Microbiology (2) (205 M)
Bacteria

by

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At the end of this lecture the student must able to:

- Describe the growth curve of bacteria.
- Compare between the different phases of bacterial growth curve.
- Describe the diauxic growth curve of bacteria.
- Discuss the importance of continuous culture of bacteria in industry.

Lecture 8
The contents

- Bacterial growth curve.
- Diaxuic growth of bacteria.
- Continuous culture of bacteria.

Growth of bacteria

- **If we with a bacterial cell reproduce by binary fission, the increase of population proceeds as follows: 1, 2, 4, 8, 32, 64, etc.**
- **The time interval required for the cell to divide is called GENERATION TIME. It is different with in the bacterial species.**
- **It ranges from 15 minutes to several hours depending on the bacterial species, type of nutrient and the physical conditions of the environment.**

- Generation time can be calculated as follows:

$$b = 1 \times 2^{n}$$

- Since we are not begin with a single cell, accordingly;

$$b = B \times 2^n$$

$$\log b = \log B + n \log 2$$

$$= \log B + (n \times 0.30103)$$

$$0.3n = \log b - \log B$$

$$n = \frac{\log b - \log B}{0.3}$$

$$n = 3.3 \log b / B$$

$$G = t / n = \frac{t}{3.3 \log b / B}$$

- Where:
- **G** = generation time
- **t** = incubation period (time)
- **n** = number of generations
- **b** = total number of bacteria at the end of “t”
- B = **number of bacteria inoculated into medium**

- **Bacterial growth curve**
- **When we inoculate a fresh medium with a given number of bacterial cells, determine the population during an incubation period, and plot the logarithms of the number of cells versus time, we obtain the bacterial growth curve.**
- **When we examine the bacterial growth curve, it is obvious that the growth passes through different stages. These stages are:**

- 1- **Lag Phase:**
- **Inoculation of a culture medium with bacteria is not followed by division at once.**
- **The number of bacteria remains unchanged for a certain period.**
- **In this period the cells increase in size. They are synthesizing new protoplasm.**

- **These enzymes must be synthesized before proceeding before proceeding the cell division.**
- **In addition, a time for adaptation to the new environment is needed.**
- **Consequently, it can be said that the organisms are metabolizing, but there is a LAG in the cell division.**

- **2- Phase of Increasing the Growth Rate:**
- **At the end of the lag phase, the cells begin to divide.**
- **However, since not all the organisms complete the lag period at the same time, there is a gradual increase in the population until the end of this period, when all the cells are capable of dividing at regular intervals.**

- 3-**Logarithmic Phase:**
- **It is also called log or exponential phase.**
- **During this phase, the bacterial cells are capable of steady division according to their specific generation time.**
- **Therefore, a straight line develops when the logarithms of the bacterial numbers are plotted against time.**
- **At appropriate conditions, the rate of growth is maximal during this phase.**

- **<u>4- Phase of Decreasing the Growth Rate:</u>**
- **It follows immediately the log phase.**
- **The cells divide slowly, the rate of death relatively increased.**
- **This phase lasts very short period.**

