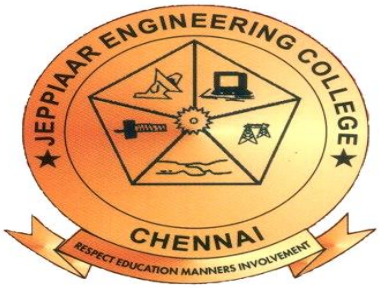
# JEPPIAAR ENGINEERING COLLEGE




## B.TECH – BIOTECHNOLOGY (R- 2013)

## BT6502 BIOPROCESS ENGINEERING

## III YEAR & V SEM

## BATCH: 2016-2020

# QUESTION BANK

## PREPARED BY

**Ms. R. SUGANYA, ASST. PROFESSOR**

## VISION OF THE INSTITUTION

- To build Jeppiaar Engineering College as an institution of academic excellence in technological and management education to become a world class University

## MISSION OF THE INSTITUTION

- To excel in teaching and **learning, research and innovation** by promoting the principles of scientific analysis and creative thinking.
- To participate in the production, **development and dissemination of knowledge** and interact with **national and international communities.**
- To equip students with **values, ethics and life skills n**eeded to enrich their lives and enable them to meaningfully contribute to the **progress of society.**
- To prepare students for **higher studies and lifelong learning,** enrich them with the **practical and entrepreneurial skills** necessary to excel as future professionals and contribute to **Nation's economy**

## PROGRAM OUTCOMES (PO)

| | |
|---|---|
| **PO 1** | **Engineering knowledge**: **Apply** the knowledge of mathematics, science, engineering fundamentals, and an engineering specialization to the solution of complex engineering problems. |
| **PO 2** | **Problem analysis**: **Identify, formulate, review** research literature, and **analyze** complex engineering problems reaching substantiated conclusions using first principles of mathematics, natural sciences, and engineering sciences. |
| **PO 3** | **Design/development of solutions**: **Design solutions** for complex engineering problems and **design** system components or processes that meet the specified needs with appropriate consideration for the public health and safety, and the cultural, societal, and environmental considerations |
| **PO 4** | **Conduct investigations of complex problems**: **Use** research-based knowledge and research methods including **design** of experiments, **analysis** and **interpretation** of data, and **synthesis** of the information to provide valid conclusions. |
| **PO 5** | **Modern tool usage**: **Create**, **select**, and **apply** appropriate techniques, resources, and modern engineering and IT tools including prediction and modeling to complex engineering activities with an understanding of the limitations. |
| **PO 6** | **The engineer and society**: **Apply** reasoning informed by the contextual knowledge to assess societal, health, safety, legal and cultural issues and the consequent responsibilities relevant to the professional engineering practice. |
| **PO 7** | **Environment and sustainability**: **Understand** the impact of the professional engineering solutions in societal and environmental contexts, and **demonstrate** the knowledge of, and need for sustainable development. |
| **PO 8** | **Ethics**: **Apply** ethical principles and commit to professional ethics and responsibilities and norms of the engineering practice. |

| | |
|---|---|
| **PO 9** | **Individual and team work**: **Function effectively** as an individual, and as a member or leader in diverse teams, and in multidisciplinary settings. |
| **PO 10** | **Communication**: **Communicate effectively** on complex engineering activities with the engineering community and with society at large, such as, being able **to comprehend** and **write** effective reports and **design** documentation, **make** effective presentations, and **give** and **receive** clear instructions. |
| **PO 11** | **Project management and finance**: **Demonstrate** knowledge and understanding of the engineering and management principles and **apply** these to one's own work, as a member and leader in a team, to **manage** projects and in multidisciplinary environments. |
| **PO 12** | **Life-long learning**: **Recognize** the need for, and have the preparation and ability to **engage** in independent and life-long learning in the broadest context of technological change. |

## VISION OF THE DEPARTMENT

❖ To pursue excellence in producing bioengineers coupled with research attributes

## MISSION OF THE DEPARTMENT

| | |
|---|---|
| **M1** | To **impart** quality education and **transform** technical knowledge into career opportunities |
| **M2** | To **establish** a bridge between the **program and society** by **fostering** technical education |
| **M3** | To **generate societal conscious technocrats** towards **community development** |
| **M4** | To **facilitate higher studies** and research in order to have an **effective career /** entrepreneurship |

## PROGRAM EDUCATIONAL OBJECTIVES (PEOS)

| | |
|---|---|
| **PEO - 1** | To impart knowledge and produce competent graduates in the field of biotechnology |
| **PEO - 2** | To inculcate professional attributes and ability to integrate engineering issues to broader social contexts. |
| **PEO - 3** | To connect the program and community by fostering technical education. |
| **PEO - 4** | To provide a wide technical exposure to work in an interdisciplinary environment |
| **PEO - 5** | To prepare the students to have a professional career and motivation towards higher education. |

| PROGRAM SPECIFIC OUTCOMES (PSOs) | |
|---|---|
| **PSO 1** | **Professional Skills:** This programme will provide students with a **solid foundation** in the field of Biological Sciences and Chemical engineering enabling them to work on engineering platforms and applications in Biotechnology as per the requirement of Industries, and facilitating the students to pursue higher studies. |
| **PSO 2** | **Problem-solving skills:** This programme will assist the students to **acquire fundamental** and **problem solving knowledge** on subjects relevant to Biotechnology thereby encouraging them to **understand** emerging and advanced concepts in modern biology. |
| **PSO 3** | **Successful Career and Entrepreneurship:** Graduates of the program will have a **strong successful career** and **entrepreneurial ability** with the blend of inputs from basic science, engineering and technology, thereby enabling them to **translate the technology and tools** in various industries and/or institutes. |

## BIO PROCESS ENGINEERING/BT6502

**OBJECTIVES:**

- To provide the students with the basics of bioreactor engineering.
- To develop bioengineering skills for the production of biochemical product using integrated biochemical processes.

**UNIT I OPERATIONAL MODES OF BIOREACTORS 9**

Fed batch cultivation, Cell recycle cultivation, Cell recycle cultivation in waste water treatment, two stage cultivation. Packed bed reactor, airlift reactor, fluidized bed reactor and bubble column reactor.

**UNIT II BIOREACTOR SCALE – UP 8**

Regime analysis of bioreactor processes, oxygen mass transfer in bioreactors –microbial oxygen demands; methods for the determination of mass transfer coefficients;mass transfer correlations. Scale up criteria for bioreactors based on oxygen transfer, power consumption and impeller tip speed.

**UNIT III BIOREACTOR CONSIDERATION IN ENZYME SYSTEMS 8**

Analysis of film and pore diffusion effects on kinetics of immobilized enzyme reactions;formulation of dimensionless groups and calculation of effectiveness factors. Design of immobilized enzyme reactors – packed bed, fluidized bed and membrane reactors.

**UNIT IV MODELLING AND SIMULATION OF BIOPROCESSES 11**

Study of structured models for analysis of various bioprocess – compartmental models,models of cellular energetics and metabolism, single cell models, plasmid replication and plasmid stability model. Dynamic simulation of batch, fed batch, steady and transient culture metabolism.

**UNIT V RECOMBINANT CELL CULTIVATION 9**

Different host vector system for recombinant cell cultivation strategies and advantages. E.coli, yeast Pichia pastoris/ Saccharomyces cereviseae, Animal cell cultivation, plant cell cultivation, Insect cell cultivation. High cell density cultivation, process strategies, reactor considerations in the above system.

**TOTAL : 45 PERIODS**

**OUTCOMES:**

Upon completion of Bioprocess Engineering course graduates will be able to

- Select appropriate bioreactor configurations and operation modes based upon the nature of bioproducts and cell lines and other process criteria.

Apply modeling and simulation of bioprocesses so as to reduce costs and to enhance the quality of products and systems. Plan a research career or to work in the biotechnology industry with strong
foundation about bioreactor design and scale-up.
Integrate research lab and Industry; identify problems and seek practical solutions for large scale implementation of Biotechnology.

**TEXT BOOKS:**

1. Jens Nielson, John Villadsen and Gunnar Liden, "Bioreaction engineering principles",2nd Edition, Kulwer Academic, 2002
2. Harvey W. Blanch, Douglas S. Clark, Biochemical Engineering, Marcel Dekker, Inc

**REFERENCES:**

1. Anton Moser, "Bioprocess Technology: Kinetics and Reactors", , Springer Verlag 2011.
2. Tapobrate Panda, "Bioreactors: Analysis and Design", Tata McGraw Hill, 2011
3. Shijie Liu "Bioprocess Engineering" Elsevier, 2013
4. Atkinson, B, Mavituna, F, "Biochemical Engineering and Biotechnology Handbook" Macmillan Publishers Ltd, New York, 1992.
5. James E. Bailey & David F. Ollis, "Biochemical Engineering Fundamentals", McGraw Hill.
6. James M. Lee, "Biochemical Engineering", PHI, USA 2002.
7. E. Heinzle, A. Biwer and C.Cooney " Development of Sustainable Bioprocesses" John Wiley & sons, 2006.

