

BIOTECHNOLOGY

UNIT 5 NOTES

- FERMENTATION METHODS
- LARGE SCALE FERMENTER
- PRODUCTION OF :
 - PENICILLINS
 - CITRIC ACID
 - VITAMIN B12 ETC.
- BLOOD PRODUCTS



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QUESTION - 1

**DEFINE FERMENTATION. EXPLAIN THE DESIGN
OF LARGE SCALE FERMENTER**

FERMENTATION

- Fermentation is a biochemical process in which microorganisms like bacteria, yeast, or fungi convert sugars and other organic substances into useful products such as alcohol, acids, gases, enzymes, or antibiotics under controlled conditions.
- It is widely used in:
 - Pharmaceutical industry
 - Food industry
 - Biotechnology
 - Alcohol production Principle of Fermentation
- Microorganisms use nutrients present in the medium for their growth and metabolism.
- During this process, they produce desired products such as:
 - Ethanol
 - Lactic acid
 - Antibiotics
 - Vitamins
 - Enzymes

COMPONENTS OF FERMENTATION

① Microorganism

- Selected organism that produces desired product.
- Examples: Yeast, Bacteria, Fungi

② Culture Medium

- Contains nutrients required for growth.
- Nutrients include:
 - Carbon source
 - Nitrogen source
 - Minerals - Vitamins

③ Fermenter

- Fermenter is a vessel/equipment in which fermentation takes place.
- It maintains sterile conditions
- Controls temperature and pH
- Supplies oxygen if needed

④ Environmental Conditions

- Proper conditions are maintained for maximum product formation such as

- Temperature
- pH
- Aeration
- Agitation

STAGES OF FERMENTATION

① Preparation of Culture Medium

- Nutrient medium is prepared for microorganism growth.
- It contains carbon source, nitrogen source, minerals, and vitamins.

② Sterilization

- Fermenter, media, and equipment are sterilized.
- Sterilization prevents contamination by unwanted microbes.

③ Inoculation

- Selected microorganism is added to the sterile medium.
- This starts the fermentation process.
- Proper aseptic conditions are maintained during inoculation.

④ Incubation

- The fermenter is maintained at suitable temperature and pH.
- Microorganisms grow and multiply rapidly.

⑤ Product Formation

- Microorganisms produce desired products during metabolism.
- Products may include antibiotics, alcohol, acids, or enzymes.

⑥ Recovery and Purification

- The final product is separated from the fermentation broth.
- Impurities are removed by purification methods.

APPLICATION OF FERMENTATION

① Pharmaceutical Industry

- Production of :
 - Antibiotics
 - Vaccines
 - Vitamins

- Enzymes
- Hormones

② Food Industry

- Production of :
 - Bread
 - Yogurt
 - Cheese
 - Vinegar

③ Alcohol Industry

- Production of :
 - Beer
 - Wine
 - Ethanol

LARGE SCALE FERMENTER

- A large-scale fermenter is a closed vessel used for the industrial production of products such as antibiotics, enzymes, alcohol, vaccines, and vitamins using microorganisms under controlled conditions.
- It is also called a Bioreactor.

FERMENTER DESIGN

① Fermenter Tank

- Main vessel where fermentation occurs
- Usually made of stainless steel
- Cylindrical in shape with rounded bottom

② Agitation System

- Contains motor and impellers
- Mixes the medium uniformly

③ Aeration System

- Supplies sterile air into the fermenter
- Air enters through sparger or aerator

④ Feeding Pump

- Adds nutrients or medium into the fermenter
- Maintains proper nutrient supply
- Used during continuous fermentation

⑤ Sensors and Probes

- Measure pH, temperature, dissolved oxygen, and foam
- Help in monitoring fermentation conditions

