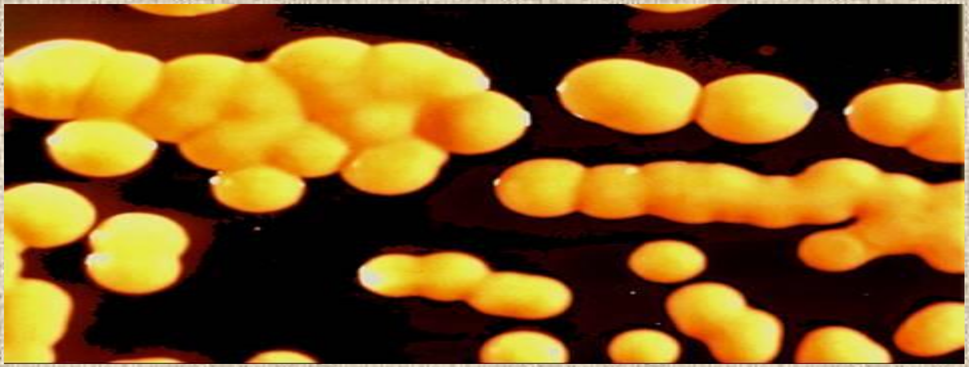
Physiology of Bacteria. Growth and reproduction of Bacteria



Presented by:
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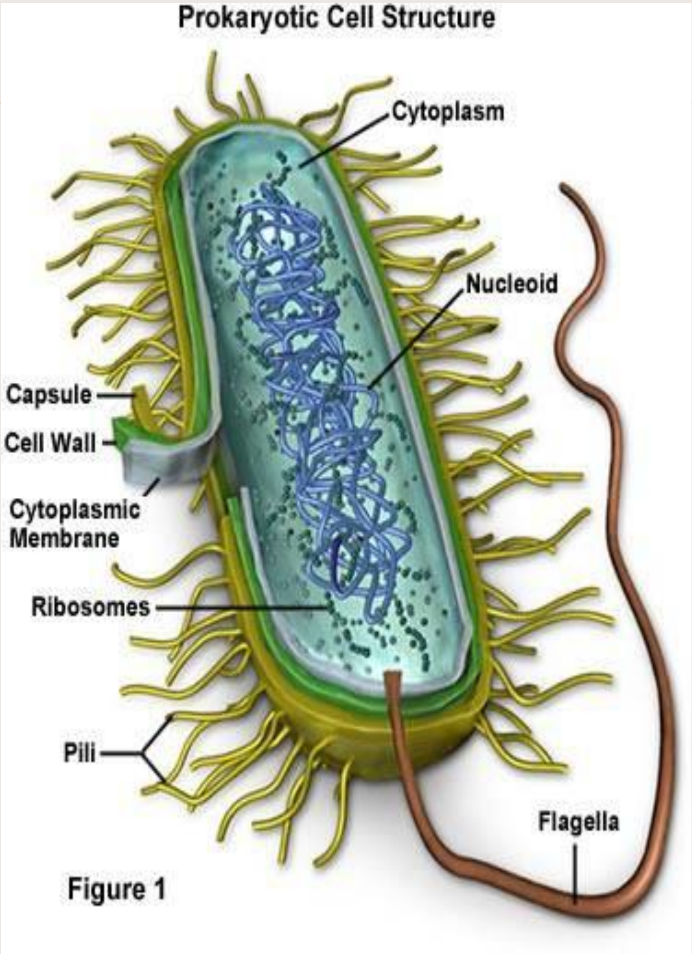
Metabolism refers to all the biochemical reactions that occur in a cell or organism.

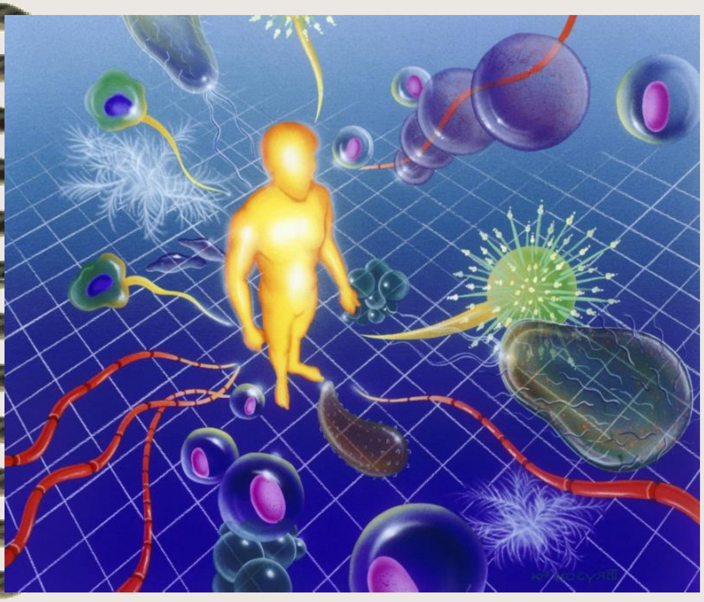
The study of bacterial metabolism focuses on the chemical diversity of substrate **oxidations and dissimilation reactions** (reactions by which substrate molecules are broken down), which normally function in bacteria to **generate energy.**



- **Metabolism** is the process of building up chemical compounds in the cell and their breaking down during activity to receive the required energy and the building elements.
- **Metabolism** comprises of ***anabolism*** (assimilation) and ***catabolism*** (dissimilation)

Chemically, bacteria consist of:

- *Water (75-85%) – bound water and free water*

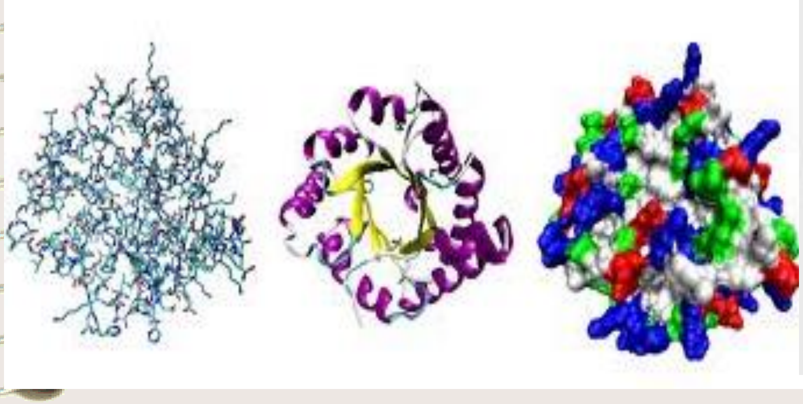


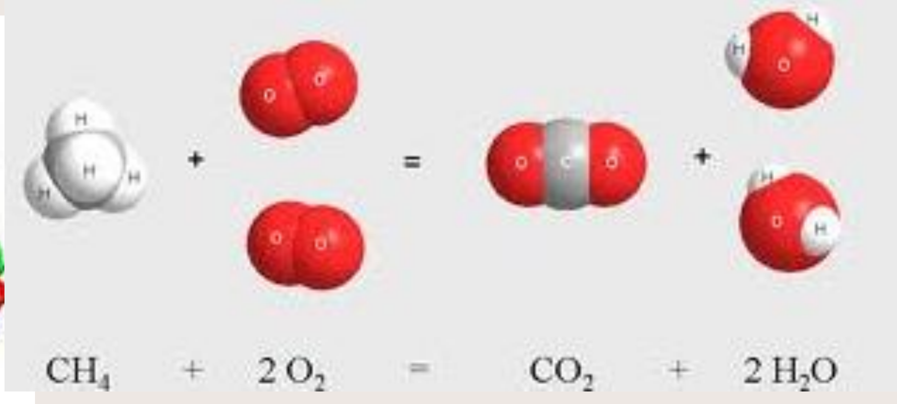
- *Dry matter (15-25%) – organic part and mineral substances (inorganic part)*

Dry matter

- ***Organic part***
- ***proteins – 50-80%***
- ***nucleic acid – 10-30%***
- ***carbohydrates – 12-18%***
- ***polysaccharides – 3-5%***
- ***lipids – 5-10%.***

- ***Inorganic part***
- ***nitrogen (N), carbon (C), oxygen (O), hydrogen (H), phosphorus (P), sulfur (S), sodium (Na), magnesium (Mg), potassium (K), calcium (Ca), iron (Fe) and other***





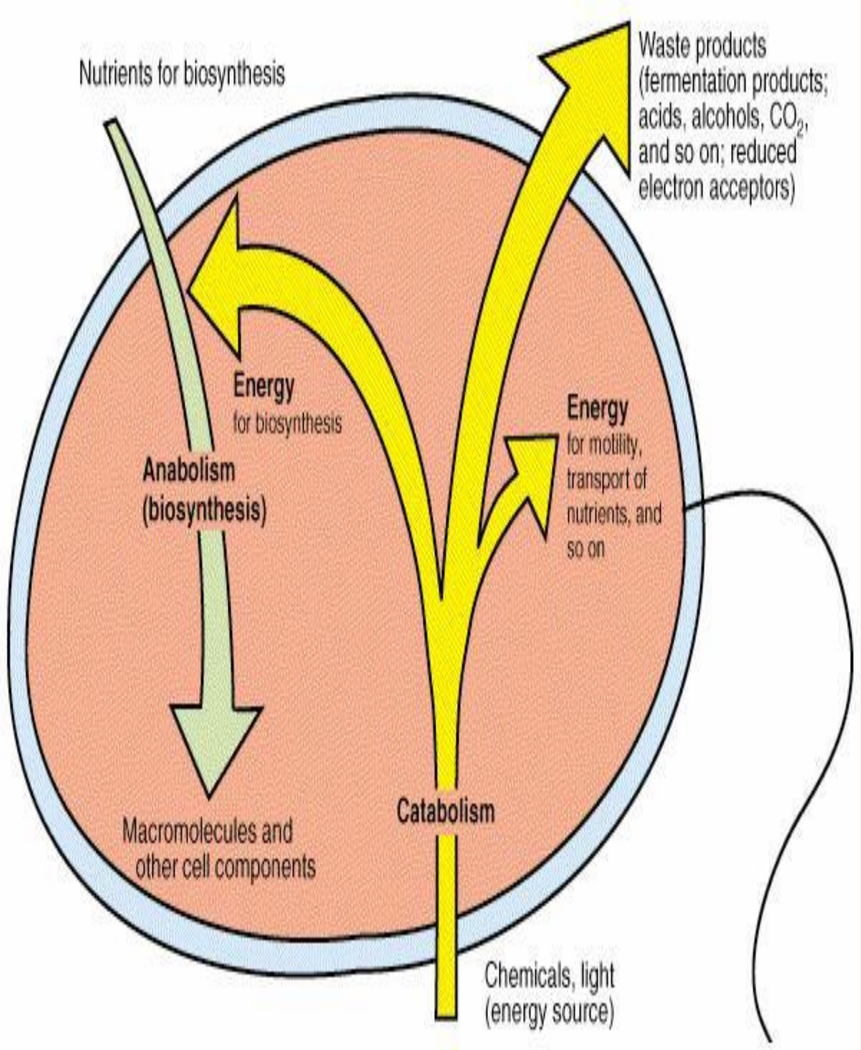
Microbial metabolism

1. Catabolism (Dissimilation)

- Pathways that **breakdown** organic substrates (carbohydrates, lipids, & proteins) to yield metabolic energy for **growth** and **maintenance**.

