Micro 260

Chapter6B

Bacterial Nutrition and Growth

Ecological association

- Influence microorganisms have on other microbes
  - Symbiotic relationship
  - Non-symbiotic relationship

Symbiotic

- Organisms that live in close nutritional relationship
- Types
  - Mutualism – both organism benefit
  - Commensalism – one organisms benefits
  - Parasitism – host/microbe relationship

An example of commensalism, where *Staphylococcus aureus* provides vitamins and amino acids to *Haemophilus influenzae.*

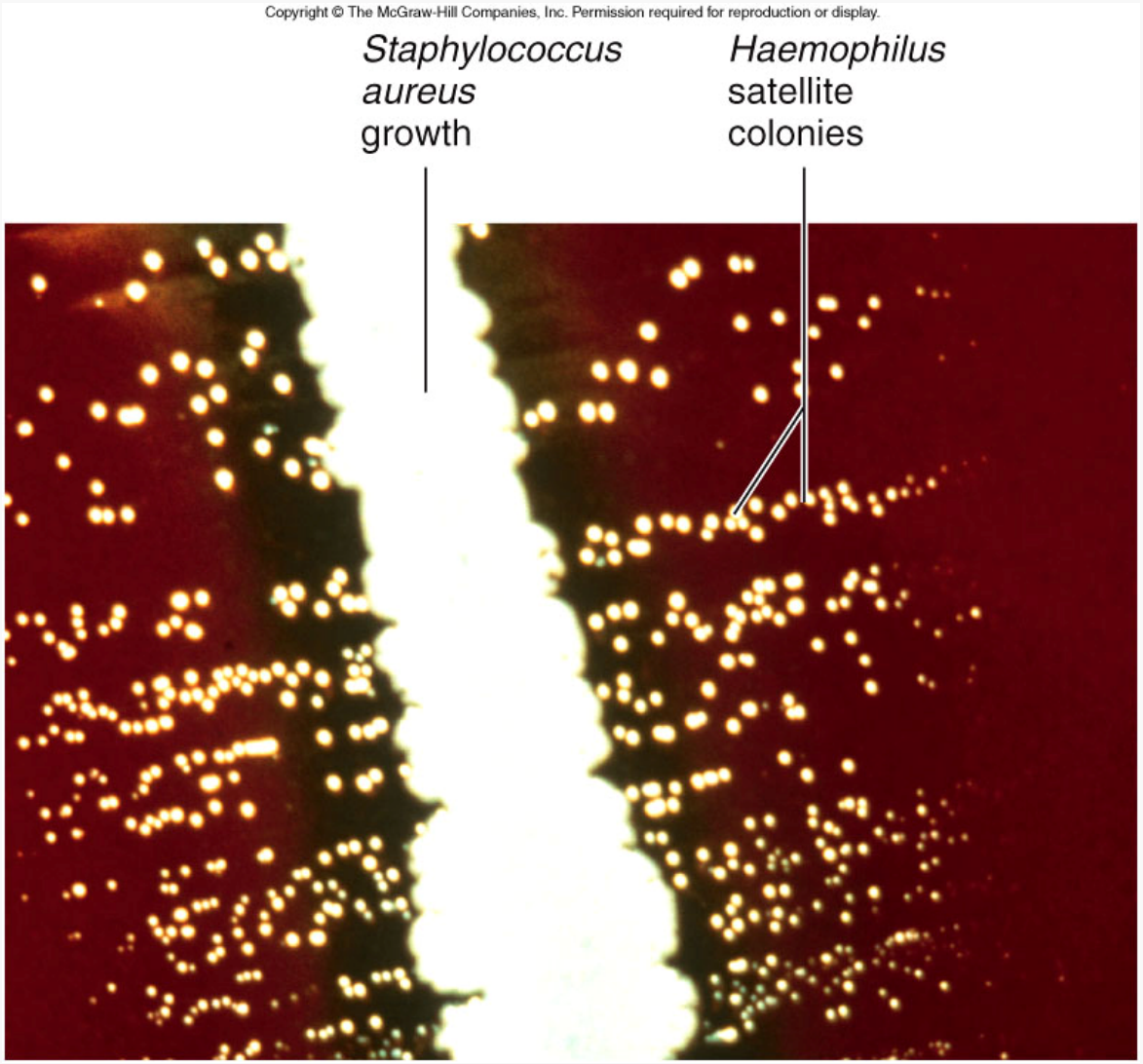


Fig. 7.12 Satellitism, a type of commensalism

Non-symbiotic

- Organisms are free-living, and do not rely on each other for survival
- Types
  - Synergism – shared metabolism, not required
  - Antagonism- competition between microorganisms

Interrelationships between microbes and humans

- Can be commensal, parasitic, and synergistic
- Ex. *E. coli* produce vitamin K for the host

