BIOSEPARATION TECHNOLOGY
(BIOT 2211)

Time Allotted : 3 hrs   **Full Marks : 70**

Figures out of the right margin indicate full marks.

*Candidates are required to answer Group A and
any 5 (five) from Group B to E, taking **at least one** from each group.*

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

Group – A
(Multiple Choice Type Questions)

1. Choose the correct alternative for the following: **10 × 1 = 10**

   (i)   Which force is involved in the Chromatography?
           (a) Hydrogen bonding   (b) Van der Wall force
           (c) Electric static force   (d) All of the above.

   (ii)   Which of the polymer is often used as solid matrix in affinity chromatography?
           (a) Dextran   (b) Agarose   (c) Cellulose   (d) All the above.

   (iii)   Which chromatographic technique depends on the highly specific interaction between pairs of biological molecules such as enzyme-substrate?
           (a) Adsorption chromatography   (b) Ion exchange chromatography
           (c) Affinity chromatography   (d) Gel permeation chromatography.

   (iv)   Protein separation technique is often based on except
           (a) charge of protein   (b) solubility of protein
           (c) viscosity of protein   (d) specific binding affinity of protein

   (v)   Concentration polarization occurs in which of the following processes
           (a) reverse osmosis   (b) ultra-filtration
           (c) micro-filtration   (d) all of the above.

   (vi)   Which of the following is not a membrane separation process?
           (a) Ultra-filtration   (b) Chromatography
           (c) Flash distillation   (d) Pervaporation.

   (vii)   At terminal velocity
           (a) $F_D = F_G + F_B$ (b) $F_G = F_D + F_B$
           (c) $F_B = F_D + F_G$ (d) None of the above.

   (viii) In filtration out of the two resistances
           (a) r_m is variable, r_c is constant   (b) r_c is variable, r_m is constant
           (c) r_c decreases over time   (d) r_m increases over time.

(ix)   The most important application of dialysis is
(a) artificial kidneys
(b) desalination of brackish water
(c) recovery of salts and sugars from natural products
(d) removal of alcohol from beer.

(x)   The only membrane separation in which the permeate undergoes a phase change is
(a) electrodialysis   (b) pervaporation
(c) ultrafiltration   (d) reverse osmosis.

Group- B

2. (a)   At the end of the filtration cycle, a total filtrate volume of 3.37 m^3 is collected in a total time of 269.7 s. The cake is to be washed by thorough washing in the plate and frame press using a volume of wash water equal to 10% of the filtrate volume. Calculate the time of washing and the total filter cycle time if cleaning the filter takes 20 min. Given: $1/K = 37.93$ s/m^6 and $2V_0/K = 16.1$ s/m^3.
[(CO1)(Analyze/IOCQ)]

   (b)   Derive Ruth's equation for constant pressure filtration.
[(CO2)(Understand/LOCQ)]
6 + 6 = 12

3. (a)   A continuous disc stack centrifuge is operated at 5000 rpm for separation of baker's yeast. At a feed rate of 60 L/min, 50% of the cells are recovered. For operation at constant centrifuge speed, solids recovery is inversely proportional to the flow rate.
(i)   What flow rate is required to achieve 90% cell recovery if the centrifuge speed is maintained at 5000 rpm?
(ii) What operating speed is required to achieve 90% recovery at a feed rate of 60 L/min?   [(CO2)(Calculate/IOCQ)]

   (b)   How are cells disrupted by non-mechanical methods?
[(CO1)(Remember/LOCQ)]
6 + 6 = 12

Group - C

4. A feed solution at 25°C contains 3500 mg NaCl/L (ρ = 999.5 kg/m^3). The permeability constant $A_w = 3.5 \times 10^{-4}$ kg solvent/s.m^2.atm and $A_s = 2.5 \times 10^{-7}$ m/s. Using a $\Delta P = 35.5$ atm, calculate the fluxes, solute rejection R and product solution concentration in mg NaCl/L (osmotic pressure = 1.937atm).   [(CO3)(Analyze/IOCQ)]
12

5. (a)   A solution containing 0.9 wt% protein is to undergo ultrafiltration using a pressure difference of 5 psi. The membrane permeability is $A_w = 1.37 \times 10^{-2}$ kg/s.m^2.atm. Assuming no effects of polarization, predict the mass flux. (1 psi = 0.068 atm).   [(CO4)(Understand/LOCQ)]

(b) Write a note on any one membrane separation process mentioning its advantages, disadvantages and applications in the field of biotechnology.

[(CO2)(Understand/LOCQ)]

6 + 6 = 12

Group - D

6. (a) How does column chromatography differs from planer chromatography? Explain with diagram. [(CO3)(Analysis/IOCQ)]

(b) The data in the following table were extracted from the chromatogram of a two-component mixture of x and y.

	Air	x	y
Retention time (sec)	5	25	30
Peak width at half height (sec)	--	2.5	2.0

(i) Calculate the capacity factors for x and y.

(ii) Calculate the number of theoretical plates N for the column for the component y and the theoretical plate height H with respect to y (assume a column 25 cm long). [(CO3, CO4)(Evaluate/HOCQ)]

4 + (4 + 4) = 12

7. (a) What variables are likely to affect the 'selectivity factor' for a pair of analytes?

[(CO3)(Analysis/IOCQ)]

(b) A chromatogram of a mixture of A and B resulted in the following data:

	Non retained	A	B
Retention time (min)	3.1	13.3	14.1
Peak width (min)	--	1.07	1.16

(i) Calculate the resolution.

(ii) Calculate the selectivity.

(iii) Calculate the length of the column necessary to achieve resolution of 1.5, if the original height of the column is 23.5 cm.

[(CO3,CO4)(Evaluate/HOCQ)]

4 + (4 + 4) = 12

Group - E

8. (a) Draw and explain the phase diagram of crystallisation of macromolecules.

[(CO6)(Analysis/IOCQ)]

(b) Define 'equilibrium moisture' and 'free moisture'.

[(CO6)(Remember/LOCQ)]

8 + (2 + 2) = 12

9. (a) During precipitation of protein, mathematically prove that $M=Mo(1+k_AN_ot)$, where

No is the initial number of solute in solution with molecular weight Mo, M is the molecular weight of the solute aggregate at time t, k_A = rate constant.

[(CO6)(Analysis/IOCQ)]

(b)   Define 'relative humidity' and 'percentage humidity'.

[(CO6)(Remember/LOCQ)]

8 + (2 + 2) = 12

Cognition Level	LOCQ	IOCQ	HOCQ
Percentage distribution	33.33	50	16.67

Course Outcome (CO):

After completion of this course, the students will be able to:
1. Acquire basic understanding of different bioseparation processes and design principle for commonly used process equipments.
2. Obtain knowledge about the basic principles and application of sedimentation, centrifugation and filtration.
3. Explain the principles of extraction and membrane based separation of bioproducts and can apply the knowledge for calculations of extraction process.
4. Understand the principle of adsorption, chromatography and relation of adsorption with chromatography.
5. Apply different chromatographic techniques for separation of different Bioproducts.
6. Comprehend the knowledge of precipitation, drying, crystallization and will be able to solve numerical problems related to these processes.

*LOCQ: Lower Order Cognitive Question; IOCQ: Intermediate Order Cognitive Question; HOCQ: Higher Order Cognitive Question