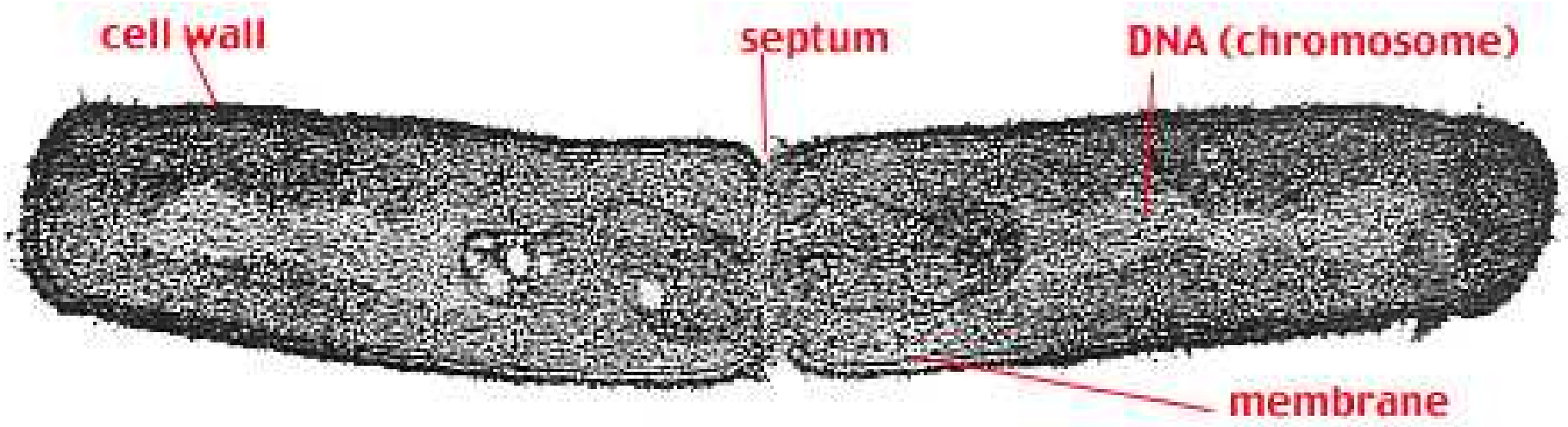
MICROBIOLOGIA GENERALE

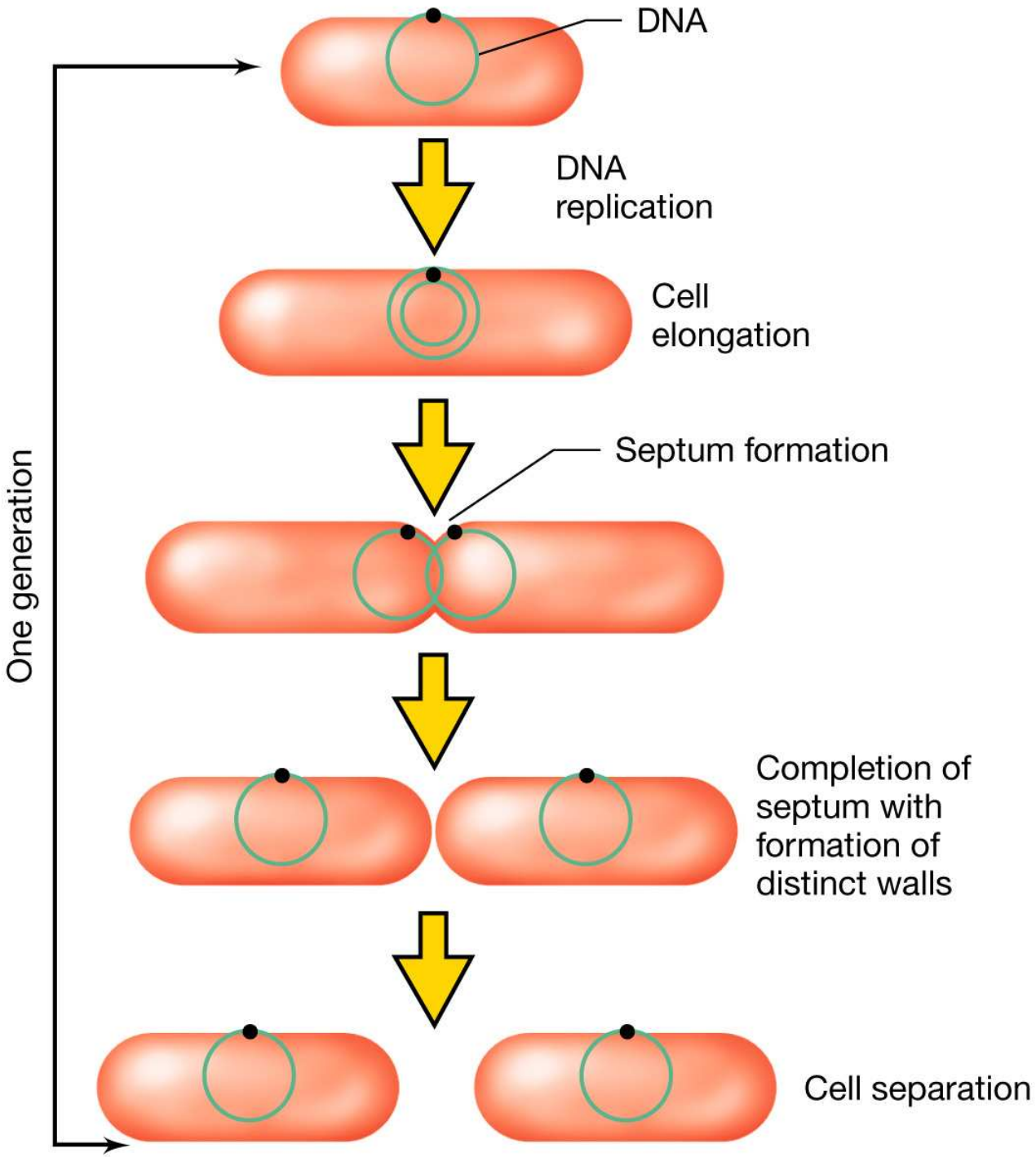
Microbial growth 1

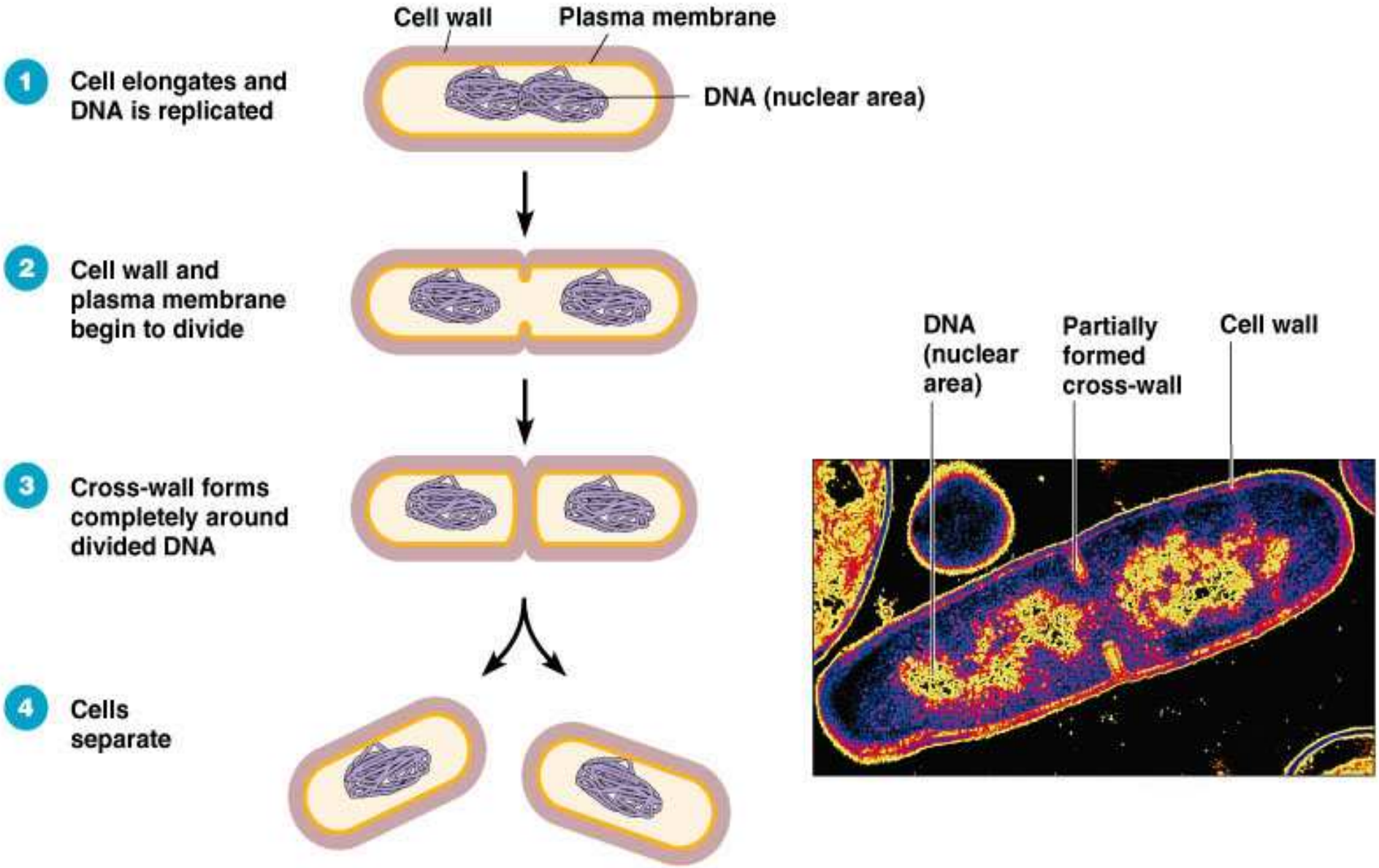
Microbial growth:

cell growth and binary fission

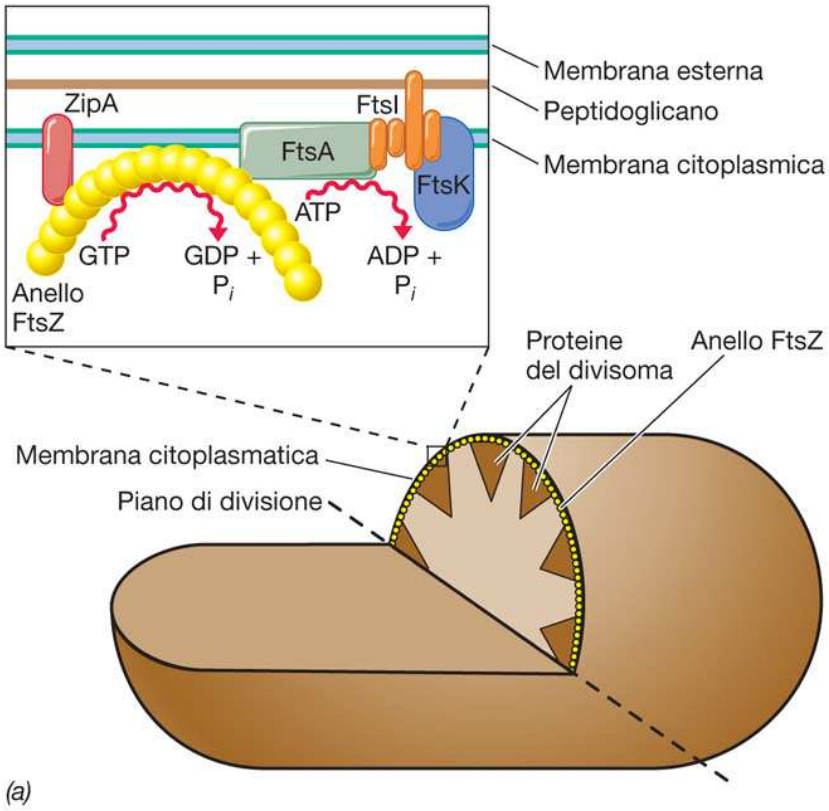
Bacterial growth by binary fission




DNA
DNA replication
Cell elongation
Septum formation
Completion of septum with formation of distinct walls
Cell separation
One generation


Cell wall
Plasma membrane
1
Cell elongates and DNA is replicated
DNA (nuclear area)
2
Cell wall and plasma membrane begin to divide
3
Cross-wall forms completely around divided DNA
4
Cells separate
DNA (nuclear area)
Partially formed cross-wall
Cell wall

The FtsZ protein and cell division

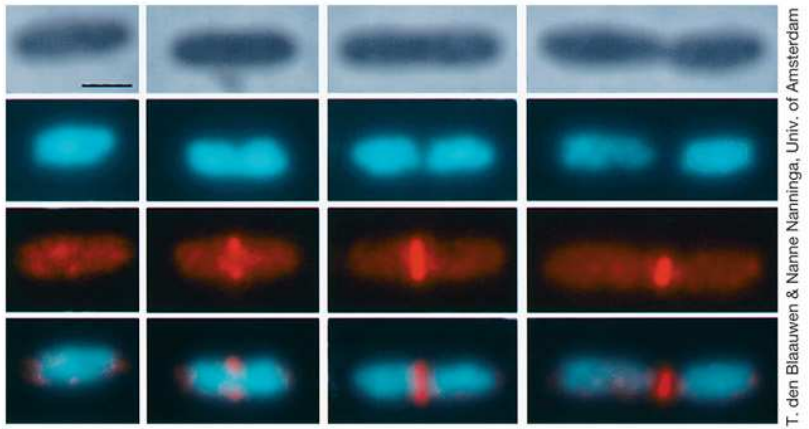


Phase contrast

Nucleoid stain

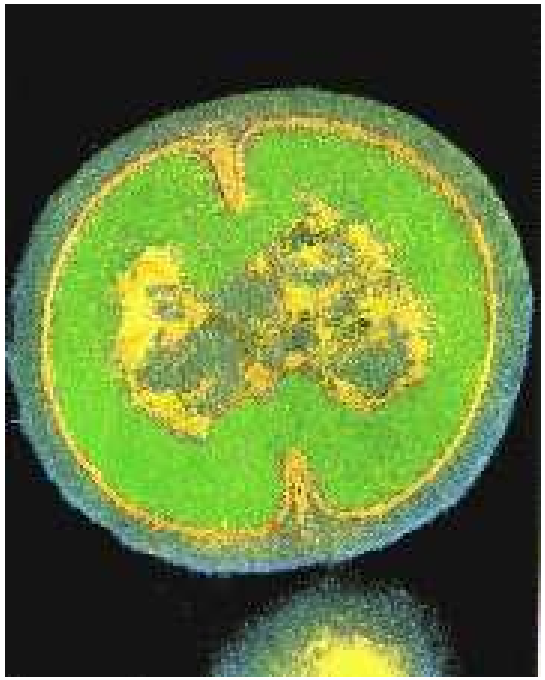
FtsZ stain

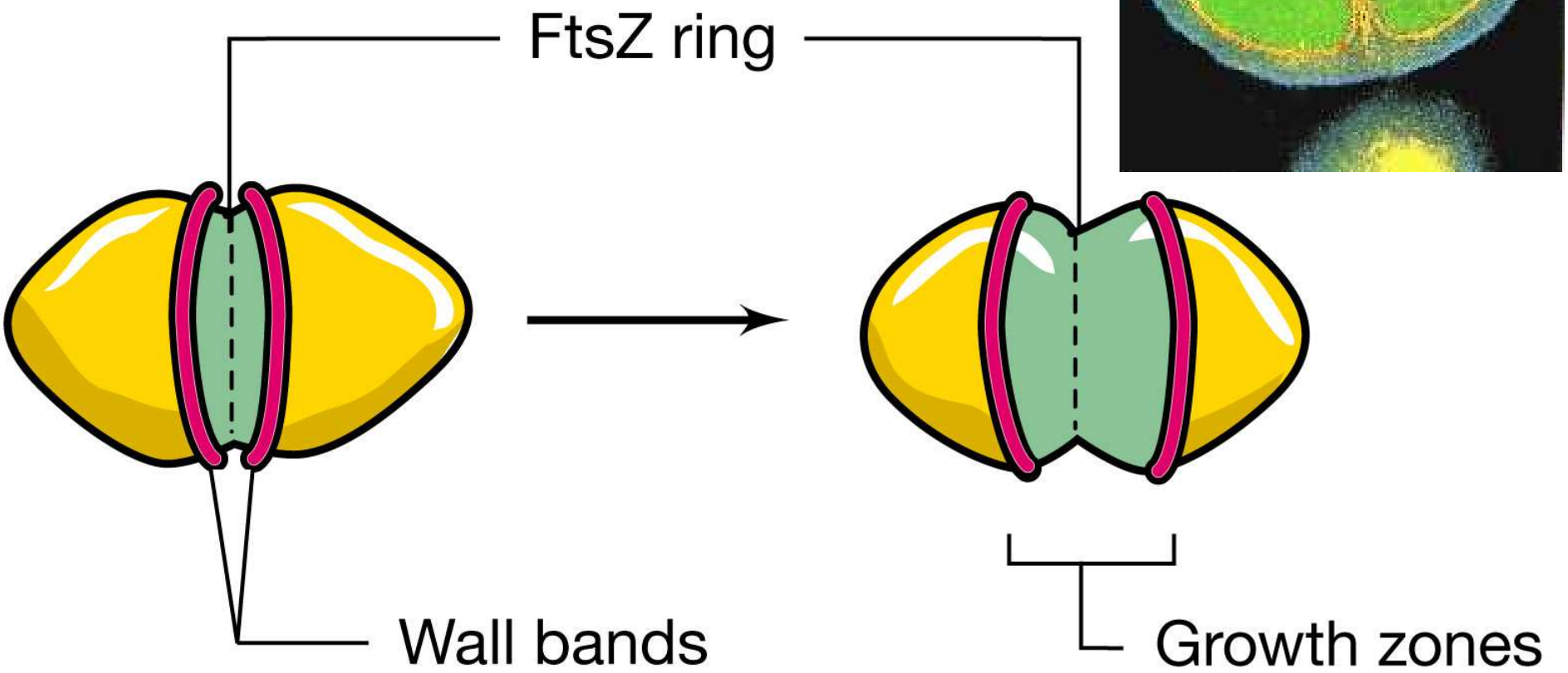
Merged



Cell wall synthesis during cell division in gram-positive Bacteria.