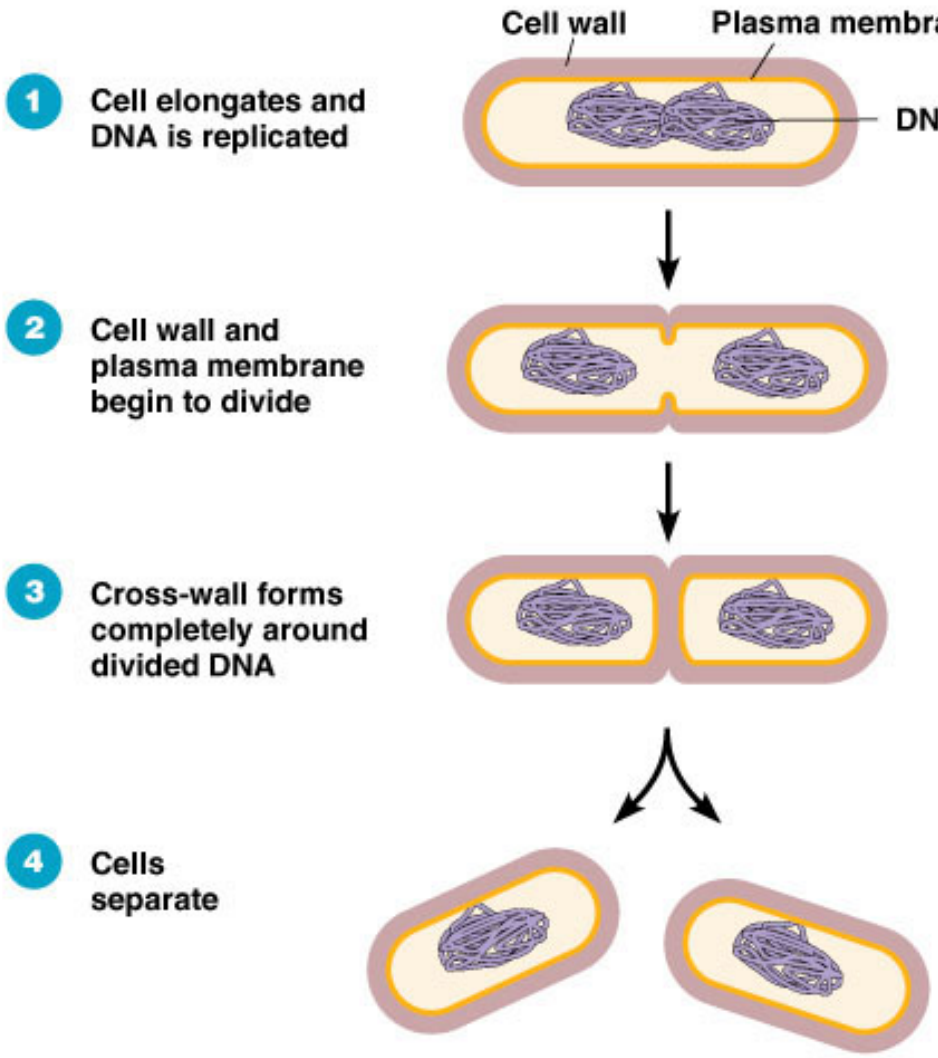
Preserving Bacteria Cultures

- Deep-freezing: -50°to -95°C
- Lyophilization (freeze-drying): Frozen (-54° to -72°C) and dehydrated in a vacuum

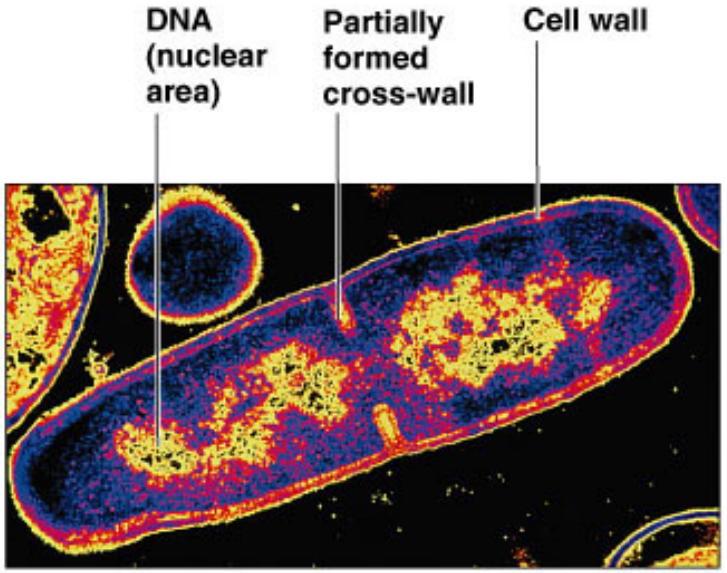
Reproduction in Prokaryotes

- Binary fission
- Budding
- Conidiospores (actinomycetes)
- Fragmentation of filaments

Binary Fission



(a) A diagram of the sequence of cell division.



(b) A thin section of a cell of *Bacillus licheniformis* starting to divide.

Figure 6.11

Generation time

- The time required for a complete division cycle (doubling)
- Length of the generation time is a measure of the growth rate
- Exponentials are used to define the numbers of bacteria after growth

Generation Number	Number of Cells	Log_{10} of Number of Cells
0	1	0
5 (2^5) =	32	1.51
10 (2^{10}) =	1,024	3.01
15 (2^{15}) =	32,768	4.52
16 (2^{16}) =	65,536	4.82
17 (2^{17}) =	131,072	5.12
18 (2^{18}) =	262,144	5.42
19 (2^{19}) =	524,288	5.72
20 (2^{20}) =	1,048,576	6.02

(b)

Figure 6.12b

If 100 cells growing for 5 hours produced 1,720,320 cells:

$$\text{Number of generations} = \frac{\text{Log number of cells (end)} - \text{Log number of cells (beginning)}}{0.301}$$

$$\text{Generation time} = \frac{60 \text{ min} \times \text{hours}}{\text{Number of generations}} = 21 \text{ minutes/generation}$$

