Exam.Code:1032
Sub. Code: 35394

2124

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. Give a brief overview of cell-removal operation.
   b. Define ideal stage concept.
   c. Define aqueous two phase liquid extraction process.
   d. Express quasi-steady state of a Fed-batch culture.
   e. Discuss the significance of low Ks value?
   f. Define specific cake resistance.
   g. Differentiate between a chemostatand aturbidostat?
   h. Justify the function of a filter aid.
   i. Enlist various polymeric materials used for membrane separation process.
   j. Define maintenance coefficient "m". (10)

SECTION-A

2. a) Describe typical unit operations required in the recovery and purification of an intracellular protein product from a bacterial source, downstream of the bioreactor. Give your recommendations.
   b) Enlist various membrane-driven processes used as a separation technique. Describe how do you determine the specific cake resistance and fictitious equivalent cake thickness in a constant-pressure filtration? (4,6)

3. a) Leucine dehydrogenase is recovered from 150 litres of *Bacillus cerus* homogenate using an aqueous two-phase polyethyleneglycol-salt system. The homogenate initially contains 3.2 units of enzyme ml^{-1}. A polyethylene glycol-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? Why is this considered standard? How does size of the DNA affect its migration through the agarose gel during electrophoresis? (5,5)

4. a) With the help of protein structure and surface chemistry, explain the following terms (i) Stern layer (ii) salting-in and salting-out.
   b) Find the g-factor of a centrifuge with an effective radius of 10 cm and rotating at a speed of 30 rps.
   c) Give a detailed account on the working and design of the *disc-bowl centrifuge*. (4,2,4)

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SECTION-B

5. a) Explain how and why 'wash-out' occurs in an ideal CSTF. Show that this problem can be overcome by separating and recycling part of the cells coming out of the reactor back to the reactor vessel.

   b) List down the factors that affect specific growth rate. If there is a mathematical description of the same, show that also. (6,4)

6. Assume the experimental measurements for a certain organism have shown that cells can convert two-thirds (wt/wt) of the substrate carbon (alkane or glucose) to biomass.

   a) Calculate the stoichiometric coefficients for the following biological reactions:

   Hexadecane: $C_{16}H_{34} + aO_2 + bNH_3 \rightarrow cC_{4.4}H_{7.3}N_{0.86}O_{1.2} + dH_2O + eCO_2$

   Glucose: $C_6H_{12}O_6 + aO_2 + bNH_3 \rightarrow cC_{4.4}H_{7.3}N_{0.86}O_{1.2} + dH_2O + eCO_2$

   b) Determine the degree of reduction for the substrate and bacteria for both reactions.

   c) Calculate the yield coefficients $Y_{X/S}$, $Y_{X/O2}$ and RQ for both reactions. Comment on the differences. (10)

7. a) Fermentation of *Candida utilis* exhibit substrate inhibition kinetics given by the following equation

$$\mu = \frac{\mu_{max}.S \; S^2=K_S{}^*I}{K_S+S+S^2/K_I}$$

   Where S is the substrate concentration and μ is the specific growth rate. For a continuous fermentation using sterile feed, derive an equation for the steady state variation of biomass concentration, substrate concentration and maximum cell productivity with dilution rate when (i) I=0(ii) I is not equal to zero.

   b) Explain different types of inhibition by toxic compounds along with the graphical representation.

   c) What is the generation time if 100 bacterial cells growing logarithmically for 5 hours produce 1.7×10^6 cells? (5,3,2)

x-x-x