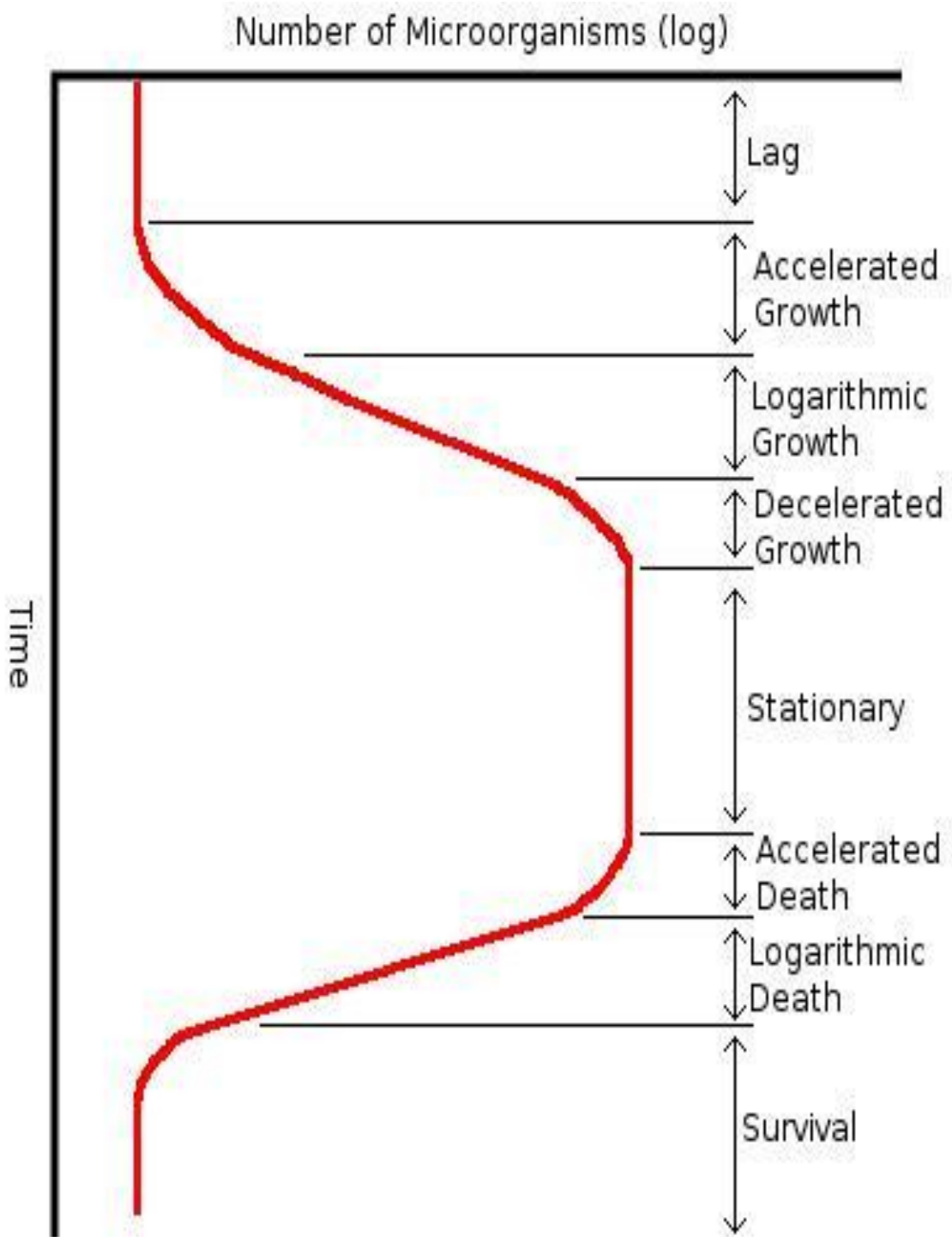
- **5- Stationary Phase:**
- **The total number of bacterial cells remains more or less constant.**
- **This cessation of growth can be attributed to the exhaustion of certain nutrients and/or the production of toxic substances.**
- **In this phase, a sort of balancing of reproduction rate by an equivalent death rate can be demonstrated.**

- **6- Phase of Increasing Death Rate:**
- **Comes after the stationary phase, the rate of death exceeds the rate of cell division.**

- **7- Decline Phase:**
- **It is also called the death phase. Here, the death rate exceeds in a manner that the bacterial number steadily lowered down.**

- **This may be due to the almost exhaustion of essential nutrients and/or the accumulation of toxic materials.**
- **The duration of this period depends largely on the bacterial species.**
- **It lasts several week or few days.**

- **The following figure shows bacterial growth curve.**


Number of Microorganisms (log)
Time
Lag
Accelerated Growth
Logarithmic Growth
Decelerated Growth
Stationary
Accelerated Death
Logarithmic Death
Survival

- **<u>Diauxic Growth Curves</u>**
- **Bacteria are liable to give normal growth curve when grown on a culture medium containing the essential elements and a single carbon source.**
- **This same curve can also be demonstrated when any pair of the following sugars is present in the medium: glucose, mannose, fructose, sucrose, or mannitol.**

- **When one of the above listed sugars is present and another sugar like maltose, arabinose or sorbitol, then two-peaked or DIAUXIC growth curve is obtained.**

- **The following figure shows the growth of *Escherichia coli* in the presence of different sugar pairs serving as carbon sources.**