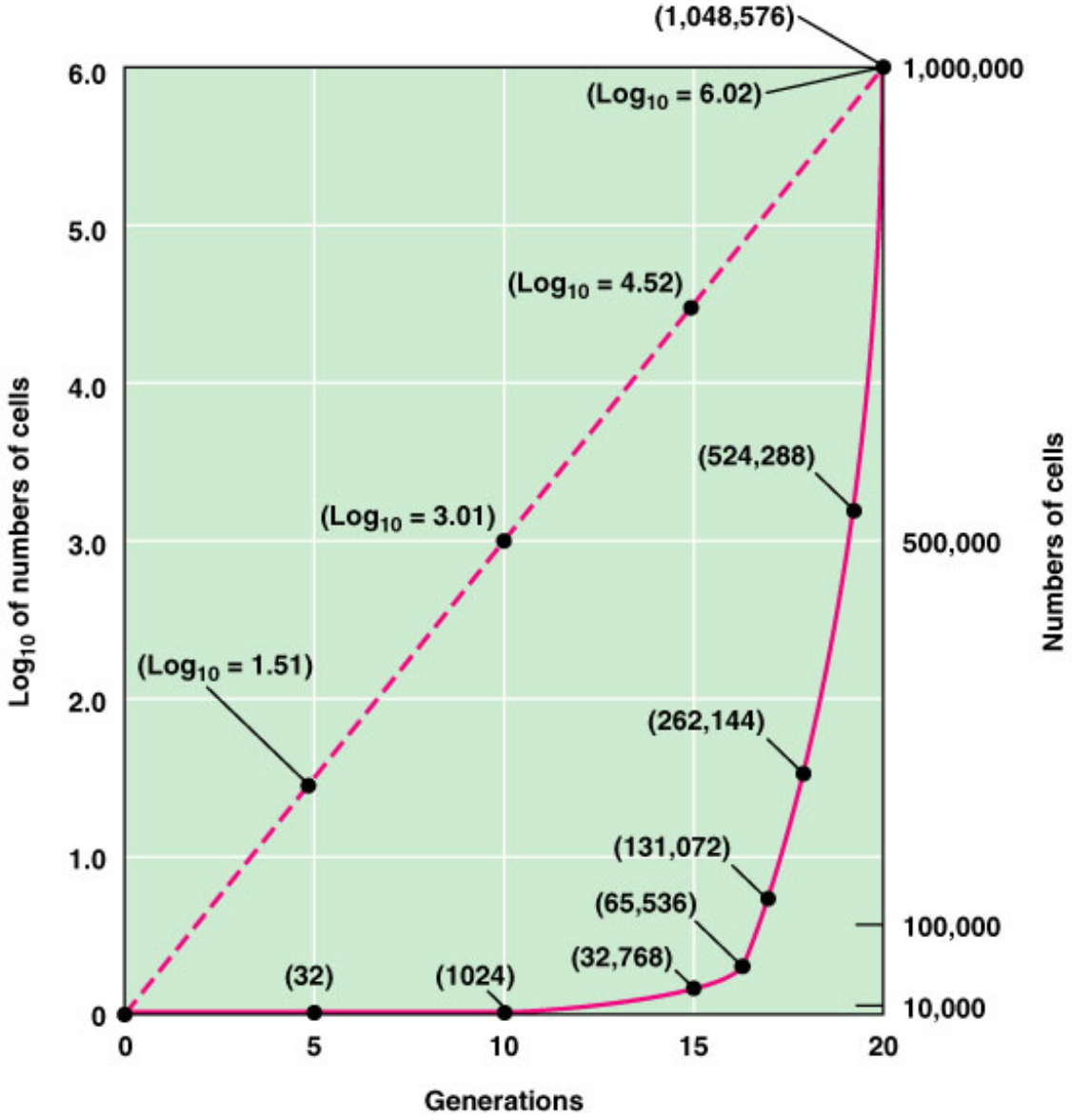
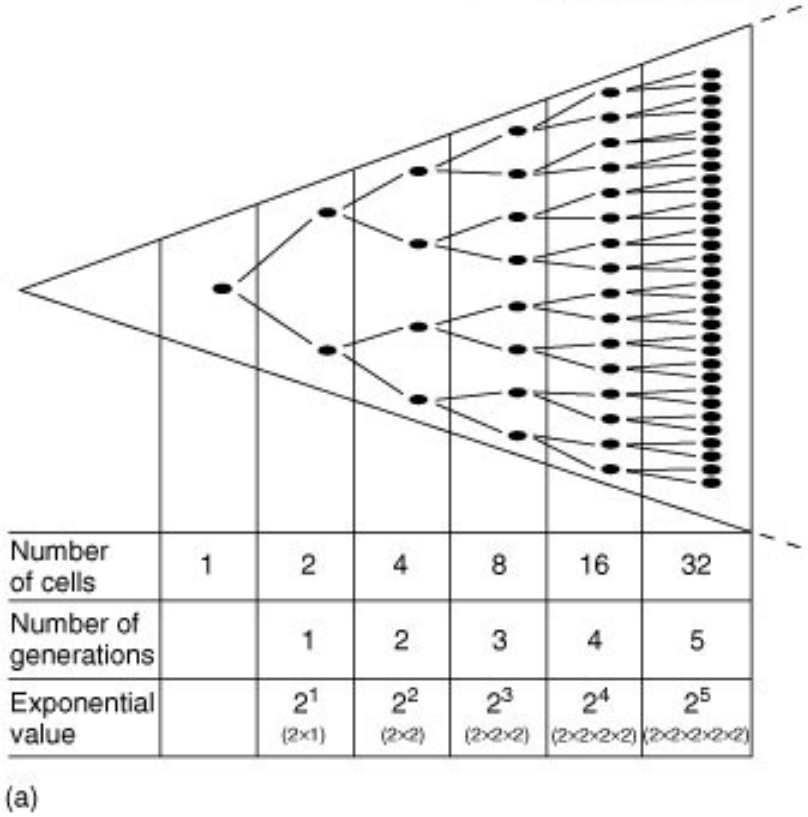

(1,048,576)
1,000,000
(Log10 = 6.02)
6.0
5.0
(Log10 = 4.52)
4.0
(524,288)
(Log10 = 3.01)
3.0
500,000
(Log10 = 1.51)
2.0
(262,144)
1.0
(131,072)
(65,536)
100,000
(32,768)
(32)
(1024)
0
10,000
0
5
10
15
20
Generations
Log10 of numbers of cells
Numbers of cells