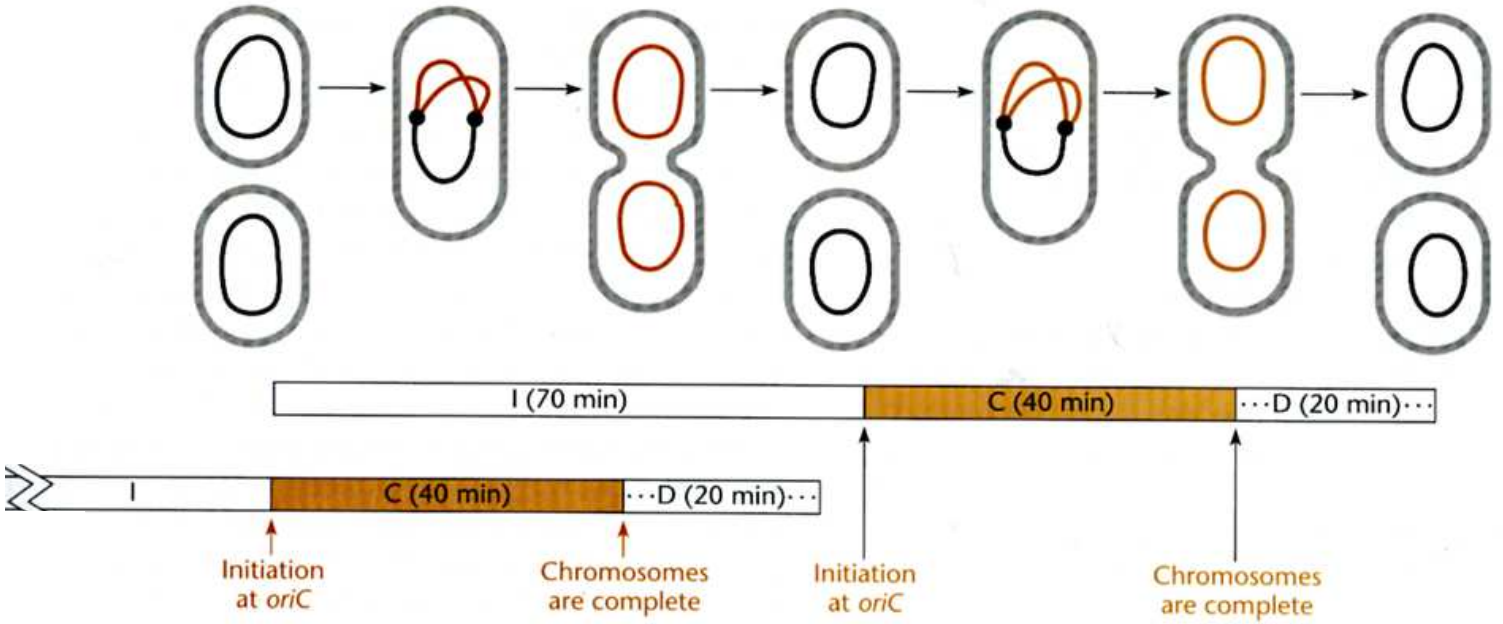
•In cocci, new cell wall synthesis (shown in green) is localized at only one point





Generation times for some bacteria under optimal conditions

Bacterium	Medium	Generation time (min)
Escherichia coli	Glucose-salts	17
Bacillus megaterium	Sucrose-salts	25
Streptococcus lactis	Milk	26
Streptococcus lactis	Lactose broth	48
Staphyloc. aureus	Heart infusion broth	27-30
Lactobacillus acidophilus	Milk	66-87
Rhizobium japonicum	Mannitol-salts-yeast extract	344-461
Mycobacterium tuberculosis	Synthetic	792-932
Treponemapallidum	Rabbit testes	1980

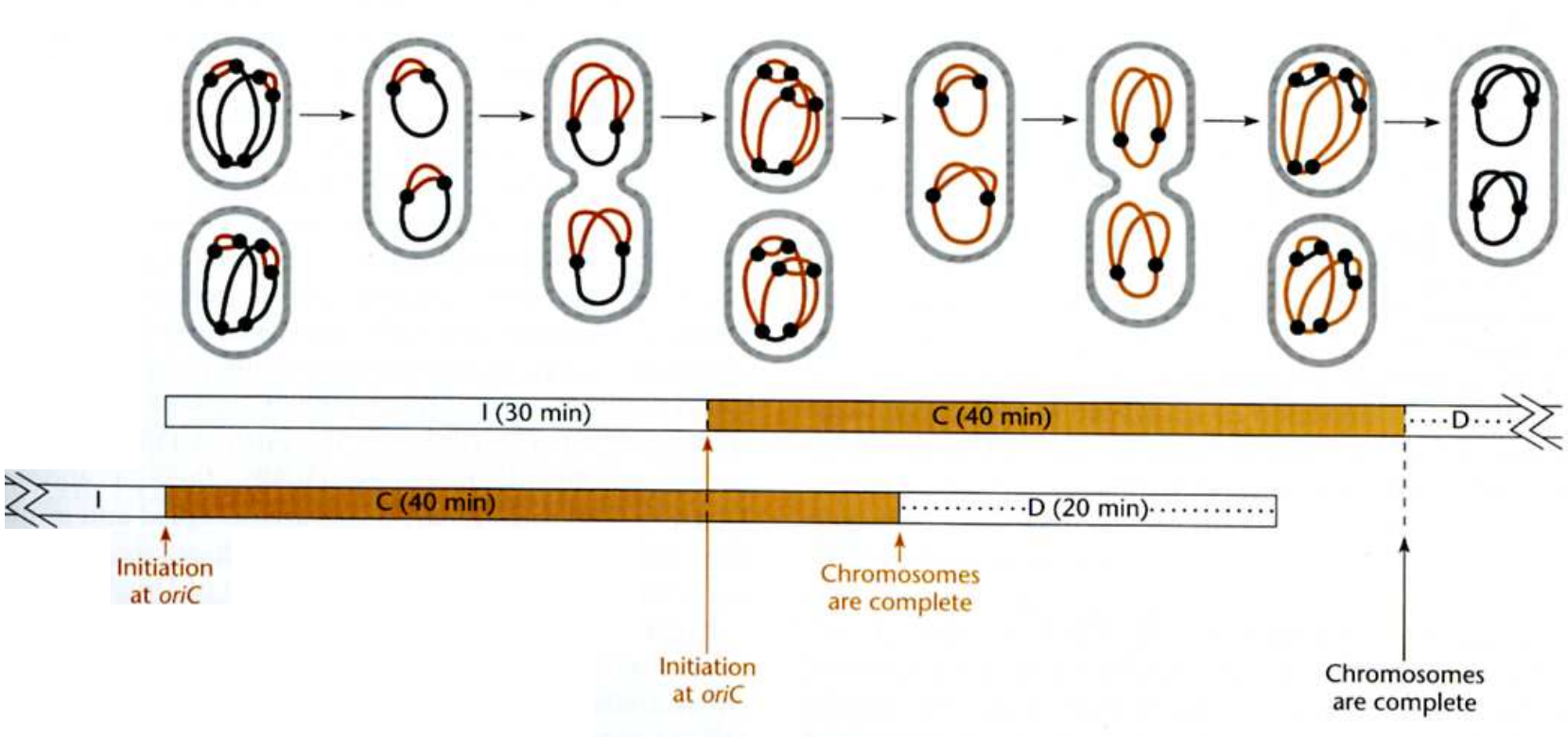


Slow growth I= C+D=70 min

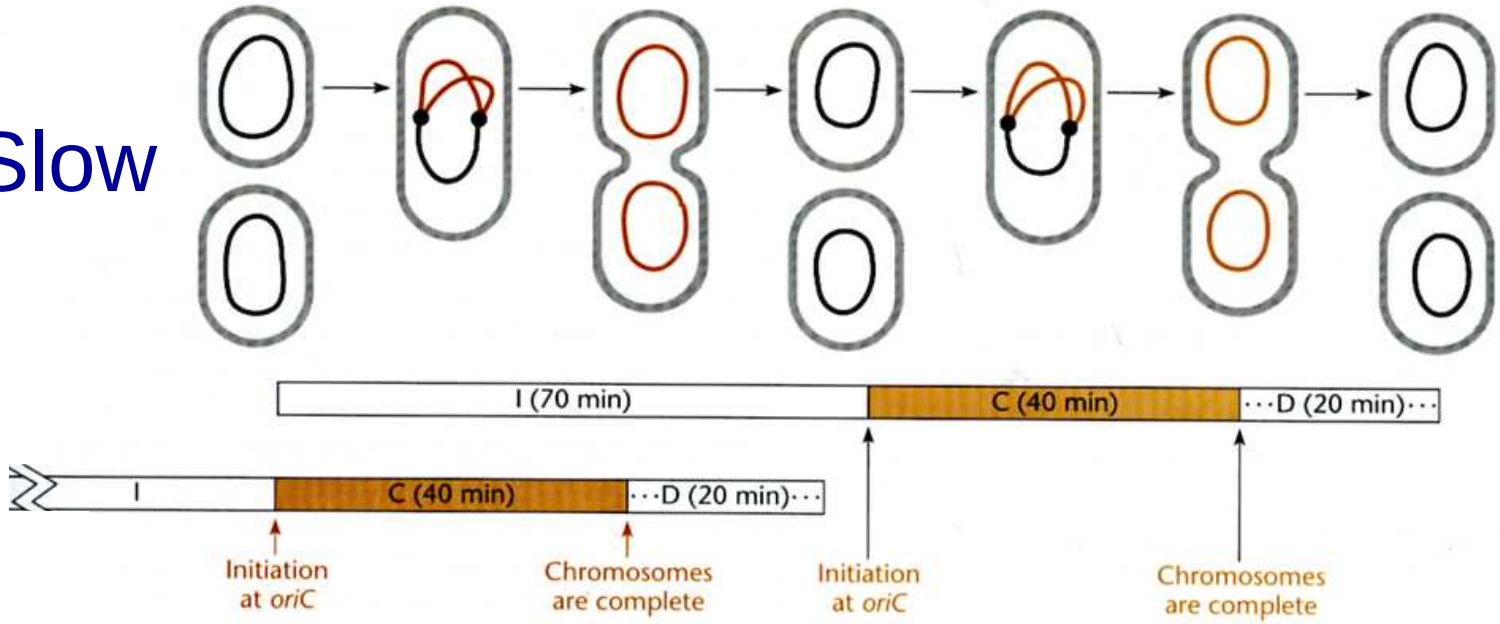
C= 40 min (DNA replication)
D= 20 min (septum formation and cell division)

