BHARATHIDASAN UNIVERSITY

Tiruchirappalli- 620024,
Tamil Nadu, India.

Programme: M.Sc., Marine Biotechnology

Course Title : Microbiology

Course Code: 21CC1

Unit-II

Microbial Nutrition

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Introduction

- Microorganisms can be grouped into nutritional classes based on how they satisfy all these (carbon energy and electrons) requirements.

- Microorganisms can be classified as either autotrophs or heterotrophs with respect to their preferred sources of carbon.

- However by definition only autotroph can use carbon dioxide as the sole or principal source of carbon.

Autotrophs

- make their own food
- include plants, some protists, and some bacteria

Heterotrophs

- CANNOT make their own food; must obtain energy from outside sources
- Includes animals, fungi, and some bacteria

https://www.careerpower.in/school/biology/nutrition-in-plants

- The reduction of carbon dioxide is a very energy explain Process.

- Thus many microorganisms cannot use carbon dioxide has their sole carbon sources must relay on the presence of more reduced complex molecules such as glucose for a supply of carbon

- Organisms that use reduce, preformed organic molecules such as carbon sources are heterotrophs these performed molecule normally come from other organisms.

- Most heterotrophs use atoms to organic compounds as source of both carbon and energy.

- For example the glycolytic pathway produces carbon skeletons for use in bio synthesis and also released energy as ATP and NADH.

- There are only 2 sources of energy available to organisms.
  - 1. Light energy
  - 2. Energy derived from oxidizing organic or in organic molecules

- Many microbes are **autotrophs** and most of the carry out photo synthesis and light as the energy source.
- **Phototrophs** use light as their sole energy source
- **Chemotrophs** obtains energy from the oxidation of chemical compounds either organic or in organic. Microorganisms also have only 2 source of electrons
- **Lithotrophs** extract electrons from organic compounds

- **Nutritional classification based on carbon, energy and electron**
  - Based on their primary source of carbon, energy, and electrons microorganisms are classified into 3 groups.

•**Carbon sources**

Autotrophs : Carbon dioxide soul or principle biosynthesis carbon source

Heterotrophs : Reduced, performed, organic molecules from other organisms

•**Energy sources**

Phototrophs : Use Light

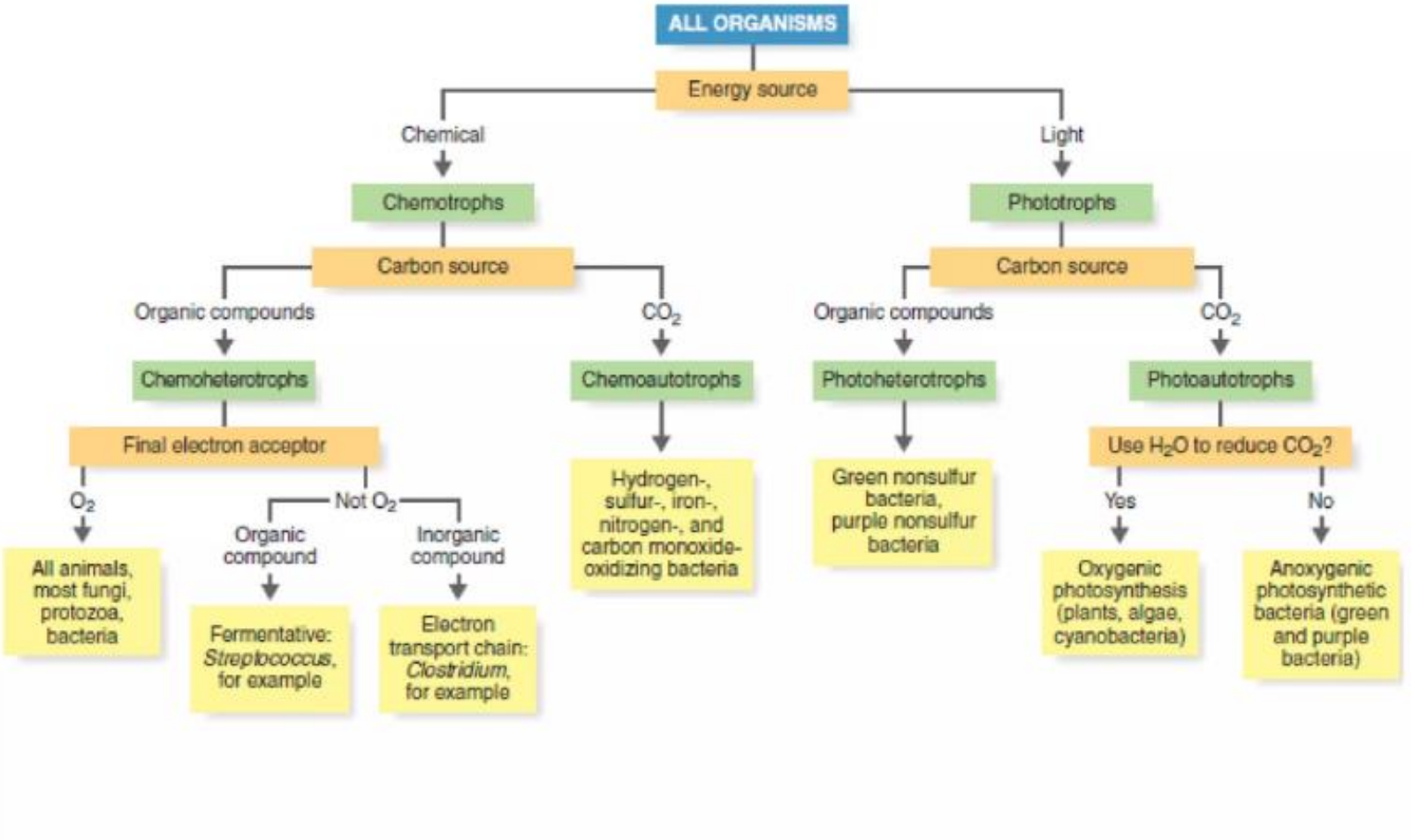
Chemotrophs : Oxidation of organic or in organic compounds

•**Electron sources**

Organotrophs : organic molecules

Lithotrophs : reduce inorganic molecules.

Nutritional classification of Microorganisms



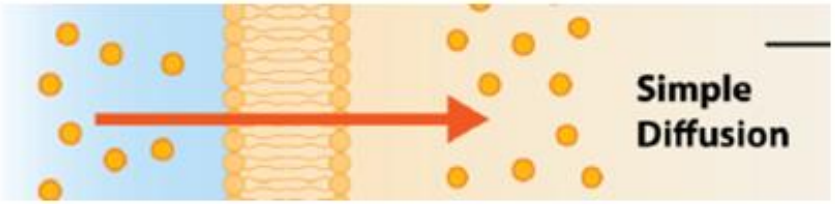
Nutritional types of microorganisms

Major Nutritional Types	Energy source	Hydrogen/ electron	carbon source	Representative Microorganisms
Photolithotrophic autotrophy (Photolithoautotrophy) (photoautotrophs)	Light energy	Inorganic hydrogen/electron (H/e–) donor	CO2 carbon source	Algae Purple and green sulfur bacteria Cyanobacteria
Photoorganotrophic heterotrophy (Photoorganoheterotrophy) (Photoheterotrophs)	Light energy	Organic H/e– donor	Organic carbon source	Purple nonsulfur bacteria Green nonsulfur bacteria
Chemolithotrophic autotrophy (Chemolithoautotrophy) (Chemoautotrophs)	Chemical energy source (inorganic)	Inorganic H/e– donor	CO2 carbon source	Sulfur-oxidizing bacteria Hydrogen bacteria Nitrifying bacteria Iron-oxidizing bacteria
Chemoorganotrophic heterotrophy (Chemoorganoheterotrophy) (Chemoheterotrophs)	Chemical energy source (organic)	Organic H/e– donor	Organic carbon source	Protozoa, Fungi, Most nonphotosynthetic bacteria (including most pathogens)

Uptake of Nutrients

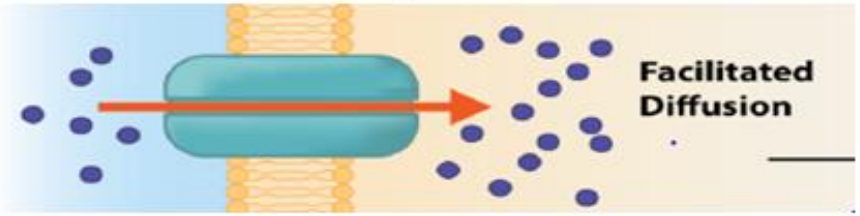
In order to support its' activities, a cell must bring in nutrients from the external environment across the cell membrane. In bacteria and archaea, several different transport mechanisms exist.

Passive Diffusion



Passive or **simple diffusion** allows for the passage across the cell membrane of simple molecules and gases, such as CO2, O2, and H2O. In this case, a concentration gradient must exist, where there is higher concentration of the substance outside of the cell than there is inside the cell. As more of the substance is transported into the cell the concentration gradient decreases, slowing the rate of diffusion.