| COURSE OUTCOMES (CO) | |
|---|---|
| **BT6502: BIOPROCESS ENGINEERING** | |
| **C302.1** | The course graduates will be able to Select appropriate bioreactor configurations and operation modes based upon the nature of bioproducts and cell lines and other process criteria |
| **C302.2** | The students will be able to Apply modeling and simulation of bioprocesses so as to reduce costs and to enhance the quality of products and systems |
| **C302.3** | The students will be able to Plan a research career or to work in the biotechnology industry with strong foundation about bioreactor design and scale-up. |
| **C302.4** | The students will be able to Integrate research lab and Industry; identify problems and seek practical solutions for large scale implementation of Biotechnology |
| **C302.5** | The students will be able to understand the design of plant cultivation reactors, animal cell cultivation reactors and recombination techniques. |

| S. No. | Title | Reference Book | Page No. |
|---|---|---|---|
| **UNIT I OPERATIONAL MODES OF BIOREACTORS (9)** | | | |
| 1. | Fed batch cultivation | James E. Bailey & David F. Ollis, "Biochemical Engineering Fundamentals", McGraw-Hill. | 600-602 |
| 2. | Cell recycle cultivation | James E. Bailey & David F. Ollis, "Biochemical Engineering Fundamentals", McGraw-Hill. | 602-603 |
| 3. | Cell recycle cultivation in waste water treatment | James E. Bailey & David F. Ollis, "Biochemical Engineering Fundamentals", McGraw-Hill. | 603-605 |
| 4. | Two stage cultivation. | James E. Bailey & David F. Ollis, "Biochemical Engineering Fundamentals", McGraw-Hill. | 605-609 |
| 5. | Packed bed reactor | James E. Bailey & David F. Ollis, "Biochemical Engineering Fundamentals", McGraw-Hill. | 609-610 |
| 6. | Airlift reactor | James E. Bailey & David F. Ollis, "Biochemical Engineering Fundamentals", McGraw-Hill. | 641-645 |
| 7. | Fluidized bed reactor | James E. Bailey & David F. Ollis, "Biochemical Engineering Fundamentals", McGraw-Hill. | 614-617 |
| 8. | Bubble column reactor. | James E. Bailey & David F. Ollis, "Biochemical Engineering Fundamentals", McGraw-Hill. | 610-614 |
| **UNIT II BIOREACTOR SCALE – UP (8)** | | | |
| 9. | Regime analysis of bioreactor processes | James M. Lee, "Biochemical Engineering", PHI, USA. | 240-247 |
| 10. | Oxygen mass transfer in bioreactors | James M. Lee, "Biochemical Engineering", PHI, USA. | 241-242 |
| 11. | Microbial oxygen demands; | James M. Lee, "Biochemical Engineering", PHI, USA. | 261-262 |
| 12. | Methods for the determination of mass transfer coefficients; | James M. Lee, "Biochemical Engineering", PHI, USA. | 264-265 |
| 13. | Mass transfer correlations. | James M. Lee, "Biochemical Engineering", PHI, USA. | 248-250 |
| 14. | Scale up criteria for bioreactors based on oxygen transfer, power consumption and impeller tip speed. | James M. Lee, "Biochemical Engineering", PHI, USA. | 272-274 |
| **UNIT III BIOREACTOR CONSIDERATION IN ENZYME SYSTEMS (8)** | | | |
| 15. | Analysis of film and pore diffusion effects on kinetics of immobilized enzyme reactions; | James M. Lee, "Biochemical Engineering", PHI, USA. | 202, 216 208 |
| 16. | Formulation of dimensionless groups and calculation of effectiveness factors. | Harvey W. Blanch, Douglas S. Clark, "Biochemical Engineering", Marcel Decker Inc. | 128, 122 |
| 17. | Design of immobilized enzyme reactors – packed bed | James E. Bailey & David F. Ollis, "Biochemical | 609-611 |

| | | | |
|---|---|---|---|
| | | Engineering Fundamentals", McGraw-Hill. | |
| 18. | Fluidized bed | James E. Bailey & David F. Ollis, "Biochemical Engineering Fundamentals", McGraw-Hill. | 614-617 |
| 19. | Membrane reactors. | James E. Bailey & David F. Ollis, "Biochemical Engineering Fundamentals", McGraw-Hill. | 610-614 |
| **UNIT IV MODELLING AND SIMULATION OF BIOPROCESSES (11)** | | | |
| 20. | Study of structured models for analysis of various bioprocess - compartmental models | James M. Lee, "Biochemical Engineering", PHI, USA. | 174-176 |
| 21. | Models of cellular energetics and metabolism, | Harvey W. Blanch, Douglas S. Clark, "Biochemical Engineering", Marcel Decker Inc. | 230-244 |
| 22. | Single cell models, | Harvey W. Blanch, Douglas S. Clark, "Biochemical Engineering", Marcel Decker Inc. | 244-246 |
| 23. | Plasmid replication | Harvey W. Blanch, Douglas S. Clark, "Biochemical Engineering", Marcel Decker Inc. | 251-257 |
| 24. | Plasmid stability model. | Harvey W. Blanch, Douglas S. Clark, "Biochemical Engineering", Marcel Decker Inc. | 251-257 |
| 25. | Dynamic simulation of batch, | Harvey W. Blanch, Douglas S. Clark, "Biochemical Engineering", Marcel Decker Inc. | 277-280 |
| 26. | Fed batch, | Harvey W. Blanch, Douglas S. Clark, "Biochemical Engineering", Marcel Decker Inc. | 305-308 |
| 27. | Steady and transient culture metabolism. | Harvey W. Blanch, Douglas S. Clark, "Biochemical Engineering", Marcel Decker Inc. | 280-296<br>309-322 |
| **UNIT V RECOMBINANT CELL CULTIVATION (9)** | | | |
| 28. | Different host vector system for recombinant cell cultivation strategies and advantages | James E. Bailey & David F. Ollis, "Biochemical Engineering Fundamentals", McGraw-Hill. | 257-259 |
| 29. | E.coli, yeast Pichia pastoris/ Saccharomyces cereviseae, | James E. Bailey & David F. Ollis, "Biochemical Engineering Fundamentals", McGraw-Hill. | 259-261 |
| 30. | Animal cell cultivation, | James E. Bailey & David F. Ollis, "Biochemical Engineering Fundamentals", McGraw-Hill. | 261-265 |
| 31. | Plant cell cultivation, | James E. Bailey & David F. Ollis, "Biochemical Engineering Fundamentals", McGraw-Hill. | 265-267 |
| 32. | Insect cell cultivation. | James E. Bailey & David F. Ollis, "Biochemical Engineering Fundamentals", McGraw-Hill. | 267-269 |

| | | | |
|---|---|---|---|
| 33. | High cell density cultivation, process strategies, reactor considerations in the above system. | James E. Bailey & David F. Ollis, "Biochemical Engineering Fundamentals", McGraw-Hill. | 269-271 |

**B.E./B.Tech. DEGREE EXAMINATION, NOVEMBER/DECEMBER 2017**
**FIFTH SEMESTER**
**BIOTECHNOLOGY**
**BT6502-BIOPROCESS ENGINEERING**
**(Regulations 2013)**

**Time: Three Hours** **Maximum: 100 Marks**

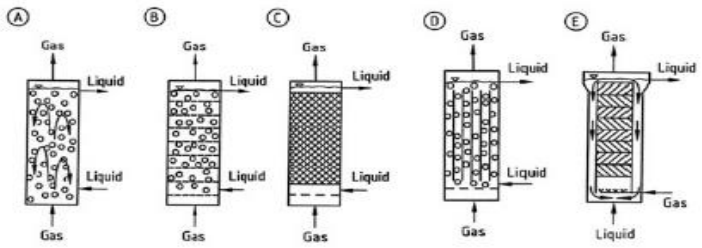
**Answer ALL questions**

**PART A (10X2=20)**

1. Give the advantages of Fed Batch culture.
   (i) Production of highest possible amount of PHA by balanced cell growth and PHA accumulation.
   (ii) No nitrogen limitation during the activegrowthphase, but it is introduced later to enhance PHA production.
   (iii) Results: maximum specific growth rate achieved compared to the max specific growth rate in batch cultivation.

2. Draw and label the parts of bubble column reactor.

## Type of Bubble Columns




A) Simple bubble column; B) Cascade bubble column with sieve trays
B) C) Packed bubble column; D) Multishaft bubble column;
C) E) Bubble column with static mixers

3. Define microbial oxygen demand.
   Biochemical **oxygen demand** is a measure of the quantity of **oxygen** used by microorganisms (e.g., aerobic bacteria) in the **oxidation** of organic matter.
4. What happens to the value of KLa when there is an increase in temperature?
   KLa increases when there is an increase in temperature. The solubility of oxygen in the liquid phase of a bioreactor was changed by a ramp change of temperature, and $k_La$ was determined from the resulting return to equilibrium of dissolved oxygen activity.
5. State down the different methods available for immobilizing bio molecules.
   (i) Self Assembly
   (ii) Self Modification
   (iii) Surface Modification
   (iv) Photochemical Immobilization
   (v) Polymer Chemistry
6. What is effectiveness factor?
   The effectiveness factor is defined as the ratio of the reaction rate actually observed to the reaction rate calculated if the surface reactant concentration persisted throughout the interior of the particle, Le., no reactant concentration gradient within the particle.
7. What is epigenetic system and metabolic system in a saturated model?
   Epigenetics is the study of heritable changes in gene function that do not involve changes in the DNA sequence. The Greek prefix *epi-* (ἐπι- "over, outside of, around") in *epigenetics* implies features that are "on top of" or "in addition to" the traditional genetic basis for inheritance. Epigenetics most often denotes changes in a chromosome that affect gene activity and expression, but can also be used to describe any heritable phenotypic change that does not derive from a modification of the genome, such as prions.
   Catabolism is the set of metabolic processes that break down large molecules. These include breaking down and oxidizing food molecules. The purpose of the catabolic reactions is to provide the energy and components needed by anabolic reactions which build molecules.

8. What is idiophase?

   The idiophase is the phase in the growth of a culture during which secondary metabolites are produced.