2. Anabolism (Assimilation)

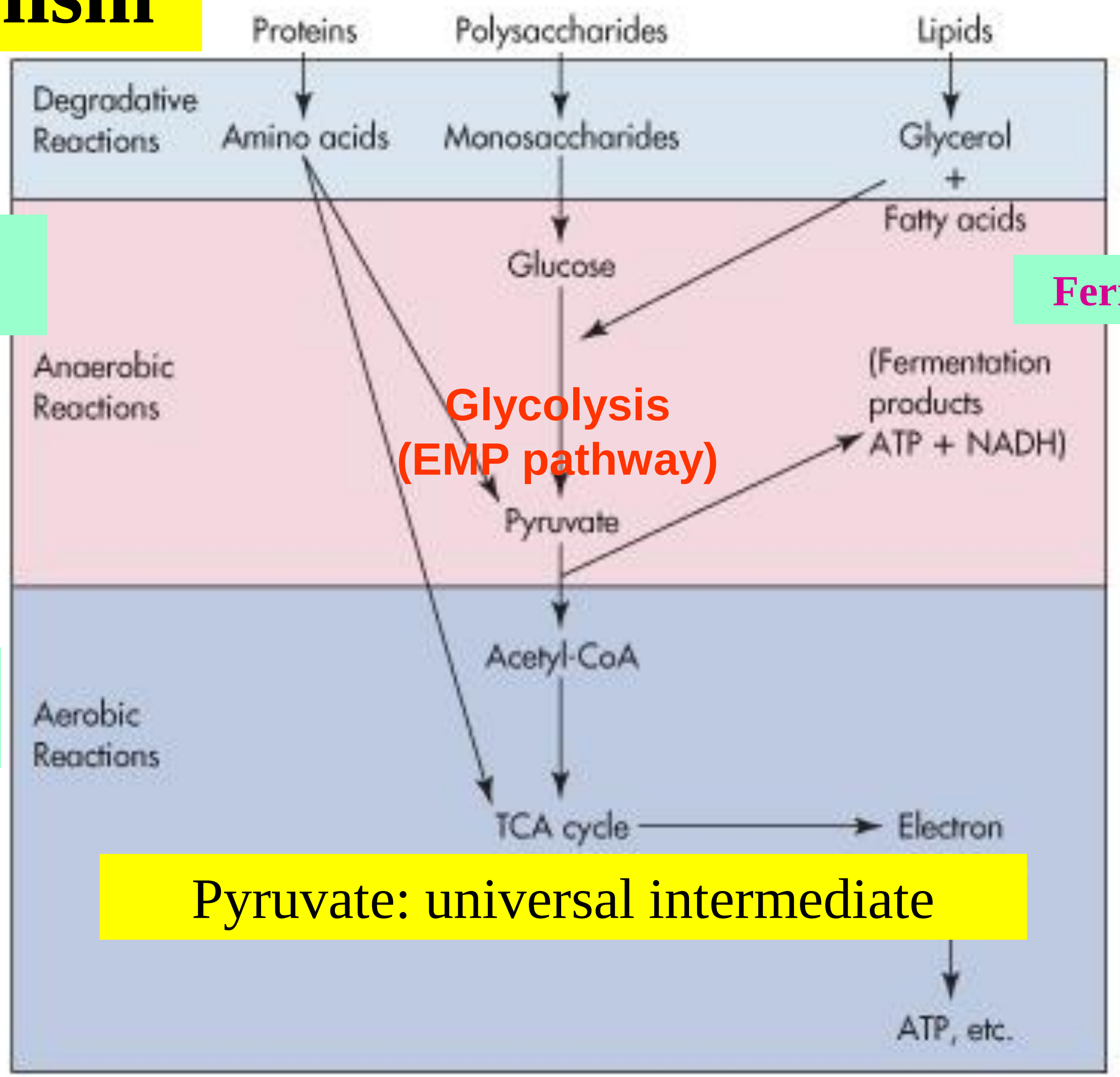
- Assimilatory pathways for the formation of key intermediates and then to end products (**cellular components).**



Catabolism

Substrate-level phosphorylation

Aerobic respiration



The bacterial cell is a highly specialized energy transformer. Chemical energy generated by substrate oxidations is conserved by formation of high-energy compounds such as adenosine diphosphate (ADP) and adenosine triphosphate (ATP) or compounds containing the thioester bond

O

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(R –C ~ S – R), such as acetyl ~ S-coenzyme A

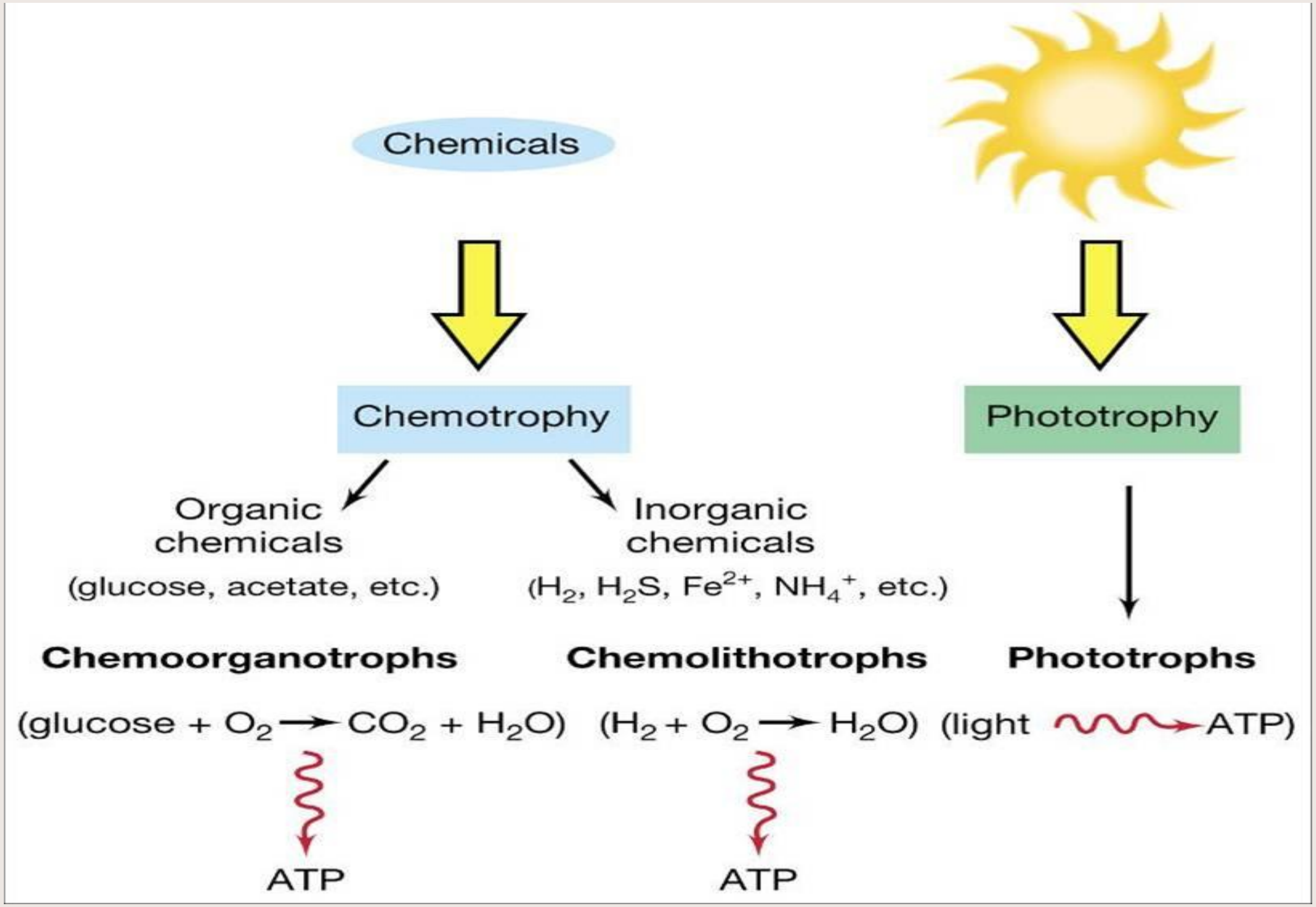
Classification of bacteria based on nutritional requirements

- ***Autotrophs*** are free-living, most of which can use carbon dioxide as their carbon source. The energy can be obtained from:
- sunlight – **protoautotrophs** (get energy from photochemical reactions)
- inorganic compounds, by oxidation – **chemoautotrophs** (get energy from chemical reactions)
- ***Heterotrophs*** are generally parasitic bacteria, requiring more complex organic compounds than carbon dioxide, e.g. sugars, as their source of carbon and energy.