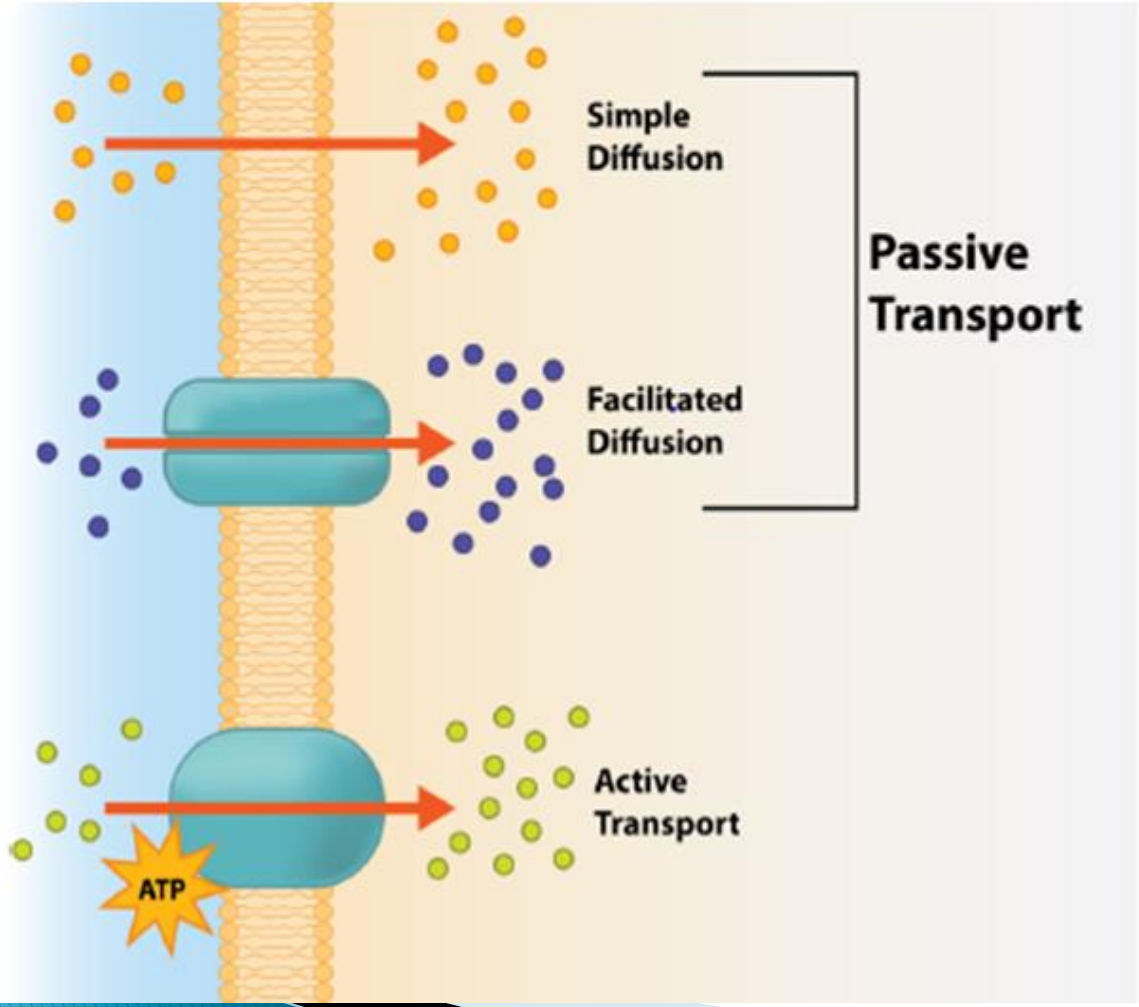
Facilitated Diffusion



Facilitated diffusion also involves the use of a concentration gradient, where the concentration of the substance is higher outside the cell, but differs with the use of **carrier proteins** (sometimes called **permeases**). These proteins are embedded within the cell membrane and provide a channel or pore across the membrane barrier, allowing for the passage of larger molecules. If the concentration gradient dissipates, the passage of molecules into the cell stops. Each carrier protein typically exhibits specificity, only transporting in a particular type of molecule or closely related molecules.

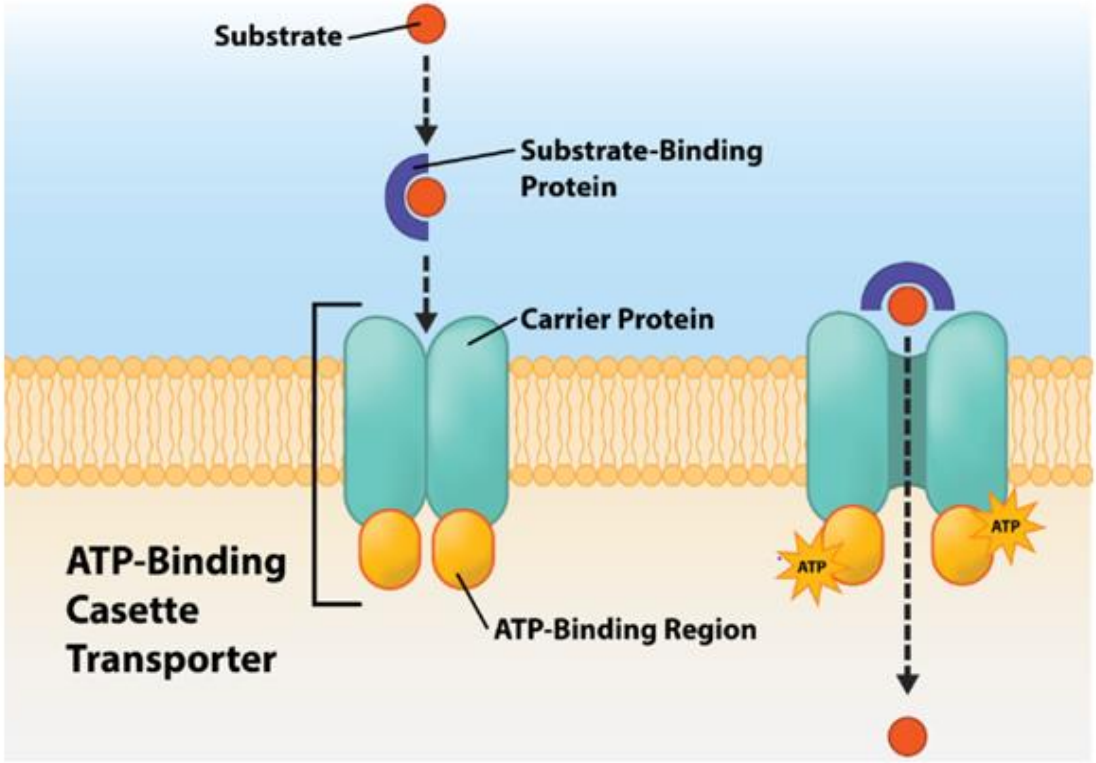
Active Transport

Many types of nutrient uptake require that a cell be able to transport substances against a concentration gradient (i.e. with a higher concentration inside the cell than outside). In order to do this, a cell must utilize metabolic energy for the transport of the substance through carrier proteins embedded in the membrane. This is known as **active transport**. All types of active transport utilize carrier proteins.



Primary active transport

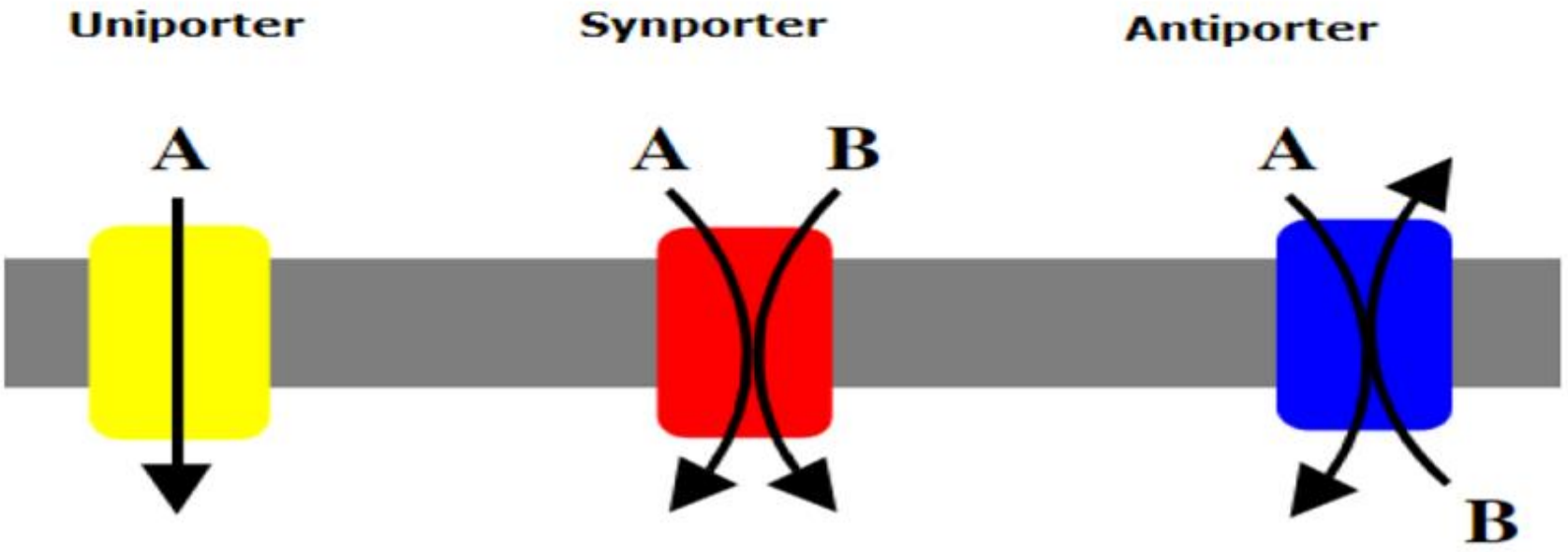
Primary active transport involves the use of chemical energy, such as ATP, to drive the transport. One example is the **ABC system**, which utilizes **ATP-Binding Cassette transporters**. Each **ABC transporter** is composed of three different components: 1) membrane-spanning proteins that form a pore across the cell membrane (i.e. carrier protein), 2) an ATP binding region that hydrolyzes ATP, providing the energy for the passage across the membrane, and 3) a substrate-binding protein, a peripheral protein that binds to the appropriate substance to be transporter and ferries it to the membrane-spanning proteins. In gram negative bacteria the substrate-binding protein is located in the cell's periplasm, while in gram positive bacteria the substrate-binding protein is attached to the outside of the cell membrane.



