GROWTH REQUIREMENT OF BACTERIA

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OBJECTIVES

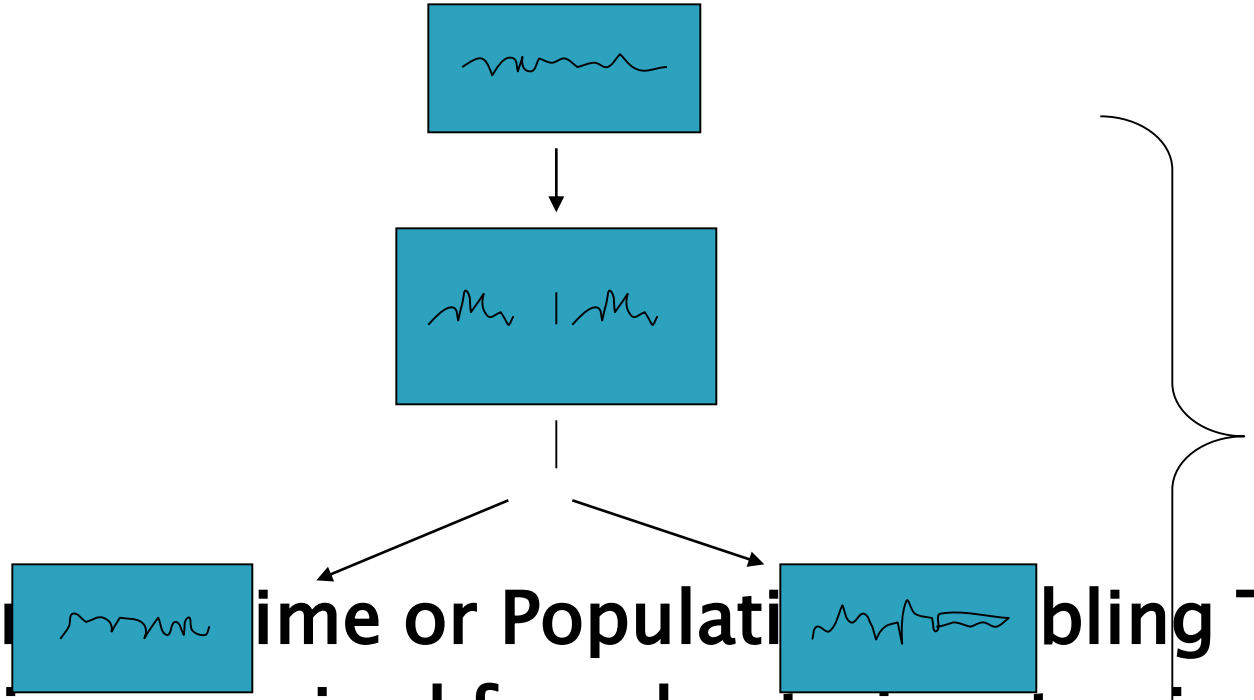
Growth
Batch culture
Continuous culture
Microbial physiology
Microbial metabolism
Oxygen requirement
Nutrients for growth
Effect of environmental factors

Growth

- Optimum temp., pH, humidity, O2, Water, minerals & nutrient require for growth of bact.

- What is growth?
  - Increase in size – after reaching certain limit, bact divide.
  - Increase in number

- Binary fission



- Gener ime or Populati bling Time
- The time required for a bacterium to give rise to two daughter cells under optimum conditions.

Generation time

Coliform bacilli is 20 min

Tuberculous bacilli is 20 hour

Lepra bacilli 20 days

Theoretically, from 1 coliform bacilli, after 24 hrs. 1×10^{21} cells.

This is not true in body. Why ?

- Because of various host defense mechanisms

-What is Colony of bacteria?

