Growth Curve

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Growth Curve

- Population growth is studied by analyzing the growth curve of a microbial culture.
- When microorganisms are cultivated in liquid medium, they usually are grown in a **batch culture or closed system—**that is, they are incubated in a closed culture vessel with a single batch of medium.
- Because no fresh medium is provided during incubation, nutrient concentrations decline and concentrations of wastes increase.
- The growth of microorganisms reproducing by binary fission can be plotted as the logarithm of the number of viable cells versus the incubation time.
- The resulting curve has four distinct phases:
  - Lag Phase
  - Exponential/ Log phase
  - Stationary phase
  - Death phase

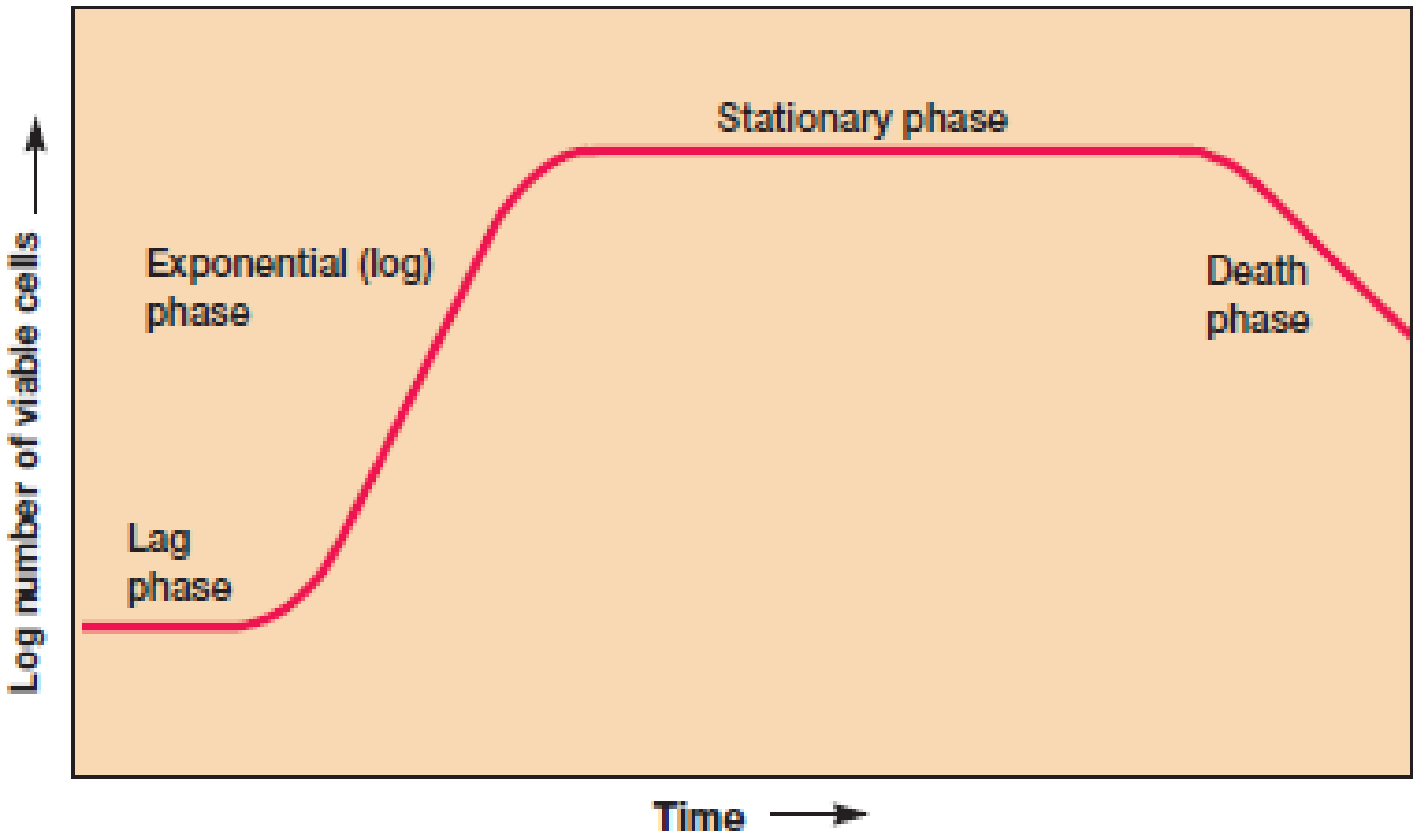

Figure 6.1 Microbial Growth Curve in a Closed System. The four phases of the growth curve are identified on the curve and discussed in the text.