Secondary active transport

Secondary active transport utilizes energy from a **proton motive force (PMF)**. A PMF is an ion gradient that develops when the cell transports electrons during energy-conserving processes. Positively charged protons accumulate along the outside of the negatively charged cell, creating a proton gradient between the outside of the cell and the inside.

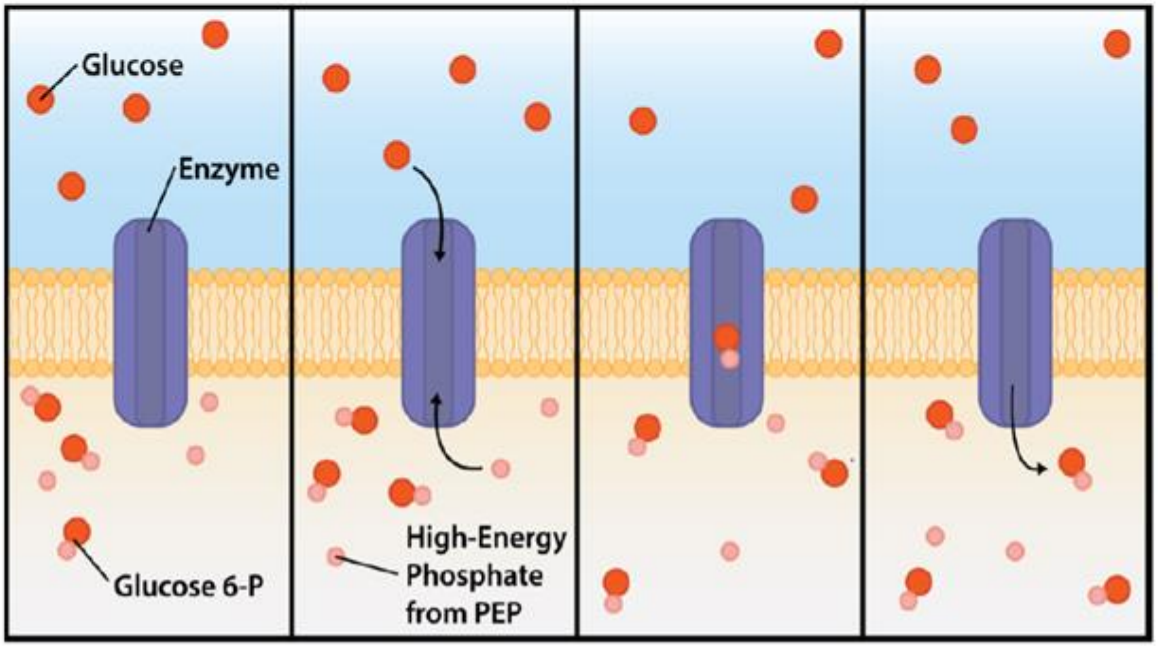
There are three different types of transport events for simple transport: **uniport**, **symport**, and **antiport** and each mechanism utilizes a different protein **porter**. **Uniporters** transport a single substance across the membrane, either in or out. **Symporters** transport two substances across the membrane at the same time, typically a proton paired with another molecule. **Antiporters** transport two substances across the membrane as well, but in opposite directions. As one substance enters the cell, the other substance is transported out.



Group Translocation

Group translocation is a distinct type of active transport, using energy from an energy-rich organic compound that is not ATP. Group translocation also differs from both simple transport and ABC transporters in that the substance being transported is chemically modified in the process.

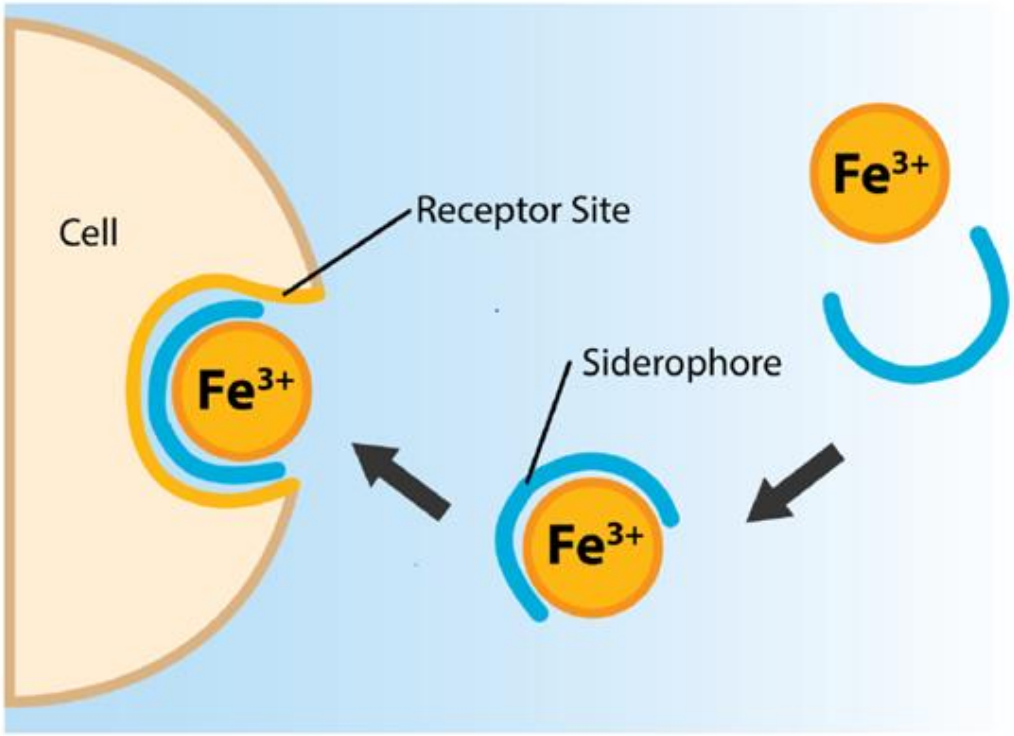
One of the best studied examples of group translocation is the **phosphoenolpyruvate: sugar phosphotransferase system (PTS)**, which uses energy from the high-energy molecule **phosphoenolpyruvate (PEP)** to transport sugars into the cell. A phosphate is transferred from the PEP to the incoming sugar during the process of transportation.



Group Translocation via PTS.

Iron is required by microbes for the function of their cytochromes and enzymes, resulting in it being a growth-limiting micronutrient. However, little free iron is available in environments, due to its insolubility. Many bacteria have evolved **siderophores**, organic molecules that chelate or bind ferric iron with high affinity. Siderophores are released by the organism to the surrounding environment, whereby they bind any available ferric iron. The iron-siderophore complex is then bound by a specific receptor on the outside of the cell, allowing the iron to be transported into the cell.

Iron Uptake



Siderophores and Receptor Sites.

Symbiotic nutrition(commensalism)

a) Mutualism: where both species are benefited. Example Rhizobium bacteria present in the leguminous plants.

b) Competition: It is the relationship between organisms where both species are harmed.

c) Predation: It is the relationship between organisms where one species is benefited and the other species is killed.

d) Parasitism: It is the relationship between organisms where one species is benefited and other species is harmed. Eg-Leech.

e) Amensalism: It is the relationship between organism where one species is harmed and other species is neutral.

f) Commensalism: It is the relationship between organism where one species is benefited and other species is neutral.

Thus Symbiotic Nutrition (Commensalism) is a type of relationship between two species where one is getting benefit from the other species and the other species derives neither benefit nor harm from former species.

5) Example of such nutrition are:

S.no.	Example	Species getting benefit	Neutral Species
1.	Orchid and Mango	Orchid	Mango
2.	Barnacles and whale	Barnacle	Whale
3.	Cattle egret and Grazing cattle	Cattle egret	Grazing Cattle
4.	Sea anemone and clown fish	Clown fish	Sea anemone

Note: Commensalism and Mutualism are different, here in commensalism one species is neutral is getting no benefit from other species but in mutualism both need to get some benefit from each other like lichen where photosynthesising algae or cyanobacteria makes food and fungi provide shelter.

Some examples of symbiotic nutrition include:

Lichens

Formed by the association of algae and fungi, where the algae provide food through photosynthesis, while fungi offer protection and moisture to the algae.

Mycorrhizae

Fungi are present in the roots of plants, called VAM (Vascular arbuscular Mycorrhizae), and absorb nutrients from the soil.

Other examples of symbiotic relationships include rhizobium and leguminous plants, and Anabaena and the azolla.

In a **symbiotic relationship**, organisms share their **shelter** and **nutrients** among them. A certain type of fungi that live inside the plant's roots provide water and nutrients to the plant, and the fungi also obtain certain nutrients from the plant.

Lichens are an association of **alga** and **fungus** living together for their mutual benefits. The **fungus** provides **water**, **minerals** and **shelter** required by the **alga**. In return, **algae** provides **food** to the **fungus**.
Note: In alga, the presence of **chlorophyll** pigment enables it to prepare its food by **photosynthesis**.