Energy Requirements

Oxidation of organic compounds - **Chemotrophs**

Sunlight - **Phototrophs**



Metabolic Requirements

Carbon source

- Autotrophs (lithotrophs): use CO_2 as the C source
  - Photosynthetic autotrophs: use light energy
  - Chemolithotrophs: use inorganics
- Heterotrophs (organotrophs): use organic carbon (eg. glucose) for growth.
- Clinical Labs classify bacteria by the carbon sources (eg. Lactose) & the end products (eg. Ethanol,…).

Nitrogen source

Ammonium (NH_4^+) is used as the sole N source by most microorganisms. Ammonium could be produced from N_2 by nitrogen fixation, or from reduction of nitrate (NO_3^-)and nitrite (NO_2).

Metabolic Requirements

Sulfur source

A component of several coenzymes and amino acids. Most microorganisms can use sulfate (SO_4^{2-}) as the S source.

Phosphorus source

- A component of ATP, nucleic acids, coenzymes, phospholipids, teichoic acid, capsular polysaccharides; also is required for signal transduction.
- Phosphate (PO_4^{3-}) is usually used as the P source.

Mineral source

- Required for enzyme function.
- For most microorganisms, it is necessary to provide sources of K^+, Mg^{2+}, Ca^{2+}, Fe^{2+}, Na^+ and Cl^-.
- Many other minerals (eg., Mn^{2+}, Mo^{2+}, Co^{2+}, Cu^{2+} and Zn^{2+}) can be provided in tap water or as contaminants of other medium ingredients.
- Uptake of Fe is facilitated by production of siderophores (Iron-chelating compound, eg. Enterobactin).

Growth factors: organic compounds (e.g., amino acids, sugars, nucleotides, vitamines) a cell must contain in order to grow but which it is unable to synthesize. **Purines and pyrimidines**: required for synthesis of nucleic acids (DNA and RNA);

Amino acids: required for the synthesis of proteins;

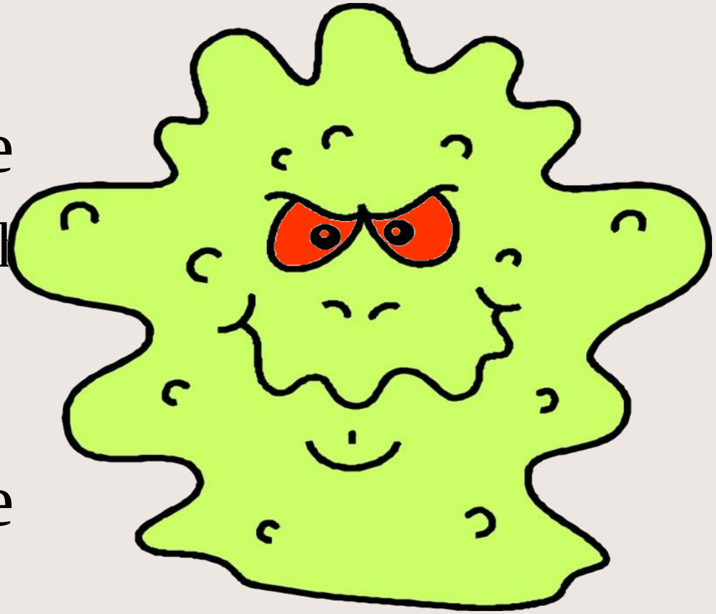
Vitamins: needed as coenzymes and functional groups of certain enzymes.

The basic requirements of culture media

- energy source;
- carbon source;
- nitrogen source;
- salts like sulphates, phosphates, chlorides and carbonates of sodium, potassium, magnesium, ferric, calcium and trace elements, like copper, etc.;
- satisfactory pH 7.2-7.6;
- growth factor like vitamins.

TEMPERATURE

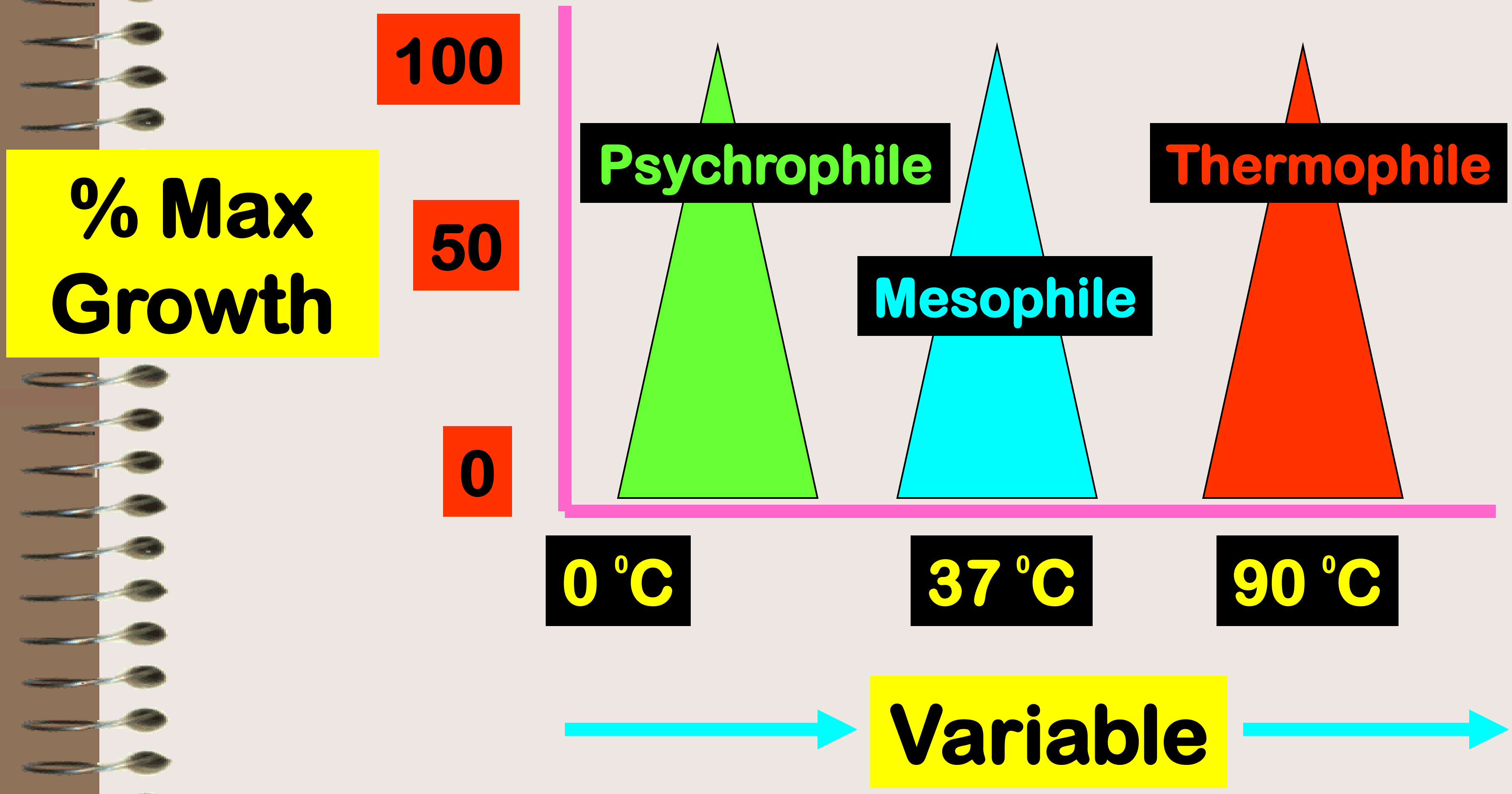
- One of the most important factors
- optimal growth temperature
  - temperature range at which th highest rate of reproduction occurs
- optimal growth temperature for human pathogens ????