Rapid growth I= C+D=30 min

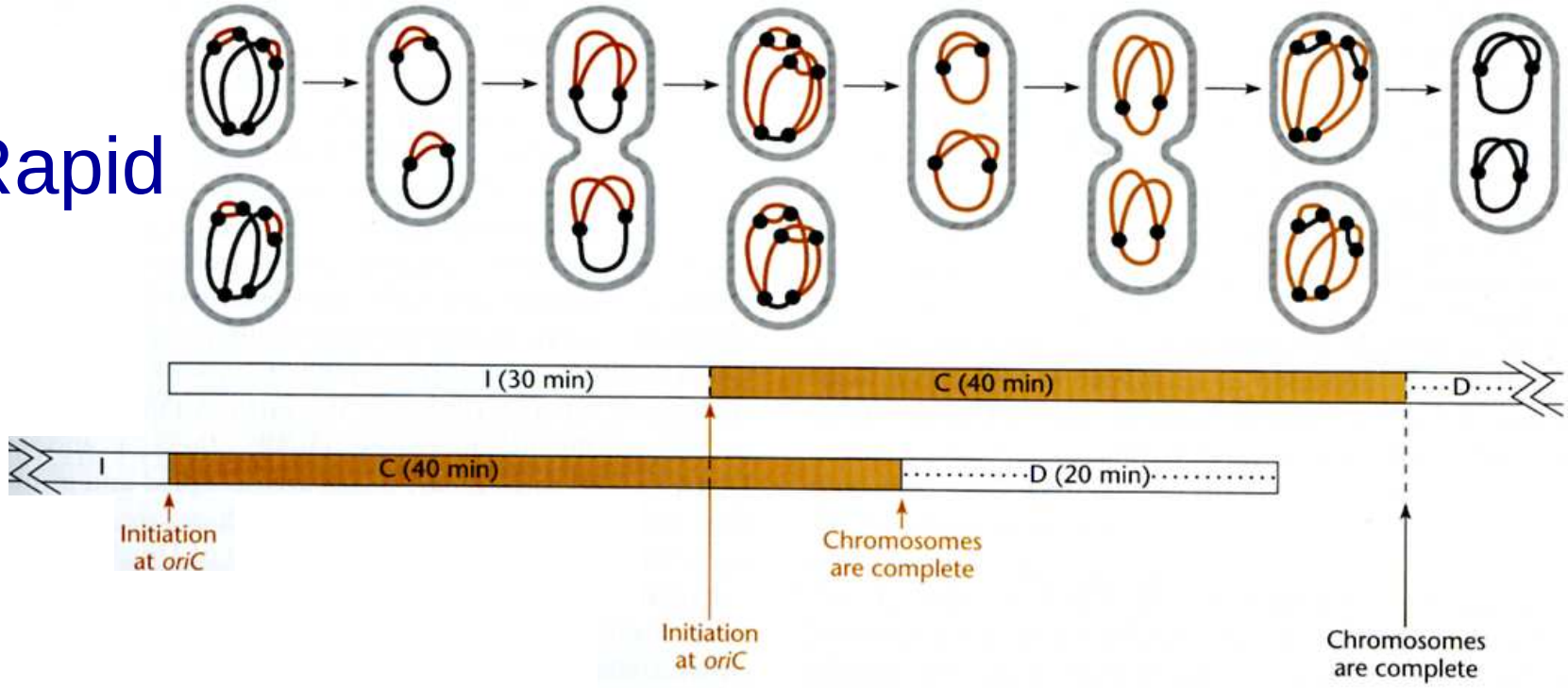
C= 40 min (DNA replication)
D= 20 min (septum formation
and cell division)



Slow


I (70 min)
C (40 min)
D (20 min)
I
C (40 min)
D (20 min)
Initiation at oriC
Chromosomes are complete
Initiation at oriC
Chromosomes are complete

Rapid


I (30 min)
C (40 min)
D
I
C (40 min)
D (20 min)
Initiation at oriC
Chromosomes are complete
Initiation at oriC
Chromosomes are complete

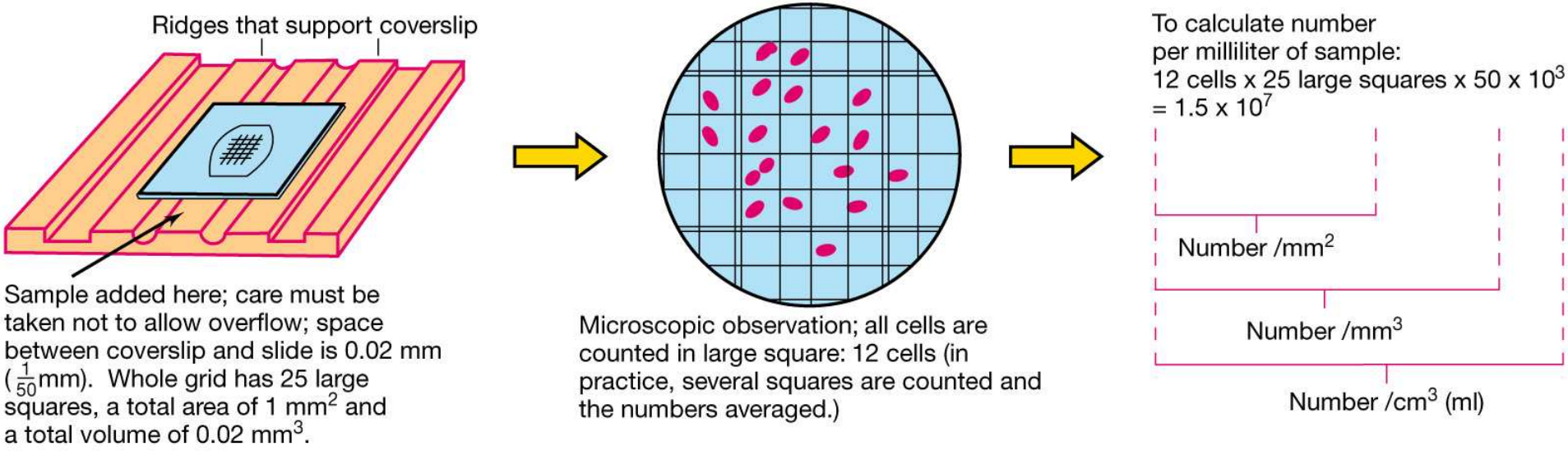
Microbial growth:

how to measure the growth

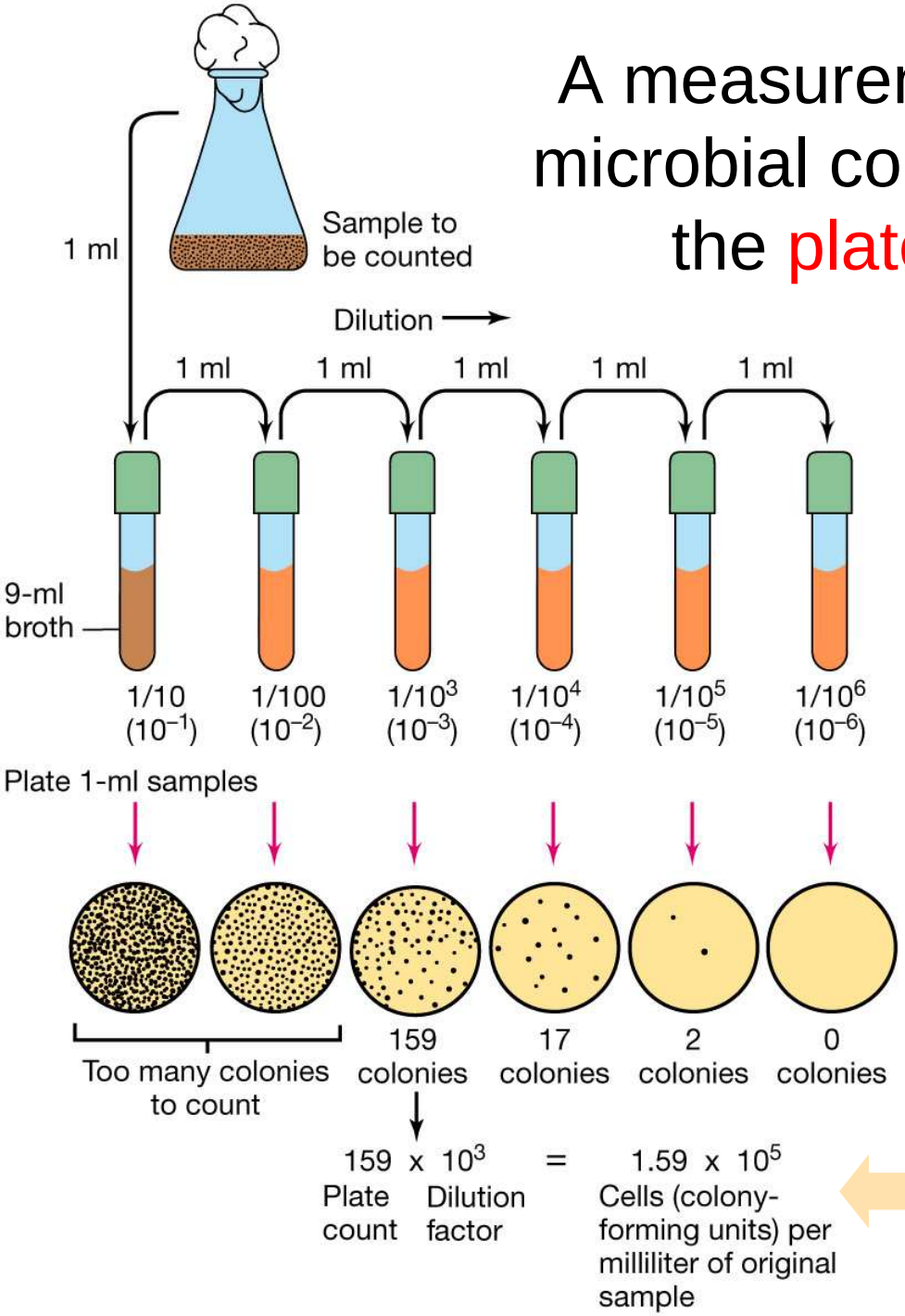
Some methods used to measure bacterial growth

Method	Application	Comments
Direct microscopic count	Enumeration of bacteria in milk or cellular vaccines	Cannot distinguish living from nonliving cells
Viable cell count (colony counts)	Enumeration of bacteria in milk, foods, soil, water, laboratory cultures, etc.	Very sensitive if plating conditions are optimal
Turbidity measurement	Estimations of large numbers of bacteria in clear liquid media and broths	Fast and nondestructive, but cannot detect cell densities less than 10^7 cells per ml
Measurement of total N or protein	Measurement of total cell yield from very dense cultures	Only practical application is in the research labs
Measurement of Biochemical activity (O2 uptake, CO_2 , ATP)	Microbiological assays	Requires a fixed standard to relate chemical activity to cell mass and/or cell numbers
Measurement of dry weight or wet weight of cells or volume of cells after centrifugation	Measurement of total cell yield in cultures	probably more sensitive than total N or total protein measurements

