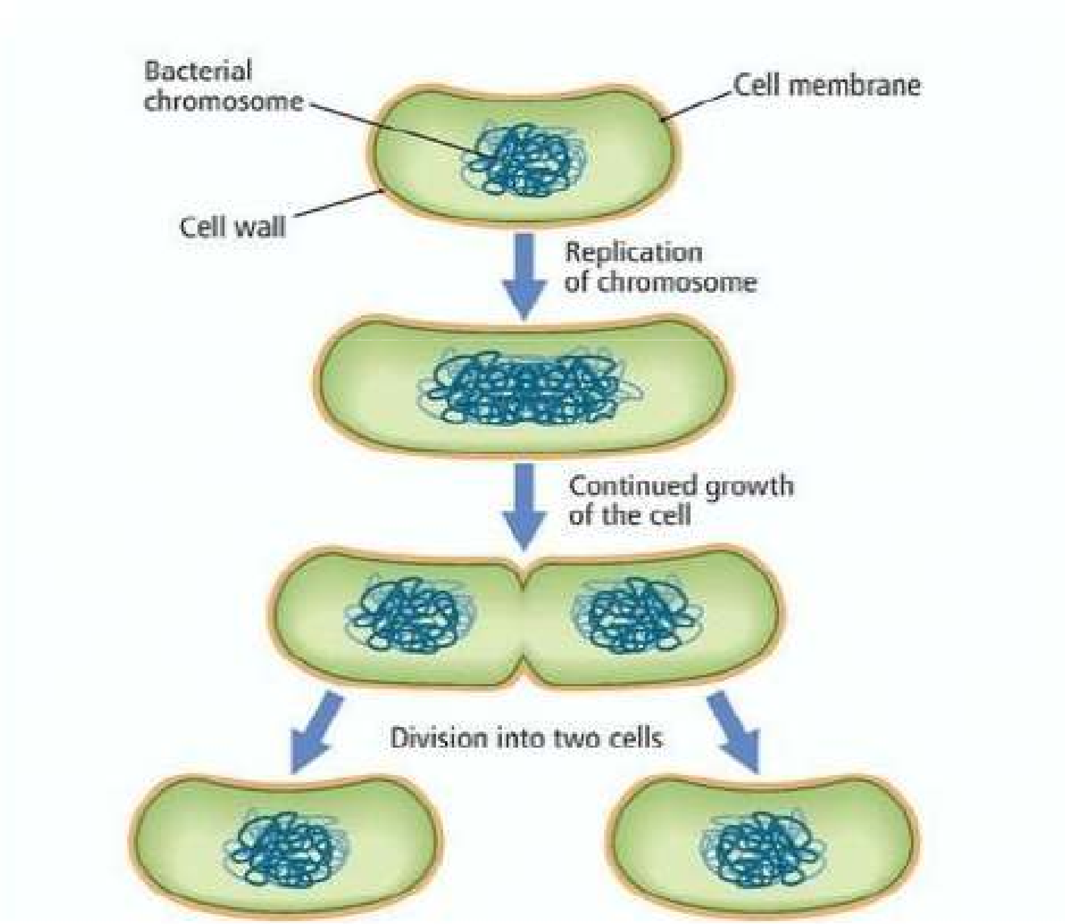
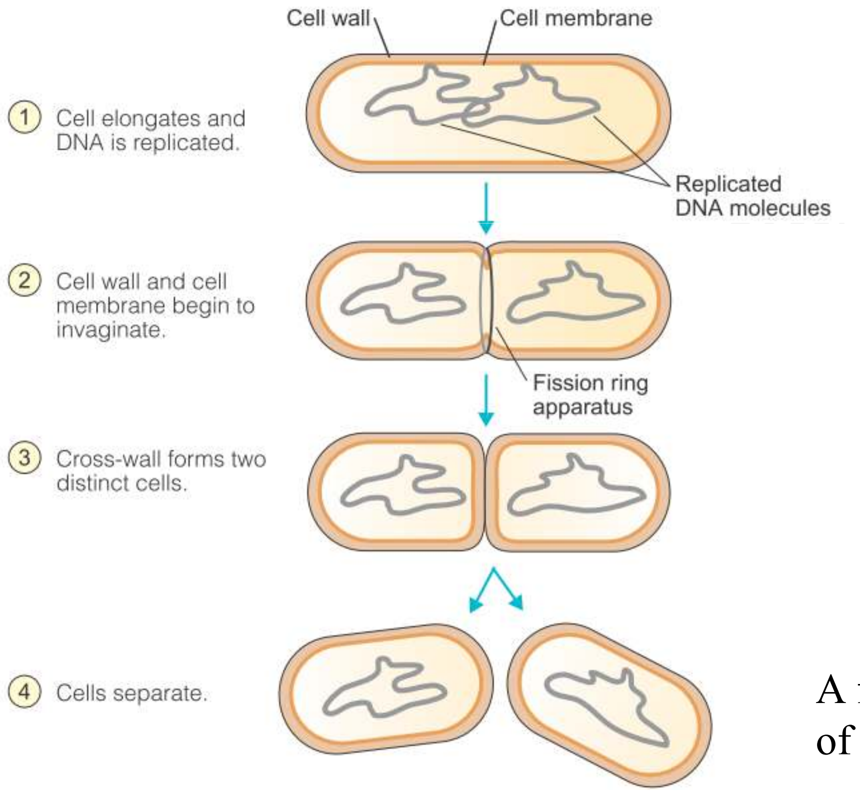
C-2: Bacteriology