⑥ Temperature Control System

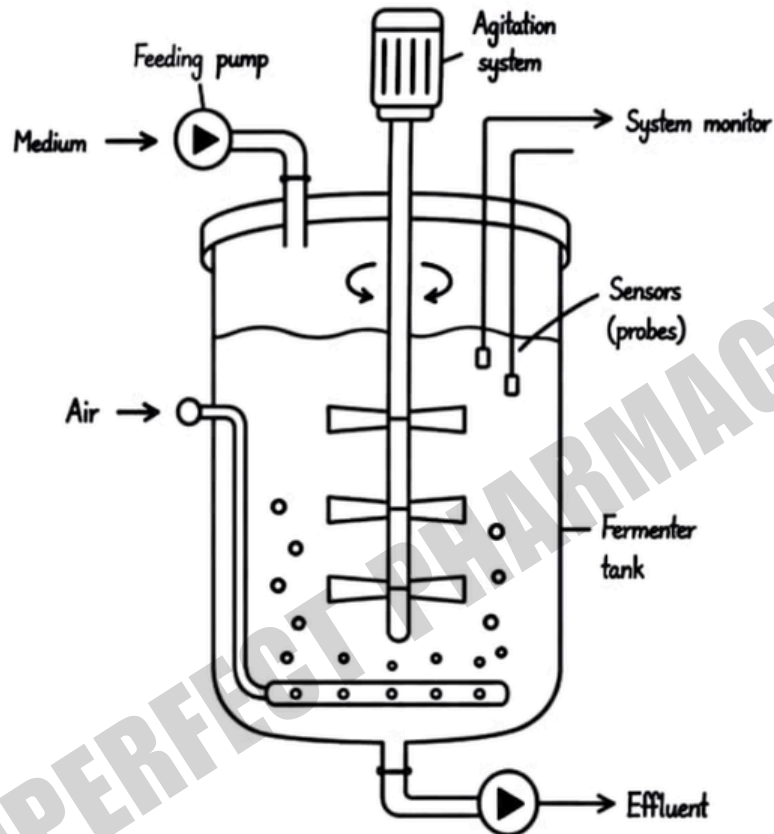
- Maintains optimum temperature
- Usually consists of cooling jacket or coils
- Prevents overheating during microbial growth

⑦ Foam Control System

- Prevents excessive foam formation
- Antifoam agents may be added
- Foam breakers may also be used

⑧ Effluent Outlet

- Removes fermented product and waste material
- Helps in product recovery
- Located at the bottom of the fermenter



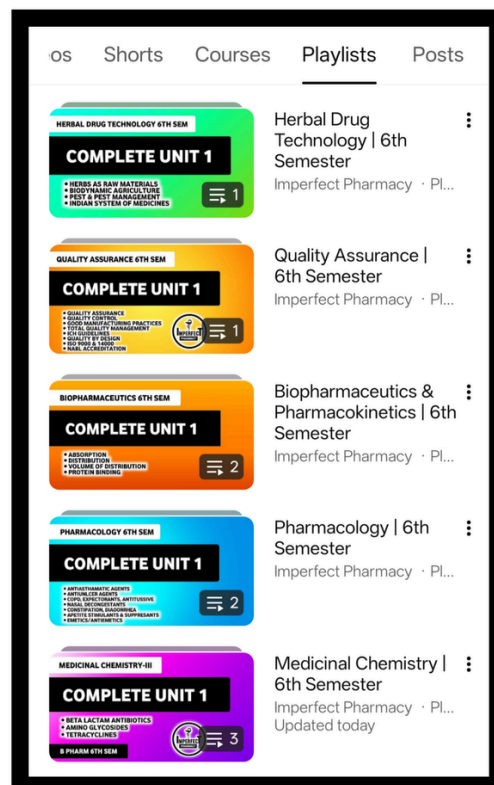
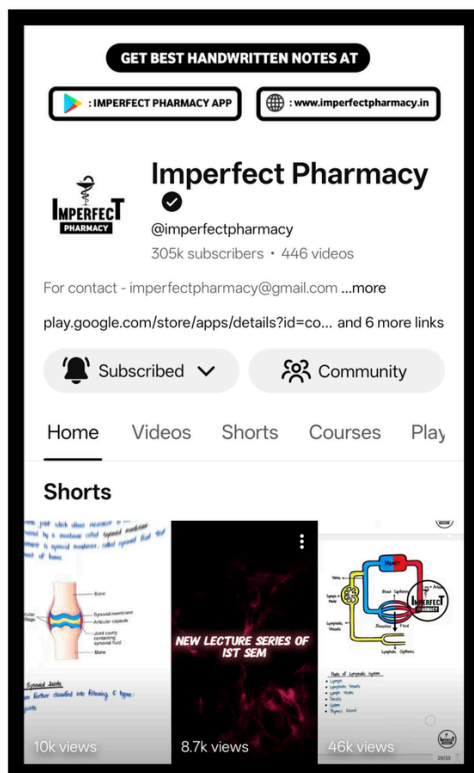
Characteristics of an Ideal Fermenter