Lichens

In **leguminous** plants, absorption of nitrogen is facilitated by **rhizobium** bacteria. This bacteria lives in the **root nodules** of leguminous plants. It converts **gaseous nitrogen** into a usable form and release it into the soil. Plants **absorb** this **nitrogen** in the soil for their **growth**. In return, **rhizobium** bacteria gets their **food** and **shelter** from the **plant**.

Nodules of the leguminous plant

References:

•A Text Book of Microbiology, revised Edition, R.C. Dubey , D.K. Maheshwari

•https://www.studocu.com

•Prescott’s Microbiology 12th Edition

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Unit-II
Microbial Growth

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What is growth

- Growth is defined as orderly increase in the quantity of all the cellular components, which is followed by cell division, resulting in increase in cell number.

Growth of Microbes

- Increase in *number* of cells, not cell size
- One cell becomes colony of millions of cells

FACTORS REGULATING GROWTH

Nutrients: All organisms need carbon ,hydrogen ,oxygen and a source of electrons for their growth.

NUTRIENTS

MACRO NUTRIENTS

C,O,H,O,N,P,K,Ca,Mg and Fe required in relatively large amount.

MICRO NUTRIENTS

Mn,Zn,Co,Mo,Ni and Cu required in trace amounts often supplied in water or media components

Environmental factor

1.TEMPERATURE
2.GASEOUS REQUIREMENTS
3.HYDROGEN ION CONCENTRATION (pH)
4.MISCELLANEOUS PHYSICAL REQUIREMENTS
(Osmotic Pressure , radiation, atmospheric pressure)

GENERATION TIME OF BACTERIA

The *time* taken by the *bacteria* to double in number during a specified *time period* is known as the *generation time*. The *generation time* tends to vary with different organisms.

Generation Times

Bacterium	Medium	Generation Time (minutes)
Escherichia coli	Glucose-salts	17
Bacillus megaterium	Sucrose-salts	25
Streptococcus lactis	Milk	26
Streptococcus lactis	Lactose broth	48
Staphylococcus aureus	Heart infusion broth	27-30
Lactobacillus acidophilus	Milk	66-87
Rhizobium japonicum	Mannitol-salts-yeast extract	344-461
Mycobacterium tuberculosis	Synthetic	792-932
Treponema pallidum	Rabbit testes	1980

Bacterial Division

- Bacteria divide by binary fission
- Alternative means
  - Budding
  - Conidiospores (filamentous bacteria)
  - Fragmentation

Nutritional Categories

- Carbon sources
  - CO_2 = autotroph
  - organic = heterotroph
- Energy sources
  - sunlight = phototroph
  - organic = chemotroph

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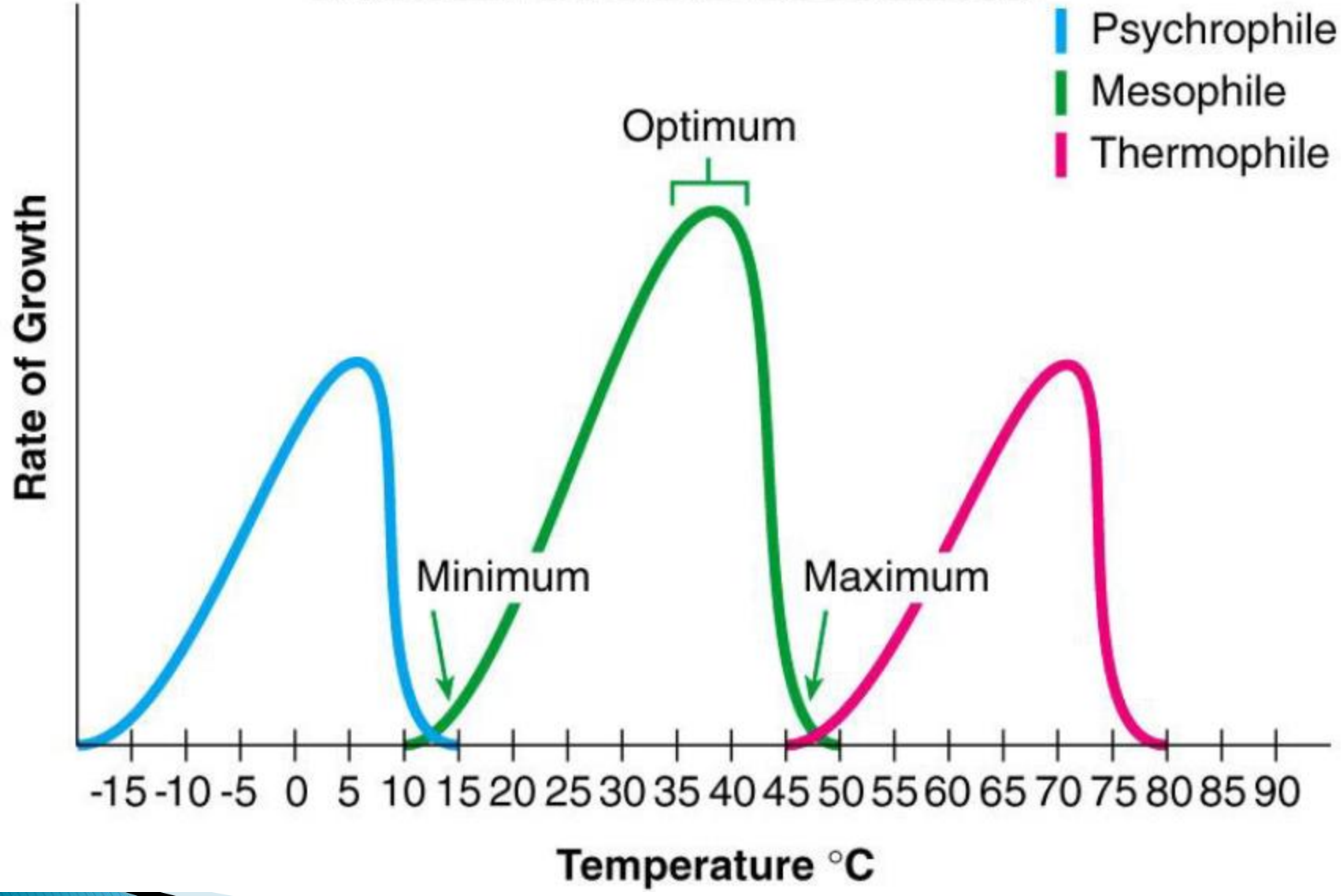

Oxygen Requirements

- Obligate aerobes – require O_2
- Facultative anaerobes – can use O_2 but also grow without it
- Obligate anaerobes – die in the presence of O_2

pH

- Most bacteria grow between pH 6.5 and 7.5
- Acid (below pH 4) good preservative for pickles, sauerkraut, cheeses
- Acidophiles can live at low pH

Measuring Bacterial Growth

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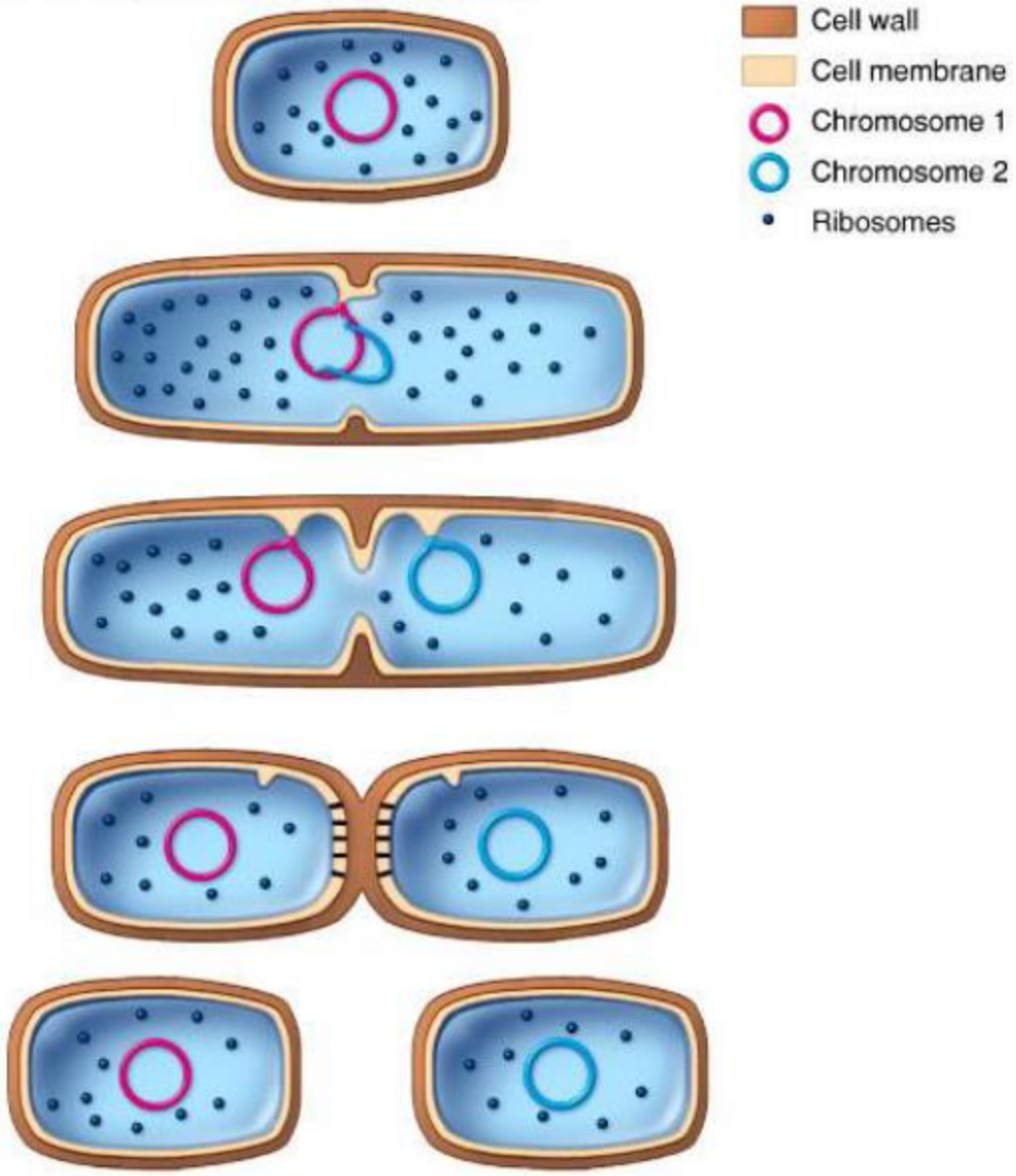
(a) A young cell at early phase of cycle.