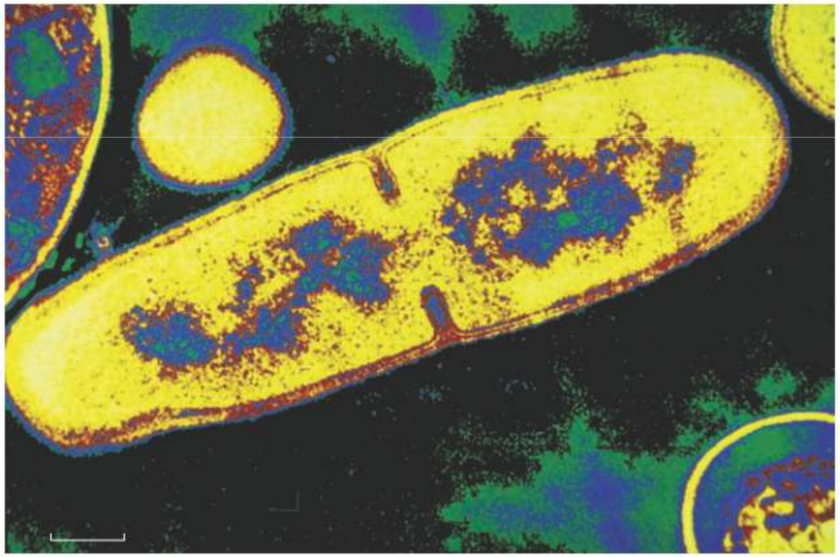
Unit 5 – Reproduction in Bacteria



Bacterial Reproduction

- Bacteria reproduce by a process called binary fission. During binary fission, the DNA is replicated and the cell separates. Each daughter cell receives a complete genome. This process represents **asexual reproduction**, as no exchange or **reassortment** of genetic information takes place.





A false-color transmission electron micrograph of a cell of *Bacillus licheniformis* undergoing binary fission.