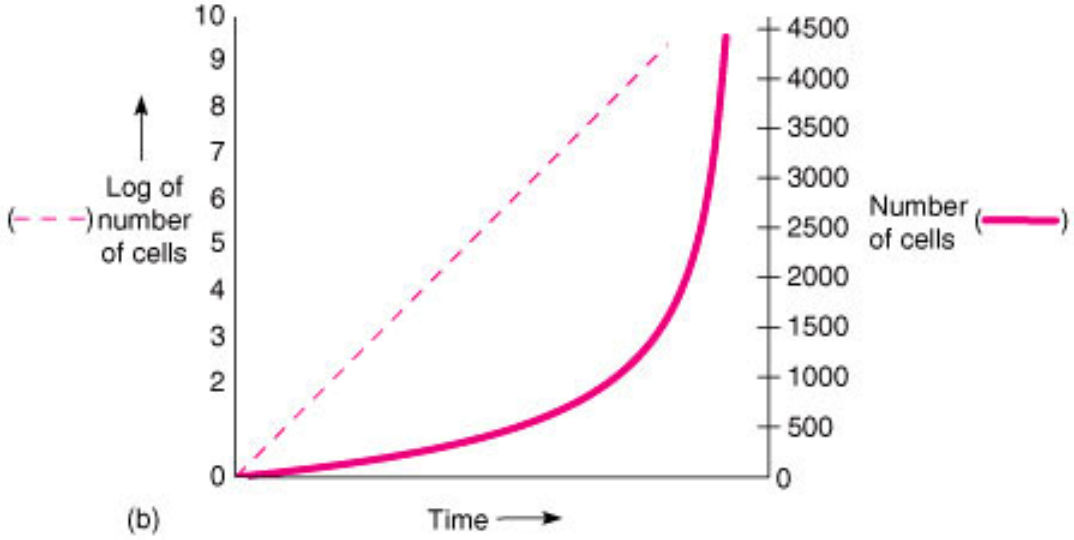
Figure 6.13

Representation of how a single bacterium doubles after a complete division, and how this can be plotted using exponentials.



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)



(b)

Fig. 7.14 The mathematics of population growth

Growth curve

- Lag phase
- Log phase
- Stationary phase
- Death phase

Lag phase

- Cells are adjusting, enlarging, and synthesizing critical proteins and metabolites
- Not doubling at their maximum growth rate

Log phase

- Maximum exponential growth rate of cell division
- Adequate nutrients
- Favorable environment

Stationary phase

- Survival mode – depletion in nutrients, released waste can inhibit growth
- When the number of cells that stop dividing equal the number of cells that continue to divide

Death phase

- A majority of cells begin to die exponentially due to lack of nutrients
- A chemostat will provide a continuous supply of nutrients, thereby the death phase is never achieved.

The four main phases of growth in a bacterial culture.

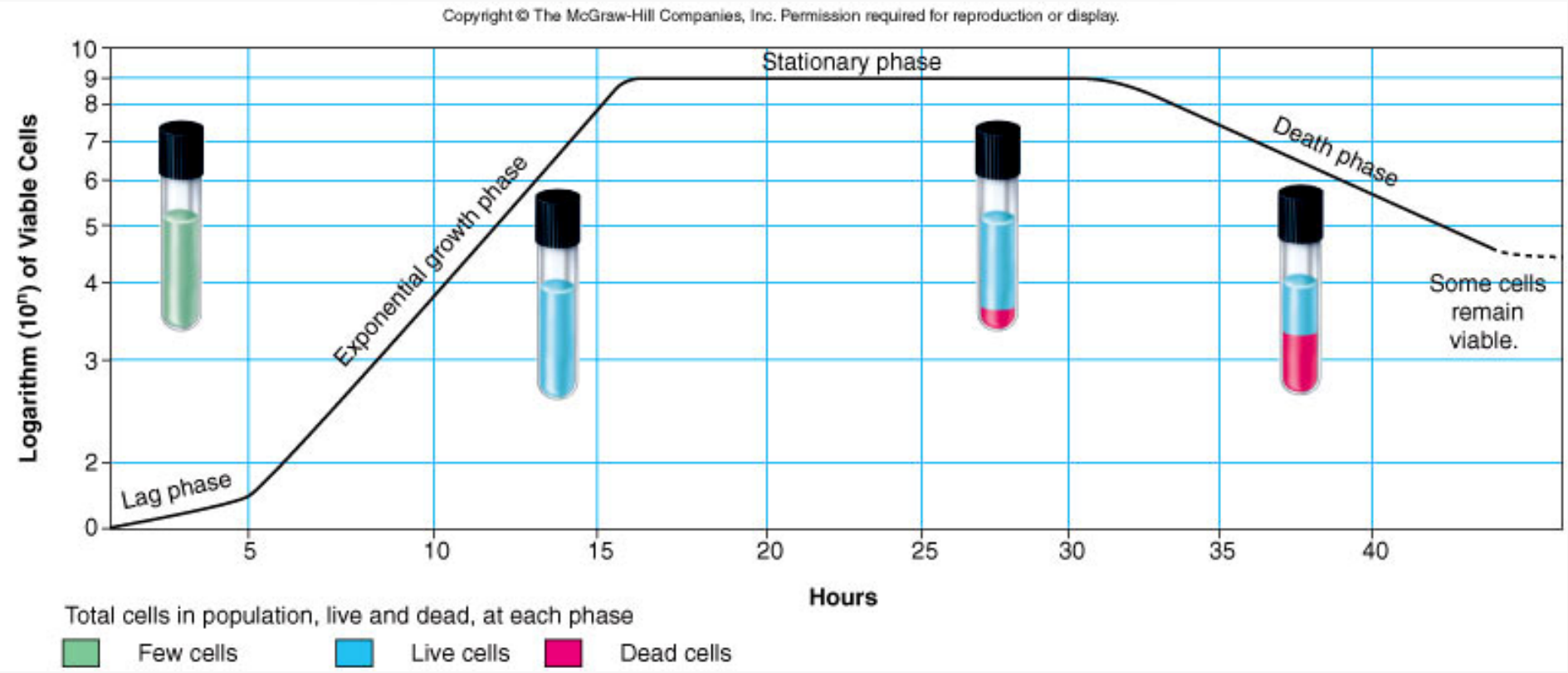


Fig. 7.15 The growth curve in a bacterial culture.