A measurement of the microbial concentration: the direct microscopic counting



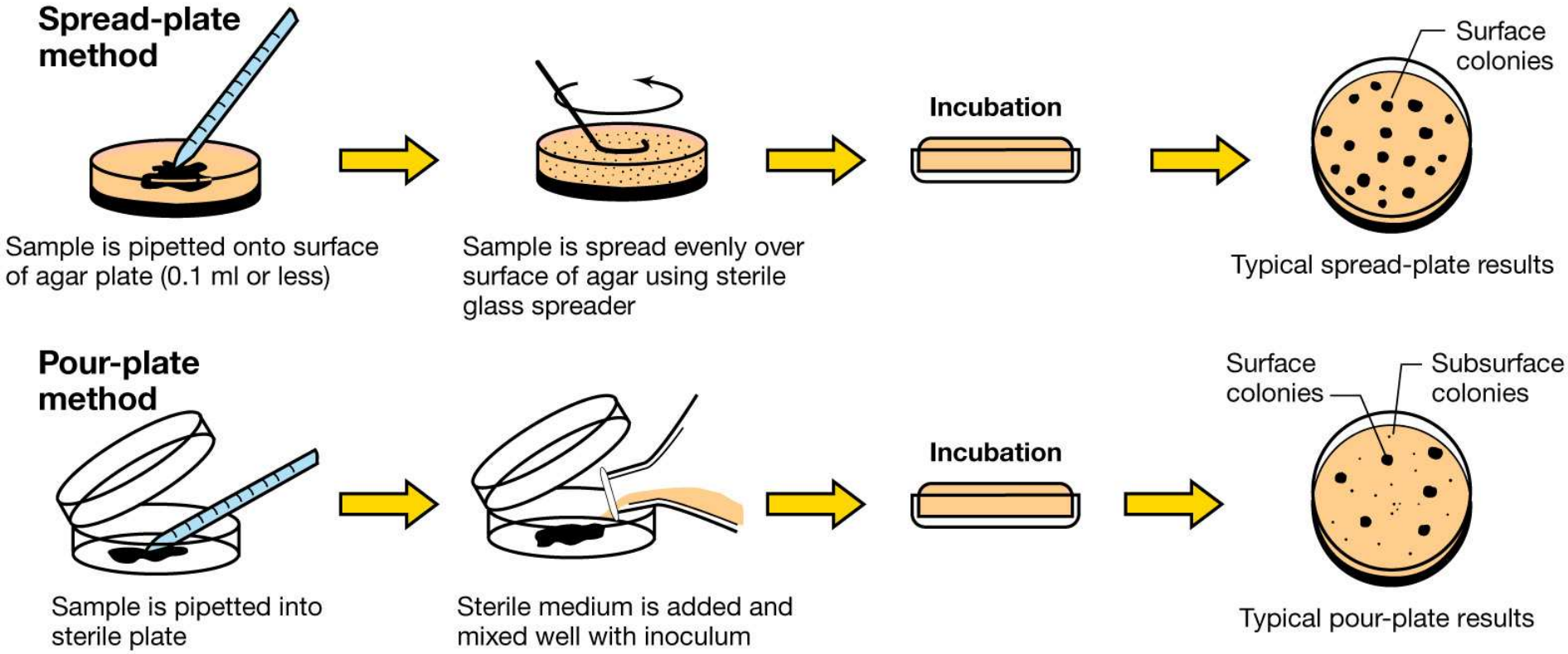
A measurement of the microbial concentration: the plate count



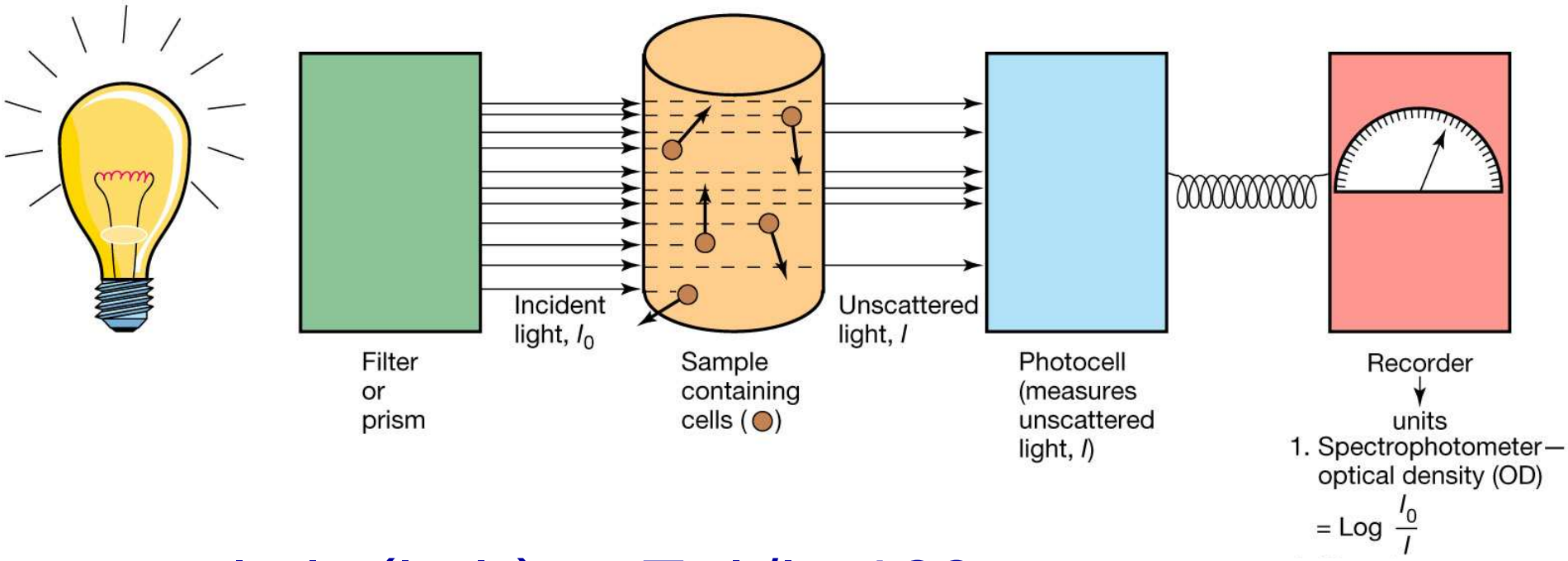
CFU/ml

Two methods of performing a viable count (plate count).

In either case the sample must usually be diluted before plating



A measurement of the microbial mass: the turbidity procedure

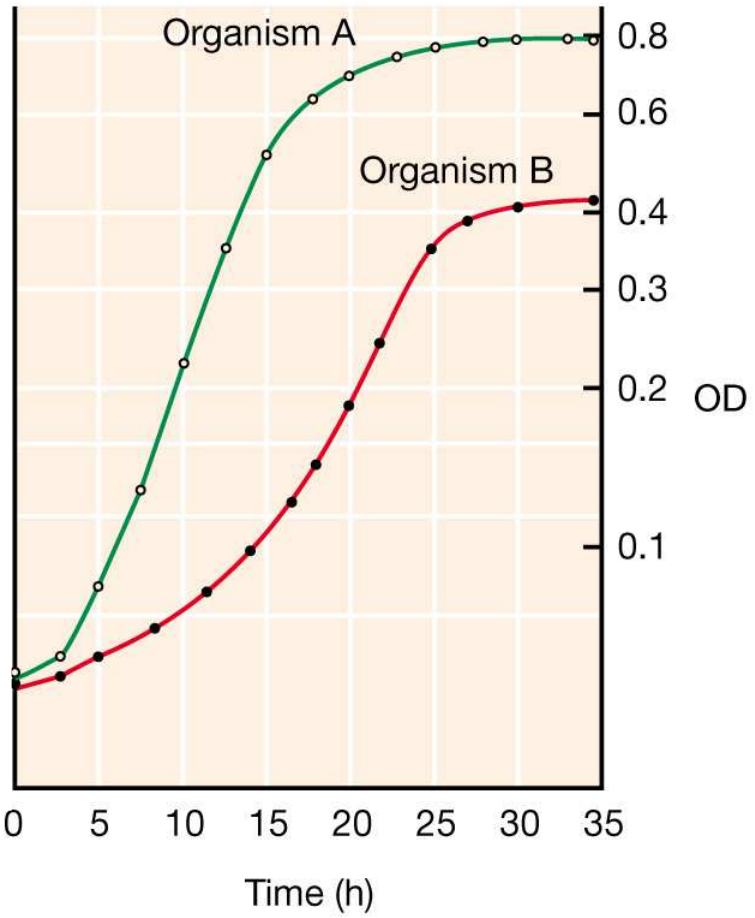


(a)

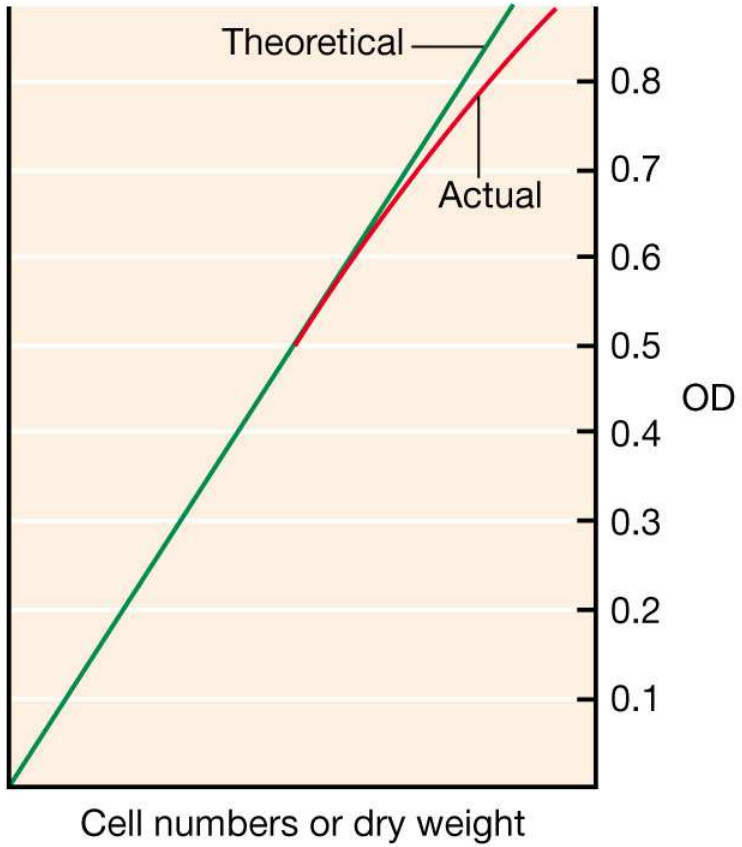
$I_t=I_0-(I_r+I_a)$ $T=I_t/I_0x100$

O.D.(Optical Density)=-logT=log I_0/I_t

Turbidity measurements of microbial growth.