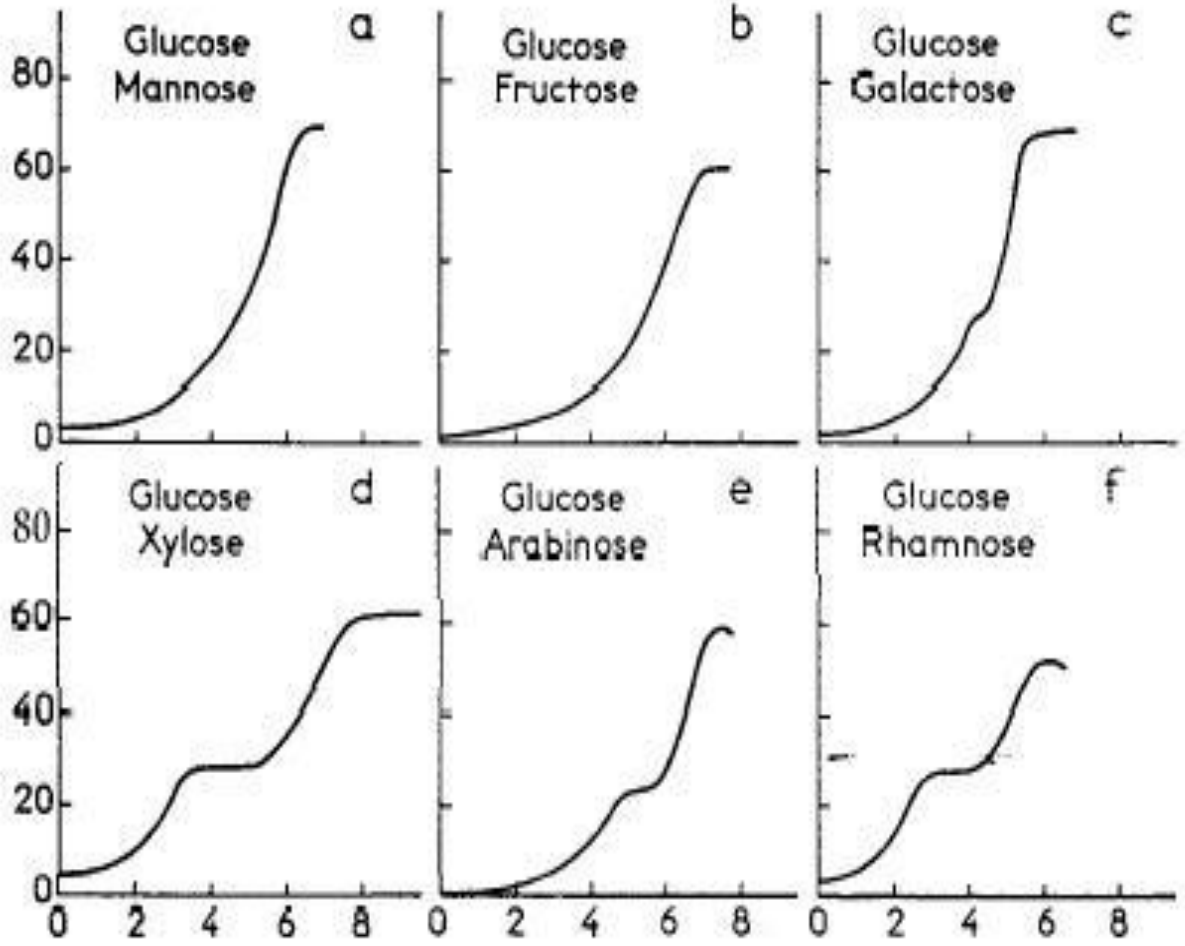
