Lect 4

I-BACTERIAL GROWTH:

1-Definition :

Growth involves an increase in the **size** and **number** of organisms"

Results from bacterial reproduction.

How can we see their growth?

In the laboratory :

In the laboratory bacterial growth, can be seen in one of two main forms.

1. Colonies development

On solid media, development of **colonies**, which are the macroscopic products of **20-30** cell divisions of a single bacterium.



Figure 1 Colonies form on solid media

2. Turbidity

In liquid media. Transformation of a clear fluid medium to Turbid suspension**.**

The turbidity of the suspension is determined by **spectrophotometer**.

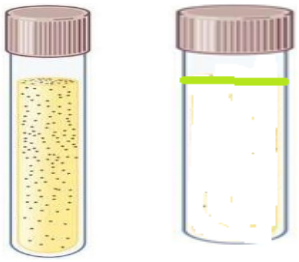


Figure **2** The turbidity of a culture

2-BACTERIAL REPLICATION

- Bacteria divide by a Process called : **Simple Binary Fission**
- Binary Fission is a type of **Asexual reproduction** in single celled organisms
- Each cell divides into 2 equal sized offspring

The process:

1) **Elongate** of the bacterial cell.
2) **Duplication** of the chromosome.
3) The two sister chromosomes attach to the **mesosome**.
4)Cell membrane and cell wall will form a **transverse septum** that divides the mother cell into 2 equal daughter cells.

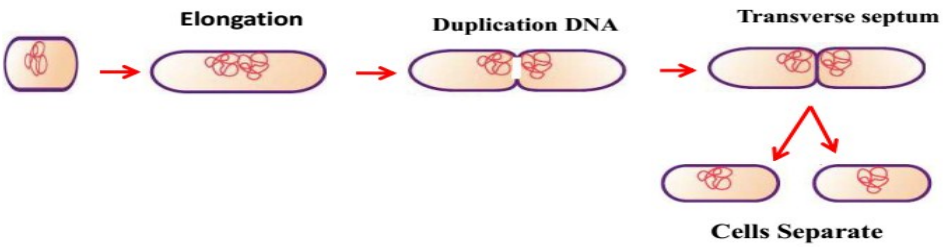


Figure 3 BACTERIAL REPLICATION

In some species, this septum splits the parent cell completely into two separate daughter cells. In other, the cell walls of the daughter cells remain continuous for some time after division giving the characteristic arrangement. e.g. pair, clusters or chains.

3-Generation time (doubling time) :

Define: The generation time is the time required for the bacteria to double. The time required for one cell to divide into two cells.

The "generation time" is often used to describe the **growth rate.**

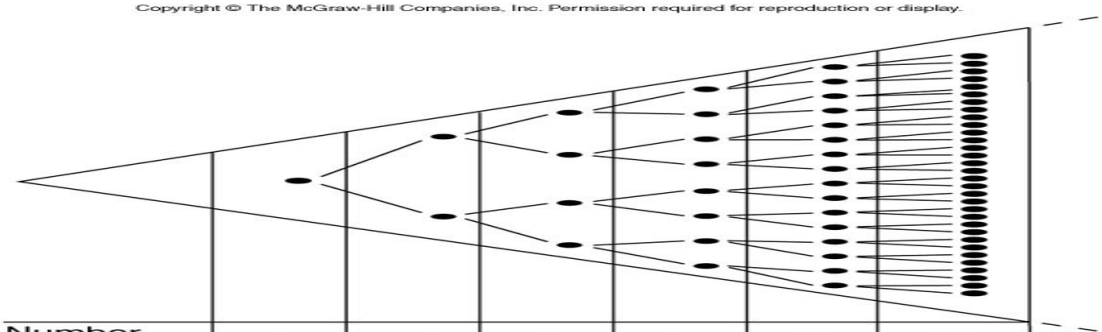
The maximal generation time varies for various organisms.

It may be as **short as**17 minutes for *Vibrio cholera* and 20 minutes for *Escherichia coli* rapidly growing pathogens and **as long as** 20 hours for others, such as *Mycobacterium tuberculosis.*

Calculation of the Growth Rate Constant and Prediction of the Amount of Growth

Bacteria reproduce by binary fission, and the average time required for the **population**, or the **biomass**, to double is known as the **generation time** or **doubling time**.