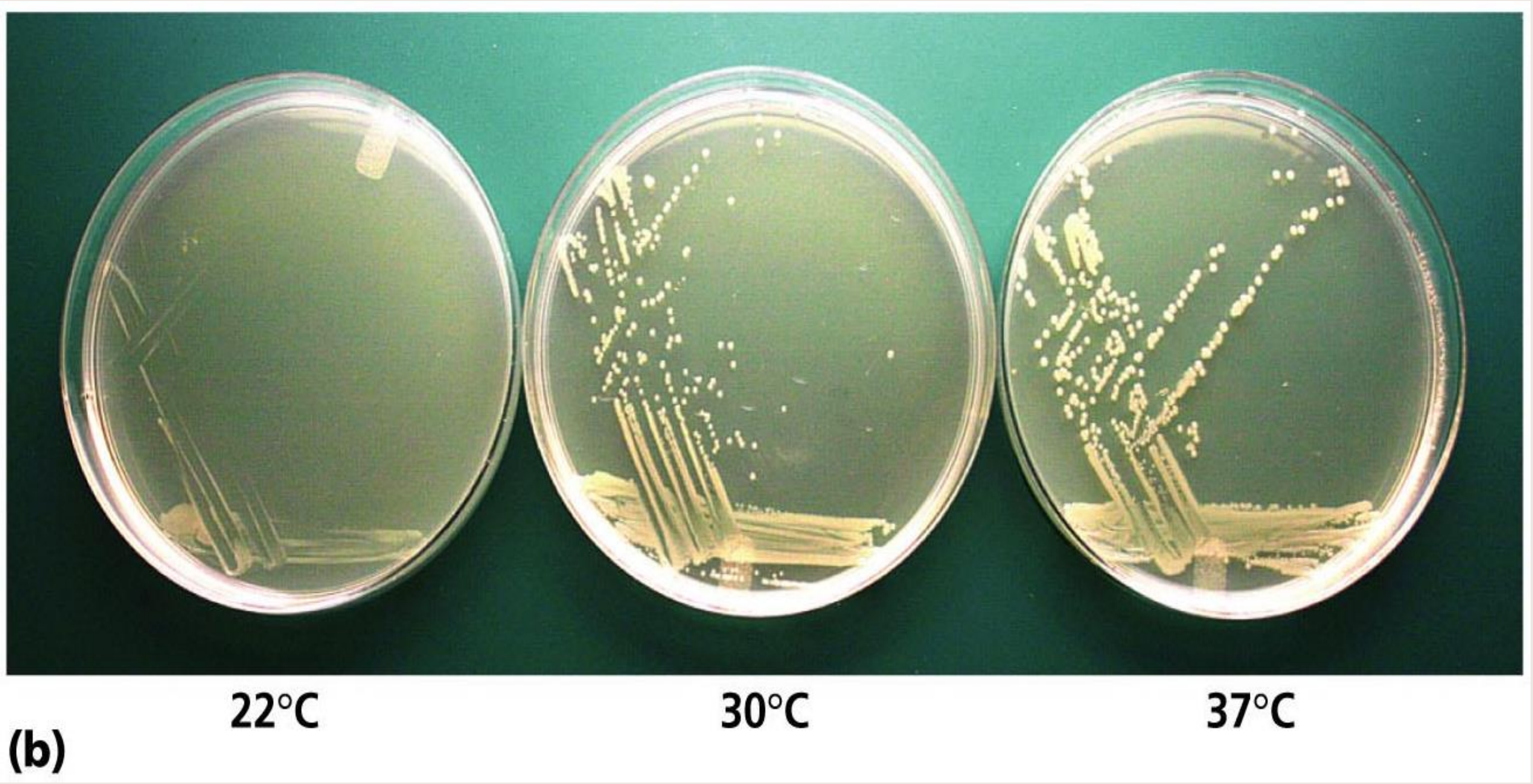
TEMPERATURE

- Microorganisms can be categorized based on their <u>optimal</u> temperature requirements
  - <u>Psychrophiles</u>
    - 0 - 20 ºC
  - <u>Mesophiles</u>
    - 20 - 40 ºC
  - <u>Thermophiles</u>
    - 40 - 90 ºC
- Most bacteria are mesophiles especially pathogens that require 37 ºC

BACTERIAL TEMPERATURE REQUIREMENTS



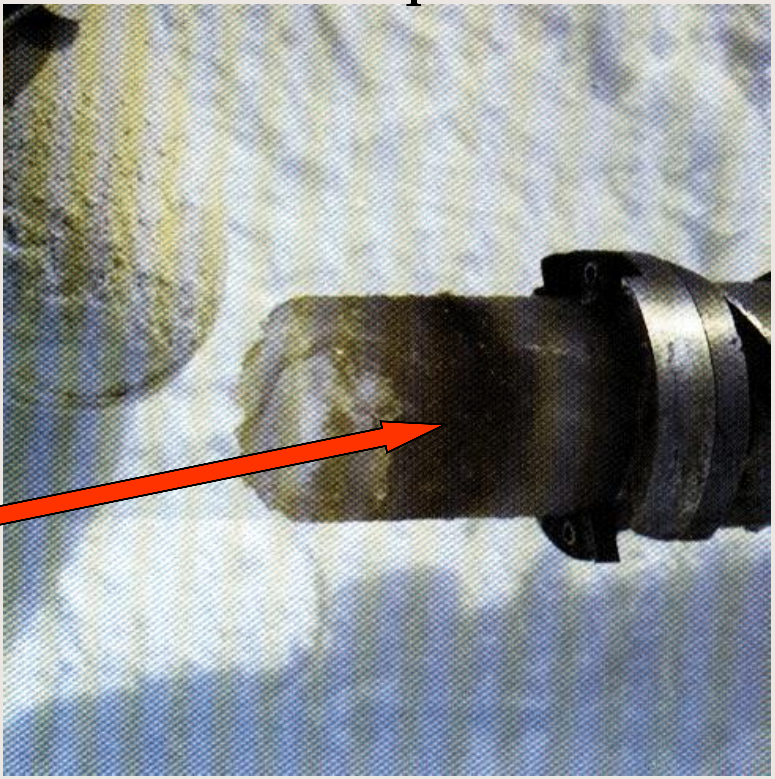
Effects of Temperature on Growth



TEMPERATURE

- <u>Psychrophiles</u>
  - some will exist below 0 ^{o}C if liquid water is available
    - oceans
    - refrigerators
    - freezers

Pigmented bacteria in Antarctic ice

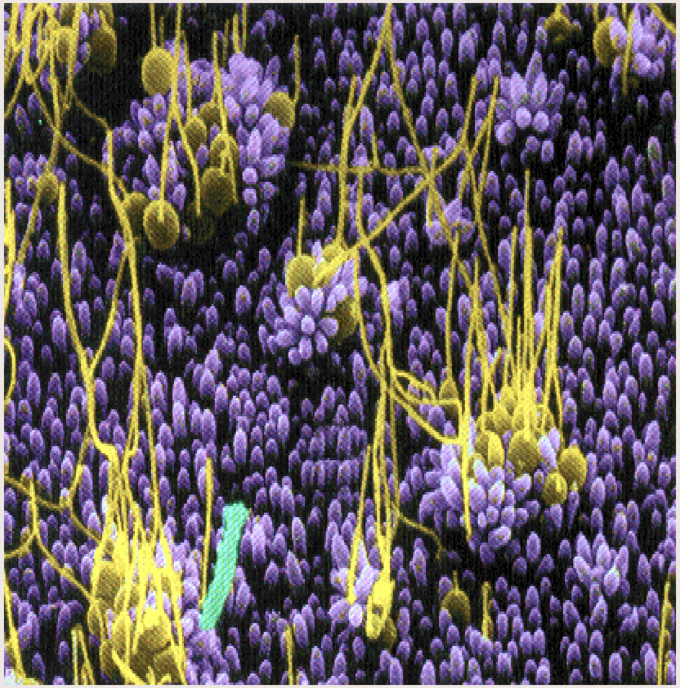


TEMPERATURE

- Mesophiles
  - most human flora and pathogens

TEMPERATURE

- Thermophiles
  - hot springs
  - effluents from laundromat
  - deep ocean thermal vents



Respiration in Bacteria

Obligate Aerobe

Microaerophile

Obligate Anaerobe

Facultative Anaerobe (Facultative Aerobe)

Aerotolerant Anaerobe

Capneic bacteria

Categories of Oxygen Requirement

Aerobe – utilizes oxygen and can detoxify it

- **obligate aerobe -** cannot grow without oxygen (Mycobacterium tuberculosis, Micrococcus spp., Bacillus spp., Pseudomonas spp.
- **facultative anaerobe** – utilizes oxygen but can also grow in its absence (Echericihia spp., Salmonella spp., Sta[phylococcus spp.)
- **microaerophylic** – requires only a small amount of oxygen (Helycobacter spp., Lactobacillus spp.)

Categories of Oxygen Requirement

Anaerobe – does not utilize oxygen

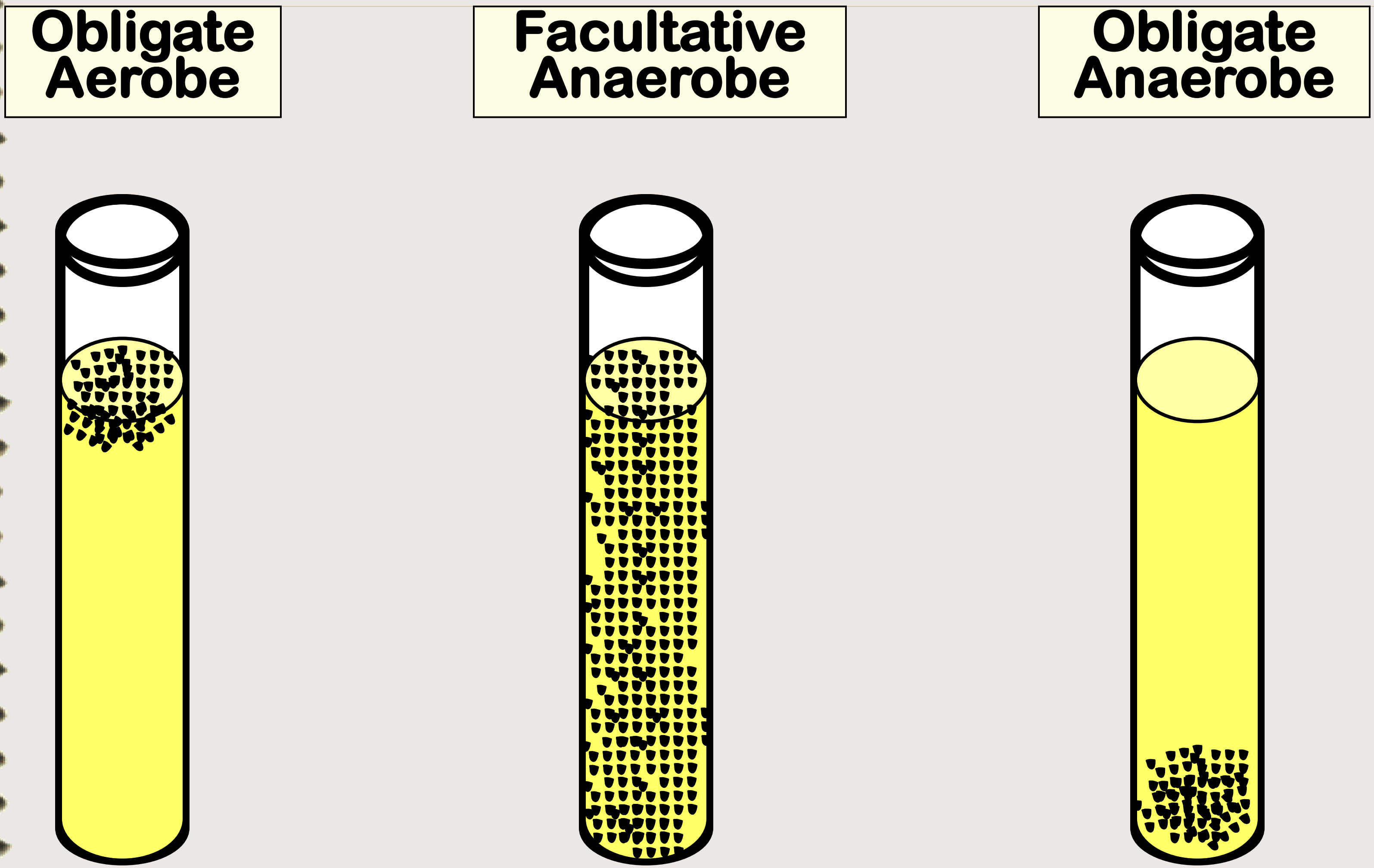
- **obligate anaerobe -** lacks the enzymes to detoxify oxygen so cannot survive in an oxygen environment (Clostridium spp., Bacteroides spp.)
- **aerotolerance anaerobes** – do no utilize oxygen but can survive and grow in its presence (Streptococcus pyogenes)