Direct Measurements of Microbial Growth

- Plate Counts: Perform serial dilutions of a sample

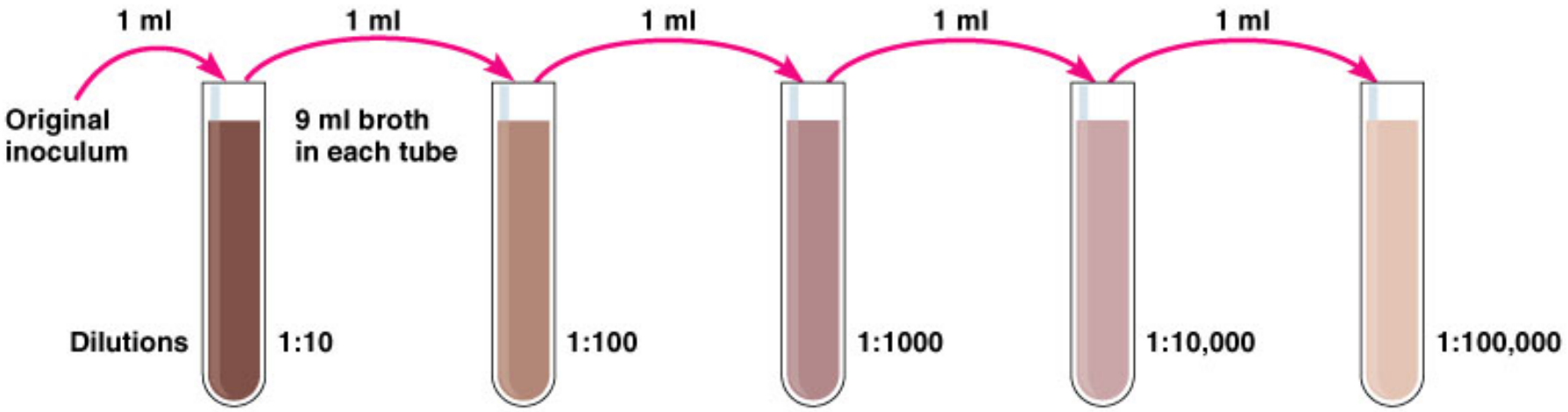


Figure 6.15, top portion

Plate Count

- Inoculate Petri plates from serial dilutions

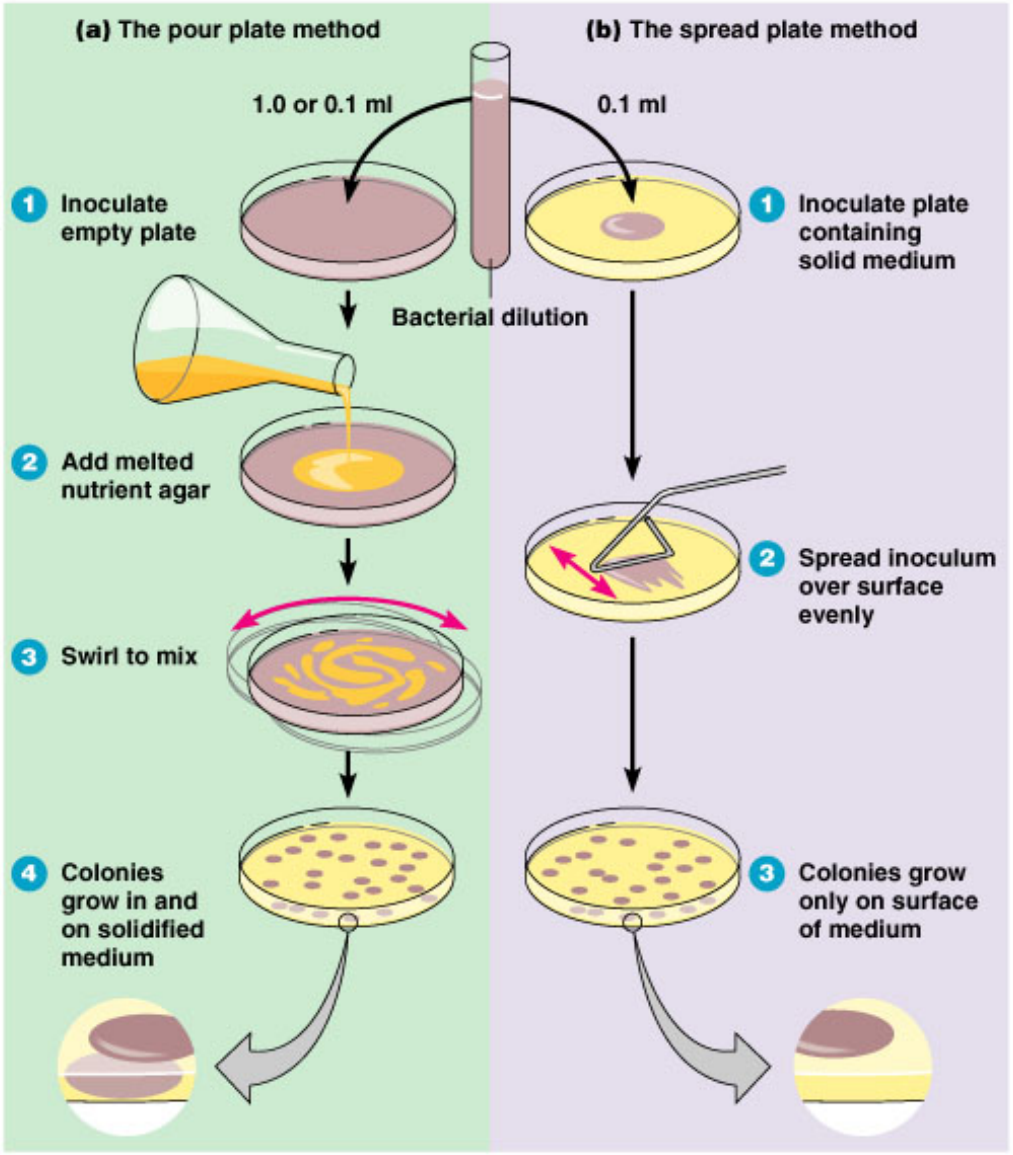
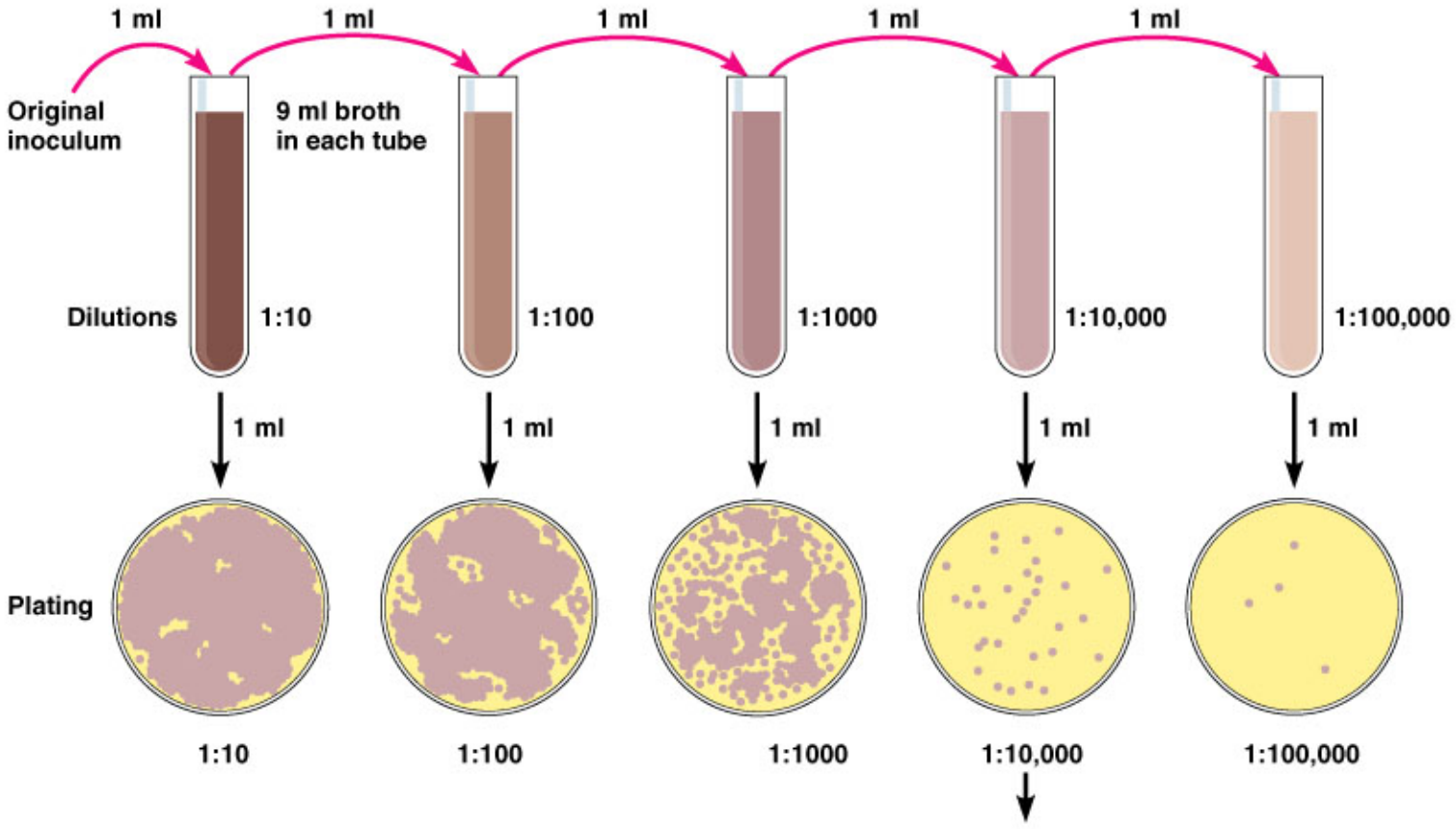


Figure 6.16

Plate Count