(b) A parent cell prepares for division by enlarging its cell wall, cell membrane, and overall volume. Midway in the cell, the wall develops notches that will eventually form the transverse septum, and the duplicated chromosome becomes affixed to a special membrane site.

(c) The septum wall grows inward, and the chromosomes are pulled toward opposite cell ends as the membrane enlarges. Other cytoplasmic components are distributed (randomly) to the two developing cells.

(d) The septum is synthesized completely through the cell center, and the cell membrane patches itself so that there are two separate cell chambers.

(e) At this point, the daughter cells are divided. Some species will separate completely as shown here, while others will remain attached, forming chains or doublets, for example.



Measuring Bacterial Growth

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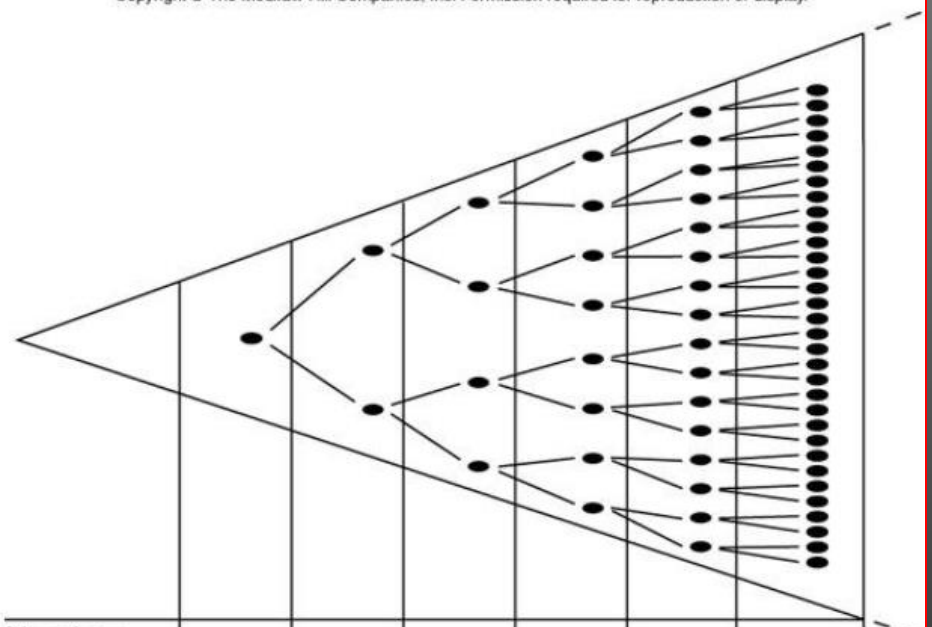

Number of cells	1	2	4	8	16	32
Number of generations		1	2	3	4	5
Exponential value		2^1 (2×1)	2^2 (2×2)	2^3 (2×2×2)	2^4 (2×2×2×2)	2^5 (2×2×2×2×2)

Plotting growth on graphs

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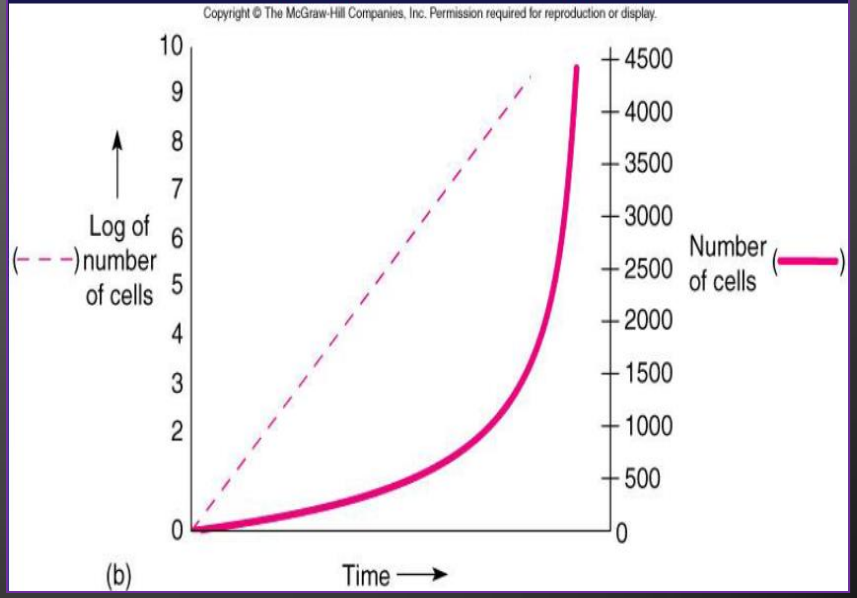
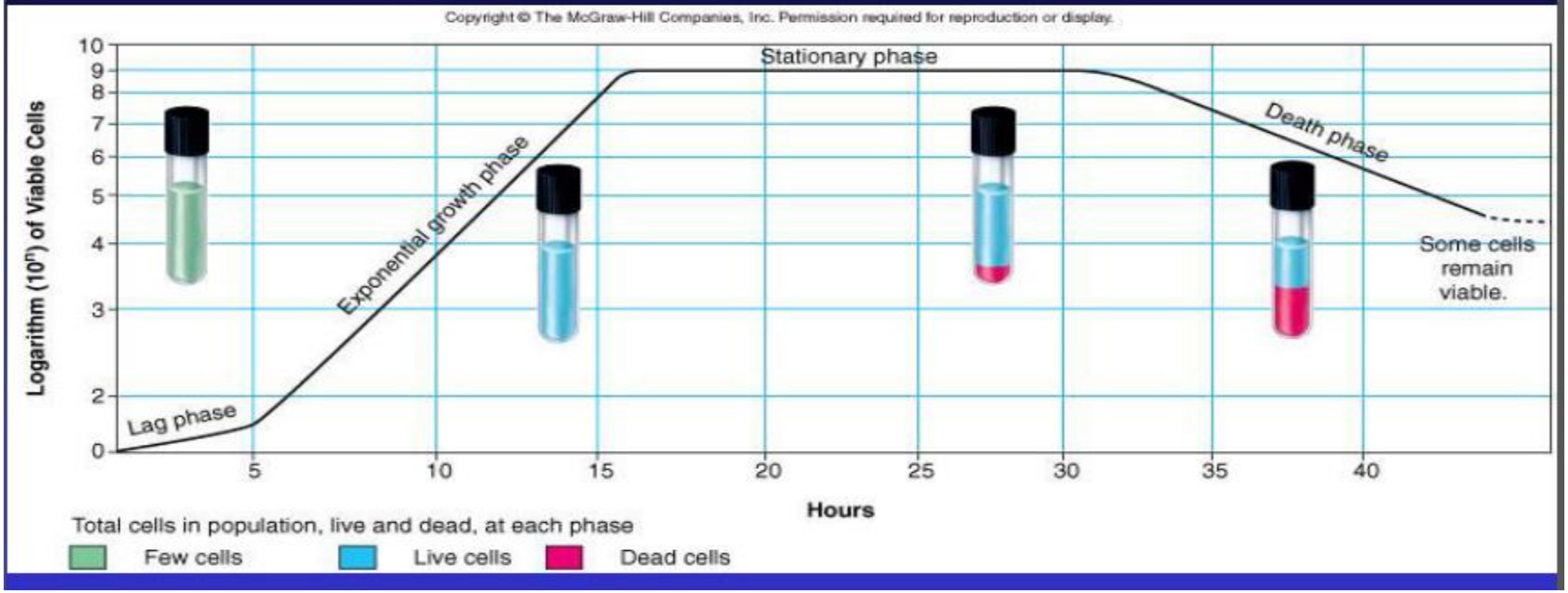


Standard Growth Curve



Phases of Growth

- Lag phase – making new enzymes in response to new medium
- Log phase – exponential growth
  - –Desired for production of products
  - –Most sensitive to drugs and radiation during this period

Phases of Growth

- Stationary phase –
  - –nutrients becoming limiting or waste products becoming toxic
  - – death rate = division rate
- Death phase – death exceeds division

previous growth

may be absent.