Carbon Dioxide Requirement

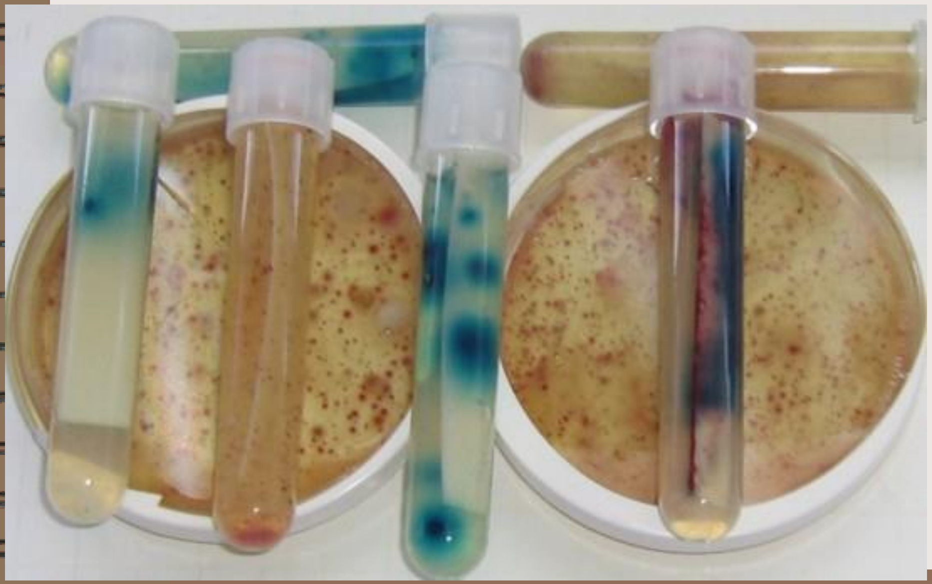
All microbes require some carbon dioxide in their metabolism.

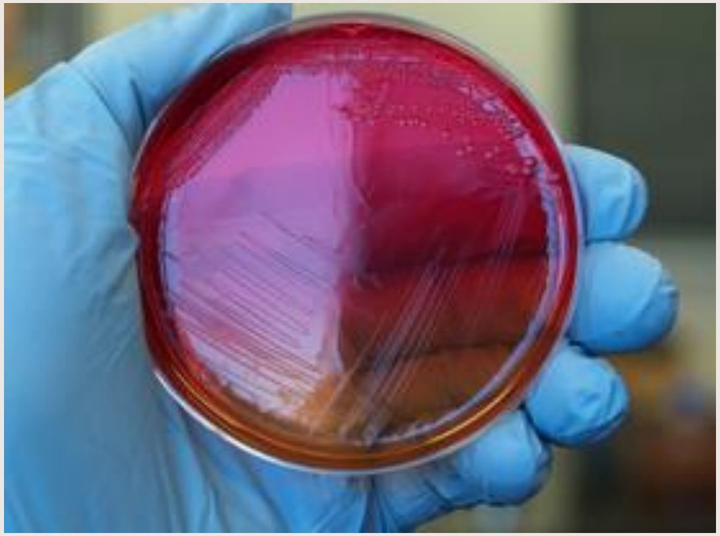
- **capneic** – grows best at higher CO_2 tensions than normally present in the atmosphere (Brucella abortus)

OXYGEN



- Aerobic and anaerobic - according to type of respiration bacteria subdivided into 4 groups:
- *Obligate aerobes* ***(Brucella)***
- *Microaerophils* ***(H.pylori)***
- *Obligate Anaerobes* ***(C.tetani)***
- *Facultative Anaerobes* ***(E.coli)***





Enzymes and Their Role in Metabolism

Enzymes, organic catalysts of a highly molecular structure, are produced by the living cell. They are of a **protein nature**, are **strictly specific in action**, and play an important part in the metabolism of micro-organisms. Their specificity is associated with active centres formed by a group of amino acids.

Some enzymes are excreted by the cell into the environment (exoenzymes) for breaking down complex colloid nutrient materials while other enzymes are contained inside the cell (endoenzymes).

Bacterial enzymes are subdivided into some groups:

1. Hydrolases which catalyse the breakdown of the link between the carbon and nitrogen atoms, between the oxygen and sulphur atoms, binding one molecule of water (esterases. glucosidases, proteases. amilases, nucleases, etc.).

2. Transferases perform catalysis by transferring certain radicals from one molecule to another (transglucosidases, transacylases. transaminases).

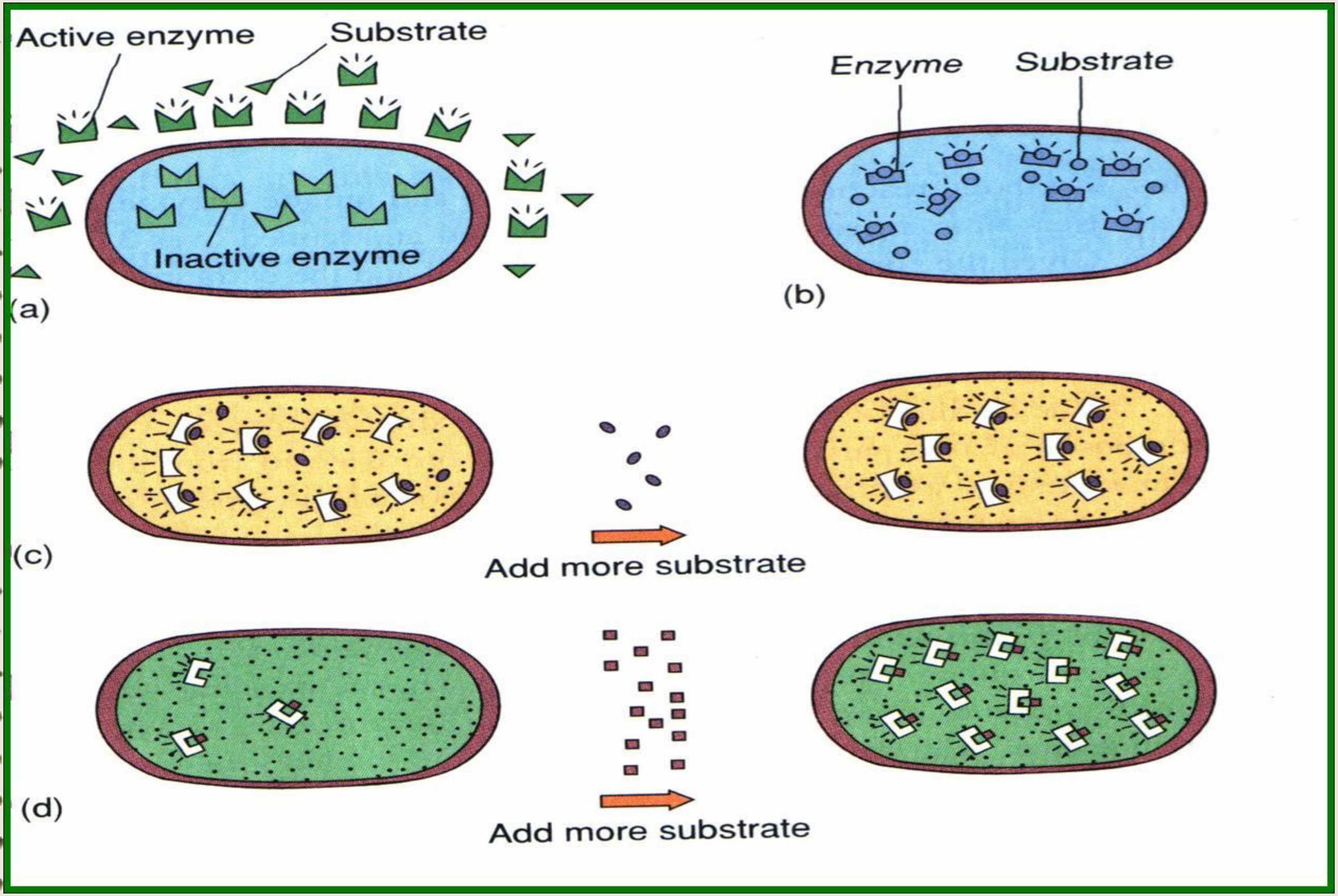
3. Oxidative enzymes (**oxyreductases**) which catalyse the oxidation-reduction processes (oxidases, dehydrogenases, peroxidases, catalases).