- After incubation, count colonies on plates that have 25-250 colonies (CFUs)

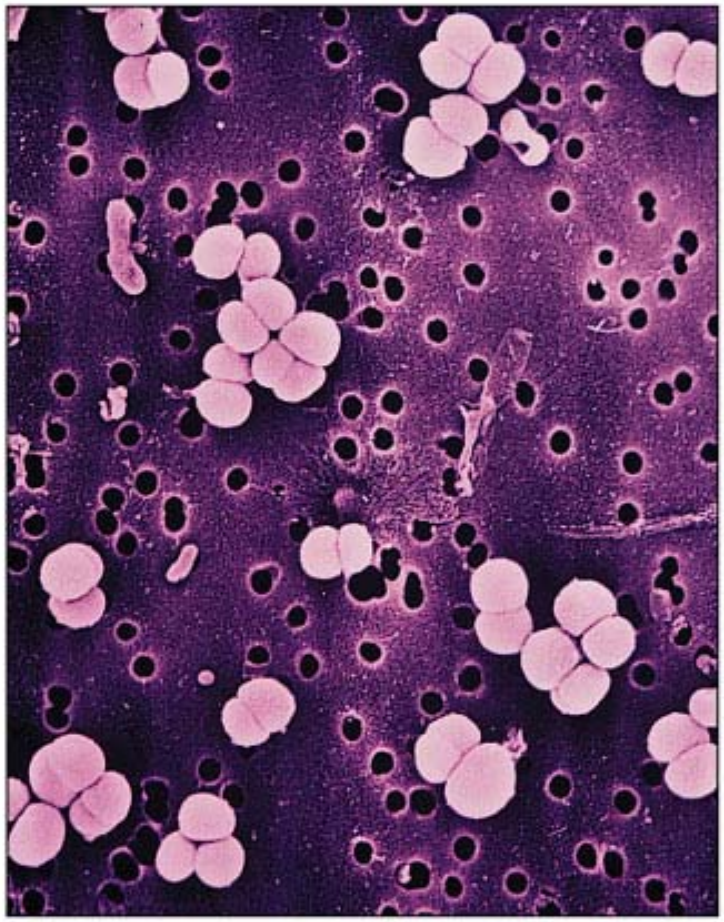


Calculation: Number of colonies on plate × reciprocal of dilution of sample = number of bacteria/ml
(For example, if 32 colonies are on a plate of $^{1}/_{10,000}$ dilution, then the count is 32 × 10,000 = 320,000/ml in sample.)