- Should be sterile and contamination-free
- Must provide proper aeration and mixing
- Should allow easy cleaning and maintenance
- Must withstand high pressure and temperature
- Should support automatic monitoring and control

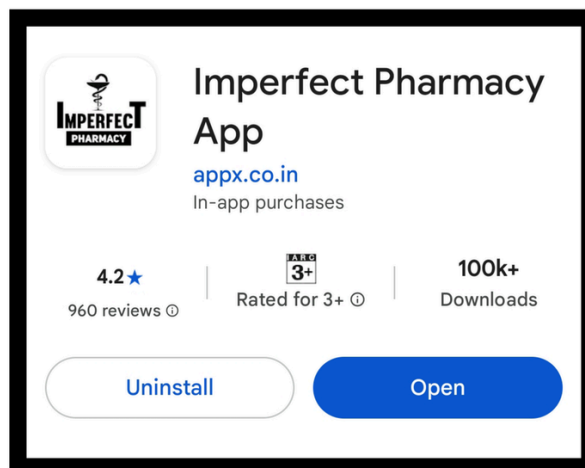
Applications of Large-Scale Fermenter

- Production of antibiotics
- Vaccine production
- Enzyme production
- Alcohol fermentation
- Production of vitamins and organic acids

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QUESTION - 2

**DEFINE THE PRODUCTION OF VITAMIN B12
THROUGH FERMENTATION**

VITAMIN B12

- Vitamin B12 is a water-soluble vitamin also known as Cobalamin.
- Vitamin B12 is mainly produced by microorganisms and is obtained from animal foods or fermentation processes.
- It is essential for:
 - Formation of red blood cells
 - Proper functioning of nervous system
 - DNA synthesis
 - Normal growth and metabolism

Sources of Vitamin B12

- Meat
- Fish
- Eggs
- Milk
- Liver
- Fermented products

Functions of Vitamin B12

- Helps in red blood cell formation
- Maintains healthy nerve cells
- Supports DNA synthesis
- Prevents megaloblastic anemia
- Helps in energy metabolism

PRODUCTION OF VITAMIN B12

Vitamin B12 is commercially produced by microbial fermentation because chemical synthesis is difficult and expensive.

Microorganisms Used

- Most commonly used microorganisms for Vitamin B12 productions are:

- *Propionibacterium shermanii*
- *Pseudomonas denitrificans*

STEPS INVOLVED IN PRODUCTION

1) Preparation of Culture Medium

- Nutrient medium containing carbon, nitrogen, minerals, and cobalt is prepared.
- Cobalt is important for Vitamin B12 synthesis.

2) Sterilization

- Fermenter and medium are sterilized.
- This prevents contamination.

3) Inoculation

- Selected microorganism is added into the sterile medium.
- Pure culture is used for better production.

4) Fermentation

- Fermentation is carried out under controlled conditions.
- Proper temperature, pH, aeration, and agitation are maintained.
- Microorganisms produce Vitamin B12 during growth.

5) Recovery of Vitamin B12

- Fermentation broth is filtered.
- Vitamin B12 is extracted from microbial cells.