numbe
(exponentially) and finally

g
Staphylococcus aureus

environment for the

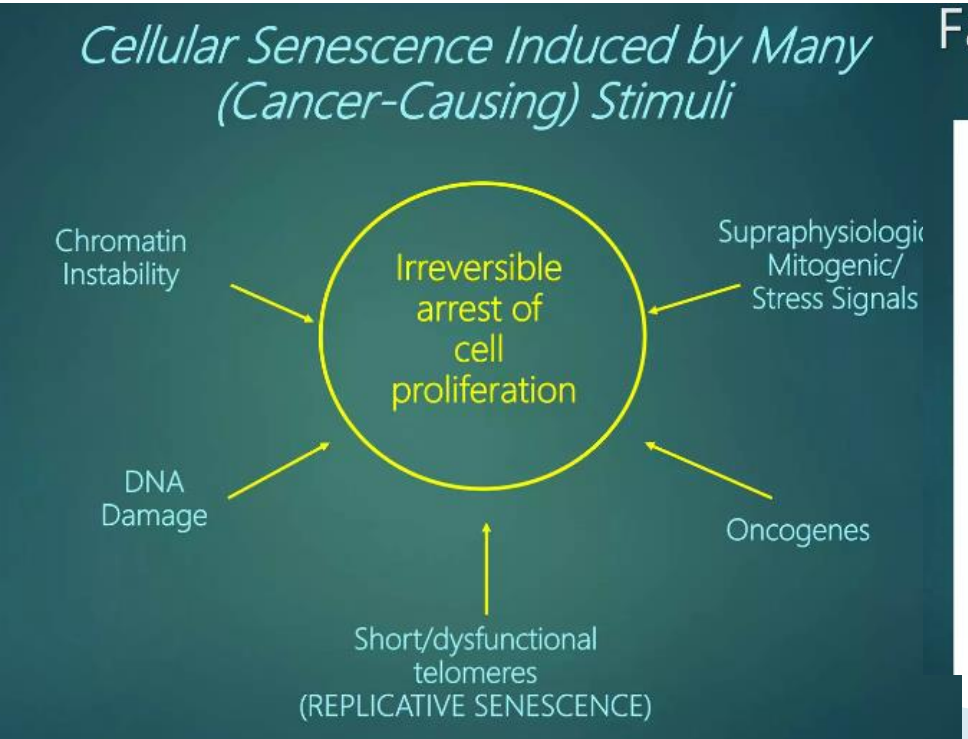
exponential ph

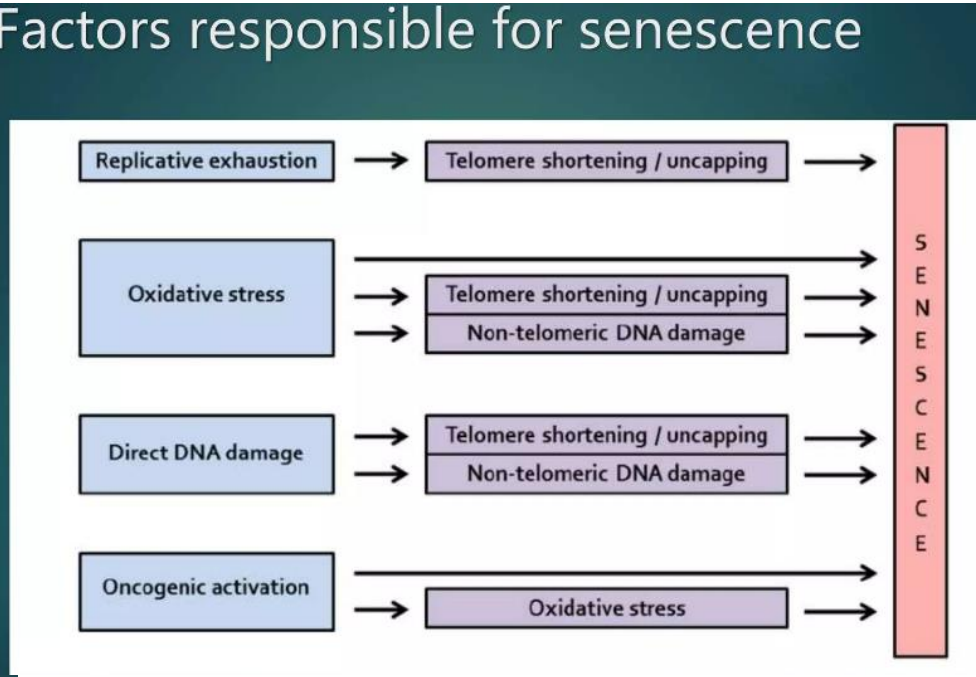
activities as usual

The number of dea

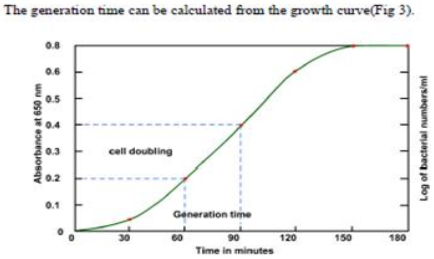
Senescence and Death Phase

- Two alternative hypotheses
  - cells are Viable But Not Culturable (VBNC)
    - cells alive, but dormant, capable of new growth when conditions are right
- Programmed cell death
  - fraction of the population genetically programmed to die (commit suicide)





Measurement of Growth Rate and Generation Time



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Calculation of the growth rate constant

Let N_0 = the initial population number

N_t = the population at time t

n = the number of generations in time t

For populations reproducing by binary fission

$$N_t = N_0 \times 2^n$$

Solving for n, the number of generations, where all logarithms are to the base 10,

$$\log N_t = \log N_0 + n \cdot \log 2, \text{ and}$$

$$n = \frac{\log N_t - \log N_0}{\log 2} = \frac{\log N_t - \log N_0}{0.301}$$

The growth rate constant (k) is the number of generations per unit time $\left(\frac{n}{t}\right)$. Thus

$$k = \frac{n}{t} = \frac{\log N_t - \log N_0}{0.301t}$$

Calculation of generation (doubling) time

If a population doubles, then

$$N_t = 2N_0$$

Substitute $2N_0$ into the growth rate constant equation and solve for

$$k = \frac{\log (2N_0) - \log N_0}{0.301g} = \frac{\log 2 + \log N_0 - \log N_0}{0.301g}$$

$$k = \frac{1}{g}$$

The generation time is the reciprocal of the growth rate constant.

$$g = \frac{1}{k}$$

Measuring Growth

- Direct methods – count individual cells
- Indirect Methods – measure effects of bacterial growth

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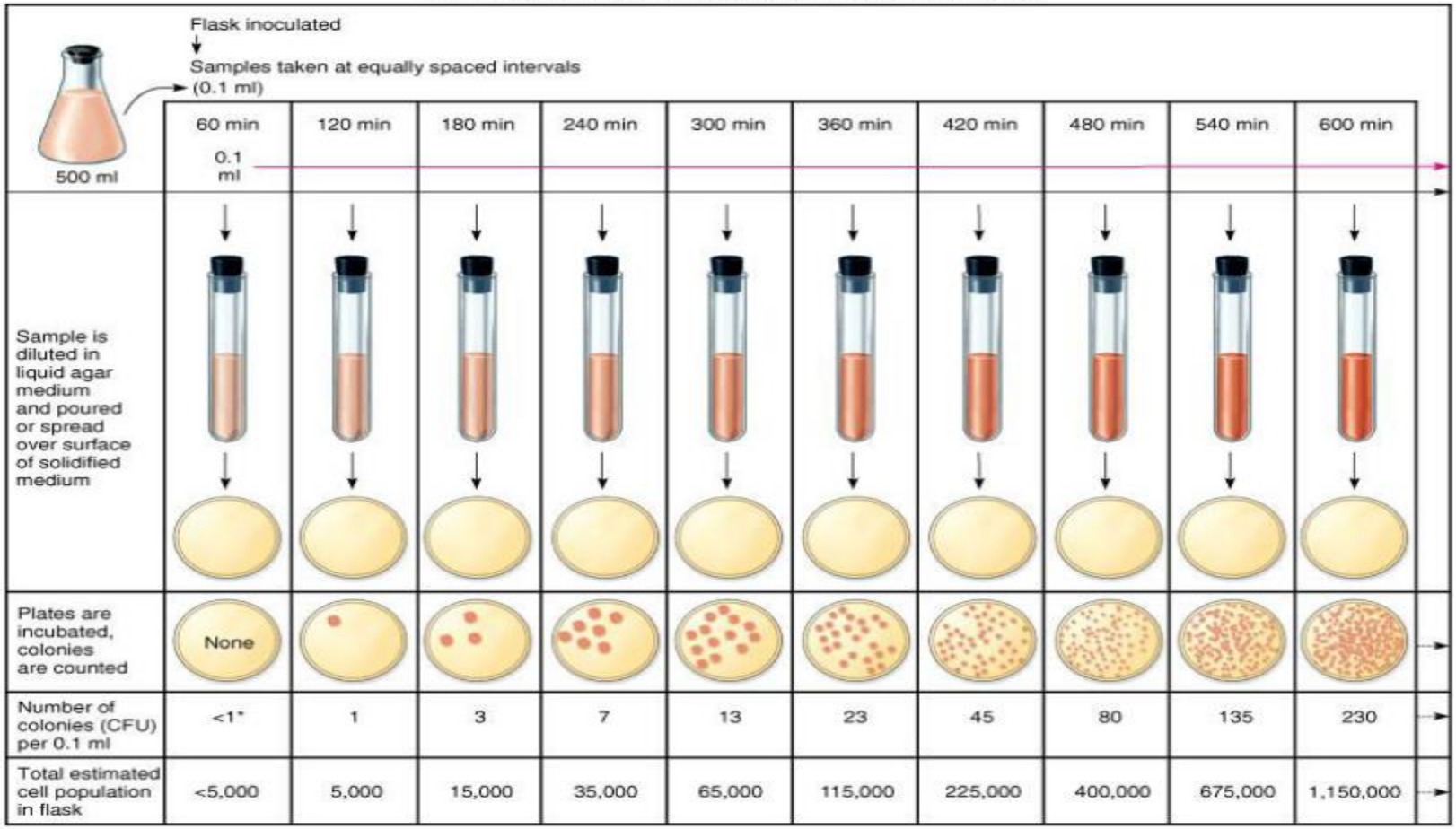

* Only means that too few cells are present to be assayed.