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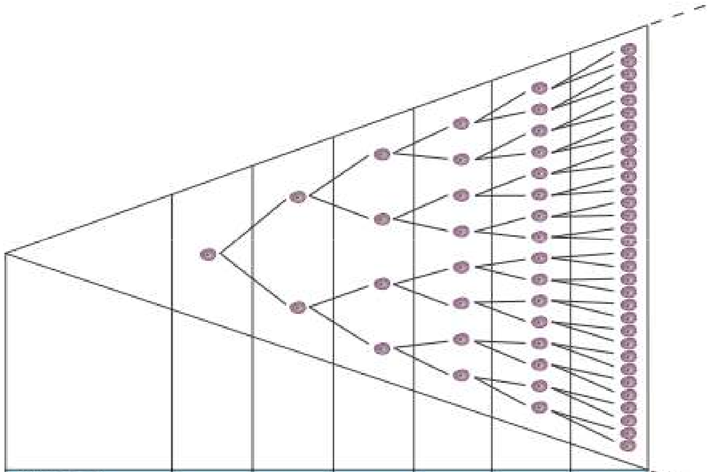
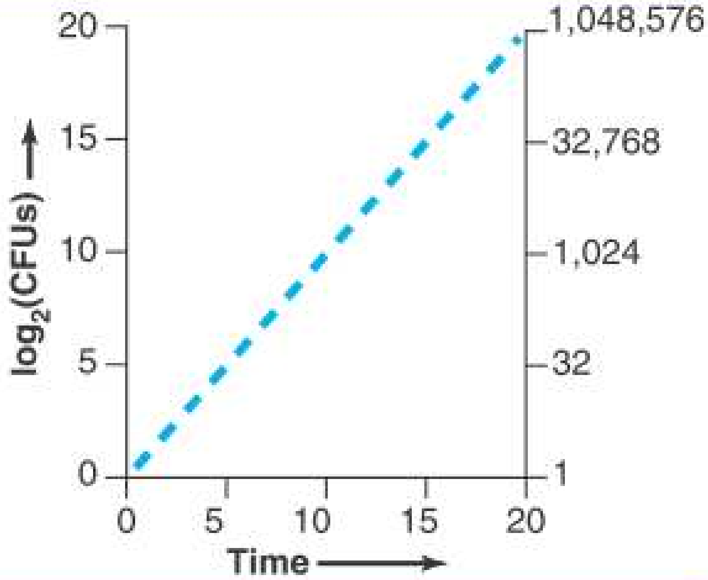
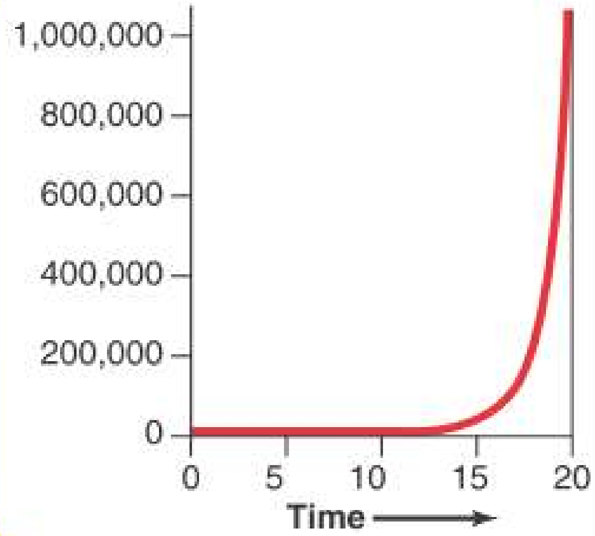
The mathematics of population growth