9. What is elicitor?

   Elicitors in plant biology are extrinsic or foreign molecules often associated with plant pests, diseases or synergistic organisms. Elicitor molecules can attach to special receptor proteins located on plant cell membranes.

10. What is the difference between transduction and transformation when discussing gene transfer to bacteria?

**Difference between transformation and transduction**

| | Transformation | Transduction |
|---|---|---|
| 1. | Transfer of genetic material takes place from donor to recipient bacterium through the liquid medium. | Transfer of genetic material takes place from donor to recipient bacterium through a Bacteriophage. |
| 2. | The enzyme deoxyribonuclease can completely check the process. | The deoxyribonuclease has no effect on trans- |

**PART B** **(5X13=65)**

11. (a) Explain in detail the design and operation of

    (i) Packed Bed Reactor

    (ii) Airlift Reactor

    Ans: Refer Reference Book 5 – Pg. No: 609, 610, 641

    (OR)

    (b) Explain in detail the kinetics of cell cycle cultivation.

    Ans: Refer Reference Book 5 – Pg. No: 603-605

12. (a) (i) Explain the static method for the determination of mass transfer coefficient in aerated bioreactor.

    Ans: Refer Reference Book 6 – Pg. No: 264-265

    (ii) Calculate the Reynolds number and power required to agitate a 10,000 litre tank filled with water. The diameter of the tank is 2.0 m and is agitated at 100 rpm by a 6-blade turbine type agitator. The agitator is half the tank diameter. (Np=4).

    (OR)

    (b) Explain in detail the regime analysis of a bioreactor process.

    Ans: Refer Reference Book 5 – Pg. No: 240-244

13. (a) Explain the effect of inhibitors, temperature and pH on immobilized enzyme catalytic activity.

    Ans: Refer Reference Book 6 – Pg. No: 209-211

    (OR)

(b) Explain the characteristics of the following bioreactors

(i) Membrane Reactors

(ii) Fluidized Bed Reactor.

Ans: Refer Reference Book 5 – Pg. No: 603-605

14. (a) Explain in detail the compartment model of William to illustrate the properties of cell growth kinetics.

Ans: Refer Reference Book 5 – Pg. No: 174-176

(OR)

(b) Explain the kinetics of plasmid replication using a generalized model.

Ans: Refer Text Book 2 – Pg. No: 251-254

15. (a) Explain the strategies for High-cell density cultivation of E.coli.

Ans: Refer text Book 2 – Pg. No: 277-280

(OR)

(b) Explain in detail the guidelines for selecting a host vector system.

Ans: Refer Reference Book 6 – Pg. No: 257-259

**PART C** **(1X15=15)**

16. (a) Explain in detail with example, how insect cells are used for protein production.

Ans: Refer Reference Book 6 – Pg. No: 267-269

(OR)

(b) Write notes on the medical and analytical applications of immobilized enzymes.

Ans: Refer Reference Book 5 – Pg. No: 208,216

## UNIT 1 OPERATIONAL MODES OF BIOREACTORS
## TWO MARKS

**1. Define residence time distribution.(APR/MAY 2017)**

Residence time distribution (RTD) describes the distribution of times required for fluid elements to pass through a continuous-flow system. RTD function and associated flow models have been widely used to characterize hydrodynamic behavior of a variety of systems encountered in agricultural, food, and biological engineering.

**2. Define pasteurization. (APR/MAY 2017, APR/MAY 2016)**

The treatment of foods or beverages with mild heat, irradiation, or chemical agents to improve keeping quality or to inactivate disease-causing microorganisms. Originally, Louis Pasteur observed that spoilage of wine and beer could be prevented by heating them a few minutes at 122–140°F (50–60°C).

**3. Define sterilization(APR/MAY 2016, APR/MAY 2015)**

A form of monetary action in which a central bank or federal reserve attempts to insulate itself from the foreign exchange market to counteract the effects of a changing monetary base. The sterilization process is used to manipulate the value of one domestic currency relative to another, and is initiated in the forex market.

**4. What is the role of draft tube? (APR/MAY 2015)**
Gas hold-up and mass transfer were examined in a column with and without a draft tube. It was found that the introduction of a draft tube increases the gas hold-up but decreases the volumetric mass transfer coefficient in Newtonian fluid systems.

**5. Write the applications of plug flow reactor.**
- Large-scale reactions
- Fast reactions
- Homogeneous or heterogeneous reactions
- Continuous production
- High-temperature reactions

**6. Write the disadvantages of plug flow reactors. (APR/MAY 2014, APR/MAY 2013)**
Disadvantages of plug flow reactors are that temperatures are hard to control and can result in undesirable temperature gradients. PFR maintenance is also more expensive than CSTR maintenance.

**7. Write the advantages of plug flow reactors(APR/MAY 2014)**
Plug flow reactors have a high volumetric unit conversion, run for long periods of time without maintenance, and the heat transfer rate can be optimized by using more, thinner tubes or fewer, thicker tubes in parallel.

**8. Define Polymerase chain reaction? (APR/MAY 2013)**
The polymerase chain reaction (PCR), a common laboratory technique, employs artificial synthesis in a cyclic manner to rapidly and specifically amplify a target DNA fragment from a pool of DNA.

**9. Define fluidization.**
Fluidization is a process similar to liquefaction whereby a granular material is converted from a static solid-like state to a dynamic fluid-like state. This process occurs when a fluid (liquid or gas) is passed up through the granular material.

**10. Define batch cultivation**
A technique used to grow microorganisms or cells. A limited supply of nutrients for growth is provided; when these are used up, or some other factor becomes limiting, the culture declines. Cells, or products that the organisms have made, can then be harvested from the culture.

**11. Define fed batch cultivation**
Fed-batch culture is, in the broadest sense, defined as an operational technique in biotechnological processes where one or more nutrients (substrates) are fed (supplied) to the bioreactor during cultivation and in which the product(s) remain in the bioreactor until the end of the run.

**12. Define cell recycle cultivation**

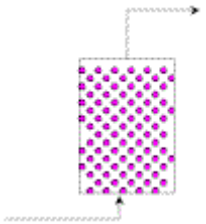
Continuous cultivation with cell recycling is used to obtain high cell concentration. Knowing that secondary metabolite formation rate is proportional to cell concentration, continuous cultivations with cell recycling is an alternative to increase the productivity

**13. What are the two stages of cultivation?**

Autotrophic
Mixotrophic

**14. Define packed bed reactor**

In packed bed reactors, cells are immobilized on large particles. These particles do not move with the liquid. Packed bed reactors are simple to construct and operate but can suffer from blockages and from poor oxygen transfer.



**15. Define airlift reactor**

Air-lift bioreactors are similar to bubble column reactors, but differ by the fact that they contain a draft tube. The draft tube is always an inner tube (this kind of air-lift bioreactor is called "air-lift bioreactor with an internal loop) or an external tube (this kind of air-lift bioreactor is called "air-lift bioreactor with an external loop) which improves circulation and oxygen transfer and equalizes shear forces in the reactor. The figure below illustrates the basic structure of an air-lift bioreactor with an internal loop

**16. Define fluidized bed reactor**

A fluidized bed reactor (FBR) is a type of reactordevice that can be used to carry out a variety of multiphase chemical reactions.

**17. Define bubble column reactor**

A bubble column reactor is an apparatus used to generate and control gas-liquid chemical reactions. It consists of a vertically-arranged cylindrical column filled with liquid, at the bottom of which gas is inserted

**18. Advantages of packed bed reactor**

1. High conversion per unit mass of catalyst
2. Low operating cost
3. Continuous operation

**19. Advantages of airlift reactor**

1. Good mixing

2. Good uniformity of temperature
3. Catalyst can be continuously regenerated with the use of an auxiliary loop

**20. Advantages of fluidized bed reactor**

- **Uniform Particle Mixing:** Due to the intrinsic fluid-like behavior of the solid material, fluidized beds do not experience poor mixing as in packed beds. This complete mixing allows for a uniform product that can often be hard to achieve in other reactor designs. The elimination of radial and axial concentration gradients also allows for better fluid-solid contact, which is essential for reaction efficiency and quality.
- **Uniform Temperature Gradients:** Many chemical reactions require the addition or removal of heat. Local hot or cold spots within the reaction bed, often a problem in packed beds, are avoided in a fluidized situation such as an FBR. In other reactor types, these local temperature differences, especially hotspots, can result in product degradation. Thus FBRs are well suited to exothermic reactions. Researchers have also learned that the bed-to-surface heat transfer coefficients for FBRs are high.
- **Ability to Operate Reactor in Continuous State:** The fluidized bed nature of these reactors allows for the ability to continuously withdraw product and introduce new reactants into the reaction vessel. Operating at a continuous process state allows manufacturers to produce their various products more efficiently due to the removal of startup conditions in batch processes.

**21. Advantages of bubble column reactor**
Provide several advantages during operation and maintenance such as high heat and mass transfer rates, compactness and low operating and maintenance costs.

