



Name :

Roll No. :

Invigilator's Signature :

CS/B.Tech/(BT-NEW)/SEM-6/BT-604A/2013

2013

BIOSEPARATION TECHNOLOGY

Time Allotted : 3 Hours

Full Marks : 70

The figures in the margin indicate full marks.

*Candidates are required to give their answers in their own words
as far as practicable.*

GROUP – A

(Multiple Choice Type Questions)

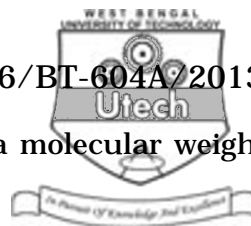
1. Choose the correct alternatives for any *ten* of the following :

10 × 1 = 10

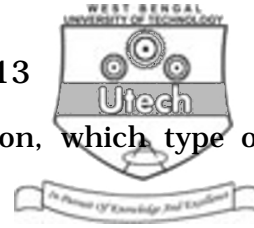
- i) Non-mechanical methods of cell disruption include
 - a) Osmotic shock
 - b) Ultrasound
 - c) Ball Mill
 - d) Homogenizer.



- ii) Liquid-Liquid extraction depends on
- a) distribution coefficient
 - b) volatility
 - c) solubility
 - d) partition coefficient.
- iii) Dialysis is a membrane separation operation used for the removal of low molecular weight solutes such as organic ions of MW range
- a) $10 < MW < 100$
 - b) $MW > 10$
 - c) $MW < 10$
 - d) $MW > 100$.
- iv) Microfiltration (MF) is used to separate species that range from
- a) 0.1 to 10 μm
 - b) 1.0 to 10 μm
 - c) 10 to 20 μm
 - d) 20 to 50 μm .



- v) UF is used for macromolecules with a molecular weight range of
- a) 2,000 to 5,00,000
 - b) 2,000 to 10,000
 - c) 5,000 to 50,000
 - d) 1,00,000 to 5,00,000.
- vi) Which of the following will help to confirm the molecular weight of the purified protein ?
- a) Isoelectric focusing
 - b) Affinity chromatography
 - c) Native-PAGE
 - d) Gel filtration.
- vii) Affinity chromatography is based on the highly specific interaction between
- a) solute molecules and ligands
 - b) solute molecules
 - c) ligands
 - d) solute molecules and ceramic beads.



viii) In rate-zonal centrifugation separation, which type of particle characteristics is taken ?

- a) Size
- b) Density
- c) Charge
- d) Volume.

ix) Extraction factor is determined during

- a) Microfiltration
- b) Solvent extraction
- c) Ultracentrifugation
- d) both (a) and (b).

x) Ultrafiltration process cannot be used for

- a) Fractionation of proteins
- b) Desalting
- c) Harvesting of cells
- d) Selective removal of solvents.

xi) Proteins are separated in the SDS – PAGE according to their

- a) Charge
- b) Hydrophobicity
- c) Size
- d) Affinity.



xii) Which one of the following proteins of different molecular weights migrates faster through a gel filtration chromatography and elutes first ?

- a) 66 kDa
- b) 100 kDa
- c) 200 kDa
- d) 30 kDa.

GROUP - B

(Short Answer Type Questions)

Answer any *three* of the following. $3 \times 5 = 15$

2. What is pervaporation ? How does it differ from distillation and evaporation ?
3. Explain the terms 'salting in' and 'salting out' of proteins. Discuss the principles and practice of salting-out of proteins by ammonium sulphate. $2 + 3$
4. What is flocculation and what is the advantage of it during filtration of a fermentation broth ? Discuss the theoretical principles of constant pressure cake filtration. $2 + 3$
5. Describe in brief the non-mechanical methods of cell disruption.
6. Define affinity chromatography. Explain basic operating principles of affinity chromatography.

**GROUP - C****(Long Answer Type Questions)**

Answer any *three* of the following. $3 \times 15 = 45$

7. a) What is an adsorption isotherm ? Give expressions of different adsorption isotherms.
- b) An enzyme produced from *Aspergillus niger* can be isolated by adsorption on a microcrystalline cellulose powder (MCCP). The adsorption follows a Langmuir Adsorption Isotherm :

$$Y^* = \frac{Y_{max} \cdot X}{K_L + X}$$

where $Y_{max} = 70 \text{ mg/cm}^3$ of adsorbent

$$K_L = 5 \times 10^{-5} \text{ g/g solution}$$

$$X = 2 \text{ g/g of solution}$$

It is desired to recover 95% of the product from 1.5 L of cell free culture filtrate of concentration 220 mg/L protein with (MCCP). Estimate the amount of solid required for the purpose ? $1 + 4 + 10$

8. a) Discuss the role of distribution coefficient in liquid-liquid extraction.
- b) A slurry with solid concentration of 23.47 kg/m^3 was filtered within a plate and frame filter press with a filter area of 0.0439 m^2 at a constant pressure drop of 330 kNm^{-2} . The data obtained are given below :

Time (t) min	4.4	9.5	16.3	24.6	34.7	46.1	59.0	73.6	89.4	107.3
Volume (V) L	0.498	1.000	1.501	2.000	2.489	3.002	3.506	4.004	4.502	5.009



The same slurry is then to be filtered in a plate and frame filter press having 10 frames and 0.873 m^2 area per frame at a constant pressure drop of 330 kNm^{-2} . For the same cake characteristics and 3 layers of filter cloth, calculate the time to recover 3300 L of filtrate. [Assume that filter cloth resistance is directly proportional to the thickness or number of layers of cloth.]

5 + 10

9. What is foam fractionation ? How is it used for the purification purposes in the area of biotechnology ? Explain the concept of spacer arm and its use. What are the advantages of aqueous two phase extraction over the other extraction processes in biotechnology ? The solubility of a protein is 15 g/litre at ammonium sulphate concentration of 2.2 M and 0.25 g/litre at 3.0 M. Calculate the solubility of the protein at 3.8 M of the salt. 2 + 2 + 2 + 5 + 4
10. Give a complete flow diagram of isolation and purification of any hormone or antibiotic in a commercial plant. Briefly describe the major operations involved in this process.
11. Write notes on any *three* of the following : 3 × 5
 - a) Ultrafiltration
 - b) Microfiltration
 - c) Reverse Osmosis
 - d) Concentration polarization.

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