Figure 6.15

Direct Measurements of Microbial Growth

- Filtration



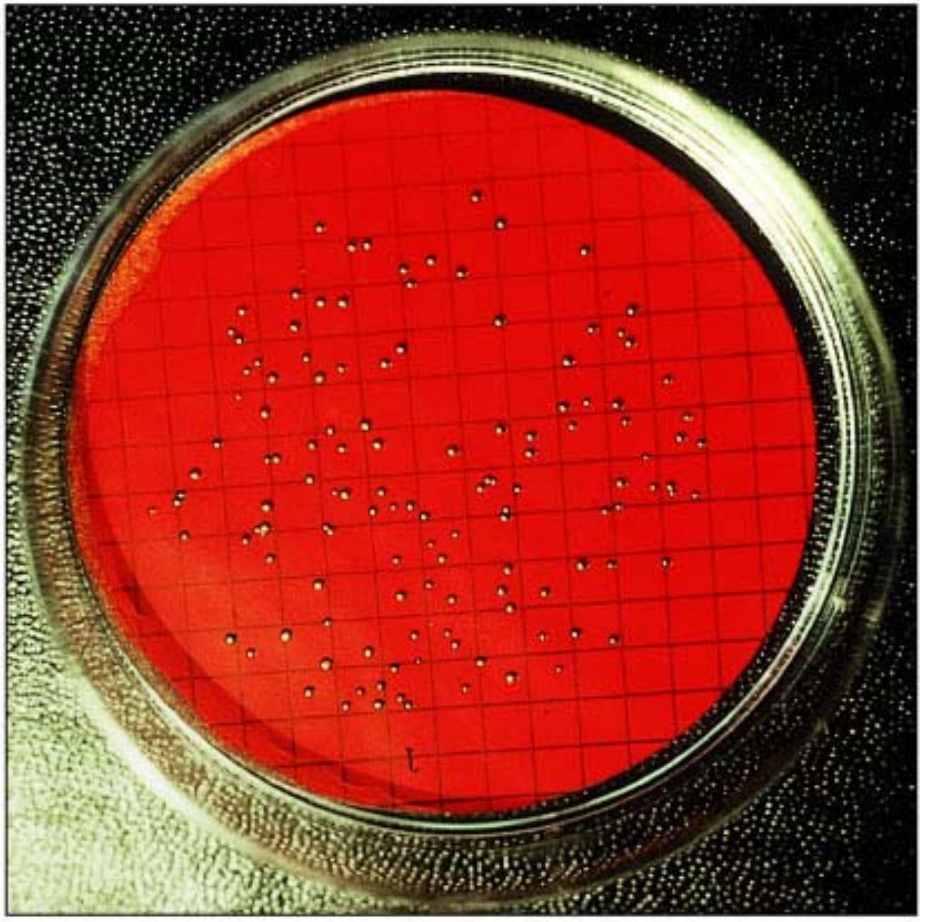


Figure 6.17a, b

Direct Measurements of Microbial Growth

- Multiple tube MPN test
- Count positive tubes and compare to statistical MPN table.

Combination of Positives	MPN Index/ 100 ml	95% Confidence Limits	
		Lower	Upper
4-2-0	22	9	56
4-2-1	26	12	65
4-3-0	27	12	67
4-3-1	33	15	77
4-4-0	34	16	80
5-0-0	23	9	86
5-0-1	30	10	110
5-0-2	40	20	140
5-1-0	30	10	120
5-1-1	50	20	150
5-1-2	60	30	180
5-2-0	50	20	170
5-2-1	70	30	210
5-2-2	90	40	250
5-3-0	80	30	250
5-3-1	110	40	300
5-3-2	140	60	360