Lag Phase

- When microorganisms are introduced into fresh culture medium, usually no immediate increase in cell number occurs, and therefore this period is called the **lag phase.**
- During lag phase the cell is synthesize new components.
- The cells may be old and depleted of ATP, essential cofactors, and ribosomes; these must be synthesized before growth can begin.
- The medium may be different from the one the microorganism was growing in previously.
- Here new enzymes would be needed to use different nutrients.
- Possibly the microorganisms have been injured and require time to recover.
- Whatever the causes, eventually the cells retool, replicate their DNA, begin to increase in mass, and finally divide.

Exponential Phase

- During the **exponential or log phase, microorganisms are** growing and dividing at the maximal rate possible given their genetic potential, the nature of the medium, and the conditions under which they are growing.
- Their rate of growth is constant during the exponential phase; that is, the microorganisms are dividing and doubling in number at regular intervals.
- Because each individual divides at a slightly different moment, the growth curve rises smoothly rather than in discrete jumps.
- Exponential growth is **balanced growth.**
- **That is, all cellular** constituents are manufactured at constant rates relative to each other. If nutrient levels or other environmental conditions change, **unbalanced growth results.**

Stationary Phase

- Eventually population growth ceases and the growth curve becomes horizontal.
- Of course final population size depends on nutrient availability and other factors, as well as the type of microorganism being cultured.
- In the stationary phase the total number of viable microorganisms remains constant.
- This may result from a balance between cell division and cell death, or the population may simply cease to divide though remaining metabolically active.
- Microbial populations enter the stationary phase for several reasons.
- One obvious factor is nutrient limitation; if an essential nutrient is severely depleted, population growth will slow.
- Aerobic organisms often are limited by O_2 availability.
- Population growth also may cease due to the accumulation of toxic waste products.
- Finally, there is some evidence that growth may cease when a critical population level is reached.
- Thus entrance into the stationary phase may result from several factors operating in concert.

Death Phase

- Detrimental environmental changes like nutrient deprivation and the buildup of toxic wastes lead to the decline in the number of viable cells characteristic of the **death phase.**
- The death of a microbial population, like its growth during the exponential phase, is usually logarithmic (that is, a constant proportion of cells dies every hour).
- This pattern in viable cell count holds even when the total cell number remains constant because the cells simply fail to lyse after dying.
- Often the only way of deciding whether a bacterial cell is viable is by incubating it in fresh medium; if it does not grow and reproduce, it is assumed to be dead.
- That is, death is defined to be the irreversible loss of the ability to reproduce.
- Although most of a microbial population usually dies in a logarithmic fashion, the death rate may decrease after the population has been drastically reduced.
- This is due to the extended survival of particularly resistant cells.
- For this and other reasons, the death phase curve may be complex.

The Mathematics of Growth

- During the exponential phase each microorganism is dividing at constant intervals.
- Thus the population will double in number during a specific length of time called the **generation time or doubling time.**
- **Suppose:** that a culture tube is inoculated with one cell that divides every 20 minutes**.**
- The population will be 2 cells after 20 minutes, 4 cells after 40 minutes, and so forth.
- Because the population is doubling every generation, the increase in population is always 2^n where n is the number of generations.
- The resulting population increase is exponential or logarithmic.

Table 6.1 An Example of Exponential Growth

Time[a]	Division Number	2^n	Population ($N_0 \times 2^n$)	$\log_{10} N_t$
0	0	$2^0 = 1$	1	0.000
20	1	$2^1 = 2$	2	0.301
40	2	$2^2 = 4$	4	0.602
60	3	$2^3 = 8$	8	0.903
80	4	$2^4 = 16$	16	1.204
100	5	$2^5 = 32$	32	1.505
120	6	$2^6 = 64$	64	1.806

[a] The hypothetical culture begins with one cell having a 20-minute generation time.

These observations can be expressed as equations for the generation time.

$$\begin{aligned} \text{Let } N_0 &= \text{the initial population number} \\ N_t &= \text{the population at time } t \\ n &= \text{the number of generations in time } t \end{aligned}$$

Then inspection of the results in table 6.1 will show that

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

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

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

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

The rate of growth during the exponential phase in a batch culture can be expressed in terms of the **mean growth rate constant** ***(k)***. This is the number of generations per unit time, often expressed as the generations per hour.