Batch culture

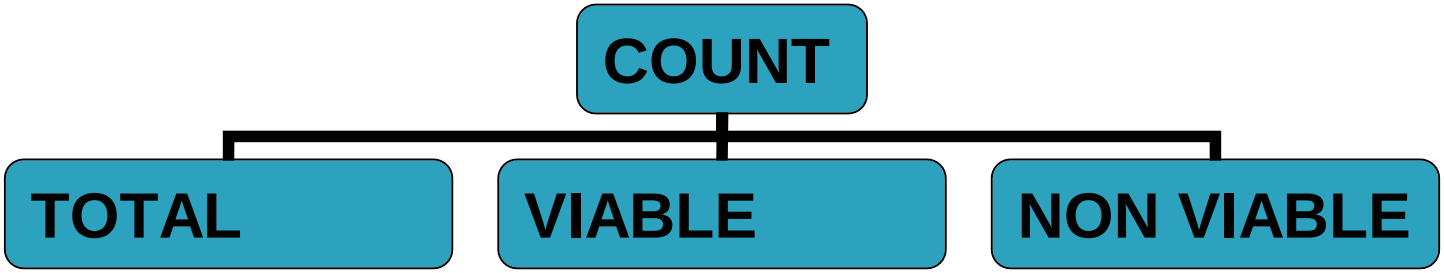
- Common method of growing bacteria in laboratory

- Bacteria are inoculated in a vessel containing liquid medium – Batch culture
- After sometime, Nutrients are depleted & Toxic metabolites are accumulated – lead to reduction in bacterial count.

Continuous culture

- Open system in which there is continuous supply of fresh nutrients into the culture vessel and a continuous removal of grown bacteria by means of a constant –level device (chemostat)
- Used in industrial sector

Bacterial cell count



Methods to count Total Count

- Counting under the microscope by counting chambers
- Counting in an electronic device – Coulter counter
- Counting in stained smears prepared by spreading a known volume of the culture

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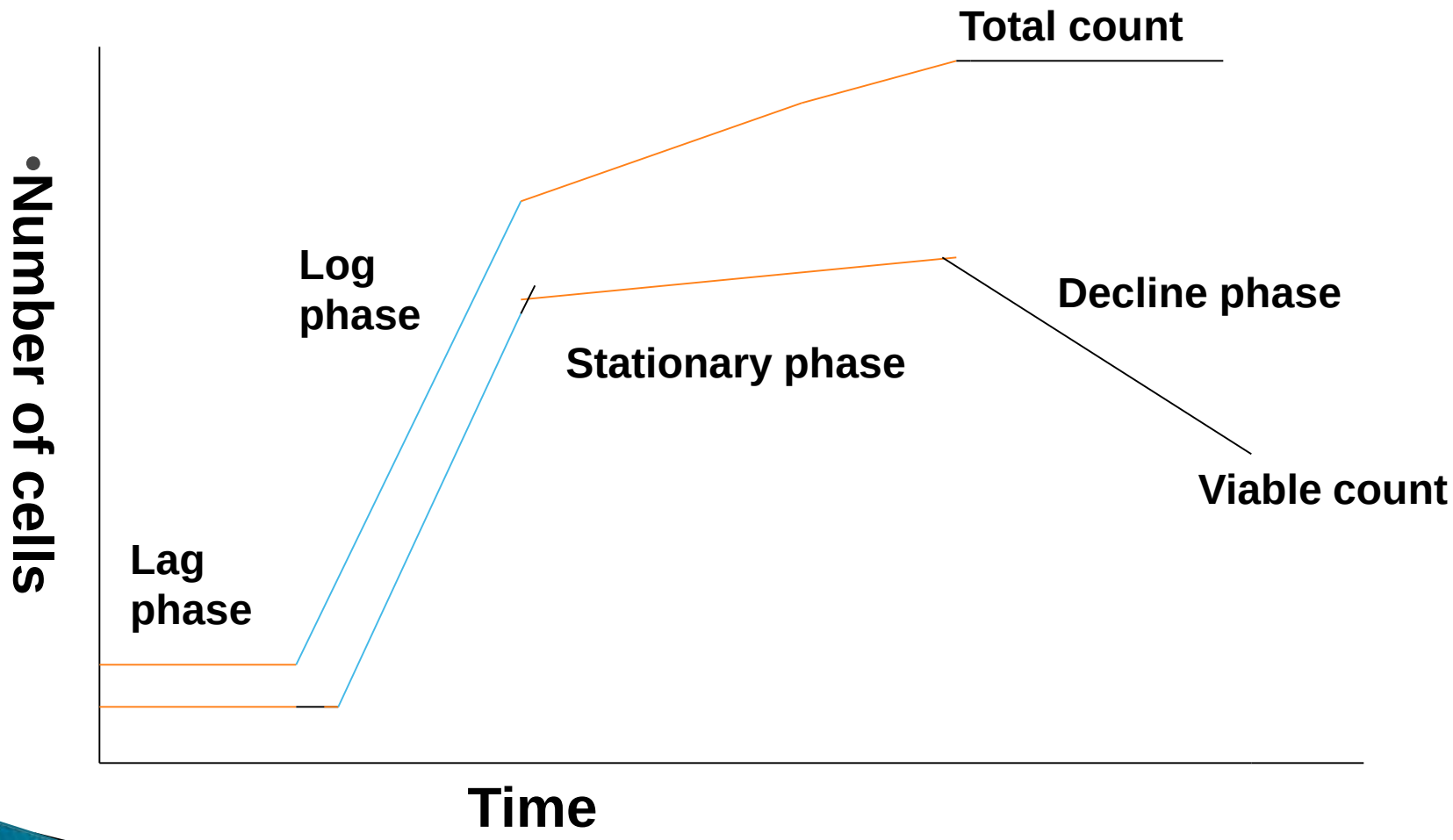
- Comparing relative numbers in smears of the culture mixed with known number of other cells
- By opacity measurement – absorptiometer or nephalometer
- By separating the cells by centrifugation or filtration & measuring their wet or dry wt.
- Chemical assay of cell components such as nitrogen