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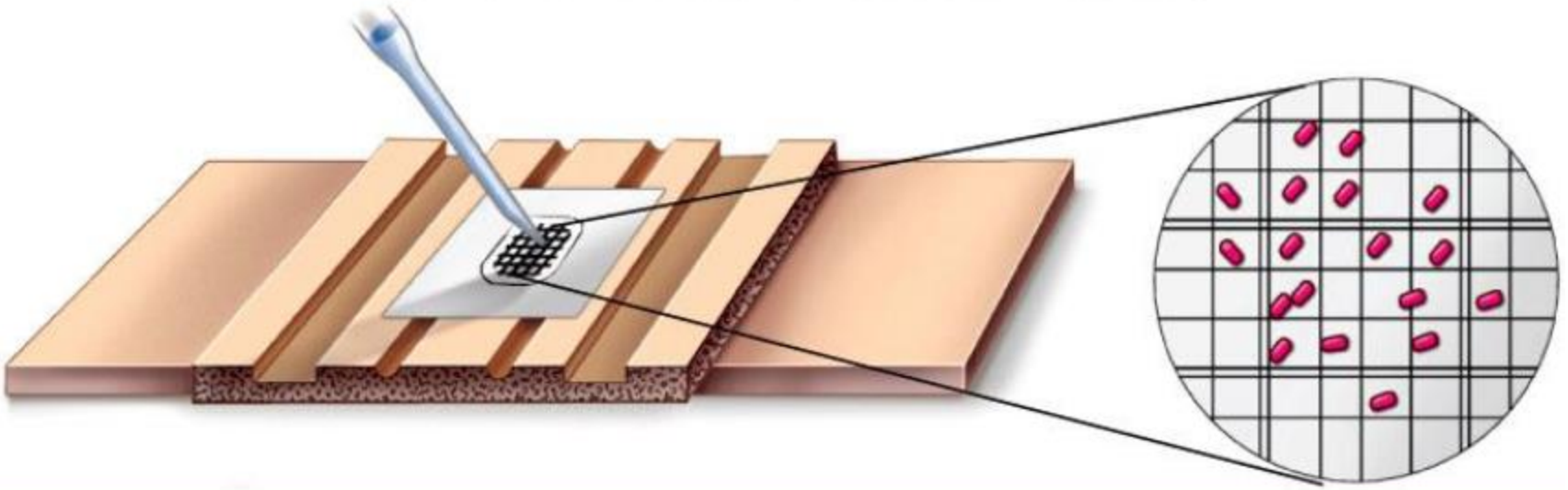

Turbidity

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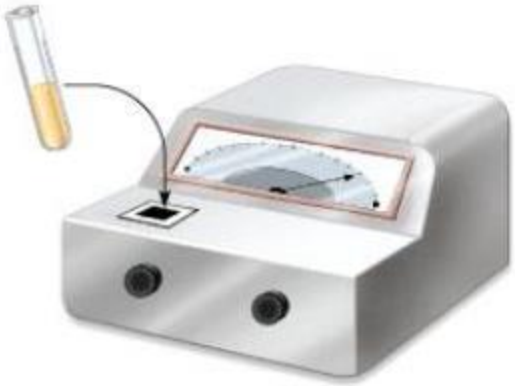
(a)



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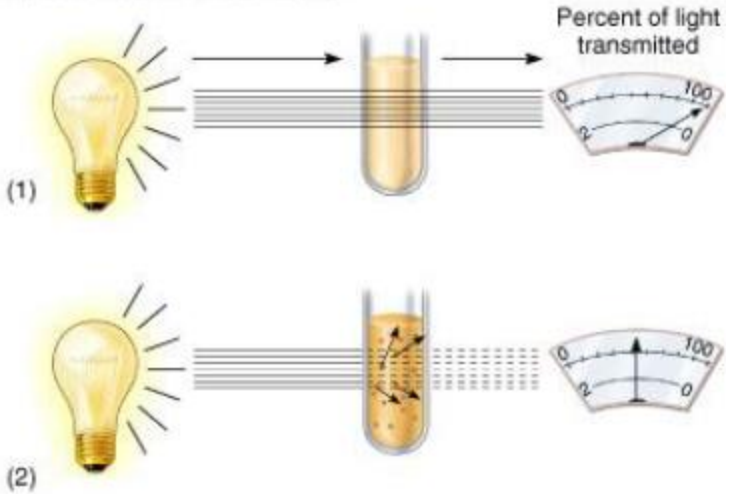


Metabolic Activity

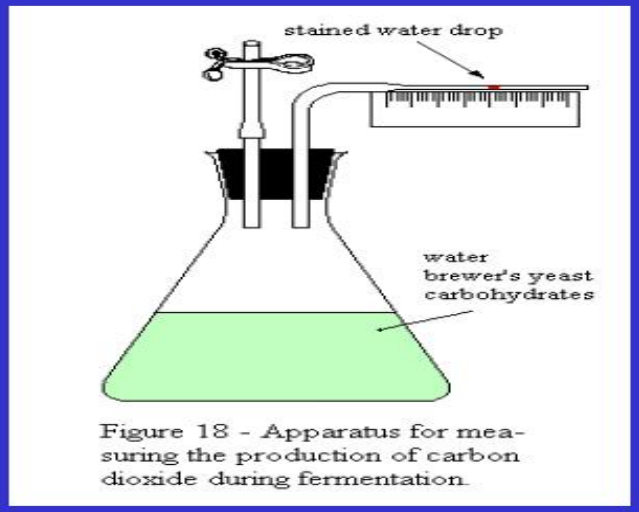
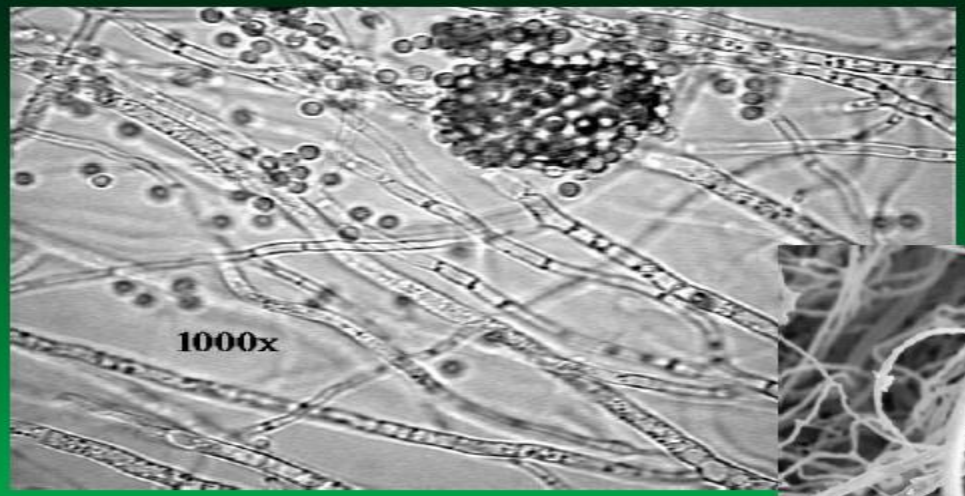
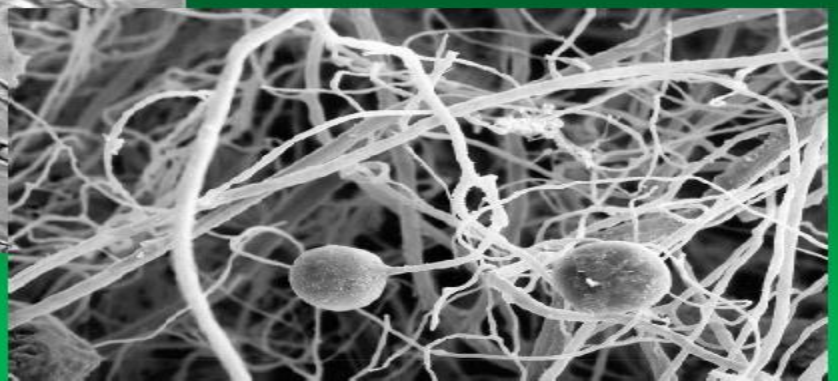
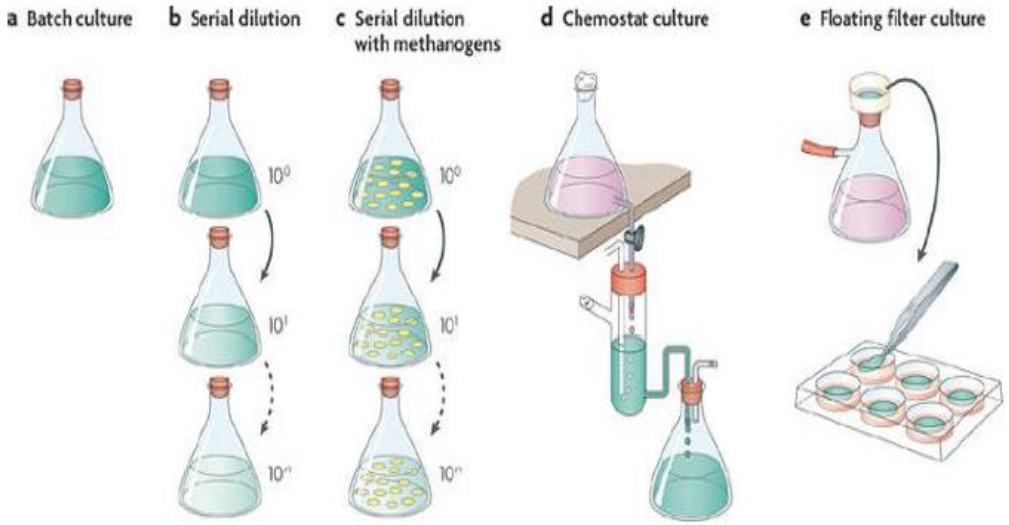


Figure 18 - Apparatus for measuring the production of carbon dioxide during fermentation.

