Exam.Code:0910
Sub. Code: 33378

2056

B.E. (Biotechnology) Sixth Semester

BIO-614: Down Stream Processing

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 Unit.

x-x-x

1. Answer the following:-

a) Differentiate between differential centrifugation and density gradient centrifugation.

b) Name any two mechanical methods of cell disruption used in industry.

c) What is the precipitation of proteins?

d) Write the applications of adsorption in downstream processing.

e) Downstream processing primarily refers to:

i) Production of microorganisms

ii) Recovery and purification of products from fermentation broth

iii) Genetic engineering of microbes

iv) Media preparation

f) The driving force for filtration is:

i) Temperature difference

ii) Pressure difference

iii) Electrical potential

iv) Concentration gradient

g) Which adsorption model describes heterogeneous surfaces?

i) Langmuir

ii) Freundlich

iii) Linear isotherm

iv) BET

h) The partition coefficient in liquid-liquid extraction is $K = \frac{C_{organic}}{C_{aqueous}}$ it represents:

i) Pressure difference

ii) Distribution of solute between phases

iii) Temperature difference

iv) Flow rate

Contd.....P/2

i) A filtration unit produces 20 L of filtrate in 40 minutes using a membrane area of 0.5 m^2. Calculate the filtration flux.

i) 30 $L/m^2 \cdot h$

ii) 60 $L/m^2 \cdot h$

iii) 120 $L/m^2 \cdot h$

iv) 10 $L/m^2 \cdot h$

j) A centrifuge has a rotor radius of 10 cm and rotates at 8000 rpm. Calculate the Relative Centrifugal Force (RCF).

i) 70 g

ii) 716 g

iii) 7160 g

iv) 71 g (10x1)

UNIT - I

2. a) Define bioseparations and explain the importance of downstream processing in biotechnological industries.

b) Compare mechanical, enzymatic, and chemical methods of cell disruption with suitable examples.

c) Discuss two methods used for protein precipitation (3,5,2)

3. An industrial fermentation produces an extracellular antibiotic. The product is soluble in organic solvents. The fermentation broth contains cells, residual nutrients, antibiotic product.

a) Propose a downstream recovery process using liquid–liquid extraction

b) Explain how the partition coefficient affects extraction efficiency

c) What are the advantages of solvent extraction compared with adsorption? And suggest how the antibiotic could be further purified. **(3,3,4)**

4. a) Define adsorption and explain the phenomenon of adsorption in bioseparation processes.

b) Explain the Langmuir adsorption isotherm and its importance in designing adsorption systems.

c) Describe the types of adsorption and different adsorbents used in bioprocessing **(3,4,3)**

UNIT - II

5. A biotechnology company produces a recombinant therapeutic enzyme using E. coli fermentation (2000 L scale). The product is used to treat a metabolic disorder and must meet FDA biopharmaceutical quality standards before release.

a) Which analytical technique is most suitable for detecting impurities, and how?

b) A filtration unit produces filtrate in 40 minutes using a membrane area of 0.5 m^2. Calculate the filtration flux with the formula $J=A \times tV$, where

J = flux ($L/m^2 \cdot h$)

V = filtrate volume

A = filtration area

t = time

c) After fermentation, explain how the following steps would be integrated in downstream processing to obtain a purified product.

i) Product separation after fermentation (mention suitable techniques and their role).

ii) Product polishing step required to achieve pharmaceutical-grade purity.

iii) Explain how these steps help meet FDA purity and safety standards. (2,2,6)

6. Membrane separation and chromatography are widely used in downstream processing for the purification of biotechnological products.

a) Explain the principle of chromatography and discuss how HPLC can be used to monitor product purity during downstream processing.

Contd.....P/3

b) Compare nanofiltration and reverse osmosis in terms of pore size, operating pressure, and industrial applications.

c) Explain the principle of membrane separation and briefly describe microfiltration, ultrafiltration, nanofiltration, and reverse osmosis. **(3,3,4)**

7. A laboratory produces several industrial bioproducts through *E. coli.* Fermentation. Each product must undergo specific downstream recovery and purification steps because its chemical nature and location differ, and the downstream recovery strategy must be carefully selected for each product. Below is a general downstream processing flow

Fermentation → Primary Separation → Cell Processing → Clarification → Product Isolation → Intermediate Step→ Product Polishing→ Final Product

a) Explain in the product isolation step the following technique (Distillation, Solvent Extraction, Precipitation, and Chromatography) used for which product (Penicillin, citric acid, ethanol, and recombinant insulin) and why?

b) Discuss the importance of crystallisation and drying in obtaining high-purity products in citric acid and penicillin production.

c) Distinguish between analytical electrophoresis and preparative electrophoresis in terms of purpose, scale of operation, and applications. **(4,3,3)**

x-x-x