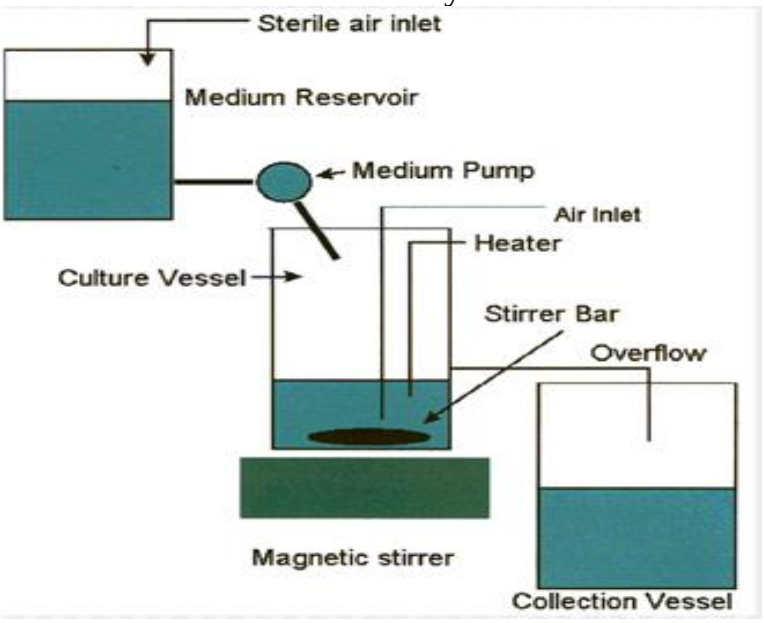
Bacterial Growth:

Growth in bacteria means increase in their number, while in eukaryotes means increase in size. Bacteria are smaller and grow faster than eukaryotes and divide by binary fission.

There are two types of microbial growth system:

1. **Batch culture:** is a closed system in broth medium in which no additional nutrient is added after inoculation of the broth.
2. **Continuous culture:** is an open system in which fresh media is continuously added to the culture at a constant rate, and old broth is removed at the same rate. This method is accomplished in a device called a **Chemostat**.

Basic Chemostat System



Growth curve

Typically, a batch culture passes through four distinct phases:
Lag phase, Logarithmic (exponential) phase, Stationary phase and Death phase

Lag phase

- Organisms are adjusting to the new environment
  - Little or no division
- Synthesizing DNA, ribosomes and enzymes
  - In order to breakdown nutrients, and to be used for growth

Exponential phase

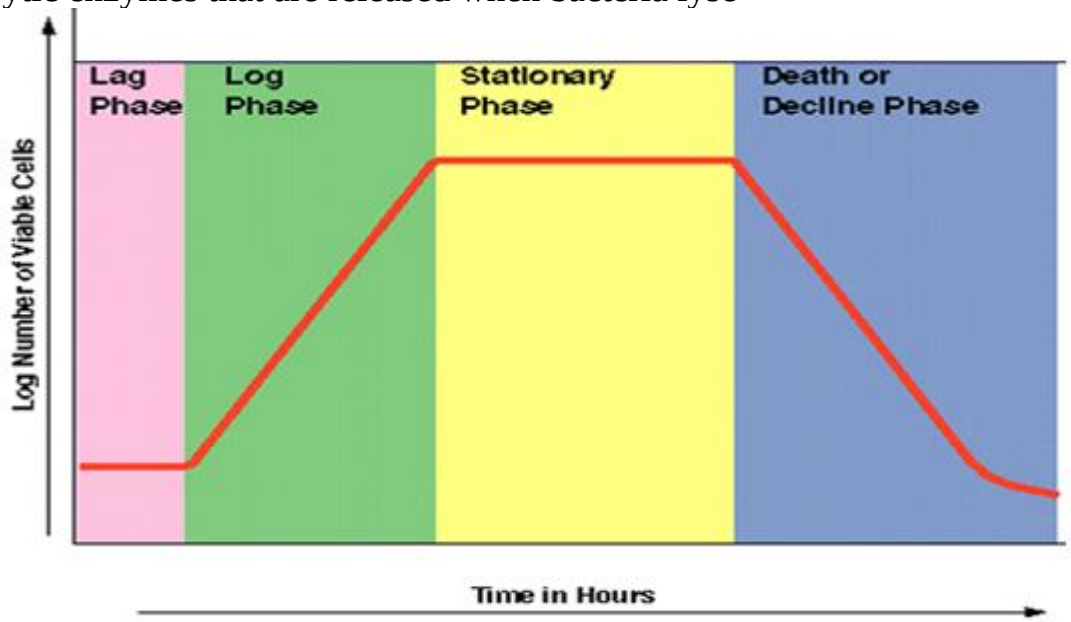
- Division is at a constant rate (generation time)
- Period of most rapid growth.
- Cells are at highest metabolic activity.
- Cells are most susceptible to inhibitors

Stationary phase

- Dying and dividing organisms are at an equilibrium
- Death is due to reduced nutrients, pH changes, toxic wastes and reduced oxygen
- Cells are smaller and have fewer ribosomes
- In some cases, cells do not die but they are not multiplying

Death phase

- The population is dying in a geometric fashion so there are more deaths than new cells
- Deaths are due to
  1) Factors in stationary phase
  2) Lytic enzymes that are released when bacteria lyse



Generation time or doubling time:

Is time required for a cell to divide and become duplicated which depends on the type of bacteria and growth condition.

Doubling time is the unit of measurement of microbial growth

Generation times for some common bacteria under optimal conditions of growth

Bacterium	Medium	Generation Time (minutes)
Escherichia coli	Glucose-salts	17 Minutes
Bacillus megaterium	Sucrose-salts	25 Minutes
Streptococcus lactis	Milk	26 Minutes
Streptococcus lactis	Lactose broth	48 Minutes
Staphylococcus aureus	Heart infusion broth	27 – 30 Minutes

Lactobacillus acidophilus	Milk	66 – 87 Minutes
Rhizobium japonicum	Mannitol-salts-yeast extract	344 – 461 Minutes
Mycobacterium tuberculosis	Synthetic	792 – 932 Minutes
Treponema pallidum	Rabbit testes	1980 Minutes

Generation time measurement

We can determine or evaluate generation time according to the following steps

1. Sterilize "1ml" of nutrient broth and "4 ml" of D.W with autoclave and use it as the **Blank**.
2. By using micropipette transfer "1 ml" of overnight bacterial culture to flask containing "99 ml" of nutrient broth, after then distribute this culture on "2" flasks each of them "50 ml".
3. Incubate these flasks in the shaker incubator (80 circle / minute) at 37 ◦C.
4. Measure optical density (O.D.) of these samples at wave length (600 nm) every 20 or 30 minutes starting with time "zero" and when reached stationary phase at 600 nm by adding "1 ml" of bacterial culture to "4 ml" of D.W with shaking
5. Calculate generation time during log phase and total number of viable cells at beginning of log phase and at the ending of it. According to the following equation:

$$g = \frac{t \log 2}{\log b - \log B}$$

g = generation time
t = time between (B) and (b)
b = total number of viable cells at the end of log phase
B = total number of viable cells at the beginning of log phase

Generation time also can be calculated by using the following equation
Gt = $t_2 - t_1$
Gt = time between two points in log phase

Calculating doubling (generation) time from an OD measurement