6) Purification

- The product is purified and concentrated.
- Final Vitamin B12 is converted into stable pharmaceutical forms.

Applications of Vitamin B12

- Treatment of Vitamin B12 deficiency
- Used in anemia treatment
- Nutritional supplements
- Pharmaceutical preparations

QUESTION - 3

**WRITE THE METHOD OF COLLECTION AND
PROCESSING OF WHOLE HUMAN BLOOD**

BLOOD

- Blood is a special type of fluid connective tissue that circulates throughout the body inside blood vessels.
- It is pumped by the heart & continuously moves to deliver important substances & nutrients to different organ & tissues.
- Haematology is the branch of science deals with study of blood & its disorders.
- It is composed of 8% of total body weight.

BASIC PROPERTIES OF BLOOD

- Adults have about 5-6 litres of blood, around 7-8% of body weight.
- Blood is slightly alkaline in Nature having pH: 7.35 - 7.45.
- Blood temperature is about 38°C.
- Blood is about 4-5 times more viscous than water.
- Blood has the ability to clot when injured.
- Blood has a slightly salty taste due to dissolved salts.
- The colour of blood varies with its oxygen content, Oxygenated Blood appears bright red, while Deoxygenated Blood appears dark red.

FUNCTIONS OF BLOOD

Blood has 3 major functions :

- ① Transportation
- ② Protection
- ③ Regulation

① TRANSPORTATION

• Blood transports :

- O_2 (lungs $\rightarrow$ body)
- CO_2 (body $\rightarrow$ lungs)
- Nutrients (intestine $\rightarrow$ tissues)
- Hormones (glands $\rightarrow$ organs)
- Waste products (tissues $\rightarrow$ kidneys / livers)

② PROTECTION

Blood protects the body by:

WBCs fighting infections & Antibodies providing immunity

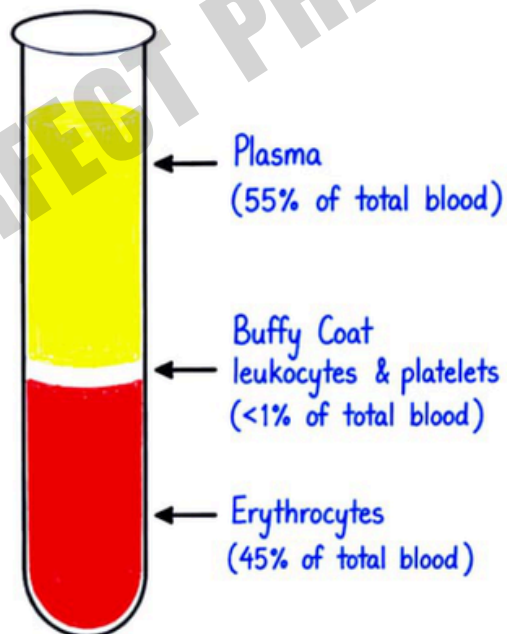
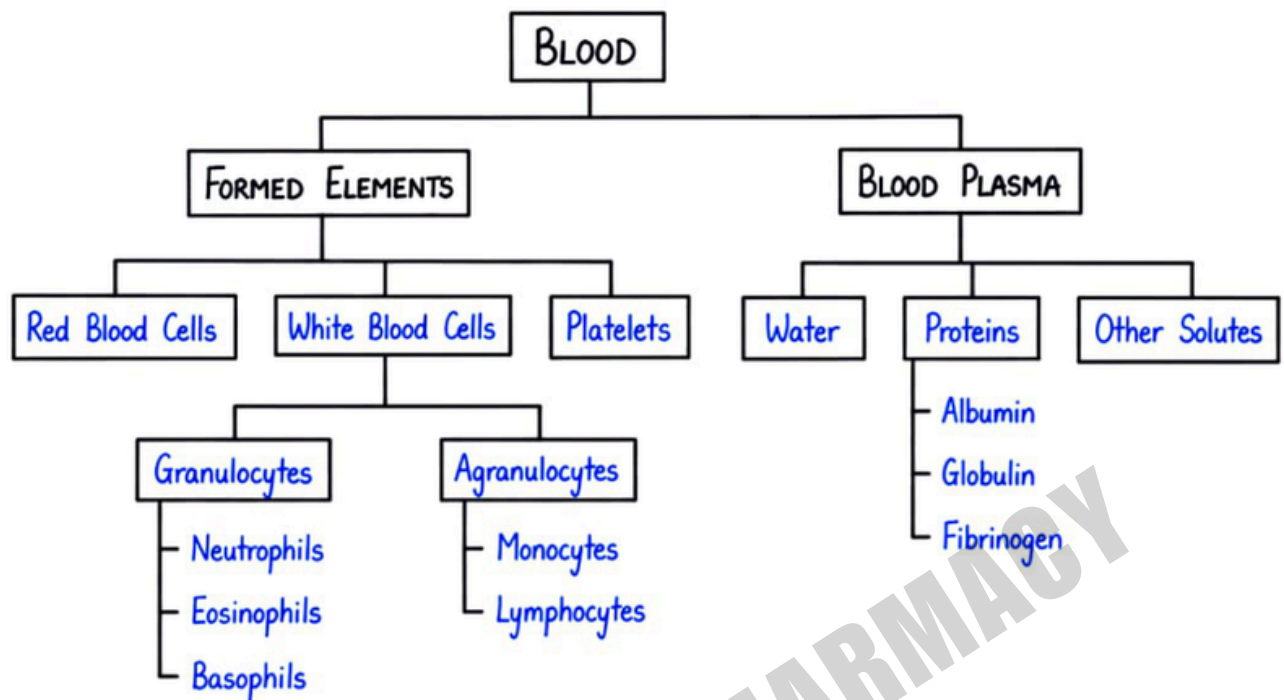
Platelets preventing bleeding & Blood Clotting