**22. Disadvantages of packed bed reactor**
1. Undesired thermal gradients may exist
2. Poor temperature control
3. Channeling may occur
4. Unit may be difficult to service and clean

**23. Disadvantages of airlift reactor**
1. Bed-fluid mechanics not well known
2. Severe agitation can result in catalyst destruction and dust formation
3. Uncertain scale-up

**24. Disadvantages of bubble column reactor**

- considerable degree of back mixing in both the liquid and the gas phase
- short gas phase residence time
- higher pressure drop with respect to packed columns
- rapid decreasing of interfacial area above values of the aspect ratio greater than, say 12, due to the increased rate of coalescence

**25. Disadvantages of fluidized bed reactor**

- **Increased Reactor Vessel Size:** Because of the expansion of the bed materials in the reactor, a larger vessel is often required than that for a packed bed reactor. This larger vessel means that more must be spent on initial capital costs.
- **Pumping Requirements and Pressure Drop:** The requirement for the fluid to suspend the solid material necessitates that a higher fluid velocity is attained in the reactor. In order to achieve this, more pumping power and thus higher energy costs are needed. In addition, the pressure drop associated with deep beds also requires additional pumping power.
- **Particle Entrainment:** The high gas velocities present in this style of reactor often result in fine particles becoming entrained in the fluid. These captured particles are then carried out of the reactor with the fluid, where they must be separated. This can be a very difficult and expensive problem to address depending on the design and function of the reactor. This may often continue to be a problem even with other entrainment reducing technologies.
- **Lack of Current Understanding:** Current understanding of the actual behavior of the materials in a fluidized bed is rather limited. It is very difficult to predict and calculate the complex mass and heat flows within the bed. Due to this lack of understanding, a pilot plant for new processes is required. Even with pilot plants, the scale-up can be very difficult and may not reflect what was experienced in the pilot trial.
- **Erosion of Internal Components:** The fluid-like behavior of the fine solid particles within the bed eventually results in the wear of the reactor vessel. This can require expensive maintenance and upkeep for the reaction vessel and pipes.
- **Pressure Loss Scenarios:** If fluidization pressure is suddenly lost, the surface area of the bed may be suddenly reduced. This can either be an inconvenience (e.g. making bed restart difficult), or may have more serious implications, such as runaway reactions (e.g. for exothermic reactions in which heat transfer is suddenly restricted).

**26. What are the reasons for non-ideality in a fluidized bed reactor? MAY/JUNE 2012**

Mass transfer,
Partitioning.

**27. Define Recycle stream**
A process stream that returns material from downstream of a process unit back to the process unit.

**28. Define bypass stream**
It is a stream that skips one or more stages of the process and goes directly to another down stream stage.

**29. Define Purge Stream**
Purge stream is a bled off to remove an accumulation of inerts or unwanted material that might otherwise buildup in the recycle stream.

**30. Explain external biomass feedback.**
A cell separator such as a centrifuge or settling tank is used to concentrate biomass leaving the reactor.Once biomass removed cell is recycled.

## PART B

1. **Explain packed bed reactor with applications and write down the design equations. APR/MAY 2016,APR/MAY 2011,MAY/JUNE 2014**
   Refer:James E. Bailey & David F. Ollis, "Biochemical Engineering Fundamentals", McGraw-Hill.(Page No-609-610)

2. **Explain fluidized bed reactor with applications and write down the design equations. APR/MAY 2011**
   Refer:James E. Bailey & David F. Ollis, "Biochemical Engineering Fundamentals", McGraw-Hill.(Page No-614-617)

3. **Explain in detail Airlift reactor, Bubble column reactor with their applications and write down the design equations. APR/MAY 2016,APR/MAY 2011,MAY/JUNE 2013,2014**
   Refer:James E. Bailey & David F. Ollis, "Biochemical Engineering Fundamentals", McGraw-Hill.(Page No-641)

4. **Describe in detail Fed batch cultivation with the design equations. APR/MAY 2008**
   Refer:James E. Bailey & David F. Ollis, "Biochemical Engineering Fundamentals", McGraw-Hill.(Page No-600)

5. **Explain in detail cell recycle cultivation.APR/MAY 2008**
   Refer:James E. Bailey & David F. Ollis, "Biochemical Engineering Fundamentals", McGraw-Hill.(Page No-602)

6. **How the cell recycle cultivation is used in waste water treatment and explain the stages of cell cultivation APR/MAY 2009**
   Refer:James E. Bailey & David F. Ollis, "Biochemical Engineering Fundamentals", McGraw-Hill.(Page No-603)

## UNIT 2 BIOREACTOR SCALE UP
## TWO MARKS

**1. Define mass transfer coefficient.**
Determinations of volumetric mass transfer coefficient were conducted in a three-phase internal-loop airlift reactor with an enlarged degassing zone. The effectof parameters such as the airflow rate (riser superficial gas velocities between 0.01 and 0.5 m/s), solids loading (up to 30%, v/v), solids density (1023 and 1048 kg/m3) and the liquid-phase properties on $kLa$ was studied.

**2. What is scale up? APR/MAY 2016**
Generally, fermenters maintain a height to diameter ratio of 2 to 1. If The height to diameter ratio remains constant, and then the surface to volume ratio decreases during scale-up.

**3. What is scale down?**
Although scale up models and the use of characteristic time analysis are potentially attractive, a more immediate approach to the rational scaling of reactors is scale down.

**4. What are the effects of scale up?**
(i) Decreases the relative contribution of surface aeration to oxygen supply.
(ii) Removal of dissolved carbon dioxide removal.

**5. Write few mass transfer correlations. APR/MAY 2016**
Correlations for the volumetric mass transfer coefficient with the riser superficial gas velocity and solids loading were determined for the two solids density and the two liquid-phases. A good agreement between experimental data and the calculated values was obtained.

**6. Write few bioreactors.**
Ideal (stirred) Batch reactor Ideal tank reactor, also named Continuous Stirred Tank Reactor (CSTR) Ideal Plug Flow Reactor (PFR)

**7. Write the features of scale up.**
Maintenance of constant power to volume ratios, constant kla, constant tip speed, a combination of mixing time and renolds number and the maintenance of a constant substrate or product level (usually dissolved oxygen concentration).

**8. Write the difference between scale up and scale down.**
**Scale up**
Generally, fermenters maintain a height to diameter ratio of 2 to 1. IfThe height to diameter ratio remains constant, then the surface to volume ratio decreases during scale-up.
**Scale down**

Although scale up models and the use of characteristic time analysis are potentially attractive, a more immediate approach to the rational scaling of reactors is scale down.

**9. Write the advantages of fed batch mode of fermentation. APR/MAY 2015**

- production of high cell densities due to extension of working time (particularly important in the production of growth-associated products)
- controlled conditions in the provision of substrates during the fermentation, particularly regarding the concentration of specific substrates as for ex. the carbon source
- control over the production of by-products or catabolite repression effects due to limited provision of substrates solely required for product formation

**10. What should be the value of impeller Reynolds number for laminar and turbulent regime in a bioreactor? MAY/JUNE 2012**

For ratios that are above 10,000 we know that the mixing regime is fully turbulent. For numbers between 1000 and 10,000 we know that the mixing environment is transitional. For numbers or ratios below 1000, the mixing regime is partially laminar or fully laminar.

**11. List out the criteria to be followed for scaling up of a bioreactor. MAY/JUNE 2012**

A. Constant power input per unit volume (P/V = constant).

B. Constant $K_La$

C. Constant mixing quality

D. Constant momentum factor (MF = ND. NWL (D – W)) = constant)

E. Similar drop size distribution ($d_s$ = constant)

F. Constant impeller tip speed ($\pi ND_i$ = constant)

G. Constant mixing rate number = $(N/K)(D_i/D_t)^{\alpha}$ constant

**12. What is meant by flooding in bioreactor? APR/MAY 2011**

When air flow dominates the flow pattern flooding is occur in the bioreactor. Flooding of fermenter occurs due to inappropriate combination of air flow rate and speed of agitation.

**13. What are the different types of impeller used in bioreactors? APR/MAY 2011**

**1) High Head Closed Channel Impeller** – high-efficiency design for pumping water and other liquids at higher head pressures
**2) Vortex Impeller** – Used for pumping stringy solids and debris-laden liquids
**3) Centrifugal Screw Impeller** – Used for pumping oils and other viscous liquids
**4) Propeller** – Used for pumping high volumes of water at low heads
**5) Shredder Impeller** – Used for chopping solids to smaller pieces when they enter the pump
**6) Closed Channel Impeller** – Used for pumping sewage and wastewater
**7) Mixed Flow Impeller** – Used for high volume water pumping at low to medium heads
**8) Semi-Open Impeller** – Used for trash and debris laden liquids
**9) Hardened Sand/Slurry Impeller** – Used for pumping abrasive liquids

**14. Write the equation for KLa. MAY/JUNE 2013**

The increase in dissolved oxygen concentration is given by –

$dCL / dt = KLa(C^*-CL)$ (ii)

Taking logarithm after Integration of equation (ii) we have

$\ln(C^*-CL) = - K_La.t$

**15. How will you calculate the power number?**

$N_P$ = Power Number = $P/(n^3D_a^5r)$

**16. What are the factors affecting the value of KLa?**

- A number of factors have been demonstrated to affect the *KLa* value. Such factors include
  - the air-flow rate employed in vessels,
  - the degree of agitation inside vessels ,
  - the rheological properties of the culture broth and
  - the presence of antifoam agents.

**17. Define oxygen mass transfer in bioreactors.**

- The majority of fermentation processes are aerobic
- Therefore oxygen is an important requirement in aerobic processes like

$$C_6H_{12}O_6 + 6O_2 = 6H_2O + 6CO_2$$

- Therefore, 192 grams of oxygen are required for the complete oxidation of 180 grams of glucose.

**18. Define microbial oxygen demands.**

Definition. Standard method for indirect measurement of the amount of organic pollution (that can be oxidized biologically) in a sample of water. BOD test procedure is based on the activities of bacteria and other aerobic microorganisms (microbes), which feed on organic matter in presence of **oxygen**.

**19. List out the methods for the determination of mass transfer coefficients.**

- Static
- Dynamic
- Sulphite oxidation

**20. Define mass transfer correlations.**

In engineering, the mass transfer coefficient is a diffusion rate constant that relates the mass transfer rate, mass transfer area, and concentration change as driving force: Where: $k_c$ is the mass transfer coefficient [mol/(s. · $m^2$)/(mol/$m^3$)], or m/s.

