# 16BT403 DOWNSTREAM PROCESSING

Hours Per Week :

L	Т	Ρ	С
2	-	2	3



Source: https://youtu.be/chA0QhnV8KQ

### Course Description and Objectives:

This course offers the importance of downstream processing in biotechnology and its problems associated with product purification. The objective of this course is to impart knowledge and skills on different separation, purification, recovery and processing techniques.

#### Course Outcomes:

Upon completion of the course, the student will be able to

- CO1: Understand and explain the bio-separation principles involved in purification of bio-products.
- CO2: Involve suitable unit operations in bioprocess industries.
- CO3: Evaluate concepts selection of membranes and assess the results of protein purification.
- CO4: Design the method for bio-separation of proteins.
- CO5: Understand the designing processes for the recovery and subsequent purification of atarget therapeutic protein.

### SKILLS:

- ✓ Different types of cell lysis.
- ✓ Separate nucleic acids and proteins.
- ✓ Identify compounds using HPLC.

#### UNIT - 1

ROLE OF DOWNSTREAM PROCESSING IN BIOTECHNOLOGY: Role and importance of downstream processing in biotechnological processes; Problems and requirements of bioproduct purification; Economics of downstream processing in biotechnology, characteristics of biological mixtures, process design criteria for various classes of bioproducts.

## UNIT - 2

PHYSICAL SEPARATION METHODS: Separation of intracellular, extra-cellular, heat and photosensitive materials; Cell disruption- chemical, mechanical and enzymatic methods; Physicochemical basis of separation; Physical separation processes- solid and liquid system, flocculation, centrifugation, precipitation, filtration and settling.

#### UNIT - 3

PRODUCT RECOVERY METHODS: Extraction, liquid-liquid extraction, aqueous two-phase extraction, absorption, adsorption and leaching; Membrane-based separations (micro, ultrafiltration, reverse osmosis, dialysis), theory, design and configuration of membrane separation equipment applications.

#### UNIT - 4

PRODUCT PURIFICATION: Chromatographic techniques- paper, TLC, adsorption, ion exchange, gel filtration, affinity chromatographic separation processes, GC, HPLC, FPLC, chromato focusing; Electrophoretic separations- electrophoresis of proteins and nucleic acids, 1D-, 2D gels, types of electrophoretic techniques (capillary and pulse field).

### UNIT - 5

PRODUCT FINISHING AND EMERGING TECHNOLOGIES: Crystallization and drying; Pervaporation, super liquid extraction and foam based separation; Case study with examples for processing of two industrial products (citric acid/penicillin and low volume high value product like recombinant proteins).

# LABORATORY EXPERIMENTS

#### LIST OF EXPERIMENTS:

- 1. Solid separation methods-filtration, sedimentation and centrifugation.
- 2. Protein precipitation methods salt precipitation (PEG, ammonium sulphate).
- 3. Dialysis for desalting the macromolecules.
- 4. Ion-exchange chromatography for separation of proteins based on charge.
- 5. Gel filtration chromatography for separation of proteins based on size.
- 6. Aqueous two-phase extraction for clarification, concentration and partial purification.
- 7. HPLC for separation of polyphenols from plant extracts.
- 8. Product preservative methods -chemical, physical and natural.
- 9. Adsorption process in batch and continuous mode in fermenters.

ACTIVITIES:

- Experiment on Paper chromatography and thin layer chromatography.
- Experiment on cell disruption methods.

 Conduct electrophoresis of nucleic acids and proteins. L-6

L-6

L-6

L-6

L-6

Total hours-30

#### TEXT BOOKS:

- 1. R.O. Jenkins, "Product recovery in bioprocess technology", Butterworth Heinemann Limited, Oxford, 1992.
- 2. B. Sivasankar, "Bioseparations Principles and Techniques", 1<sup>st</sup> edition, PHI Publications, 2009.
- 3. A.K. Scopes, "Protein Purification ", IRL Press, 1993.
- 4. J. Jayaraman, "Laboratory Manual in Biochemistry", 1<sup>st</sup> Edition, New Age International, 1993.

#### REFERENCE BOOKS:

- 1. S.N. Mukhopadhyay, "Process Biotechnology Fundamentals", 2<sup>nd</sup> edition, Viva Books Private Limited, 2005.
- 2. P. F. Stanbury and A. Whitaker, "Principles of Fermentation Technology", 2<sup>nd</sup> edition, Elsevier Publication, 2008.
- 3. K. Wilson and J. Walker, "Principles and Techniques of Practical Biochemistry", 7<sup>th</sup> edition, Cambridge Publication, 2010.
- 4. R. Eisenthal and N.J. Danson, "Enzyme Assays- A Practical Approach", 1<sup>st</sup> edition, IRI Press, Oxford, UK, 1992.