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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)

(a)

Figure 4 generation time

- **Number of bacteria= 2^n n = generation time**

II-Growth Requirement

In order to grow and **divide, bacteria** need the growth requirements:

Growth Requirement

In order to grow bacteria have certain requirements:

- Nutrients
- Growth factors
- Suitable temperature
- Suitable pH
- Oxygen
- Carbon dioxide
- Moisture.

1- Nutrition: According to the means by which a particular organism **obtains energy** and raw material to sustain its growth, bacteria are classified into:

a-Autotrophs: (auto= self/ troph= feeding)

- ☒ Free-living, non-parasitic bacteria

They can utilize simple **inorganic** materials

e.g. **Co2** as a source of **carbone** and **ammonium salts** as source of **nitrogen**. They can synthesize complex organic substances from the simple inorganic material carbon and ammonium salts as a source of nitrogen.

The energy required for their metabolism is predominantly derived from **light** or **simple chemical reactions**.

Autotrophs are no medical importance (not pathogenic).

- ☒ Do not require **organic** carbon
- ☒ Use **photosynthetic** energy to reduce CO2 to H_2O

b-Heterotrophs: (hetero= other)
These bacteria, on the other hand, require **organic** sources or carbon, as they can't synthesize complex organic substances from simple inorganic source.
Most bacterial of medical important are **Heterotrophs**

2-Growth factors

Growth factors are essential substances (organic compounds) that are required by the bacteria but can't synthesize by them. ()

• Factor 'V' and factor 'X' necessary for the growth of *Haemophilus influenzae*.

Fastidious bacteria: are those which require unusually complex nutrients.

-Oxygen (O_2) requirements

According to Oxygen requirements, bacteria are classified into:

1- **Obligate (strict) aerobe: Grow only in the presence of Oxygen**

e.g. -*Pseudomonas aeruginosa*
-*Mycobacterium tuberculosis*

2-Obligate (strict) anaerobe : Require complete a absence of Oxygen

- E.g. *Bacteroides fragilis*
  Clostridium tetanus

3-Facultative anaerobe : Generally grow better in presence of oxygen (but)still are able to grow in its absence,

- e.g. staphylococci .
- *E.coli*

-Can grow in the presence or absence of O2. Most pathogenic bacteria of medical

4-Microaerophilic bacteria

Micro, Aero, Philic

- **Organism required reduced or low O2 tension**

e.g. Helicobacter pylori
campylobacter

5-Aerotolerant anaerobes:

-Anaerobic bacterial

- Do not utilize oxygen, but can **tolerate low amount of oxygen** because they have **superoxide dismutase.** (no catalase)

Eg..*Lactobacilli*

-Carbon dioxide (CO2) requirements

The Small amount of CO_2 is required by all bacteria.

- Some organisms grow best at higher amounts of CO_2 and are called **Capnophillic**

e.g.:

- *Neisseria* species require 5-10%.
- *Brucella abortus* require 20% CO2.

-Temperature requirements

1) Mesophilic:

- ✓ Are organisms able to grow within temperature range of 20-40 C^o
- -Grow best or optimum temperature at 37 C^o.
- -Are mostly pathogenic bacteria.
- E.g. *Escherichia coli*
- *Staphylococcus aureus*

2-Psychrophilic: (cold-Loving)

- Bacteria are able of growth at refrigeration temperature (0-8)
  - e.g. *Flavobacterium Spp.*
- Grow best at low temp (15 -20°C)

- Can grow in refrigerator temperature.

3-Thermophilic forms: (Heat-loving)

Grow best at high temperature > 60°C

e.g. Bacillus stearothermophilus

-Hydrogen ion concentration (pH)

- Most bacteria grow at neutral or slightly alkaline (pH 7.2)
- pH can harm organisms by disrupting the plasma membrane , inhibiting the activity of enzymes and membrane proteins and ionization of nutrient molecules.
- Classification

- **1-Neutralophiles :**
  - Most bacteria of medical importance grow at a pH of 6-8
    - Eg. Many pathogenic bacteria

- **2-Acidophiles :**
  - Some forms have optima as low as pH 3.0.
    - e.g *lactobacilli*.