4. Isomerases and racemases play an important part in carbohydrate metabolism. Rearrangement atoms of a molecule.

5. Lyases (remove chemical groups from molecules without adding water).

6. Lygases (join two molecules together and usually require energy from ATP).

Enzymes



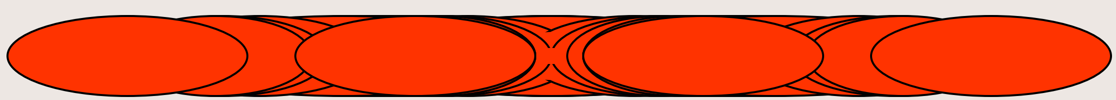
Active enzyme
Substrate
Inactive enzyme
(a)
Enzyme
Substrate
(b)
(c)
Add more substrate
(d)
Add more substrate

Metabolism Results in Reproduction

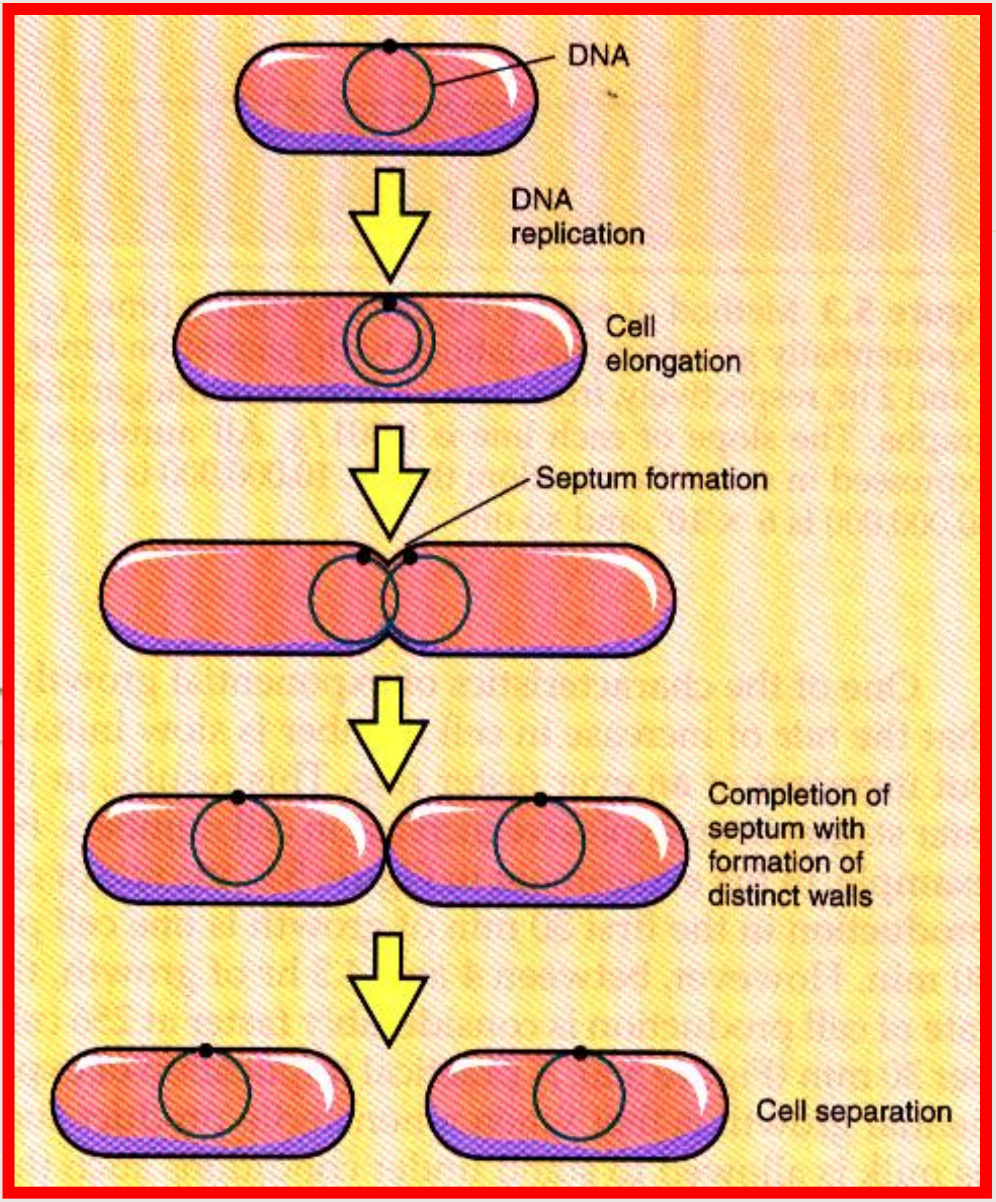
- Microbial growth – an increase in a population of microbes rather than an increase in size of an individual
- Result of microbial growth is discrete colony – an aggregation of cells arising from single parent cell
- Reproduction results in growth

BINARY FISSION

- division exactly in half
- most common means of bacterial reproduction
  - forming two equal size progeny
  - genetically identical offspring
  - cells divide in a geometric progression doubling cell number



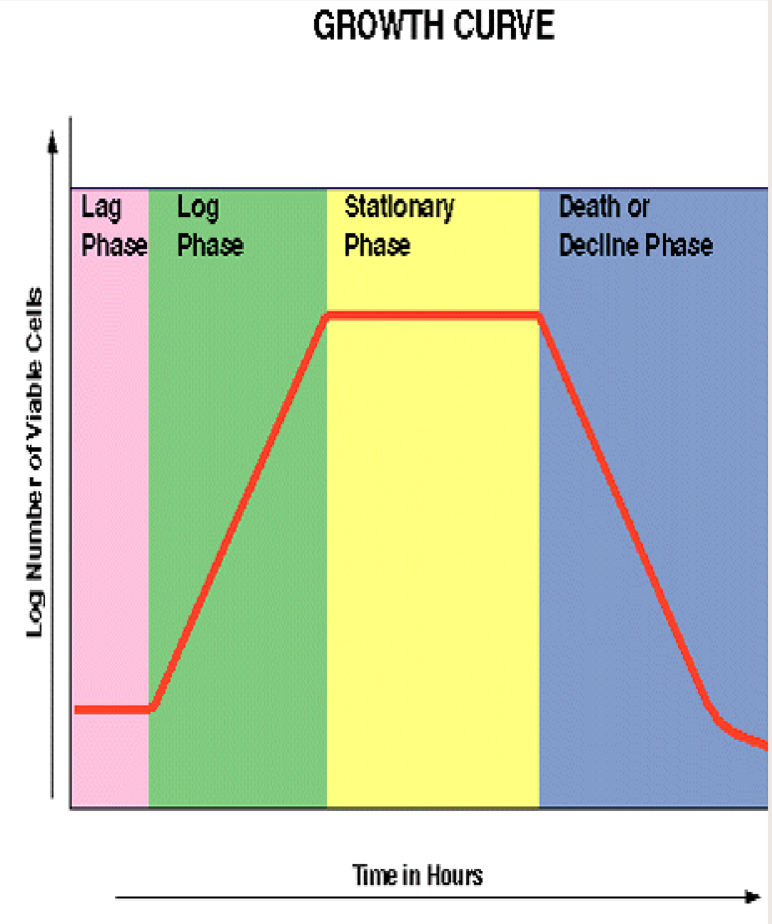
BINARY FISSION



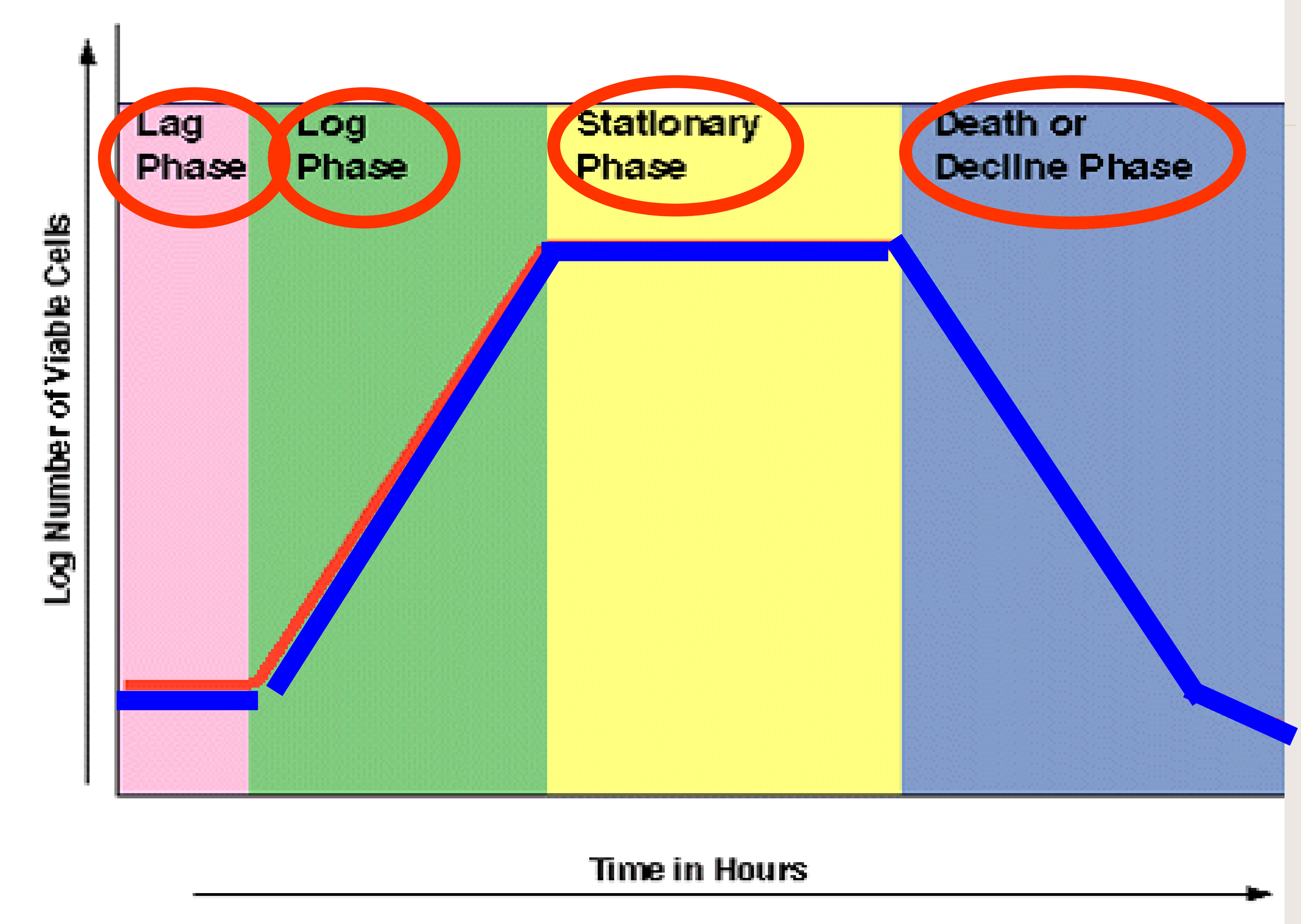
- ***Doubling time* is the unit of measurement of microbial growth**