③ REGULATION

Blood helps maintain :

- Body temperature
- pH balance (acid-base balance)
- Water balance
- Blood pressure

COMPONENTS OF BLOOD



COLLECTION & PROCESSING OF WHOLE HUMAN BLOOD

- Whole human blood is collected from healthy donors and processed to prepare blood and blood products for transfusion.
- Blood collection and processing must be done carefully to maintain safety, sterility, and quality.

COLLECTION OF WHOLE HUMAN BLOOD

1) Selection of Donor

- Before blood donation, the donor is checked to ensure the blood is safe.
- Requirements of a Healthy Donor
 - Age: usually 18-65 years
 - Weight: minimum about 45-50 kg
 - Hemoglobin level should be normal
 - Normal blood pressure and pulse - Free from infectious diseases

2) Blood Collection

- Sterile disposable blood bags are used.
- The vein area (usually arm) is cleaned with antiseptic solution.
- A sterile needle is inserted into the vein.
- About 350-450 mL of blood is collected.
- Blood bag contains anticoagulant solution such as CPDA (Citrate Phosphate Dextrose Adenine) to prevent clotting.

After Collection

- Needle is removed and bandage is applied.
- Donor is advised to rest and take fluids.

PROCESSING OF WHOLE HUMAN BLOOD

1) Labeling and Documentation

- Each blood bag is labeled properly.
- Donor information, blood group, and collection date are recorded.

2) Blood Group Testing

- Blood is tested for:
 - ABO blood group

- Rh factor
- This helps in compatible transfusion.

3) Screening for Infectious Diseases

- Blood samples are tested again for:
 - HIV
 - Hepatitis viruses
 - Syphilis
 - Malaria
- Unsafe blood is discarded.

4) Separation of Blood Components

- Whole blood is separated by centrifugation into:
 - Packed Red Blood Cells (RBCs)
 - Platelets
 - Plasma
 - Cryoprecipitate
- This allows one unit of blood to help multiple patients.

5) Storage

- Different blood components are stored at different temperatures:

Component	Storage Temperature
Whole blood / RBCs	2-6°C
Platelets	20-24°C
Plasma	-18°C or below

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