**21. Scale up criteria for bioreactors based on oxygen transfer.**

$$Q/V\alpha ND_i^3/Di^3=N$$

**22. Scale up criteria for bioreactors based on power consumption**

$$P/V\alpha N^3 D_i^5/Di^3=N^3Di^2$$

**23. Scale up criteria for bioreactors based on impeller tip speed.**

$$ND_i$$

**24. What are the effects on scale up?**

The objective of a successful scale up is a homogeneous environment (temperature, pH and $pO_2$, nutrients, metabolites, cell density, etc.). This is achieved using optimisations based on:

- Agitation based parameters (mixing times, power input, tip speed)
- Gas based parameters (gassing rates)
- Heat transfer

**25. write down the benefit of scale down.**

- Production of data at much smaller effort and time
- Improved Safety ( Smaller is safer )
- Improve the knowledge of Process Characteristics

**26. Define dilution rate?**

At steady state the growth rate (μ) of the micro-organism is equal to the dilution rate (D). The dilution rate is defined as the rate of flow of medium over the volume of culture in the bioreactor.

**27. Define Maximal growth rate?**

Each microorganism has a maximal growth rate (μMAX) (the rate of growth observed if none of the nutrients are limiting). If a dilution rate is chosen that is higher than the maximal growth rate, the culture will not be able to sustain itself in the bioreactor, and will wash out.

**28. What are the types of bioreactors?**

A bioreactor may be classified as batch, fed batch or continuous (e.g. continuous stirredtank reactor model). An example of a continuous bioreactor is the chemostat

**29. Write the advantages of fed batch mode of fermentation.**

 production of high cell densities due to extension of working time (particularly important in the production of growth-associated products)

 controlled conditions in the provision of substrates during the fermentation, particularly regarding the concentration of specific substrates as for ex. the carbon source

 control over the production of by-products or catabolite repression effects due to limited provision of substrates solely required for product formation

**30.Mension theReuse potential of MBR effluent**

- The UMBR system can provide Good Quality effluent that is completely acceptable for reuse.
- The reclaimed water can be directly reused for municipal watering, toilet flushing and car washing. After the softening treatment, the reclaimed water can be used as a cooling water supply or processing water.

- Therefore, lots of urban waste water can be effectively harnessed, and moreover, large quantities of water can be saved. As a result, the water industry would move towards a more sustainable future.

## PART B

1. **Explain the resistance involved in transport of oxygen from a bubble to biochemical reaction site. Explain clearly the assumptions made and explain the importance of oxygen mass transfer determination for aerobic fermentation with suitable examples. MAY/JUNE 2012**
   Refer:James M. Lee, "Biochemical Engineering", PHI, USA.(Page No-240-247)

2. **Discuss the methods used for finding kLa in bioreactors. APR/MAY 2011,MAY/JUNE 2013**
   Refer:James M. Lee, "Biochemical Engineering", PHI, USA.(Page No-264-265)

3. **Scale up criteria for bioreactors based on oxygen transfer, power consumption and impeller tip speed. APR/MAY 2011**
   Refer:James M. Lee, "Biochemical Engineering", PHI, USA.(Page No-272-274)
4. **Derive the expression for power requirement in an aerated bioreactor and in an ungassed bioreactors. MAY/JUNE 2012**
   Refer:James M. Lee, "Biochemical Engineering", PHI, USA.(Page No-274)

5. **What are the various factors which were followed while scaling up process in the bioreactor? And comment on its significance. MAY/JUNE 2013**
   Refer:James M. Lee, "Biochemical Engineering", PHI, USA.(Page No-255)

6. **Problem based on finding kLa in bioreactors. APR/MAY 2015, NOV/DEC 2015**
   Refer:James M. Lee, "Biochemical Engineering", PHI, USA.(Page No-267)

## UNIT 3 BIOREACTOR CONSIDERATION IN ENZYME SYSTEMS
## TWO MARKS

**1. Define metabolism. APR/MAY 2016**

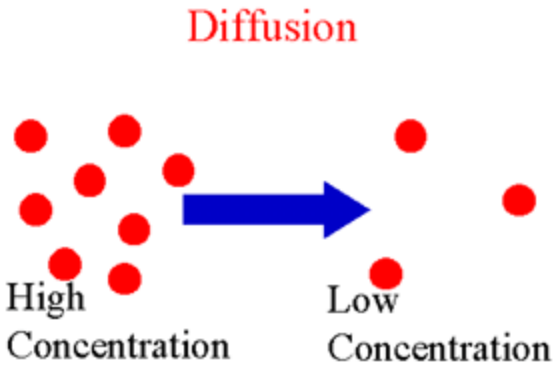
The term metabolism refers to all of the chemical reactions by which complex molecules taken into an organism are broken down to produce energy and by which energy is used to build up complex molecules.

**2. What is PFR? APR/MAY 2016**

The plug flow reactor (PFR) model is used to describe chemical reactions in continuous, flowing systems. The PFR model is used to predict the behavior of chemical reactors, so that key reactor

variables, such as the dimensions of the reactor, can be estimated. PFRs are also sometimes called as Continuous Tubular Reactors (CTRs).

**3.Define diffusion.**




Diffusion is the movement of molecules from a high concentration to a low concentration.
Many molecules diffuse across cell membranes.

**4. Define kinetics. APR/MAY 2015**

That part of classical mechanics which deals with the relation between the motions of material bodies and the forces acting upon them. It is synonymous with dynamics of material bodies.

**5. Define enzyme.**

Enzymes are biological catalysts, or chemicals that speed up the rate of reaction between substances without themselves being consumed in the reaction.

**6. What is dimensionless number?**

A ratio of various physical properties (such as density or heat capacity) and conditions (such as flow rate or weight) of such nature that the resulting number has no defining units of weight, rate, and so on. Also known as nondimensional parameter.

**7. What is Damkohler numbers? APR/MAY 2015**

The Damköhler numbers (Da) are dimensionless numbers used in chemical engineering to relate chemical reaction timescale to other phenomena occurring in a system.

**8. What is Data acquisition?**

Data acquisition is the sampling of the real world to generate data that can be manipulated by a computer. Sometimes abbreviated DAQ or DAS, data acquisition typically involves acquisition of signals and waveforms and processing the signals to obtain desired information.

**9. Define thiele modulus. APR/MAY 2014, APR/MAY 2011**

The Thiele modulus was developed by E.W. Thiele in his paper 'Relation between catalytic activity and size of particle' in 1939. Thiele reasoned that with a large enough particle, the reaction rate is so rapid that diffusion forces are only able to carry product away from the surface of the catalyst particle.

**10. What is known as internal mass transfer resistance? APR/MAY 2011**

The internal mass transfer resistance to either the products or reactants that occurs between the external pellet surface and the interior of the pellet.

**11. Write down the disadvantage of sulphite oxidation method?**

Disadvantages i) slow,

ii) effected by surface active agents

iii) Rheology of soln not like media

**12. Name any four polymers used in enzyme immobilization. MAY/JUNE 2012**

**(a). Alginate:** A natural polymer derived from the cell wall of some algae. Calcium or magnesium alginate is the most commonly used matrix. They are inert and have good water holding capacity.

**(b). Chitosan and chitin:** They are structural polysaccharides occurring naturally in the cell wall of fungi and the exoskeleton of Arthropods. The various functional groups in enzymes can bind to the – OH group of chitin and can form covalent bonds.

**(c). Collagen:** It is the protenaceous support with good porosity and water holding capacity. The side chains of the amino acids in the collagen and that of enzyme can form covalent bonds to permanently hold the enzyme to the support.

**(d). Carrageenan:** It is a sulfated polysaccharide obtained from some red algae. Their good gelling properties together with its high protein holding capacity makes it good support for immobilizing enzymes.

**13. What are the distinct advantages of membrane reactors?**

Making a gaseous product in a membrane reactor generally affects the way that pressure affects the extent of reaction at thermodynamic pseudo-equilibrium. In an ordinary flow reactor, the composition of the exhaust gas is determined by the composition of the feed gas and the extent of reaction. As a result, at pseudo-equilibrium, the extent of reaction is entirely determined by the feed composition and the exhaust equilibrium constant, the latter being determined by the temperature and pressure of the exhaust. In a membrane reactor, the partial pressure of the components at psueudo-equilibrium are not uniquely determined by the total pressure, exit temperature, and feed composition. There is also a significant (and beneficial) effect that derives from the controlled removal of a product or addition of reactant.

**14. What is the mechanism of entrapment?**

In this method enzymes are physically entrapped inside a porous matrix. Bonds involved in stabilizing the enzyme to the matrix may be covalent or non-covalent. The matrix used will be a water soluble polymer. The form and nature of matrix varies with different enzymes. Pore size of matrix is adjusted to prevent the loss of enzyme. Pore size of the matrix can be adjusted with the concentration of the polymer used. Agar-agar and carrageenan have comparatively large pore sizes. The greatest disadvantage of this method is that there is a possibility of leakage of low molecular weight enzymes from the matrix.

Examples of commonly used matrixes for entrapment are:

**(1).** Polyacrylamide gels

**(2).** Cellulose triacetate

**(3).** Agar

**(4).** Gelatin

**(5).** Carrageenan

**(6).** Alginate

**15. Write down the factors which influence the pore diffusion of immobilized enzyme.**

Analysis of the effect of internal diffusion is complicated by such factors as the shape of the particles, the distribution in size and shape of the pores, the total volume of the pores with respect to the particle volume (porosity, h), the depth to which the pores penetrate the particles (e.g. pellicular particles)

**16. Compare the role of enzymes and cells in the manufacture of a biochemical product.**

The basic function of an enzyme is to increase the rate of a reaction. Most cellular reactions occur about a million times faster than they would in the absence of an enzyme. Second, most enzymes act specifically with only one reactant (called a substrate) to produce products.

