the proteins

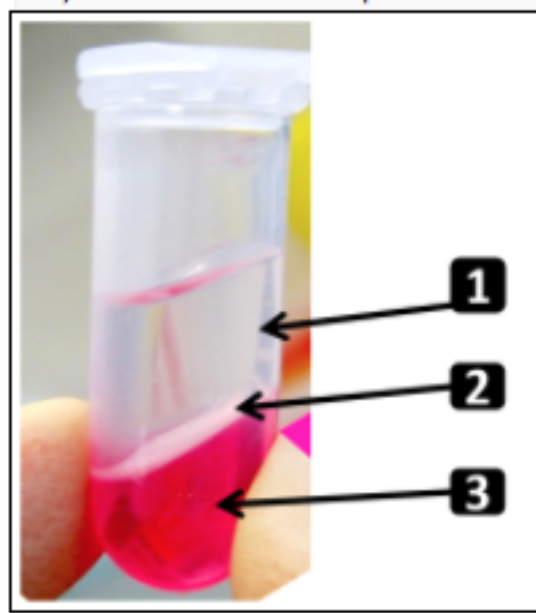
- 5) While performing a proteomic experiment using blood samples which of the following method should be preferred for initial cell homogenisation? **1 point**
- Osmotic lysis
 - Sonication
 - Bead beating
 - Grinding in liquid Nitrogen

No, the answer is incorrect.

Score: 0

Accepted Answers:
Osmotic lysis

- 6) Given below is a picture showing the three layers formed during the process of protein extraction in Trizol method. **0 points**



Which of the following is a correct match for labels 1 to 3 based upon the figure?

- 1-Protein, 2-DNA and 3-RNA
- 1- RNA, 2- DNA and 3- Protein
- 1- Protein, 2- Salt and 3- RNA
- 1- RNA, 2- Protein and 3- DNA

No, the answer is incorrect.

Score: 0

Accepted Answers:
1- RNA, 2- DNA and 3- Protein

- 7) Information for Q7 and 8 **1 point**

Sudesh wants to quantify the amount of protein in three unknown protein samples (A, B and C) using Bradford assay quantification method. He used BSA as a standard protein to plot the standard curve. Given below is the figure showing the absorbance values for increasing amount of BSA in the standard sample and the standard curve obtained by plotting the average absorbance values (on y-axis) against respective amount of BSA (on x-axis).

BSA amt.	OD 1	OD 2	Avg. OD
0	0	0	0
2	0.173	0.154	0.164
4	0.264	0.27	0.267
6	0.443	0.359	0.401
8	0.511	0.522	0.517
10	0.657	0.609	0.633

The average absorbance values obtained for samples A, B and C were 0.622, 0.232 and 0.487, respectively. Based on this information, answer the questions 7 and 8.

What would be the concentration (ug/ul) of proteins in unknown samples A and C?

- 7.6 and 3.5 respectively
- 9.7 and 7.5 respectively
- 9.7 and 3.5 respectively
- 11.6 and 7.6 respectively

No, the answer is incorrect.

Score: 0

Accepted Answers:
9.7 and 7.5 respectively

- 8) Sample _____ got the lowest concentration and sample _____ has the highest concentration. **1 point**

- A and B respectively
- B and C respectively
- B and A respectively
- C and A respectively

No, the answer is incorrect.

Score: 0

Accepted Answers:
B and A respectively

- 9) Given below are few statements about 1D SDS-PAGE and 2-DE. Which of the following is correct? **1 point**

- In 2-DE, stacking gel is not required.
- 1-D SDS-PAGE resolves the proteins on the basis of their charge only whereas 2-DE separates on the basis of Isoelectric point and molecular weight
- Isoelectric focussing is a part of 2-DE only
- None of the above

No, the answer is incorrect.

Score: 0

Accepted Answers:
In 2-DE, stacking gel is not required.
Isoelectric focussing is a part of 2-DE only

- 10) While preparing solution for SDS-PAGE, Ammonium Persulfate (APS) is usually added. What is the purpose of adding APS? **1 point**

- It helps to trace the migration of proteins on the gel
- It helps in polymerisation of acrylamide and bis-acrylamide
- It denatures the proteins
- None of the above

No, the answer is incorrect.

Score: 0

Accepted Answers:
It helps in polymerisation of acrylamide and bis-acrylamide