

**DOWNSTREAM PROCESSING
(BIOT 6133)**

Time Allotted : 2½ hrs

Full Marks : 60

Figures out of the right margin indicate full marks.

*Candidates are required to answer Group A and
any 4 (four) from Group B to E, taking one from each group.*

*Candidates are required to give answer in their own words as far as practicable.
Symbols are of usual significance*

Group – A

1. Answer any twelve:

12 × 1 = 12

Choose the correct alternative for the following

- (i) Microfiltration membranes have pore size in the range
(a) 0.1 to 10 µm (b) 1 to 20 µm
(c) 10 to 20 µm (d) 50 to 80 µm.
- (ii) The most common ion-exchange resin used in aqueous two-phase extraction is
(a) Polyvinylidene difluoride (b) Polyethylene glycol
(c) Polysulfone (d) Polytetrafluoroethylene.
- (iii) Absolute alcohol from fermentation broth may be obtained by the following methods
(a) Liquid-liquid extraction (b) adsorption
(c) pervaporation (d) azeotropic distillation.
- (iv) Proteins are separated in the SDS-PAGE according to their
(a) Charge (b) Hydrophobicity
(c) Size (d) Polarity.
- (v) Gel permeation chromatography separates protein on the basis of
(a) molecular size (b) charge
(c) PI (d) molecular shape.
- (vi) Cell lysis by sonication the frequency of sound waves used is
(a) More than 20,00 kHz (b) less than 20,000 kHz
(c) less than 10 kHz (d) any frequency.
- (vii) Penicillin is more soluble in organic phase at pH
(a) 2 to 3 (b) 4 to 5 (c) 6 to 7 (d) 8 to 9.
- (viii) Which of the following organism is used for ethanol production?
(a) *S. cerevisiae* (b) *E. coli* (c) *Vibrio cholerae* (d) *K. fragillis*.

- (ix) Medical oxygen is best prepared by a packed bed of adsorbent of the type
 (a) Activated carbon (b) Sintered alumina
 (c) Molecular sieve (d) ZSM-5.
- (x) Inhibitory fermentation product is best separated by
 (a) liquid-liquid extraction (b) aqueous two phase extraction
 (c) adsorption (d) ultrafiltration.

Fill in the blanks with the correct word

- (xi) If $C_w / C_b = 10$ concentration polarization is _____.
 [C_w = Solute concentration at membrane surface, C_b = Bulk solute concentration]
- (xii) $\Delta_{PTM} =$ _____
 Δ_{PTM} is transmembrane pressure drop
- (xiii) When particle density and solvent are equal, the sedimentation velocity is zero, and the process is called _____.
- (xiv) In case of sedimentation Reynold's number for spherical particles is _____.
- (xv) Fourier's law of heat conduction applied in conductive drying, $Q =$ _____.

Group - B

2. (a) Define polarization modulus. [[CO2](Remember/LOCQ)]
 (b) It is desired to use a cross flow filtration system to desalt 1000 litres of a protein solution containing NaCl. The system is capable of operating at a transmembrane flux of 30 lit/m² hr. To remove 99.99 % of the salt, determine the time required and the volume of water required using a cross flow filtration unit with a membrane area of 100 m². Given equation $-V(dC_s/dt) = -Q C_s (1 - r_s)$.
[[CO2](Analyse/IOCQ)]
4 + 8 = 12
3. (a) Estimate how long it would take to completely clarify a suspension of ribosomes in a high speed centrifuge operating at 10,000 rpm with a tube containing ribosome suspension in which the maximum distance of travel of particles radially outward is 1.0 cm and the initial distance from the centre of rotation to the particles nearest the centre of rotation is 4.0 cm.
 Data Given : sedimentation coefficient, $S = 70 \times 10^{-13}$ sec. [[CO3](Analyse/HOCQ)]
 (b) If bacterial cell debris has $Gt = 54 \times 10^6$ sec, how large must the centrifuge bowl be and centrifuge speed is needed to effect a full sedimentation in two hours.
 (Given data : Centrifuge diameter is 10 cm , reasonable time 2 hrs.)
[[CO4](Evaluate/IOCQ)]
6 + 6 = 12

Group - C

4. (a) Explain Aqueous two-phase extraction. [[CO3](Remember/LOCQ)]
 (b) A column 20 cm long, with an internal diameter of 5 cm, gives sufficient

purification to merit scale-up. The column produces 3.2 gm of purified protein per cycle, and a cycle takes 6 hour from equilibrium through regeneration. You want a throughput of 10 gm/hr. What are the new column's dimensions if linear velocity is held constant?

[[CO3](Evaluate/HOCQ)]

3 + 9 = 12

5. (a) Explain the function of columns in chromatography. [[CO4](Understand/LOCQ)]
(b) A column 20 cm long, with an internal diameter of 5 cm, gives sufficient purification to merit scale-up. The column produces 3.2 gm of purified protein per cycle, and a cycle takes 6 hr, from equilibrium through regeneration. You want a throughput of 10 gm/hr. Available standard column diameters are 20 cm and 25 cm. Calculate bed depths to each of these columns?

[[CO5](Evaluate/HOCQ)]

3 + 9 = 12

Group - D

6. (a) Explain function of solvent in chromatography. [[CO3](Analyse/LOCQ)]
(b) It is desire to scale up a batch crystallization of an antibiotic based experiments with a 1-liter crystallizer. The use of a 3 cm diameter impeller at a speed of 800 rpm led to good crystallization results. For maintaining power per volume constant upon scale-up to 300 liters, what should be the diameter and speed of the large scale impeller? The solvent has the same density and viscosity as water.

[[CO2](Apply/HOCQ)]

2 + 10 = 12

7. (a) Define protein structure (primary and secondary). [[CO3](Analyse/HOCQ)]
(b) Explain primary nucleation and secondary nucleation. [[CO4](Remember/IOCQ)]

6 + 6 = 12

Group - E

8. (a) Estimate the dimension of a drying chamber for a spray dryer that has an output of 1000 kg/hr of a heat-sensitive biological material at 60°C containing 5% moisture and having a mean particle size of 100 μm. The feed contains 40% solids by weight in an aqueous solution at 4°C. The inlet air has a humidity of 0.01 kg/kg dry air and is at 150°C, while the outlet air is at 80°C. Assume Specific heat of dry solid is 0.3 K cal/kg. C°. [[CO3](Analyse/HOCQ)]

- (b) Differentiate between wet bulb and dry bulb temperature. [[CO6](Remember/IOCQ)]

10 + 2 = 12

9. (a) What do you understand by bound moisture and unbound moisture. [[CO6](Remember/IOCQ)]
(b) Explain the operation of Freeze dryer. [[CO6](Remember/IOCQ)]

4 + 8 = 12

| Cognition Level | LOCQ | IOCQ | HOCQ |
|-------------------------|-------|-------|-------|
| Percentage distribution | 12.50 | 35.42 | 52.08 |

Course Outcome (CO):

After the completion of the course students will be able to

1. Acquire basic understanding of different bio-separation processes and design principle for commonly used process equipment.
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.*