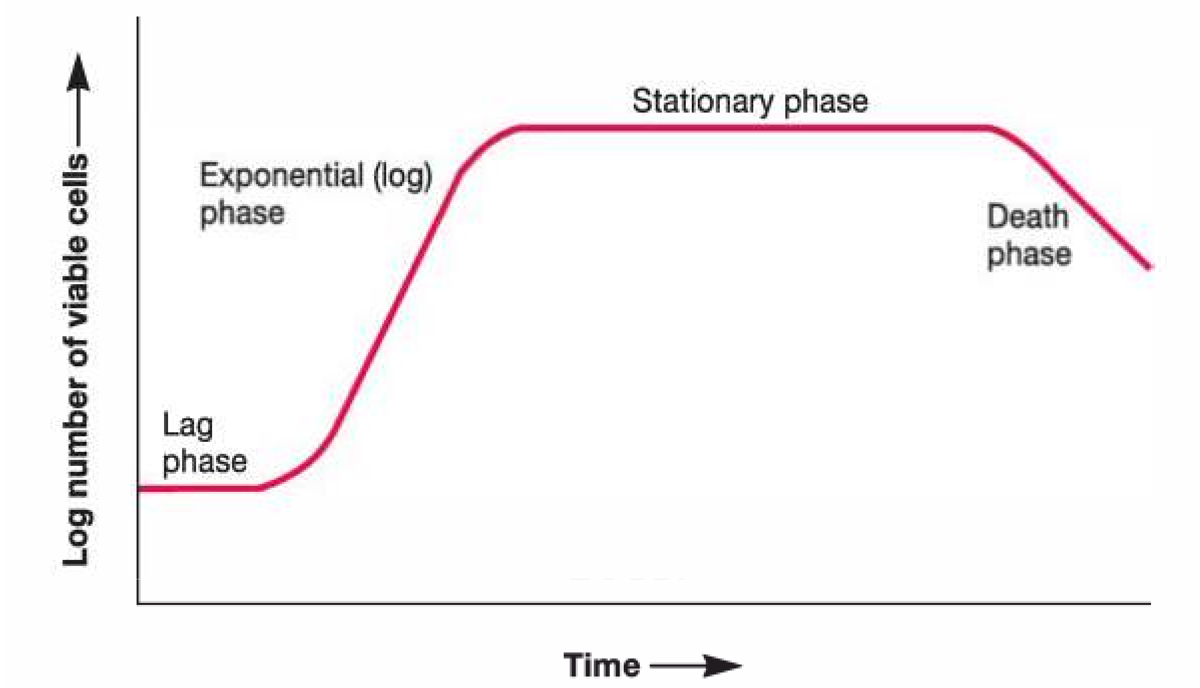


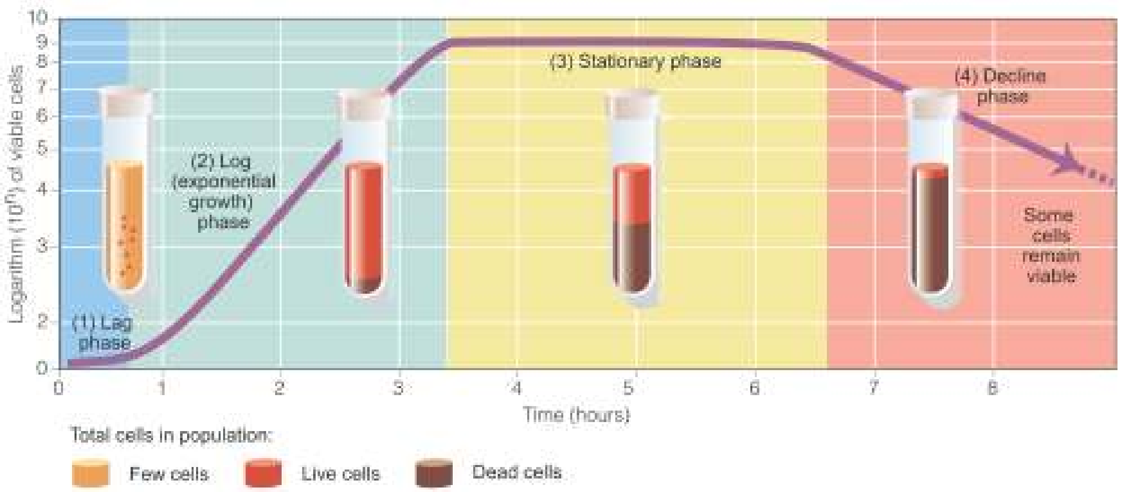
Number of cells	1	2	4	8	16	32
Number of generations	0	1	2	3	4	5
Exponential representation	2^0 (1)	2^1 (2)	2^2 (2 × 2)	2^3 (2 × 2 × 2)	2^4 (2 × 2 × 2 × 2)	2^5 (2 × 2 × 2 × 2 × 2)
Logarithm (base 2)	0	1	2	3	4	5