Methods to count Viable Count

- **Dilution method – eg. Presumptive coliform count in drinking water. Viable counts are calculated statistically.**
- **Plating method**
  - **Streaking method**
  - **Pouring method**

❖ **Colony forming units**

Bacterial (batch) growth curve



Lag phase

- Increase in size of cells, Not in number
- Adapt a new environment & to built up necessary enzymes and metabolites
- Duration of period varies with –
  - species,
  - size of inoculation,
  - nature of culture medium &
  - environmental factors like temp.

Log phase

- Continuous cell growth
- Cell divides continuously
- Straight line on plot
- Depend on generation time of the bacterium
- Cells are smaller and stain uniformly
- Not possible in vivo

Stationary phase

- Decrease rate of multiplication
- No growth no death
- Balance between reproduction & death
- Why ?
  - Exhaustion of an essential nutrients in the medium
  - Accumulation of toxic waste products (e.g. organic acids – $p^{H)}$

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Contd.

- Cells
  - are gram variable
  - have high level of Intracellular storage granules
- Many species produce secondary metabolites such as antibiotics, exotoxins
- Sporogenesis occur during this period

Decline or Death phase

- Cell population decreases due to cell death
- Why ?
  - Accumulation of toxic waste products
  - Autolysis
  - Involution forms are common

Bacterial cell – Chemical composition

Principle constitute of bacterial cell is **water**.

- 80% of the total wt.

Rest 20 % :

Proteins,
Polysaccharides,
lipids,
Peptidoglycan,
Nucleic acid,

Nutritional classification

- Depends on Carbon, Energy, Hydrogen/Electron source

- C source
  - CO_2 → **AUTOTROPHS**
  - Reduced, preformed, organic molecules from other organisms → **HETEROTROPHS**
    - Medically imp. bacteria