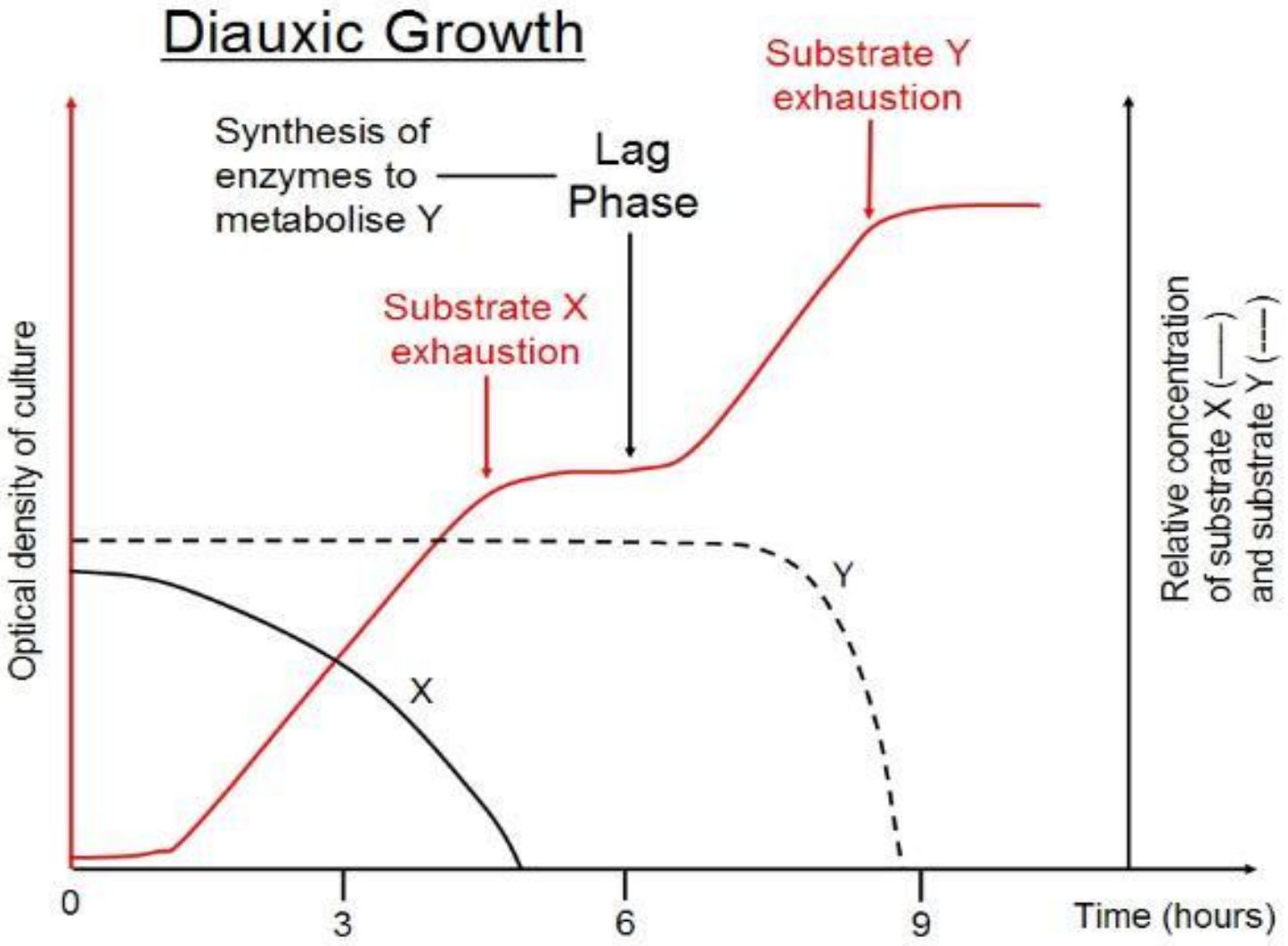