MICROBIAL GROWTH CYCLE





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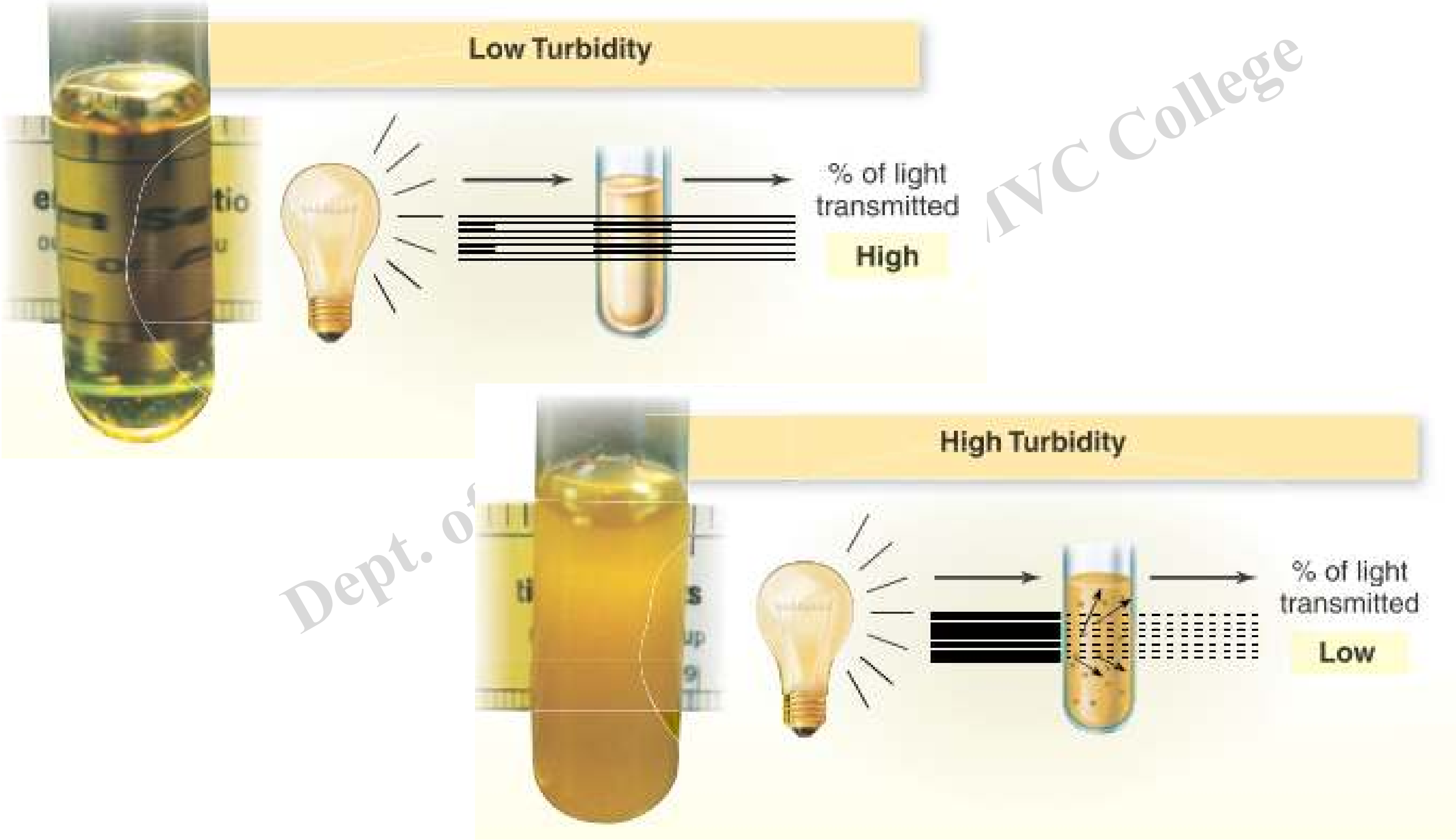
Turbidity measurements as indicators of growth



Lag and Exponential Phases

- When a microbial culture is inoculated into fresh growth media growth begins only after a period of time called the lag phase. This interval may be brief or extended depending on the history of the cells used as inoculum and the composition of the growth medium and growth conditions.

- If an exponentially growing culture is transferred into the same medium under the same conditions of growth, there will be essentially no lag and exponential growth will begin immediately.

- However, if the inoculum is taken from an old culture, there is usually a lag because the cells are depleted of various essential constituents and time is required for their biosynthesis.

- In order to grow, cells must have a complete complement of enzymes for synthesis of the essential metabolites not present in that medium. Hence, when transferred to a nutrient-poor medium, time is needed for the biosynthesis of new enzymes and for these to produce a sufficient pool of required metabolites before growth can actually begin. These events occur during the lag period.

