

KATHMANDU UNIVERSITY
End Semester Examination
18, January 2024

Level : B.Tech.
Year : III
Time : 2 hrs. 30 mins.

Course : BIOT 301
Semester : I
F.M. : 55

SECTION "C"

[3 Q. × 8 = 24 marks]

Attempt *ANY THREE* questions.

1. List the various properties of biological molecules based on which bio separation can be performed. Explain any two properties.
2. List the various physical and chemical cell disruption techniques used in downstream processing. Explain any two techniques.
3. Explain the principles of precipitation using organic solvents and anti-chaotropic salts with an example.
4. Ammonium sulphate being used to precipitate a humanized monoclonal antibody from 10 liters of cell culture media, the initial concentration of the antibody in this liquid being 0.5 mg ml^{-1} . Solid ammonium sulphate is added to the liquid such that the concentration of the salt is $1.5 \text{ kgmoles m}^{-3}$. This results in the precipitation of 90 % of the antibody. When the ammonium sulphate concentration of the mixture is raised to $1.75 \text{ kgmoles m}^{-3}$, a further 76.5 % of the remaining antibody is precipitated. Predict the ammonium sulphate concentration needed for total antibody precipitation.
5. A suspension of yeast cells is to be filtered at a constant filtration rate of 50 L min^{-1} . The suspension has a solid content of 70 kg m^{-3} of suspension and the yeast cells have a bulk density of 800 kg m^{-3} . The specific cake resistance is 40 m kg^{-1} and the viscosity of the filtrate is $2.9 \times 10^{-3} \text{ kg m}^{-1}\text{s}^{-1}$. The filter has an area of 0.1 m^2 and the medium offers negligible resistance. How long can the filtration rate be maintained before the pressure drop exceeds 1 N m^2 ?

SECTION "D"

Attempt *ANY SIX* questions. (Question N.O 6 is compulsory)

6. Write short notes on [3+3=6]
 - a. Cohn equation
 - b. Microfiltration
7. Explain the working principle of disc stack centrifuge with a diagram. [5]
8. Explain briefly the steps involved in extraction. [5]
9. Write down the general filtration rate equation and define all the variables in the equation. [5]
10. Explain briefly the principle of ion exchange adsorption. [5]
11. How biological molecules can be separated based on their molecular weight? [5]

12. A batch of yeast cells was disrupted using ultrasonic vibrations to release an intracellular product. The concentration of released product in the solution was measured during the process as shown in the table. If the ultrasonic cell disruption were carried out for 200 seconds, predict the product concentration. [5]

Time (s)	Concentration (mg/ml)
60	3.49
120	4.56

13. Chlorella cells are being cultivated in an open pond. We plan to harvest this biomass by passing the dilute stream of cells through an available disc bowl centrifuge. The settling velocity (v_g) for these cells has been measured as $1.07 \times 10^{-4} \text{ cm s}^{-1}$. The centrifuge has 80 discs with an angle of 40° , an outer radius of 15.7 cm and an inner radius of 6 cm. We plan to operate the centrifuge at 600 rpm. Estimate the volumetric capacity (Q) for this centrifuge. [5]

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Date :

SECTION "A"

[20Q × 0.5 = 10 marks]

Choose and mark [X] the most appropriate alternative from each set of choices.

1. Which of the following statement for "Bio separation" is **NOT TRUE**?
 Dilute product streams have to be processed
 Selective in nature
 Carried out at sub-ambient temperature
 Based on single step separation
2. Biomolecules can be separated based on shape by
 Extraction Electrophoresis Precipitation Centrifugation
3. Stokes-Einstein diameter of a protein refers to the particle diameter with equal
 Dielectric constant Diffusivity
 Polarity Viscosity
4. Molecular weights of macromolecules are usually determined using all the following techniques except
 Hydrodynamic chromatography Size-exclusion chromatography
 Ultracentrifugation Conventional Filtration
5. Osmotic shock is used to remove
 Intracellular protein Periplasmic protein
 Membrane proteins Mitochondrial protein
6. Typical rotation speed of rotor-stator mill is in the range of _____ rpm.
 1000-5000 5000-20000 15000-40000 10000-50000
7. Kick's coefficient depends on
 Size Viscosity Rigidity Operating condition
8. Cohn's fractionation of plasma proteins is based on
 Centrifugation Precipitation Extraction Filtration
9. Solvents precipitate proteins by decreasing
 Dielectric constant Polarity
 Chemical Potential pH
10. Chaotropic salts are used for the purification of
 Nucleic acids Proteins Lipids Carbohydrates
11. Ultrasonic disruption of cells is commonly performed at frequency
 25 kHz 20kHz 30kHz 35kHz
12. Typical G values employed in laboratory centrifuges range from
 1000 - 20000 500-1000 1000-15000 1000-30000
13. Which rotors are preferred for centrifuging substances with high sedimentation coefficients such as cells?
 Fixed angled rotor Swing out rotors
 Vertical rotors All of the above