Fig.1. Growth of *Escherichia coli* in the presence of different carbohydrate pairs serving as the only source of carbon in a synthetic medium[50].

- **In the diauxic curve, the first cycle corresponds to the utilization of the first mentioned carbohydrates.**
- **When this carbohydrate is almost used up, and a stationary of decline phase begins, the second sugar is attacked.**
- **This may be due to the inductive formation of adaptive enzymes responsible for the utilization of the second sugar.**
- **In other words, the first sugar is attacked by constitutive enzymes while the second is assimilated by adaptive enzymes.**


Diauxic Growth
Substrate Y exhaustion
Synthesis of enzymes to metabolise Y
Lag Phase
Substrate X exhaustion
Optical density of culture
Relative concentration of substrate X (——) and substrate Y (- - -)
Y
X
0
3
6
9
Time (hours)

The previous figure shows Diauxic growth curve of bacteria.

- **In the same manner a TRIAUXIC growth curve may be obtained with three growth cycles and two lag periods as a result of the presence of three sugars like glucose, sorbitol and glycerol.**
- **The three growth cycles correspond the three carbohydrates, respectively.**

Continuous Cultivation of Bacteria:

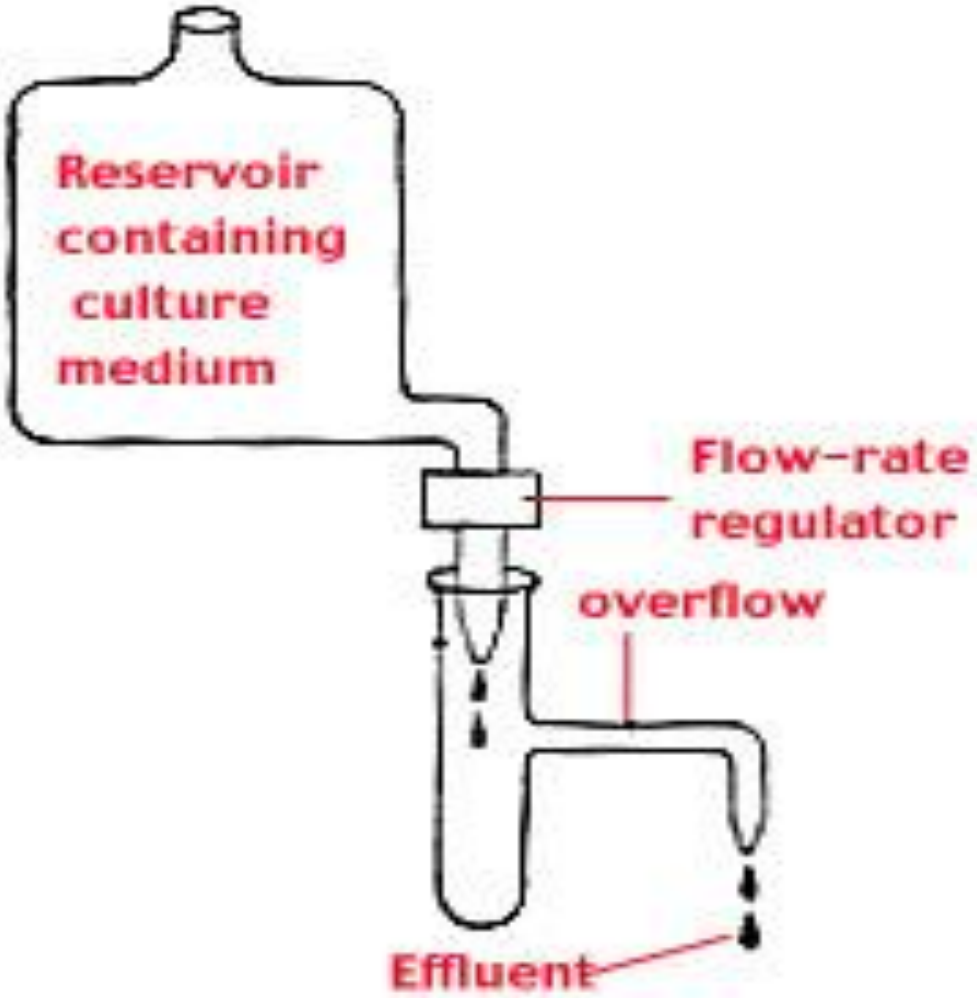
- As mentioned before, the reasons for the organism to enter the decline phase is the exhaustion of essential nutrients and the production of toxic materials.
- In industry, it is desirable to maintain a constant population (or log phase) throughout a fermentation process.

- This is carried out in two ways:

- a- Turbidostat: It is based upon regulation of the growth by measuring the turbidity of the cultured medium. The turbidity is corresponding to the number of the bacterial cells.
- b- Chemostat: It is the regulation of growth by the regulation of the inflow and outflow of the cultured medium, on the bases of limiting the concentration of particular nutrient (e.g. carbon source).

- In both devices, the culture is kept at a constant volume by allowing fresh medium to enter at the same rate at which fermented medium is removed from the fermentation vessel.

- **The following figure shows the continuous culture of bacteria.**


Reservoir containing culture medium
Flow-rate regulator
overflow
Effluent

Synchronous growth:

- **It is the adjustment of a culture so as all the cells divide at the same time, grow for a specific generation time and again divide at the same time.**
- **The culture, in this case is called synchronized culture.**
- **Synchronous growth can be achieved by adjustment of physical conditions like pH and temperature as well as the chemical composition of the medium.**

- **The culture may be held at a sub optimal temperature. At this temperature, the cells metabolize but not dividing.**
- **Upon rising the temperature, the cells divide uniformly.**
- **However the population remains in synchronous conditions for a short time then randomized.**

Questions

- 1- Why are continuous culture systems so useful to microbiologists?
- 2- Describe the bacterial growth curve and explain how can you benefit from it in industry?
- 3- Draw diauxic bacterial growth curve. Label and define each of the different phases. Give examples.

References

1- Mansour, F.A. Principle of bacteriology. Mansoura University.

2-http:www.google.com (search for growth of bacteria).