Typical growth curves of two bacteria growing at different growth rates



Relationship between cell number or dry weight and turbidity readings

Microbial growth:

the growth curve of a bacterial population

Time (h)	Total number of cells	Time (h)	Total number of cells
0	1	4	256
0.5	2	4.5	512
1	4	5	1,024
1.5	8	5.5	2,048
2	16	6	4,096
2.5	32	.	.
3	64	.	.
3.5	128	10	1,048,576

(a)

I=30 min

The rate of growth of a bacterial culture

Time (h)	Total number of cells	Time (h)	Total number of cells
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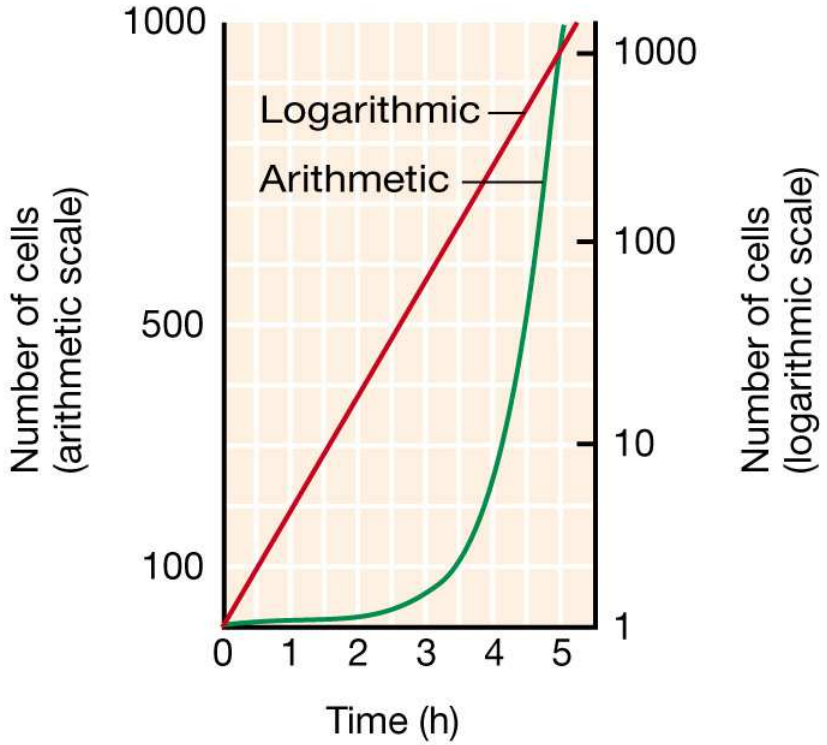
(a)