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

The time it takes a population to double in size—that is, the **mean generation time** or mean doubling time (g), can now be calculated. If the population doubles ($t = g$), then

$$N_t = 2\,N_0.$$

Substitute $2N_0$ into the mean growth rate equation and solve for k.

$$k = \frac{\log(2N_0) - \log N_0}{0.301g} = \frac{\log 2 + \log N_0 - \log N_0}{0.301g}$$

$$k = \frac{1}{g}$$

The mean generation time is the reciprocal of the mean growth rate constant.

$$g = \frac{1}{k}$$

Examples

- For example, suppose that a bacterial population increases from 10^3 cells to 10^9 cells in 10 hours.

$$k = \frac{\log 10^9 - \log 10^3}{(0.301)(10\ \text{hr})} = \frac{9 - 3}{3.01\ \text{hr}} = 2.0\ \text{generations/hr}$$

$$g = \frac{1}{2.0\ \text{gen./hr}} = 0.5\ \text{hr/gen. or 30 min/gen.}$$

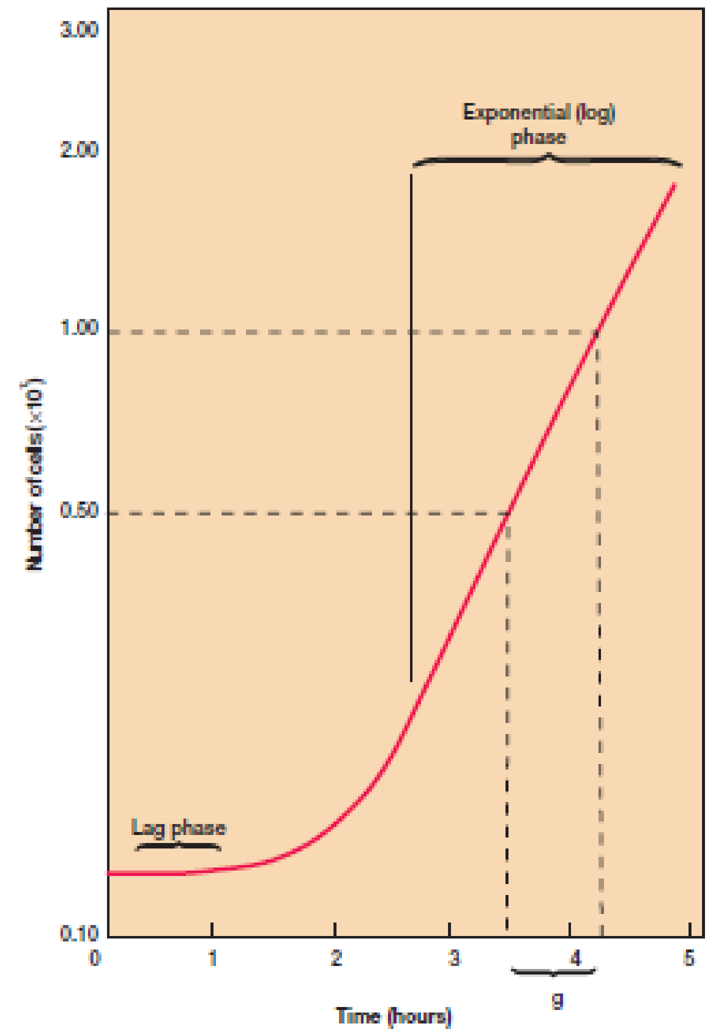

Figure 6.4 Generation Time Determination. The generation time can be determined from a microbial growth curve. The population data are plotted with the logarithmic axis used for the number of cells. The time to double the population number is then read directly from the plot. The log of the population number can also be plotted against time on regular axes.

Specific Growth Rate

- Specific growth rate (S_g) The specific growth rate period is defined as **the rate of increase of biomass of a cell population per unit of biomass concentration**.
- In the sigmoid curve it occurs between the lag and stationary phases where cell growth follows a straight-line equation as follows:

$$\ln F_m = S_g\, t + \ln I_m$$

$$S_g = \ln F_m - \ln I_m/t$$

- Where S_g is specific growth rate, F_m is the biomass (or cell density) at time t, and I_m is the initial biomass (or cell density).