- **3-Alkaliphiles :**
  - Others have optima as high as pH > 8.0.
    - e.g. *Vibrio cholerae.*

5-Moisture

- Four-fifth of bacteria cell weight consists of water, and moist is necessary for growth.

Osmotic pressure

- Bacteria can withstand wide range of external osmotic pressure because of protective cell wall.
- 0.5% of NaCl is added in most culture medium to maintain suitable environment

III-Growth Phases (Bacterial Growth Curve)

If a small number of an organism is placed in a suitable fluid nutrient medium under appropriate **physical** and **chemical conditions**, then the number of viable cells per milliliter **is determined periodically**, and **plotted**, **a characteristic growth curve with four phases obtained** (Fig. 5):

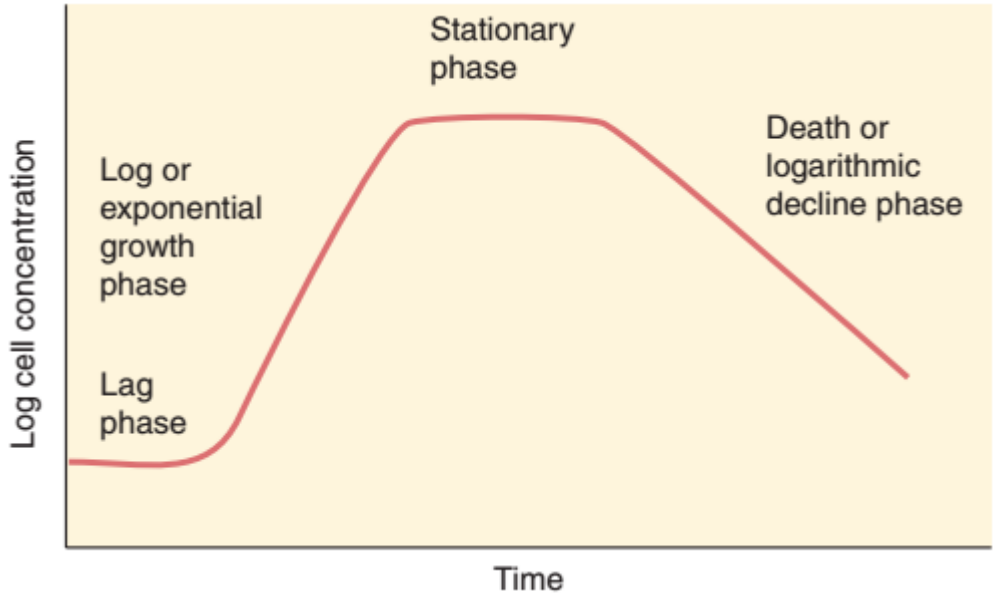


Figure 5 A bacterial growth curve.

1- Lag phase: The initial number of bacterial cells remain **constant**. During this period, **the cells adapt to their new environment**. Enzymes and intermediates are formed to permit growth. **No cell division** so number is constant. C**linical significance- incubation period of disease**

2- Exponential (Logarithmic) phase: There is marked **increase in cell number** and **its rate is accelerated exponentially** with time giving a **characteristic linear plot** on a logarithmic scale. In this phase, the organism shows **typical morphology**.
-Bacteria are most sensitive to antimicrobial substances.
-Clinical significance- symptoms & signs appears

3-Stationary phase: Exhaustion of nutrients and **accumulation of toxic products** cause growth **to decrease**. There is slow loss of cells through death which is **just balanced** by formation of new cells through growth and division. The **number of viable bacteria remains constant** death = replication so number constant due to balance
-Number of viable and death bacteria counts equal

-Production of spore (Sporulation), release of toxin

4. Decline-phase : At the end of the stationary phase**, the death rate increases and exceeds the multiplication rate** due **to nutrient exhaustion** and **accumulation of toxic metabolic end products**. So, the **number of viable bacteria decreases**.
-Clinical significance- phase of convalescence