Enzymes are what make all the chemical reactions in the cell possible. The human body is made up of trillions of cells, and there are different cells for differentfunctions. Cells are little bundles of chemical reactions. They reproduce, they create energy, and they break molecules down and build them up.

**17. Define packed bed reactor**

A packed bed is a hollow tube, pipe, or other vessel that is filled with a packing material. The packing can be randomly filled with small objects like Raschig rings or else it can be a specifically designed structured packing. Packed beds may also contain catalyst particles or adsorbents such as zeolite pellets, granular activated carbon, etc.

The purpose of a packed bed is typically to improve contact between two phases in a chemical or similar process. Packed beds can be used in a chemical reactor, a distillation process, or a scrubber, but packed beds have also been used to store heat in chemical plants. In this case, hot gases are allowed to escape through a vessel that is packed with a refractory material until the packing is hot. Air or other cool gas is then fed back to the plant through the hot bed, thereby pre-heating the air or gas feed.

**18. Define fluidized bed reactor**

A fluidized bed reactor (FBR) is a type of reactor device that can be used to carry out a variety of multiphase chemical reactions.

**19. Advantages of packed bed reactor**

1. High conversion per unit mass of catalyst

2. Low operating cost

3. Continuous operation

**20. Advantages of fluidized bed reactor**

1. Good mixing

2. Good uniformity of temperature

3. Catalyst can be continuously regenerated with the use of an auxiliary loop

**21. Disadvantages of packed bed reactor**

1. Undesired thermal gradients may exist

2. Poor temperature control

3. Channeling may occur

4. Unit may be difficult to service and clean

**22. Disadvantages of fluidized bed reactor**

1. Bed-fluid mechanics not well known

2. Severe agitation can result in catalyst destruction and dust formation

3. Uncertain scale-up

**23. What are the reasons for non-ideality in a fluidized bed reactor? MAY/JUNE 2012**
ψ, the "sphericity," which is a measure of a particle's nonideality in both shape and roughness. It is calculated by visualizing a sphere whose volume is equal to the particle's, and dividing the surface area of this sphere by the actually measured surface area of the particle.

**24. Application of membrane filters**
Membrane filtration is a technique that uses a physical barrier, a porousmembrane or filter, to separate particles in a fluid. Particles are separated on the basis of their size and shape with the use of pressure and specially designedmembranes with different pore sizes.

**25.Define membrane reactor.**

Membrane bioreactor (MBR) is the combination of amembrane process like microfiltration or ultrafiltration with a suspended growth bioreactor, and is now widely used for municipal and industrial wastewater treatment with plant sizes up to 80,000 population equivalent (i.e. 48 million liters per day).

**26.What is the significance of Thiele modulus in porous catalyst?**

Thiele modulus

$$\Phi = \sqrt{\frac{ka^2}{D_A}} \qquad \frac{\text{reaction rate}}{\text{diffusion rate}}$$

Effectiveness factor

$$\eta = \frac{1}{\Phi}\left[\frac{1}{\tanh 3\Phi} - \frac{1}{3\Phi}\right] \qquad \eta \equiv \frac{R_{Ap}}{R_{As}}$$

η = 1 : the entire pellet volume is reacting at the same high rate because reactant is able to diffuse quickly through the pellet,

η = 0 : the pellet reacts at a slow rate, since the reactant is unable to penetrate into the pellet interior.

**27. Name any four polymers used for enzyme immobilization.**

The polymers used for enzyme immobilization include vinylsulfone-hydroxymethyl methacrylate, poly(vinylalcohol) beads, polyacrylamide gels, 2-fluoro-1-methylpyridinium salt-activated Fractogel, hexamethylacrylate polymer (Sepharon Herma E, an anion exchange polymer), chlormethylated polystyrene beads, and beaded cellulose. Natural polymers, which are very hydrophilic, are popular support materials for enzyme immobilization since the residues in these polymers contain hydroxyl groups, which are ideal functional groups for participating in covalent bonds.

**28.Effectiveness factor**

Overall productivity for a first order reaction in a spherical pellet

$$R_{Ap} = -\eta k c_{As}$$

Effectiveness factor

$$\eta = \frac{1}{\Phi}\left[\frac{1}{\tanh 3\Phi} - \frac{1}{3\Phi}\right] \qquad \eta \equiv \frac{R_{Ap}}{R_{As}}$$

η = 1 : the entire pellet volume is reacting at the same high rate because reactant is able to diffuse quickly through the pellet,

η = 0 : the pellet reacts at a slow rate, since the reactant is unable to penetrate into the pellet interior.

**29. DefineDamköhler number**

**Damköhler number** (Da)is used to determine the significant effect of external diffusion resistance on the rate of enzyme catalytic reaction rate.

$$Da = \frac{\text{maximum rate of reaction}}{\text{maximum rate of diffusion}} = \frac{V_m'}{k_L[S_b]}$$

$V_m'$ is the maximum reaction rate per unit of external surface area (e.g. g/cm$^2$-s)

$k_L$ is the liquid mass transfer coefficient (cm/s)

$[S_b]$ Is the substrate concentration in bulk solution (g/cm$^3$)

- When Da >> 1, the external diffusion rate is limiting;
- Da << 1, the reaction rate is limiting;
- Da ≈ 1, the external diffusion and reaction resistances are comparable.

**30. Immobilization technique :Adsorption Explain**

Oldest method of enzyme immobilization
Simplest method of enzyme immobilization
Nelson &Griffin used charcoal to adsorb invertase
Enzymes are adsorbed to external surface of support
Support/carrier may be:

1. Mineral support (aluminium oxide, clay)
2. Organic support(starch)
3. Modified sepharose and ion exchange resins

## PART B

1. **Describe in detail the analysis of film and pore diffusion effects in enzyme immobilized in porous matrix. APR/MAY 2011,NOV/DEC 2015**
   Refer:James M. Lee, “Biochemical Engineering”, PHI, USA.(Page No-208-216)

2. **Explain in detail immobilized reactor. NOV/DEC 2015**
   Refer:James E. Bailey & David F. Ollis, “Biochemical Engineering Fundamentals”, McGraw-Hill.(Page No-606)

3. **Explain in detail membrane reactor. NOV/DEC 2015**
   Refer:James E. Bailey & David F. Ollis, "Biochemical Engineering Fundamentals", McGraw-Hill.(Page No-610)

4. **How will you design the packed bed reactor for immobilized enzyme reaction? APR/MAY 2011**
   Refer:James E. Bailey & David F. Ollis, "Biochemical Engineering Fundamentals", McGraw-Hill.(Page No-609)

5. **How will you design the fluidized bed reactor for immobilized enzyme reaction? APR/MAY 2011**
   Refer:James E. Bailey & David F. Ollis, "Biochemical Engineering Fundamentals", McGraw-Hill.(Page No-614)

6. **Derive the expression for effectiveness factors. MAY/JUNE 2014**
   Refer:Harvey W. Blanch, Douglas S. Clark, "Biochemical Engineering", Marcel Decker Inc.(Page No-122)

## UNIT 4 MODELLING AND SIMULATION OF BIOPROCESSES
## TWO MARKS

**1. What is plasmid? APR/MAY 2016**

A plasmid is an extra-chromosomal DNA molecule separate from the chromosomal DNA which is capable of replicating independently of the chromosomal DNA.[1] In many cases, it is circular and double-stranded. Plasmids usually occur naturally in bacteria, but are sometimes found in eukaryotic organisms (e.g., the *2-micrometre-ring* in *Saccharomyces cerevisiae*).

**2. What are the types of plasmids? APR/MAY 2016**

*Fertility-F-plasmids*, which contain tra-genes. They are capable of conjugation. *Resistance-(R) plasmids*, which contain genes that can build a resistance against antibiotics or poisons. Historically known as R-factors, before the nature of plasmids was understood. *Col-plasmids*, which contain genes that *code for* (determine the production of) bacteriocins, proteins that can kill other bacteria. *Degradative plasmids*, which enable the digestion of unusual substances, e.g., toluene or salicylic acid. *Virulence plasmids*, which turn the bacterium into a pathogen

**3. Define Simulation of plasmids**

The use of plasmids as a technique in molecular biology is supported by bioinformatics software. These programmes record the DNA sequence of plasmid vectors; help to predict cut sites of restriction enzymes, and to plan manipulations. Examples of software packages that handle plasmid maps are Lasergene, GeneConstructionKit, and Vector NTI.

**4. What is compartmental model? APR/MAY 2015**

Compartmental models are, in theory, instantaneously mixed throughout, which means that the outflow concentration for all solutes is equal to the concentration within the flowing fluid. This

allows ordinary differential equations (ODE's) to be used, for example, to calculate the change in concentration, C, within the unit and at its outflow:
dC/dt = Flow*(Cin - C)/V, where Flow = volume of flow in and out, the volume, V, is a constant and Cin is the concentration of solute in the entering flow.

**5. What is DNA replication? APR/MAY 2015, APR/MAY 2014**
DNA replication is the process of copying a double-stranded DNA molecule to form two double-stranded molecules. The process of DNA replication is a fundamental process used by all living organisms as it is the basis for biological inheritance.