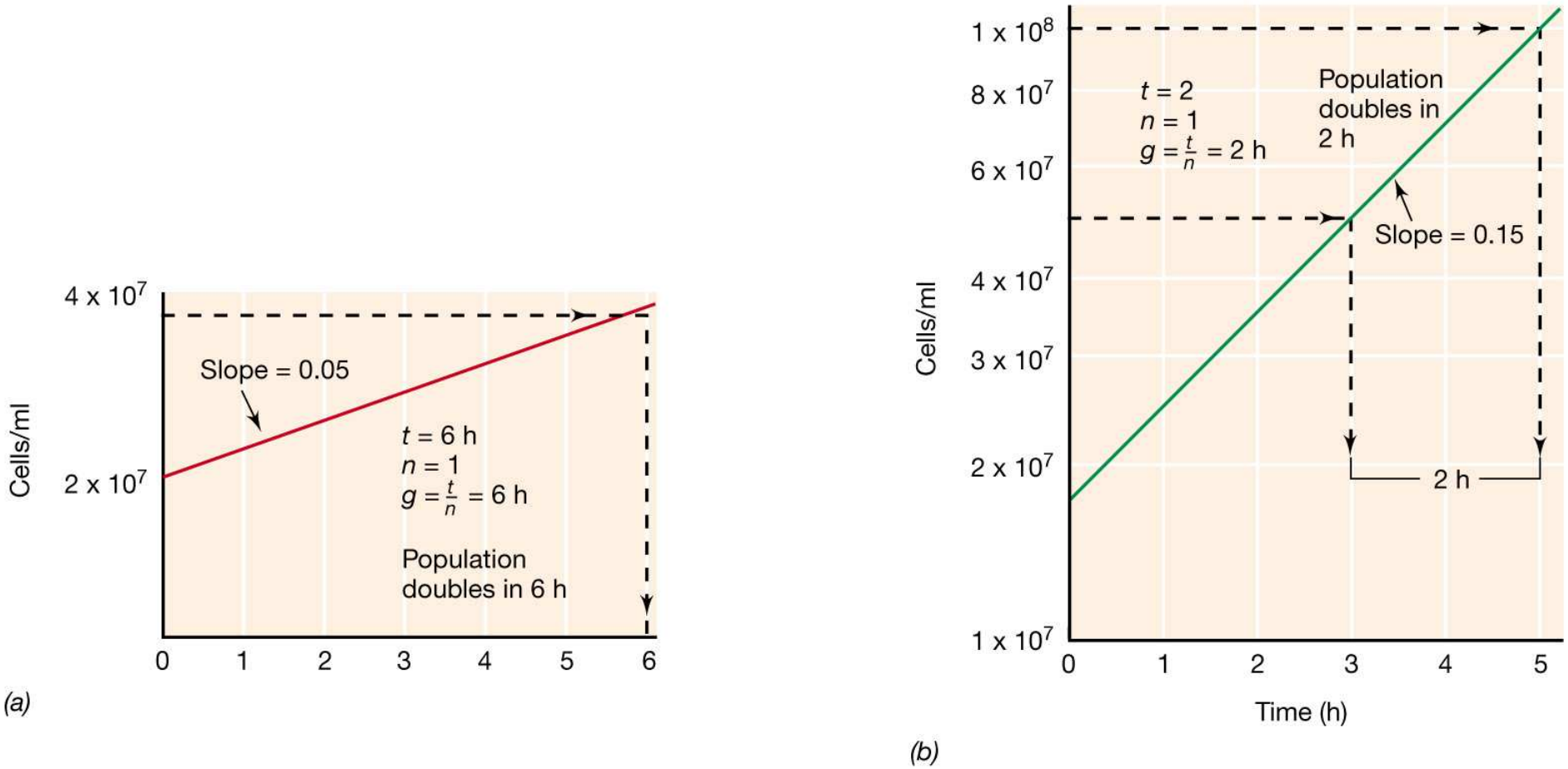
I=30 min

The rate of growth of a bacterial culture

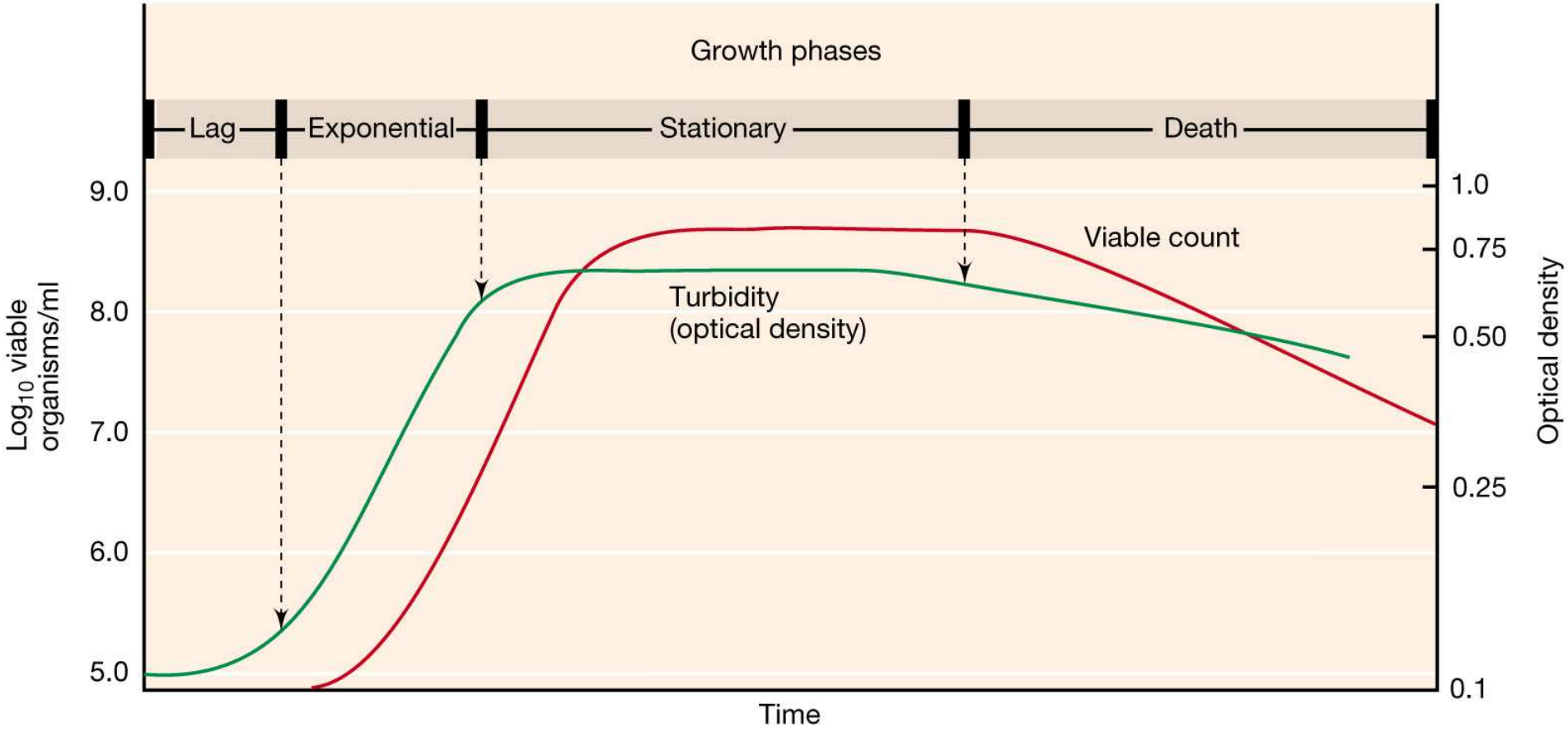


(b)

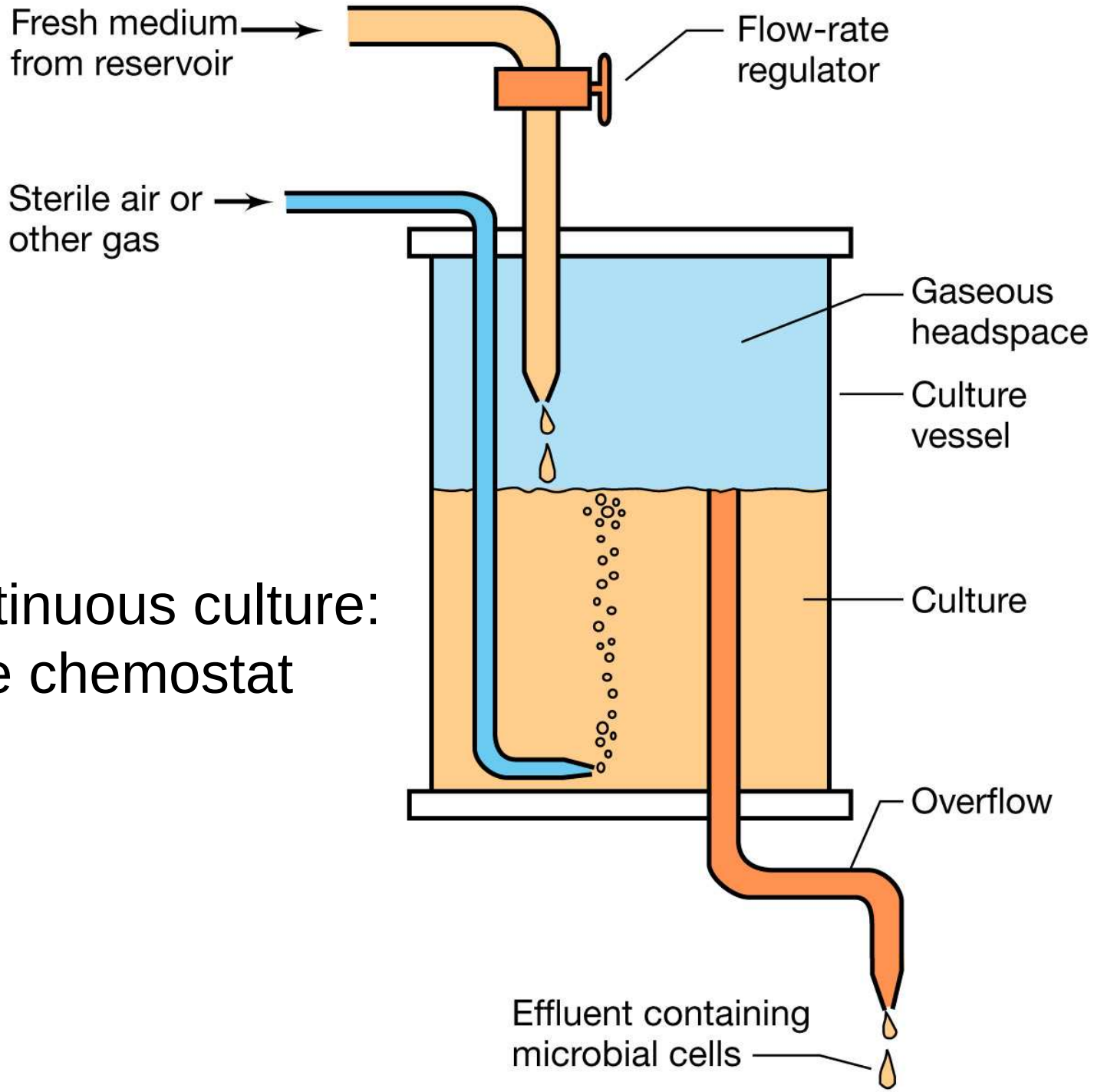
Method of estimating the generation times (g)



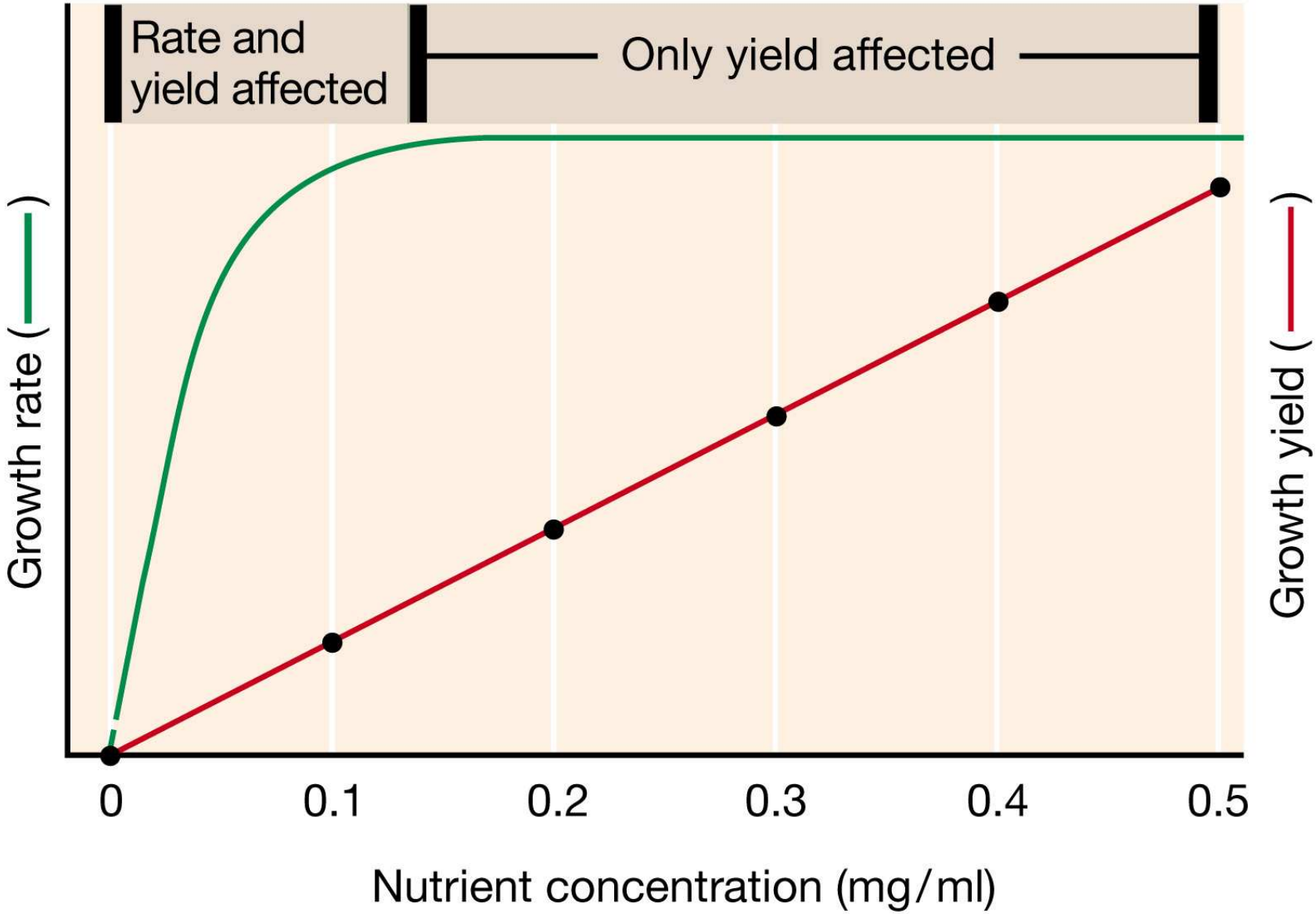
Typical **growth curve** for a bacterial population