- Energy source

Light → **PHOTOTROPHS**

Oxidation of inorganic/organic compounds → **CHEMOTROPHS**

Nutrients required

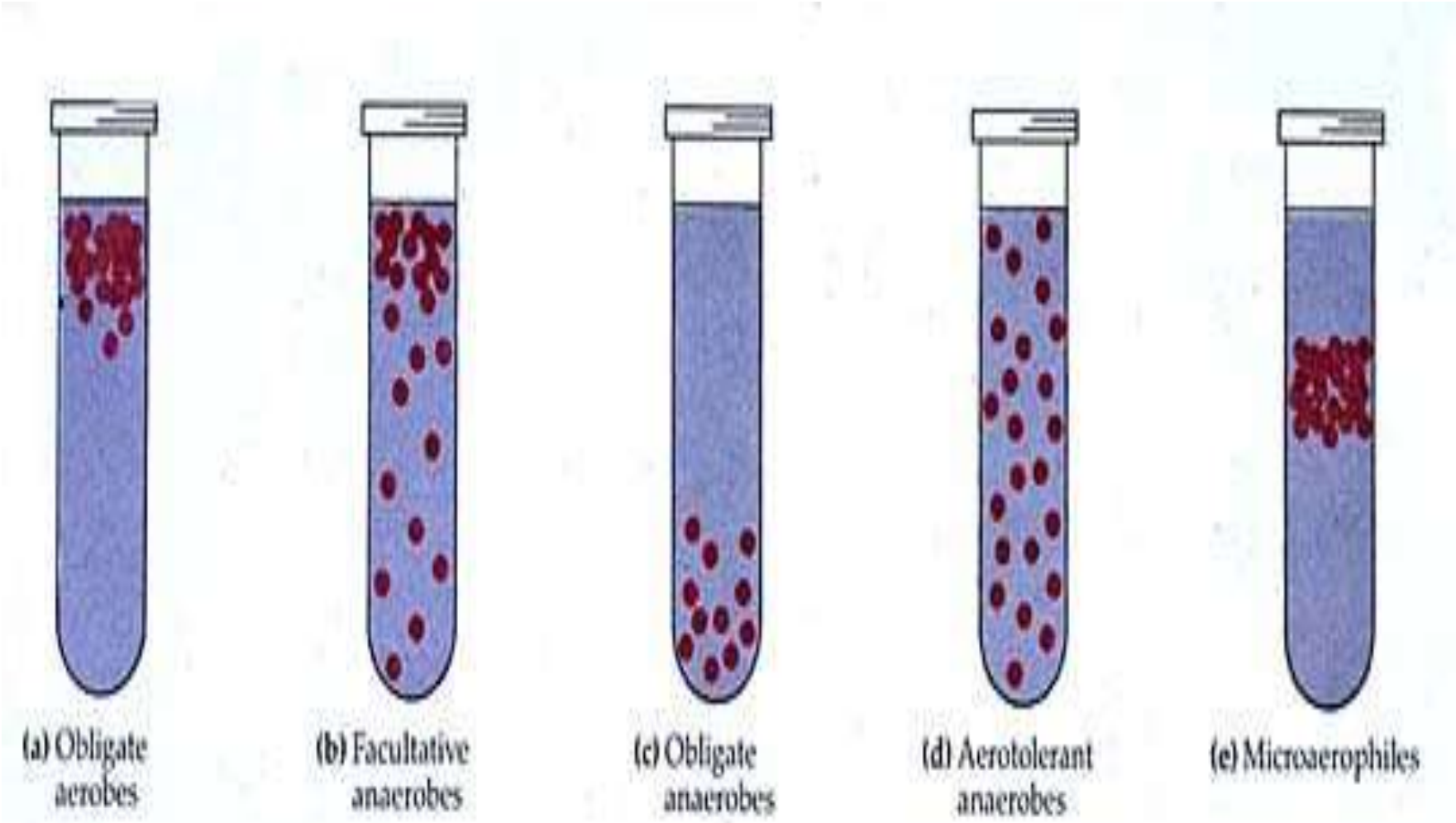
- Water
- Macro elements – C, O, N, S, H, P
- Microelements – K, Ca, Mg, Iron
- Trace amt. of manganese, cobalt, copper etc.
- Growth factors– essential, accessory

Oxygen requirement & Metabolism

- Aerobic bacteria – require oxygen for their growth
- Obligate aerobe – grow only in presence of oxygen
- Facultative anaerobe – are ordinarily aerobic but can grow anaerobically, though less abundantly

Contd.

- Anaerobic bacteria – grow in absence of oxygen
- Obligate anaerobe – grow only in absence of oxygen, may even killed in its presence
- Microaerophilic – grow in presence of a trace amount of free oxygen & often prefer an increased conc. of CO_2.


(a) Obligate aerobes
(b) Facultative anaerobes
(c) Obligate anaerobes
(d) Aerotolerant anaerobes
(e) Microaerophiles

Physical conditions

1. Temperature

- Psychrophiles (cold-loving microbes) bacteria – grows below 20^0 C,soil and water saprophytes
- Mesophiles (moderate-temp.-loving) bacteria – grow at temp. of 25-40^0 C
- Thermophiles (heat-loving) bacteria – grow at high temp., 55-80^0 C

2.pH

* pH refers to the acidity or alkalinity of a solution.
* most of the pathogenic bacteria grow at neutral or slightly alkaline pH. (except V. cholerae & Lactobacilli)

3. Light

* Bacteria grow well in dark except phototropic spp. (Pigment production in M. kansasii & M. marinum only in presence of light)
* Sensitive to UV light, sunlight, radiations

4. Osmotic effect

Plasmolysis – in hypertonic solution, osmotic withdrawal of water & shrinkage of protoplasm.

Plasmoptysis – sudden transfer from concentrated solution to distilled water leads to cell swelling & rupture.

Any Question

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THANK YOU