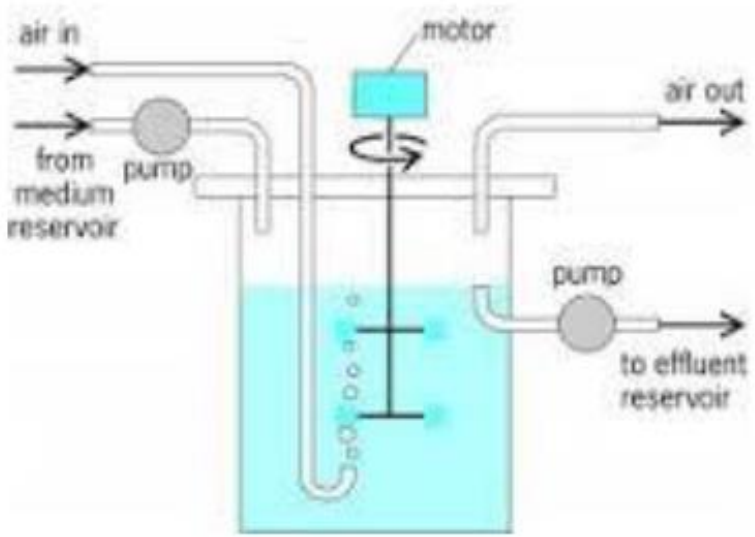
Dry Weight







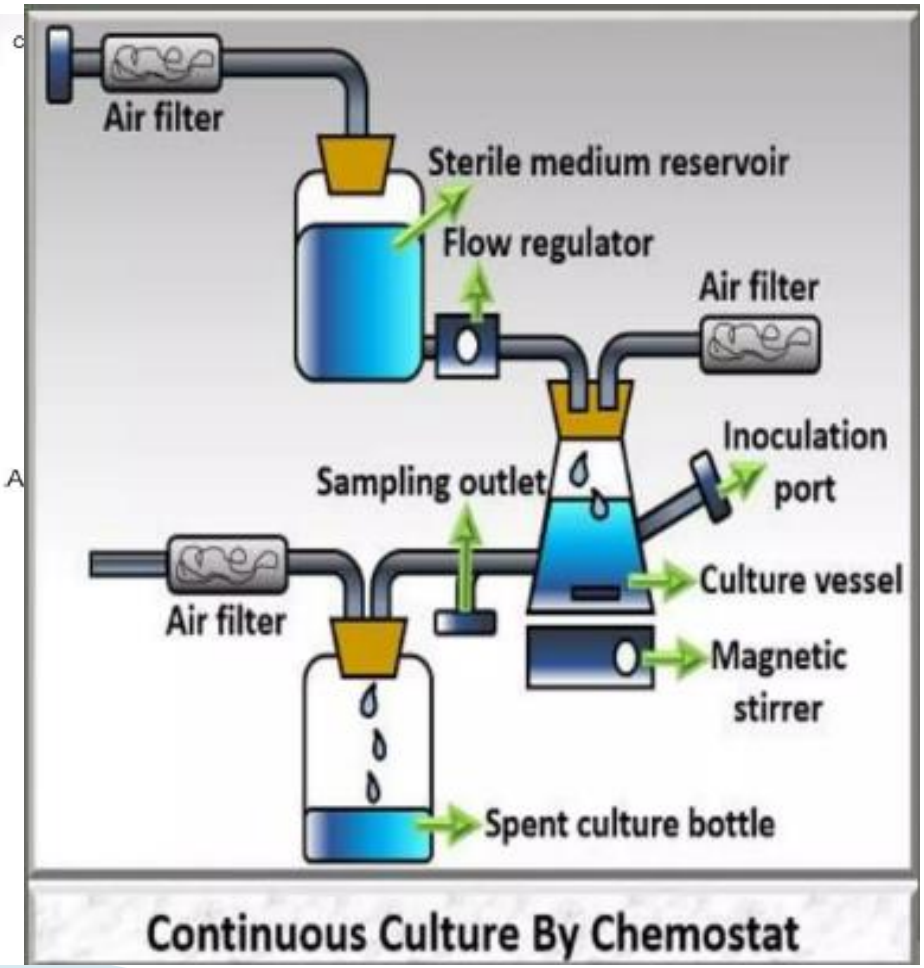
Nature Reviews | Microbiology



constant.

The Chemostat

- Rate of incoming medium = rate of removal of medium from vessel
- An essential nutrient is in limiting quantities



Continuous Culture By Chemostat

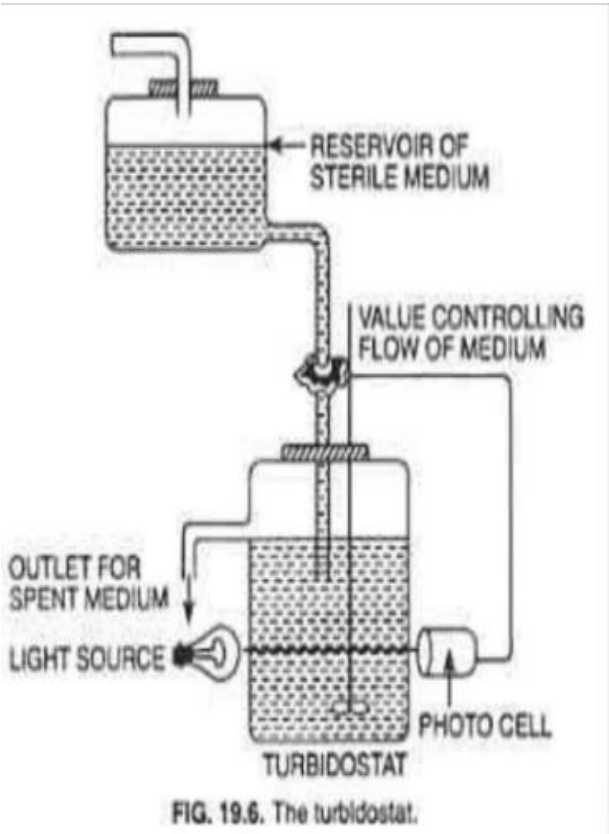
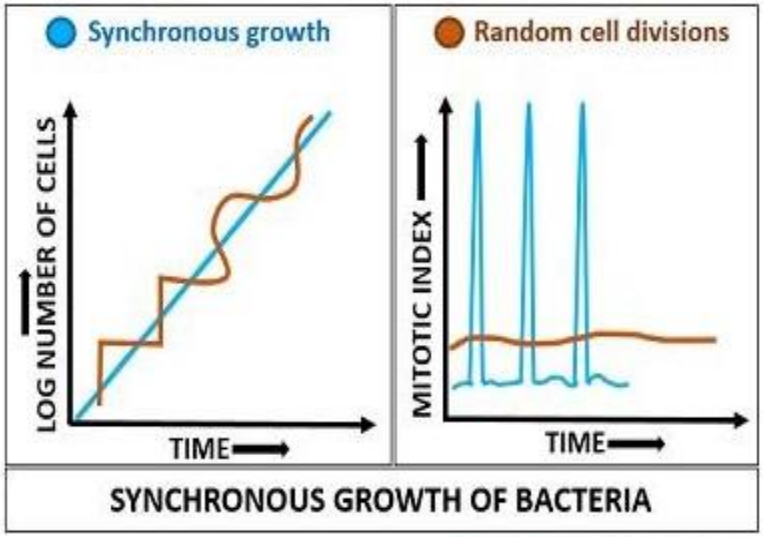


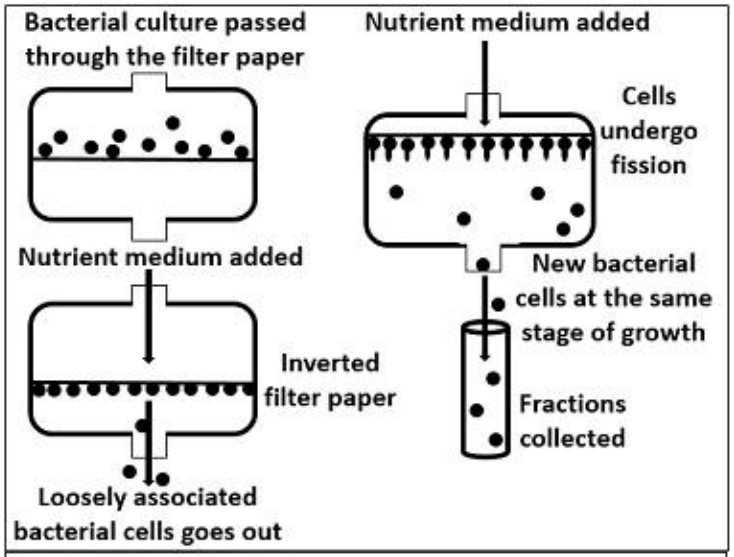
FIG. 19.6. The turbidostat.

density for
such as dielectric permittivity.

Cas
advantage of the



SYNCHRONOUS GROWTH OF BACTERIA



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