CULTURE GROWTH

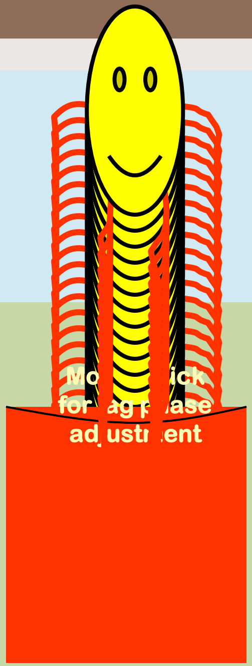
- Growth of culture goes through **four phases** with time
- 1) **Lag phase**
- 2) **Log or Logarithmic phase**
- 3) **Stationary phase**
- 4) **Death or Decline phase**



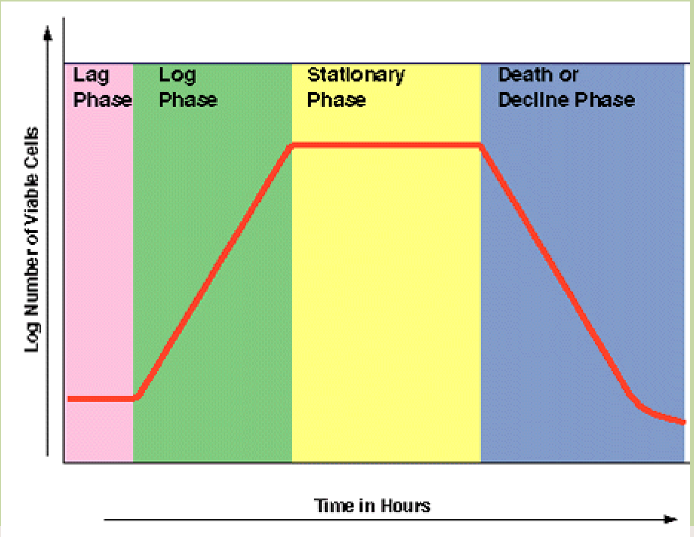
BACTERIAL GROWTH CURVE



LAG PHASE

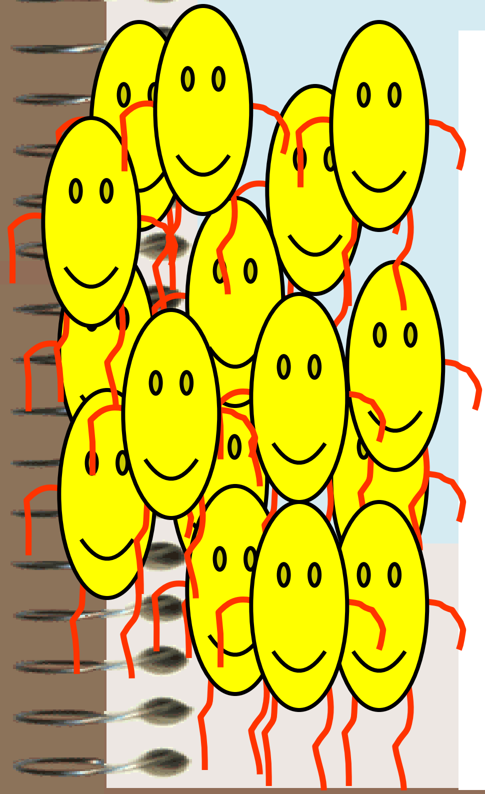


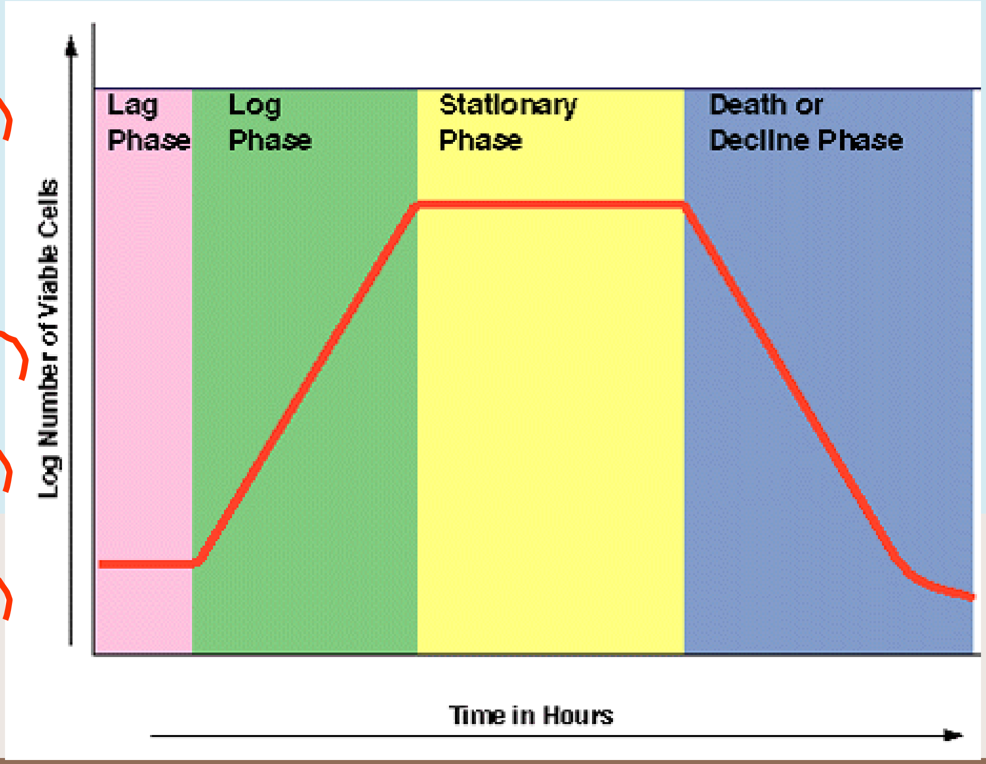
- Organisms are adjusting to the environment
  - little or no division
- <u>synthesizing DNA</u>, ribosomes and enzymes
  - in order to breakdown nutrients, and to be used for **growth**



LOGARITHMIC PHASE

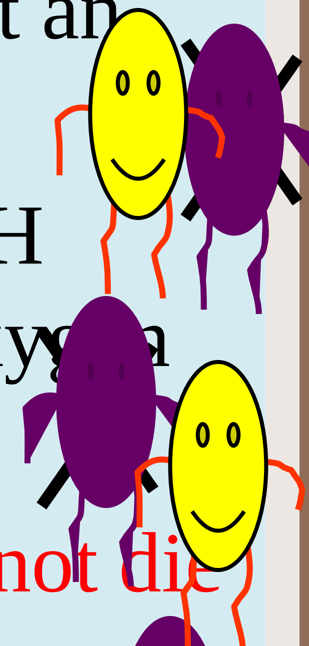
- Division is at a constant rate (**generation time)**
- Cells are most **susceptible** to inhibitors



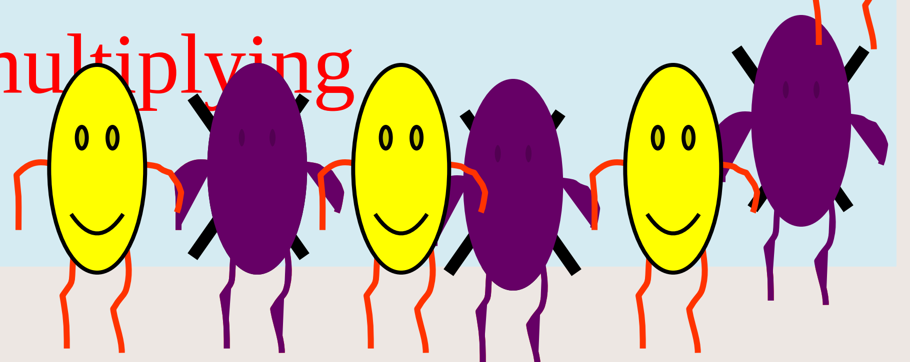


STATIONARY PHASE

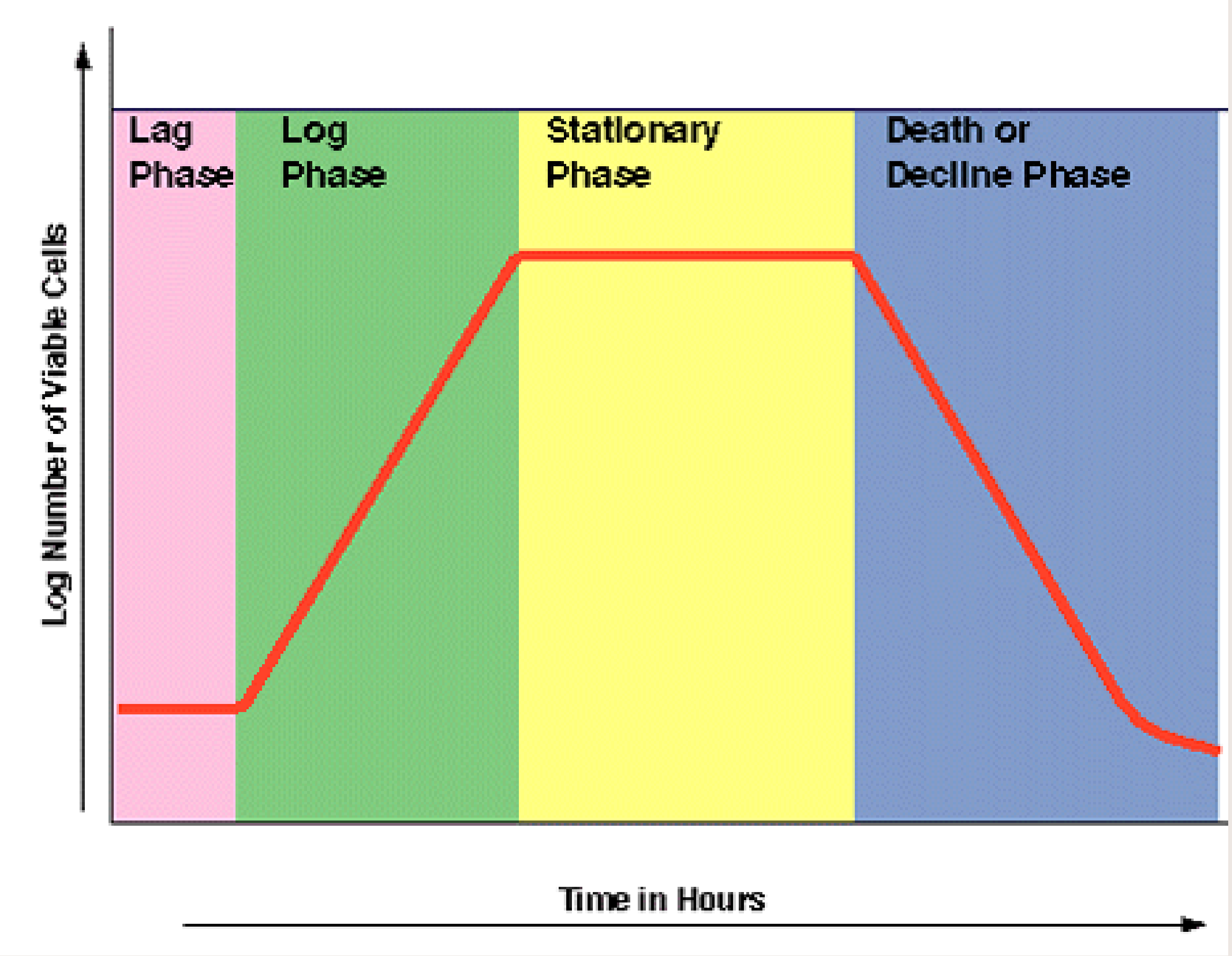
- <u>**Dying**</u> and <u>**dividing**</u> organisms are at an equilibrium