**6. What is dynamic simulation? APR/MAY 2014**
A dynamic simulation can be used to estimate or illustrate the response, over time, to a change in the process. This primary concern of this site is dynamic models

**7. What types of simulations used in process engineering?**
***Static***
Static simulations, typically used in process design, simulate the process at steady state conditions, usually at the design operating conditions. Time is not a variable.
***Dynamic***
Dynamic models consider time as a variable and simulate the process over a period of time. A dynamic simulation can be used to estimate or illustrate the response, over time, to a change in the process. This primary concern of this site is dynamic models

**8. Define bioprocess engineering.**
Bioprocess engineering include the production of biofuels, design and operation of fermentation systems, development of food processing systems, application and testing of product separation technologies, design of instrumentation to monitor and control biological processes.

**9. Application of bioprocess engineering?**
Application areas commonly associated with bioprocess engineering include the production of biofuels, design and operation of fermentation systems, development of food processing systems, application and testing of product separation technologies,design of instrumentation to monitor and control biological processes.

**10. What is semi-structured model?**
The semi-structured model is a database model. In this model, there is no separation between the data and the schema, and the amount of structure used depends on the purpose.

**11. What is fed batch reactor model?**
The plug flow reactor (PFR) model is used to describe chemical reactions in continuous, flowing systems. The PFR model is used to predict the behavior of chemical reactors, so that key reactor variables, such as the dimensions of the reactor, can be estimated. PFRs are also sometimes called as Continuous Tubular Reactors (CTRs).

**12. Write the advantages of semi structured model.**
It can represent the information of some data sources that cannot be constrainedby schema.

It provides a flexible format for data exchange between different types ofdatabases.
It can be helpful to view structured data as semi-structured (for browsingpurposes).
The schema can easily be changed
. The data transfer format may be portable.

**13. What are semi structured model?**

The semi-structured model is a database model where there is no separation between the data and the schema, and the amount of structure used depends on the purpose. The advantages of this model are the following: It can represent the information of some data sources that cannot be constrained by schema.

**14. Write down the constituents of k and g compartments of the cell in Williams's model.**

k compartments consists of RNA pools of small metabolites and g compartments consist of DNA and proteins.

**15. What is meant by transient culture? MAY/JUNE 2012**

Transientculture means it is short-lived culture (usually less than 24 hours). Ex: I have observed that in daily life people come together in groups for a short time and then disperse. Example, a bus queue or strangers on a train or in an aircraft. Individuals bring their own cultural attributes to this situation and build common ground with their fellow temporary group members. They form, in my terns, 'transient group culture'.

**16. What is called promiscuous plasmids?**

Plasmids that have transfer systems that allow transfer of DNA to unrelated species are called promiscuous plasmids.

**17. How will you determine the KL value in growth associated model with inhibition. MAY/JUNE 2012**

The K parameter, defined as the ratio of the concentration of undissociated weak acid to the number of microorganisms in the medium, was determined. The K value is related to the degree of growth inhibition and provides new insight into the mode of action of weak organic acids against the studied yeasts.

**18. Define single cell model.**

The cell, itself, is the biochemical reactor. The engineer often faces the challenge of designing macroscopic bioreactors, operating polices and control strategies for the macroscopic reactor. However, the engineer must recognize that the macroscopic reactor only 'houses' the cellular bioreactors; the reactions take place under the control of the cell's own internal regulatory system. Thus, the design of the macroscopic reactor and its operating policy requires a quantitative and explicit linkage between the abiotic and biotic environment; between the macroscopic control policy and the cellular regulatory system.

**19. What is plasmid replication?**

The replication strategy that a plasmid uses directly affects its copy number, host range, and incompatibility group.

• Plasmid replication relies on the normal host DNA replication machinery.

• Plasmids will carry distinct origins of replication (oriV) and genes that enable and control the frequency of their replication.

**20. Define plasmid stability.**
Structural plasmid stability exists, when all generated plasmids have the correct base sequence.

**21. Define metabolism.**
**Metabolism** is a term that is used to describe all chemical reactions involved in maintaining the living state of the cells and the organism. **Metabolism** can be conveniently divided into two categories: Catabolism - the breakdown of molecules to obtain energy.

**22. Explain fed batch culture**
**Fed-batch culture** is, in the broadest sense, defined as an operational technique in biotechnological processes where one or more nutrients (substrates) are **fed**(supplied) to the bioreactor during cultivation and in which the product(s) remain in the bioreactor until the end of the run.

**23. Explain batch culture**
batch culture A technique used to grow microorganisms or cells. A limited supply of nutrients for growth is provided; when these are used up, or some other factor becomes limiting, the culturedeclines. Cells, or products that the organisms have made, can then be harvested from the culture.

**24. Differentiate steady state and unsteady state culture.what is the difference between transient and periodic flow?**
Steady state flow implies a uniform flow field that has no change in properties, e.g. velocity,thermal properties (such as temperature), etc.With unsteady flow, properties of the flow arechanging.Transient flow implies an unsteady state flow that is changing or developing as afunction of time – often observed in flow start up. With periodic flows, the flow field is unsteadybut oscillates around a mean.

**25. What are the origin of plasmid replication?**
The origin of replication (also called the replication origin) is a particular sequence in a genome at which replication is initiated. This can either involve thereplication of DNA in living organisms such as prokaryotes and eukaryotes, or that of DNA or RNA in viruses, such as double-stranded RNA viruses.

**26.Define static method**
Static simulations, typically used in process design, simulate the process at steady state conditions, usually at the design operating conditions. Time is not a variable.

***27. Define Dynamic method***
Dynamic models consider time as a variable and simulate the process over a period of time. A dynamic simulation can be used to estimate or illustrate the response, over time, to a change in the process. This primary concern of this site is dynamic models

**28. Define bioprocess engineering.**

Bioprocess engineering include the production of biofuels, design and operation of fermentation systems, development of food processing systems, application and testing of product separation technologies, design of instrumentation to monitor and control biological processes.

**29. Application of bioprocess engineering?**

Application areas commonly associated with bioprocess engineering include the production of biofuels, design and operation of fermentation systems, development of food processing systems, application and testing of product separation technologies, design of instrumentation to monitor and control biological processes.

**30. Write the advantages of semi structured model.**

- It can represent the information of some data sources that cannot be constrained by schema.
- It provides a flexible format for data exchange between different types of databases.
- It can be helpful to view structured data as semi-structured (for browsing purposes).
- The schema can easily be changed.
- The data transfer format may be portable.

**PART B**

1. **Describe the design consideration to be taken in account during the bio processing of animal cell culture. NOV/DEC 2015**

   Refer: Harvey W. Blanch, Douglas S. Clark, "Biochemical Engineering", Marcel Decker Inc.(Page No-203)

**Explain in detail plasmid replication. APR/MAY 2016, APR/MAY 2015,MAY/JUNE 2012**

Refer:Harvey W. Blanch, Douglas S. Clark, "Biochemical Engineering", Marcel Decker Inc.(Page No-251-257)

**Discuss in detail compartmental model. APR/MAY 2016, MAY/JUNE 2013,2014**

Refer:James M. Lee, "Biochemical Engineering", PHI, USA.(Page No-174)

2. **Explain in detail the structured model for analysis of various bioprocess.APR/MAY 2011**

   Refer:James M. Lee, "Biochemical Engineering", PHI, USA.(Page No-174)

3. **Discuss in detail Single cell model. NOV/DEC 2015**

   Refer:Harvey W. Blanch, Douglas S. Clark, "Biochemical Engineering", Marcel Decker Inc.(Page No-244-246)

4. **Explain in detail dynamic simulation of batch, fed batch, steady and transient culture metabolism MAY/JUNE 2013,2014 APR/MAY 2011**

Refer:Harvey W. Blanch, Douglas S. Clark, “Biochemical Engineering”, Marcel Decker Inc.(Page No-277-280)

## UNIT 5 RECOMBINANT CELL CULTIVATION
## TWO MARKS

**1. Define Animal cell culture? APR/MAY 2016, APR/MAY 2015**

Animal cell culture (ACC) is the process of culture of animal cells outside the tissue (*ex vivo*) from which they were obtained. The process of ACC is carried out under strict laboratory conditions of asepsis, sterility and controlled environment involving temperature, gases and pressure.

**2. Define cell culture?**

Cell culture is the process by which prokaryotic, or eukaryotic cells are grown under controlled conditions. In practice the term "cell culture" has come to refer to the culturing of cells derived from multicellular eukaryotes, especially animal cells.

**3. What are the applications of cell culture?**

Mass culture of animal cell lines is fundamental to the manufacture of viral vaccines and many products of biotechnology. Biological products produced by recombinant DNA (rDNA) technology in animal cell cultures include enzymes, synthetic hormones, immunobiologicals (monoclonal antibodies, interleukins, lymphokines), and anticancer agents.

**4. Explain high cell density cultivation? APR/MAY 2015**

In particular, microbial high cell density cultures have a high metabolic oxygen demand. In these cultures the oxygen transfer rate of the bioreactor determines the maximum biomass concentration. Unfortunately, the solubility of oxygen is even decreasing with increasing cell densities due to a higher viscosity of the cell suspension. Therefore, in order to enhance the existing oxygen transfer limitation for aerobic high cell density cultivation in disposable bioreactors, an aeration membrane or disposable spargers and baffles have been inserted or the medium was vibrated.

**5. What is meant by host?**

An animal or plant on or in which a parasite or commensal organism lives.

**6. Define recombinant cell cultivation? APR/MAY 2014**

Relating to or denoting an organism, cell, or genetic material formed by recombination.

**7. Advantages of recombinant cell cultivation?**

Precise control over growth conditions, batch-to-batch product consistency, a high level of containment and the ability to produce recombinant proteins in compliance with good manufacturing practice.

