

2125

M. E. (Bio-Technology)  
First Semester

ME-BIO-104: Bio-Separation and Bio-Process Technology

Time allowed: 3 Hours

Max. Marks: 50

*NOTE: Attempt five questions in all, including Question No. I which is compulsory and selecting two questions from each Section.*

x-x-x

1. Attempt the following:-

- a) Define the ideal stage concept.
- b) Justify the use of monovalent salts for precipitation.
- c) Express graphically the concept of washout with respect to dilution factor.
- d) Give the relevance of substrate utilization constant in microbial fermentations.
- e) Differentiate between a chemostat and a turbidostat?
- f) Justify the role of a filter aid.
- g) Discuss the importance of aqueous two phase liquid extraction in bioseparations.
- h) What are the different important factors for improving rate of filtration?
- i) How a multistage system is useful in the culture of genetically engineered cells?
- j) Define sigma factor and g-number of a centrifuge. (10)

**Section-A**

2. A) Summarize in a table form various size range and applications of filtration processes by giving examples.  
B) With the help of appropriate expressions deduce relationship between solubility and salt concentration during precipitation procedure. Showing protein structure and surface chemistry, explain the following terms i) Stern layer ii) salting-out. (4,6)
3. A) Enlist the different types of cell-disruption methods. Explain in detail cell-disruption in a high-pressure homogenizer and deduce expression to quantify extent of cell-disruption achieved under certain operating conditions.  
B) A tubular bowl centrifuge is used to concentrate a suspension of genetically engineered yeast containing a new recombinant protein. At a speed of 12,000 rpm, the centrifuge treats 3 l of broth per min with satisfactory results. It is proposed to use the same centrifuge to separate cell-debris from the homogenate produced by mechanical disruption of yeast. If the average size of the debris is one-third that of the yeast and the viscosity of the homogenate is five times greater than the cell-suspension, what flow rate can be handled if the centrifuge is operated at the same speed? (5,5)

(2)

4. A) Leucine dehydrogenase is recovered from 150 litres of *Bacillus cerus* homogenate using an aqueous two-phase PEG-salt system. The homogenate initially contains 3.2 units of enzyme  $\text{ml}^{-1}$ . A PEG-salt mixture is added and two phases form. The enzyme partition coefficient is 3.5. i) What volume ratio of upper and lower phase must be chosen to achieve 80% recovery of enzyme in a single extraction step? ii) If the volume of the lower phase is 100 litres, what is the concentration factor for 80% recovery?
- B) What is a marker or a ladder? How does size of the DNA affect its migration through the agarose gel during electrophoresis? (6,4)

### Section-B

5. A) With the help of suitable plot and equations, express the metabolite productivity of any one basic cell-cultivation strategy.
- B) Describe quasi steady-state of a fed-batch culture. Include in your answer relevant time-profiles for the important process parameters. (5,5)
6. *Tetrahymena thermophile* protozoa have a minimum doubling time of 6.5 hours when grown using bacteria as the limiting substrate. The yield of protozoal biomass is 0.33 g per g of bacteria and the substrate constant is  $12 \text{ mg l}^{-1}$ . The protozoa are cultured at steady state in chemostat using a feed stream containing  $10 \text{ g l}^{-1}$  of nonviable bacteria.
- A) What is the maximum dilution rate for operation of the chemostat?
- B) What is the concentration of *T. thermophile* when the operating dilution rate is one-half of the maximum?
- C) What is the concentration of the bacteria when the dilution rate is three-quarters of the maximum?
- D) What is the biomass productivity when the dilution rate is one-third of the maximum? (10)
7. A) Give a brief account on the growth stoichiometry and elemental balances. Deduce relevant expressions for respiratory quotient and degree of reduction of substrate.
- B) *Klebsiella aerogens* ( $\text{CH}_{1.75}\text{O}_{0.43}\text{N}_{0.22}$ ) is produced from glycerol ( $\text{C}_3\text{H}_8\text{O}_3$ ) in aerobic culture with ammonia as nitrogen source. The biomass contains 8% ash, 0.40 g biomass is produced for each g of glycerol consumed, and no major metabolic products are formed. Degree of reduction of glycerol relative to ammonia is 4.67. What is the oxygen requirement for this culture in mass terms? (4,6)