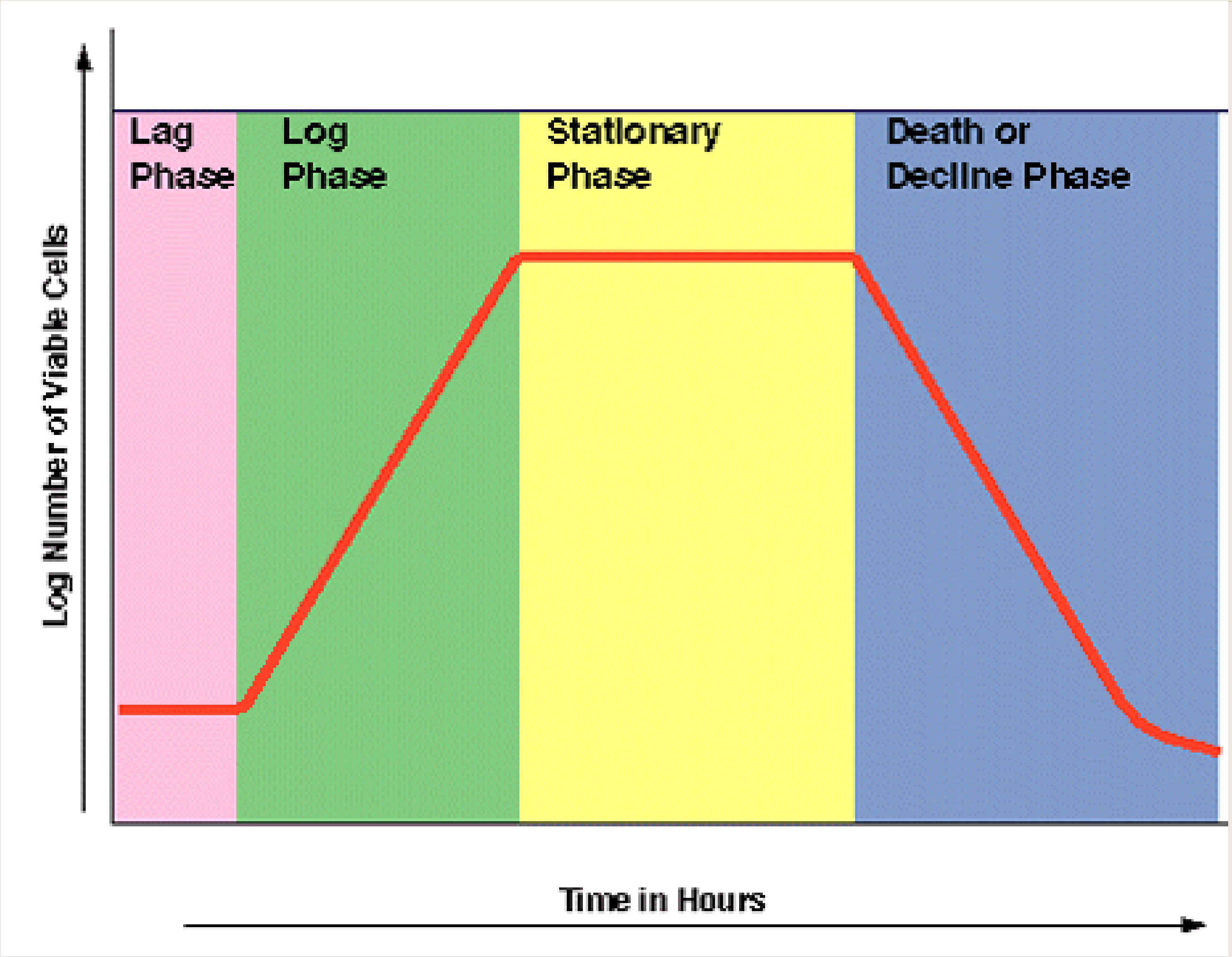
- **Death** is due to reduced nutrients, pH changes, toxic waste and reduced oxygen
- Cells are <u>**smaller**</u> and have fewer ribosomes @In some cases cells do not die <u>**but**</u> they are not multiplying



STATIONARY PHASE

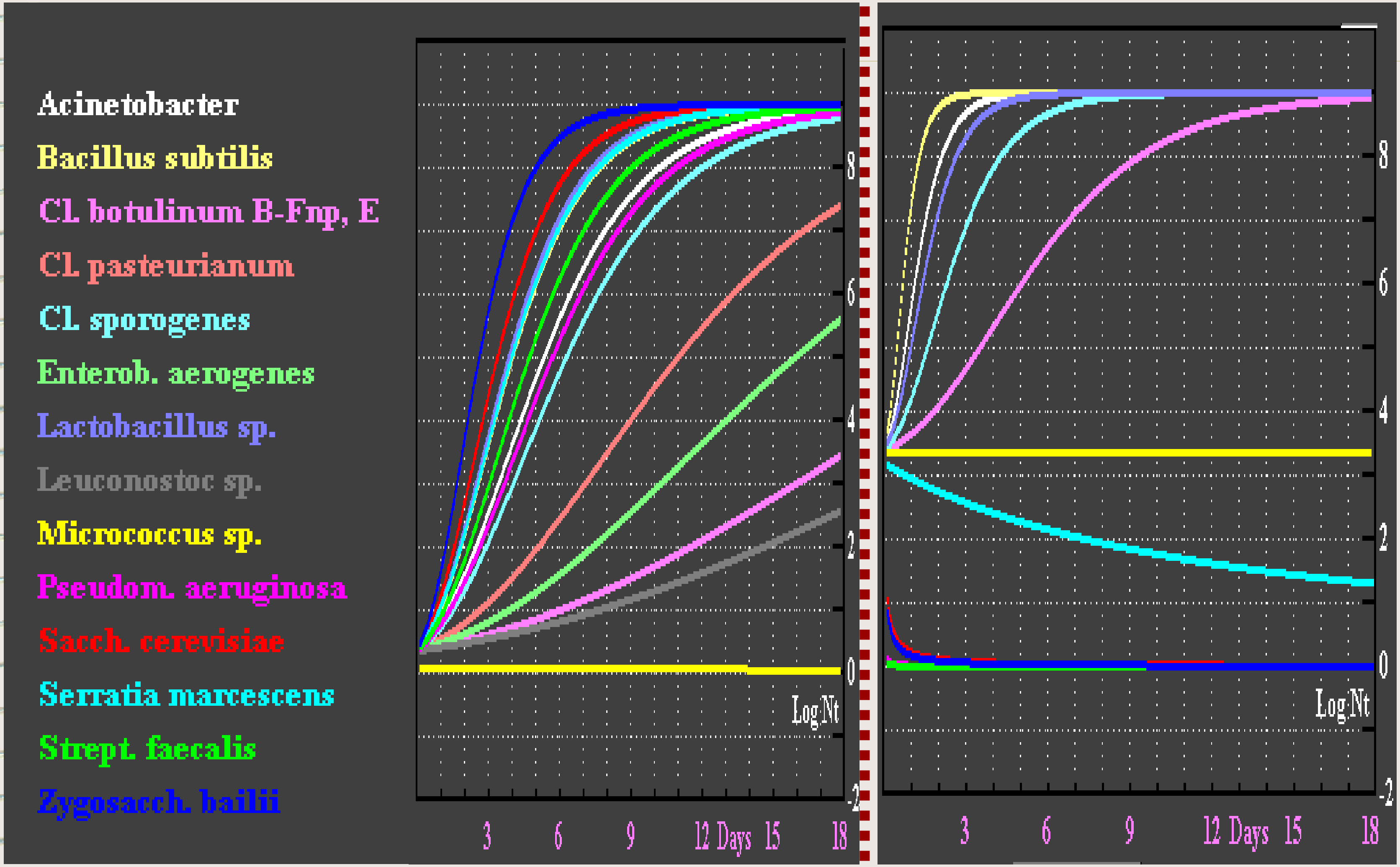


DEATH PHASE


Lag
Phase
Log
Phase
Stationary
Phase
Death or
Decline Phase
Log Number of Viable Cells
Time in Hours

In bioreactors

in 37ºC, pH 5.1 ; in 45ºC, pH 6.2



Thank you