Lag and Exponential Phases

- When a growing cell population doubles at regular intervals the cells are said to be in the exponential phase of growth. Exponential phase cells are typically in their healthiest state and are thus most desirable for studies of their enzymes or other cell components. Rates of exponential growth vary greatly. Exponential growth rates are influenced by the growth conditions an organism is experiencing as well as genetic characteristics of the organism itself.

Stationary and death Phases

- When exponential growth ceases, the population enters **stationary phase**. In the stationary phase, **there is no net increase or decrease in cell number and thus the growth rate of the population is zero**. Despite growth arrest, energy metabolism and biosynthetic processes in stationary phase cells may continue, but typically at **a greatly reduced rate**. Some cells may even divide during stationary phase, but no net increase in cell number occurs. This is because some cells in the population grow while others die, the two processes balancing each other out (**cryptic growth**).

- Eventually, the population will enter the death phase of the growth cycle occurs as an exponential function. Typically, however, the **rate of cell death is much slower than the rate of exponential growth and** viable cells may remain in a culture for months or even years.

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The Mathematics of Bacterial Growth

The increase in cell number in an exponentially growing bacterial culture can be expressed mathematically as a geometric progression of the number 2. A fixed relationship exists between the initial number of cells in a culture and the number present after a period of exponential growth, and this relationship is expressed as

$$N_t = N_0 \times 2^n$$

where N_t is the final cell number, N_0 is the initial cell number, and n is the number of generations during the period of exponential growth.

Generation time: The interval of time between successive binary fissions of a cell or population of cells is known as the generation time (or doubling time). The generation time of a given bacterial species is variable and depends on nutritional and genetic factors, and on temperature.

Under the best nutritional conditions, the generation time of a laboratory culture of *E. coli* is about 20 min.

The Mathematics of Bacterial Growth

Calculation of the growth rate constant

Let N_0 = the initial population number

N_t = the population at time t

n = the number of generations in time t

For populations reproducing by binary fission

$$N_t = N_0 \times 2^n$$

Solving for n, the number of generations, where all logarithms are to the base 10,

$$\log N_t = \log N_0 + n \cdot \log 2, \text{ and}$$

$$n = \frac{\log N_t - \log N_0}{\log 2} = \frac{\log N_t - \log N_0}{0.301}$$

$n = [3.3(\log N_t - \log N_0)]$

The growth rate constant (k) is the number of generations per unit time $\left(\frac{n}{t}\right)$. Thus

$$k = \frac{n}{t} = \frac{\log N_t - \log N_0}{0.301\,t}$$

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The Mathematics of Bacterial Growth

- The equation $N_t = N_0 \times 2^n$ can be expressed in terms of n by taking the logarithms of both sides and doing some simple algebra to yield the expression

$$n = [3.3(\log N_t - \log N_0)]$$

- Using this expression, it is possible to calculate generation times in terms of measurable quantities, N_t and N_0

- The **generation time** (g) of the exponentially growing population is t/n, where t is the duration of exponential growth expressed in days, hours, or minutes.

  - $g = t / n = t / [3.3(\log N_t - \log N_0)]$

  - $g = t / 3.3 \log N_t / N_0$

- **Specific growth rate** is defined as the rate of increase of biomass of a cell population per unit of biomass concentration. It can be calculated in batch cultures, since during a defined period of time, the rate of increase in biomass per unit of biomass concentration is constant and measurable.

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Bacterial Growth

- The **generation time**, which varies among bacteria, is controlled by many **environmental conditions** and by the **nature of the bacterial species**. For example, *Clostridium perfringens*, one of the fastest-growing bacteria, has an optimum generation time of about 10 minutes; *Escherichia coli* can double every 20 minutes; and the slow-growing *Mycobacterium tuberculosis* has a generation time in the range of 12 to 16 hours.

- The **composition of the growth medium** is a major factor controlling the growth rate. The growth rate increases up to a maximum when the **medium provides a better energy source** and more of the biosynthetic intermediates that the cell would otherwise have to make for itself.

- When bacteria are placed in a **medium that provides all of the nutrients** that are necessary for their growth, the population exhibits **four phases of growth** that are representative of a typical **bacterial growth curve**.

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