**8. Write down the Application of animal cell cultivation. APR/MAY 2014**

Mass culture of animal cell lines is fundamental to the manufacture of viral vaccines and other products of biotechnology.

many simpler proteins can be produced using rDNA in bacterial cultures, more complex proteins that are glycosylated(carbohydrate-modified) currently must be made in animal cells

**9. What is meant by plant cell cultivation?**

Plant cell cultures are typically grown as cell suspension cultures in a liquid medium or as callus cultures on a solid medium. The culturing of undifferentiated plant cells and calli requires the proper balance of the plant growth hormones auxin and cytokinin.

**10. Write down the Application of high cell density cultivation.**

In this system the amount of an enzyme determines the glucose release rate and all optimisation can be done at the microwell or deepwell plate stage. Here we show at the example of ADH that such preoptimised processes simply can be transferred to a rocking-motion-system, without the need for setting up any extra control. This would provide a simple scale up to the 100 scale. On top of this, further modifications can be done for improving the volumetric yield further

**11. Explain the reactor considerations in animal cell cultivation.**

- It is critical to find a 'HAPPY' environment for cell cultures.
- Happy environment = allows cells to increase in number by undergoing cell division (mitosis).
- Provide the cells with appropriate temp, good substrate for attachment and proper culture medium.
- Temperature:Cold-blooded vertebrates – 18-25°C,Mammalian cells – 36-37°C

**12. Explain the common features of animal cell bioreactor?**

1) Reactor should be gently agitated and aerated. Agitation speed ≈20rpm.
   Bubble-column & airlift reactor operating at high aeration may cause damage of cells
2) Supply of $CO_2$-enriched air
3) Removal of toxic products from metabolism eg lactic acid, ammonium

**13. List out the advantages of animal cell bioreactor?**

- Ability to perform post-translational modifications and achieve a product close to that produced in vivo
- The used of other expression systems (yeast, plant or insect cell) more limited because despite their potential economic advantages for culture and higher yields of these systems, their glycosylation capacities do not resemble those of the mammalian cells.
- Prokaryotes cells can be easily manipulated and grown in large scale but they lack the necessary glycosylation machinery and so the proteins produced are not glycosylated.
- Lower eukaryotes cells produced differ significantly from those present in human glycoproteins.
- So mamalian cells are the chosen host for the production of human glycoproteins because it has been recognized that they meet the criteria for an appropriate glycosylation of recombinant human glycoprotein.
- The most commonly used cell lines are hamster-derived Chinese hamster ovari (CHO and baby hamster kidney (BHK) cells
- These cells are chosen because of their favorable growth characteristics in culture both as anchorage-dependent or suspension cells.

**14. Define E.coli?**

A bacterium commonly found in the intestines of humans and other animals, some strains of which can cause severe food poisoning.

**15. What is yeast?**

A microscopic fungus consisting of single oval cells that reproduce by budding, and capable of converting sugar into alcohol and carbon dioxide.A greyish-yellow preparation of the yeast fungus obtained chiefly from fermented beer, used as a fermenting agent, to raise bread dough, and as a food supplement.

**16. What is Pichia pastoris?**

Pichia pastoris is a species of methylotrophic yeast. Pichia is widely used for protein production using recombinant DNA techniques. Hence it is used in biochemical and genetic research in academia and the biotechnical industry.

**17. What is Saccharomyces cereviseae?**

Saccharomyces cerevisiae is a species of yeast. It has been instrumental to winemaking, baking, and brewing since ancient times.

**18. Process strategies of animal cell cultivation.**

(1) cell lines capable of synthesizing the required molecules at high productivities that ensure low operating cost;
(2) culture media and bioreactor culture conditions that achieve both the requisite productivity and meet product quality specifications;
(3) appropriate on-line and off-line sensors capable of providing information that enhances process control; and
(4) good understanding of culture performance at different scales to ensure smooth scale-up. Successful implementation also requires appropriate strategies for process development, scale-up and process characterization and validation that enable robust operation and ensure compliance with current regulations

**19. Process strategies of plant cell cultivation.**

In order to successfully cultivate the plant cells at large scale, several engineering parameters such as, cell aggregation, mixing, aeration, and shear sensitivity are taken into account for selection of a suitable bioreactor. The media ingredients, their concentrations and the environmental factors are optimized for maximal synthesis of a desired metabolite. Increased productivity in a bioreactor can be achieved by selection of a proper cultivation strategy (batch, fed-batch, two-stage etc.), feeding of metabolic precursors and extraction of intracellular metabolites. Proper understanding and rigorous analysis of these parameters would pave the way towards the successful commercialization of plant cell bioprocesses.

**20. Process strategies of insect cell cultivation.**

1. Seed the experimental culture at between 0.5-1 x 106 cells per ml and allow to grow overnight.
2. Confirm log growth of cells by observation of increase cell counts. If the culture did not grow overnight, discard and start again.

3. Infect the culture at your desired MOI Volume of inoculum needed = MOI ( iu/cell ) X number of cells Titer of virus ( iu/ml )

**21. Process strategies of high cell density cultivation.**

the application of more advanced control strategies, such as exponential feeding procedures and feed-back control of the $pO_2$. Pulsing of extra oxygen provides some advantage in overcoming the low oxygen transfer rates.

**22. List out the host vector system.**

Prokaryotes

1) Gram Negative
2) Gram Positive

Eukaryotes

1) Yeast
2) Fungi
3) Insect Cells
4) Plant Cell Lines
5) Animal Cell lines
6) Transgenic Animals
7) Transgenic Plants

**23. What is a recombinant cell?**

**Recombinant** DNA (rDNA) molecules are DNA molecules formed by laboratory methods of genetic recombination (such as molecular cloning) to bring together genetic material from multiple sources, creating sequences that would not otherwise be found in biological organisms.

**24. What is a primary culture?**

Primary culture refers to the stage of the culture after the cells are isolated from the tissue and proliferated under the appropriate conditions until they occupy all of the available substrate (i.e., reach confluence).

**25. What does meant to culture cells?**

**Cell culture** is the complex process by which **cells** are grown under controlled conditions, generally outside of their natural environment. In practice, the term "**cell culture**" now refers to the culturing of **cells** derived from multi-cellular eukaryotes, especially animal **cells**.

**26. Define Cell strain?**

A cell strain is derived either from a primary culture or a cell line by the selection or cloning of cells having specific properties or characteristics which must be defined. Cell strains are cells that have been adapted to culture but, unlike cell lines, have a finite division potential. Non-immortalized cells stop dividing after 40 to 60 population doublings and, after this, they lose their ability to proliferate (a genetically determined event known as senescence).

**27.How will you Determine the Cell Viability (for all cell types)**

After the cells have been suspended into the medium, 0.2 ml of the cell suspension can be mixed with 0.3 ml PBS and 0.5 ml trypan blue (final concentration, 0.2% w/v) in a small test tube. An

aliquot is then placed on a hemacytometer and counted with the compound microscope. The number of viable cells (those not taking up the stain) can be determined and used for initiating the new culture with a precise number of viable cells. I personally find this is a time consuming step that does not greatly improve the probability of maintaining healthy cultures but this is largely because I feel confident in recognizing healthy cells just by examining them in the flask with the inverted microscope. Beginners may want to include this step until they gain confidence in their visual inspection of cells.

**28.Explain Micro Propagation**

It is in vitro asexual propagation of crop plants. This technique is advantages over the conventional practice of asexual propagation as only a small amount of plant is needed, species highly resistant to conventional bulk propagation can be propagated by this method and it is non season dependent. Micro propagation is used for rapid multiplication of stocks, elimination of diseases, germ plasm preservation, and induction of mutation.

**29.Define Embryo Culture**

The embryos are isolated from young seeds and placed on a solid medium containing nutrients and vitamins. Embryos are cultured at 25°c,first in dark until seedlings are about 2 cm long and root formation has started, and than in light until the seedlings can be planted in soil.

**30.Short notes on Protoplast Culture**

It is one of the most significant and recent developments in the field of plant tissue culture. the protoplast are usually isolated from cultured cell or leaf mesophyll cell by treating them with enzyme solutions. the isolated protoplast may be used to regenerate the plants directly, or for the production of somatic hybrids through fusion.

## PART B

**Discuss animal cell cultivation and explain the products of animal cell culture. APR/MAY 2016, NOV/DEC 2015**

Ref: James E. Bailey & David F. Ollis, “Biochemical Engineering Fundamentals”, McGraw-Hill.(Page No-261)

1. **Discuss plant cell cultivation.**
   Ref:James E. Bailey & David F. Ollis, “Biochemical Engineering Fundamentals”, McGraw-Hill.(Page No-265)

2. **Explain in detail Different host vector system for recombinant cell cultivation. NOV/DEC 2015,MAY/JUNE 2016, 2014**
   Ref:James E. Bailey & David F. Ollis, “Biochemical Engineering Fundamentals”, McGraw-Hill.(Page No-257)

3. **Explain in detail insect cell cultivation APR/MAY 2015**
   Ref:James E. Bailey & David F. Ollis, "Biochemical Engineering Fundamentals", McGraw-Hill.(Page No-267)

4. **How the high cell density cultivation was processed explain in detail?**
   Ref:James E. Bailey & David F. Ollis, "Biochemical Engineering Fundamentals", McGraw-Hill.(Page No-269)

5. **In what way recombinant cell cultivated in E.coli explain clearly. APR/MAY 2014**
   Ref:James E. Bailey & David F. Ollis, "Biochemical Engineering Fundamentals", McGraw-Hill.(Page No-259)