Figure 6.18b

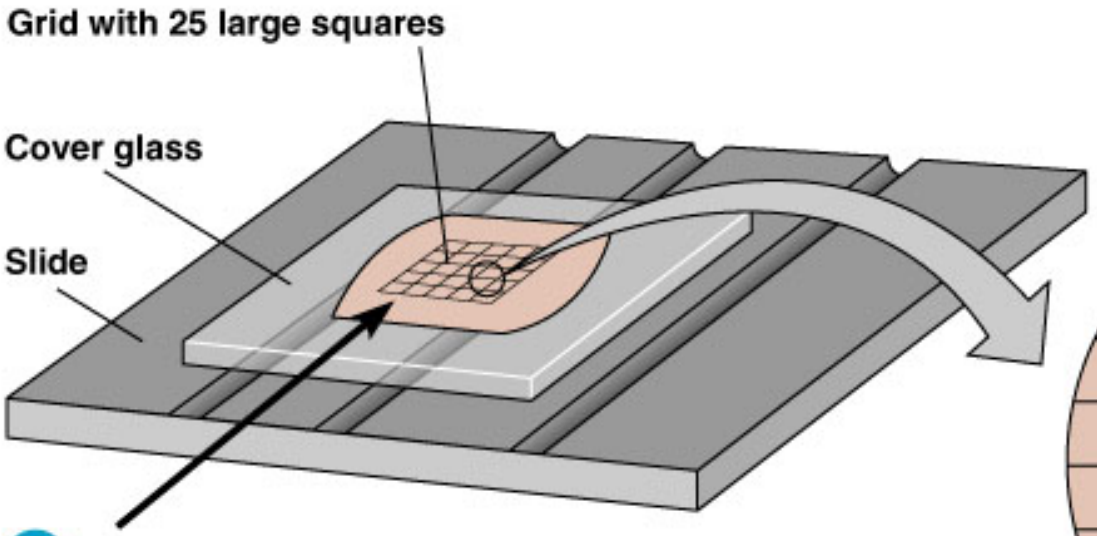
Direct Measurements of Microbial Growth

- Direct Microscopic Count

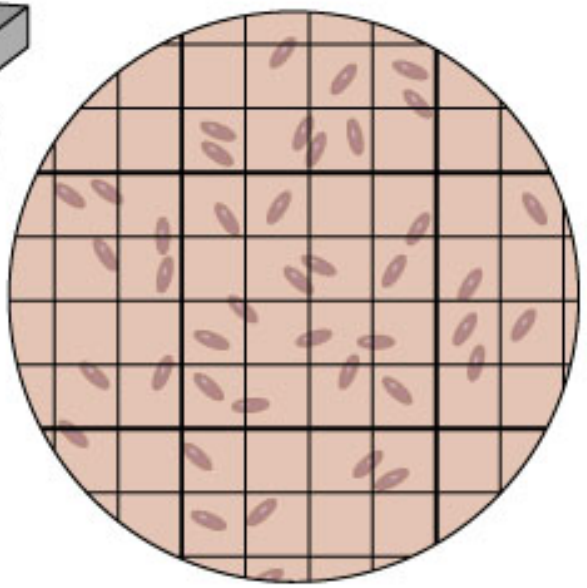
$$\text{Number of bacteria/ml} = \frac{\text{number of cells counted}}{\text{volume of area counted}}$$

$$\frac{14}{8 \times 10^{-7}} = 17{,}500{,}000$$

Direct Measurements of Microbial Growth



1 Bacterial suspension is added here and fills the shallow volume over the squares by capillary action.



3 Microscopic count: All cells in several large squares are counted, and the numbers are averaged. The large square shown here has 14 bacterial cells.

Figure 6.19

Estimating Bacterial Numbers by Indirect Methods

- Turbidity

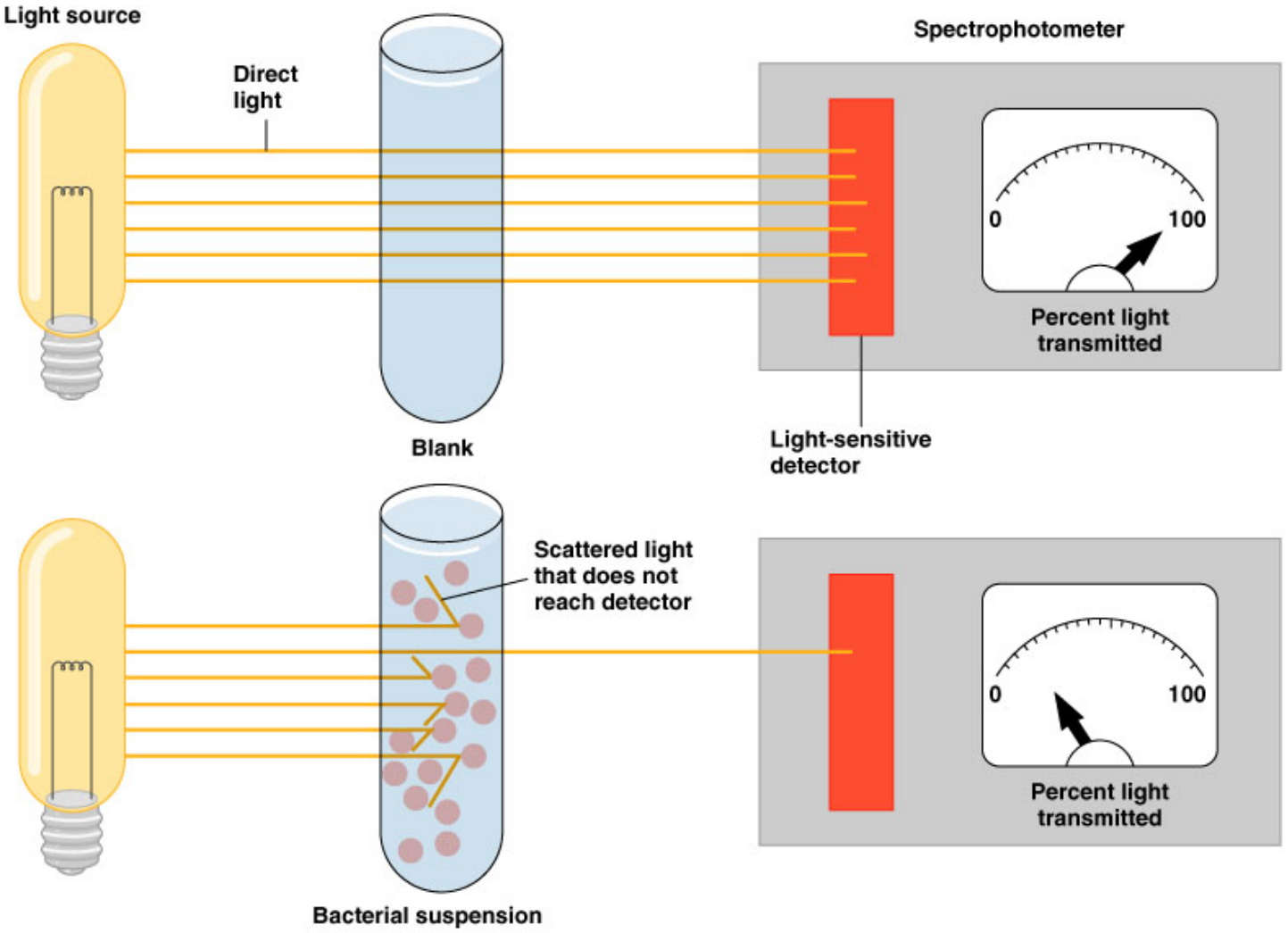


Figure 620

Estimating Bacterial Numbers by Indirect methods

- Metabolic activity
- Dry weight