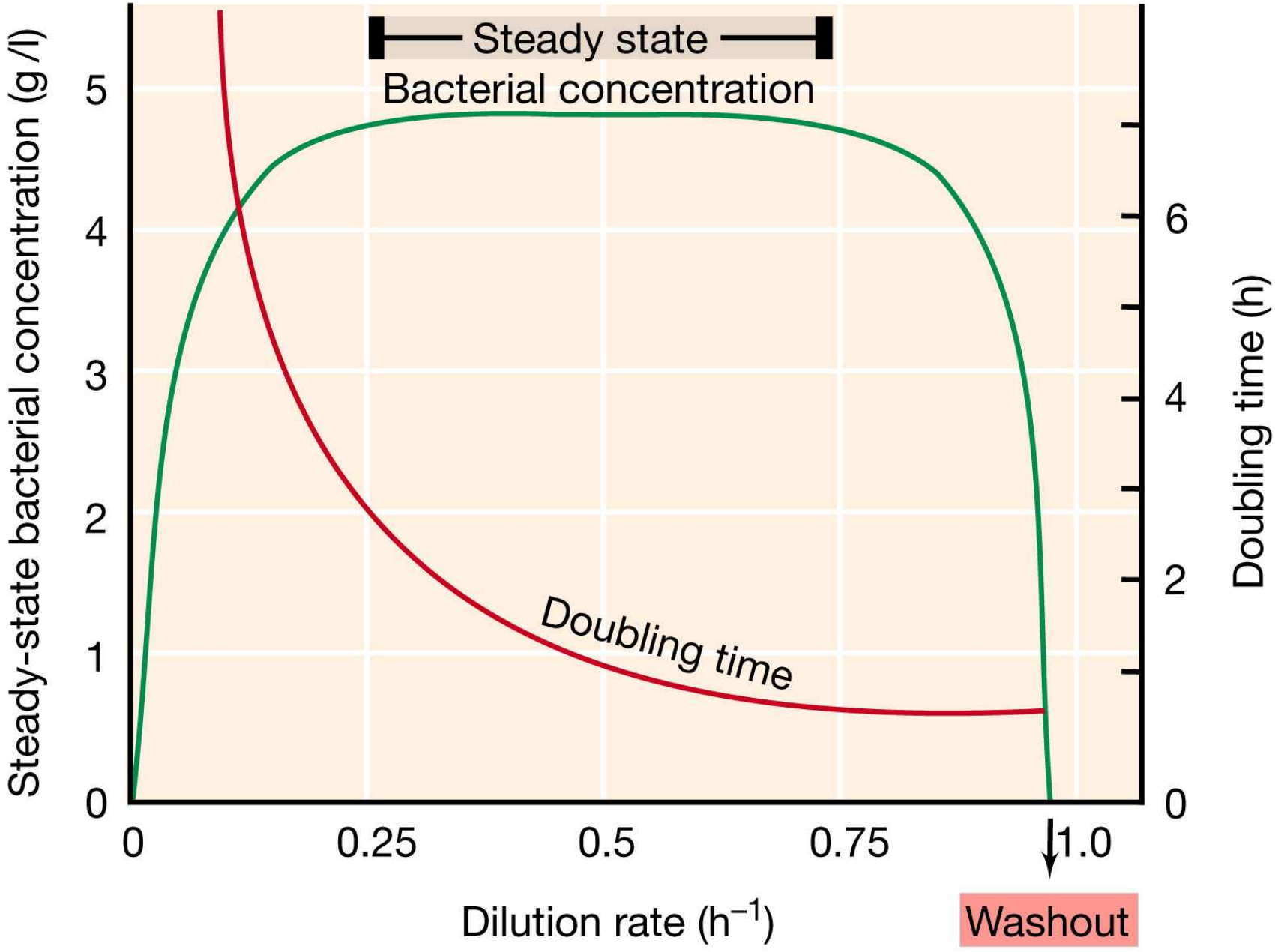
A continuous culture: the chemostat



Relationship between nutrient concentration, growth rate (green curve), and growth yield (red curve)



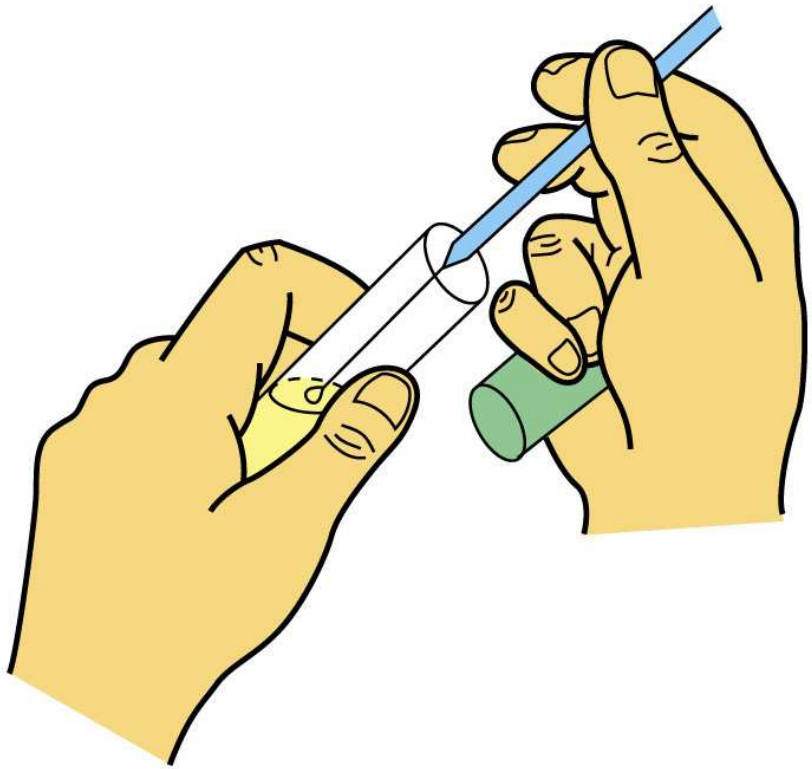
Steady-state relationships in the chemostat



Microbial growth:

the growth on solid media

Growth on solid media: **colonies** and **pure cultures**



(a)

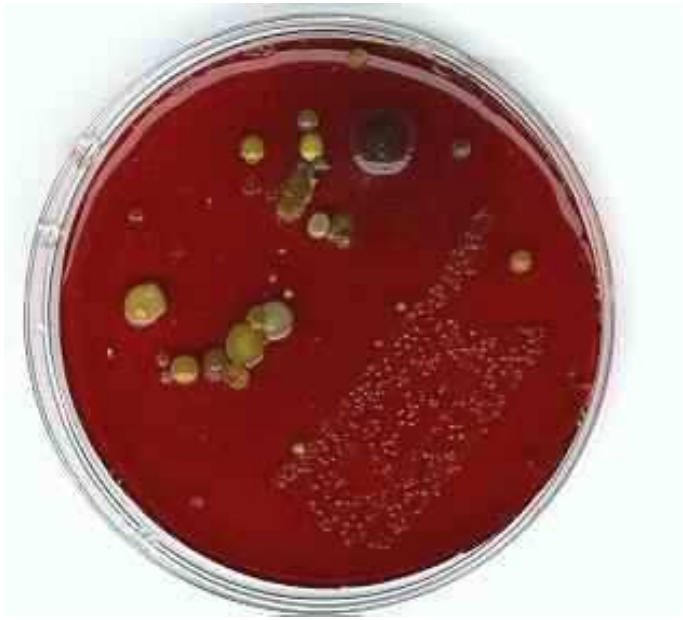
(b)

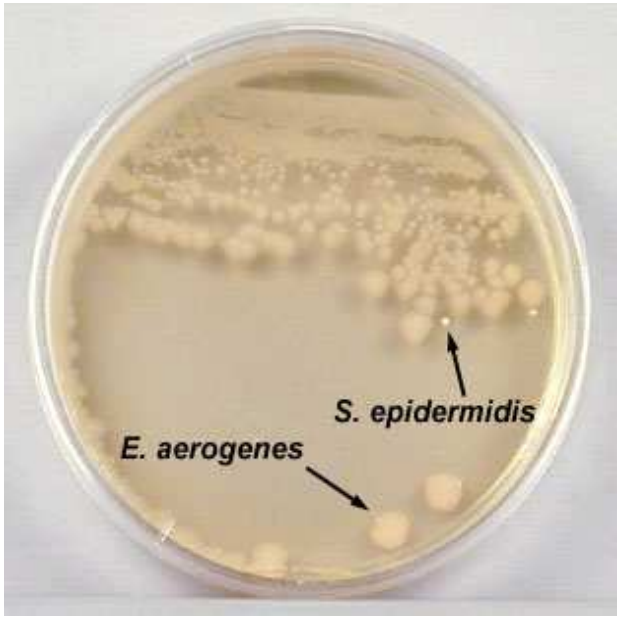
A bacterial colony contains 1-10 x10^6 cells

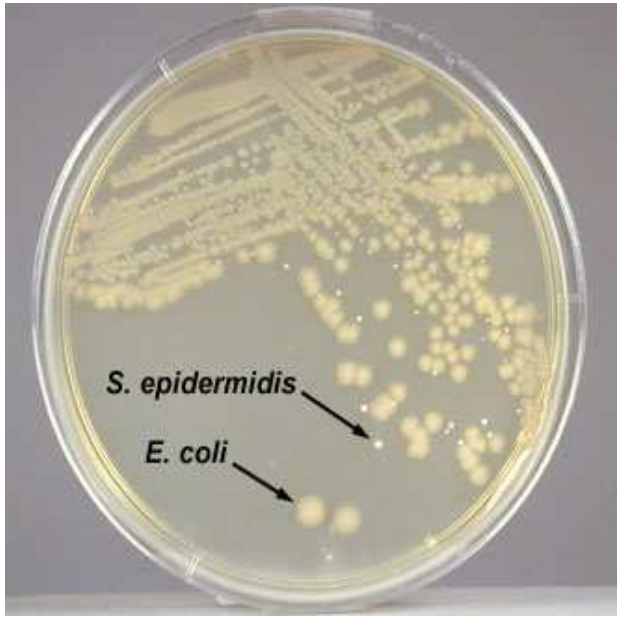
Procedure:

1. Flame the loop and wire and streak a loopful of broth as at **A** in the diagram.
2. Reflame the loop and cool it.
3. Streak as at **B** to spread the original inoculum over more of the agar.
4. Reflame the loop and cool it.
5. Streak as at **C.**
6. Reflame the loop and cool it.
7. Streak as at **D**.
8. Label the plate and incubate it inverted

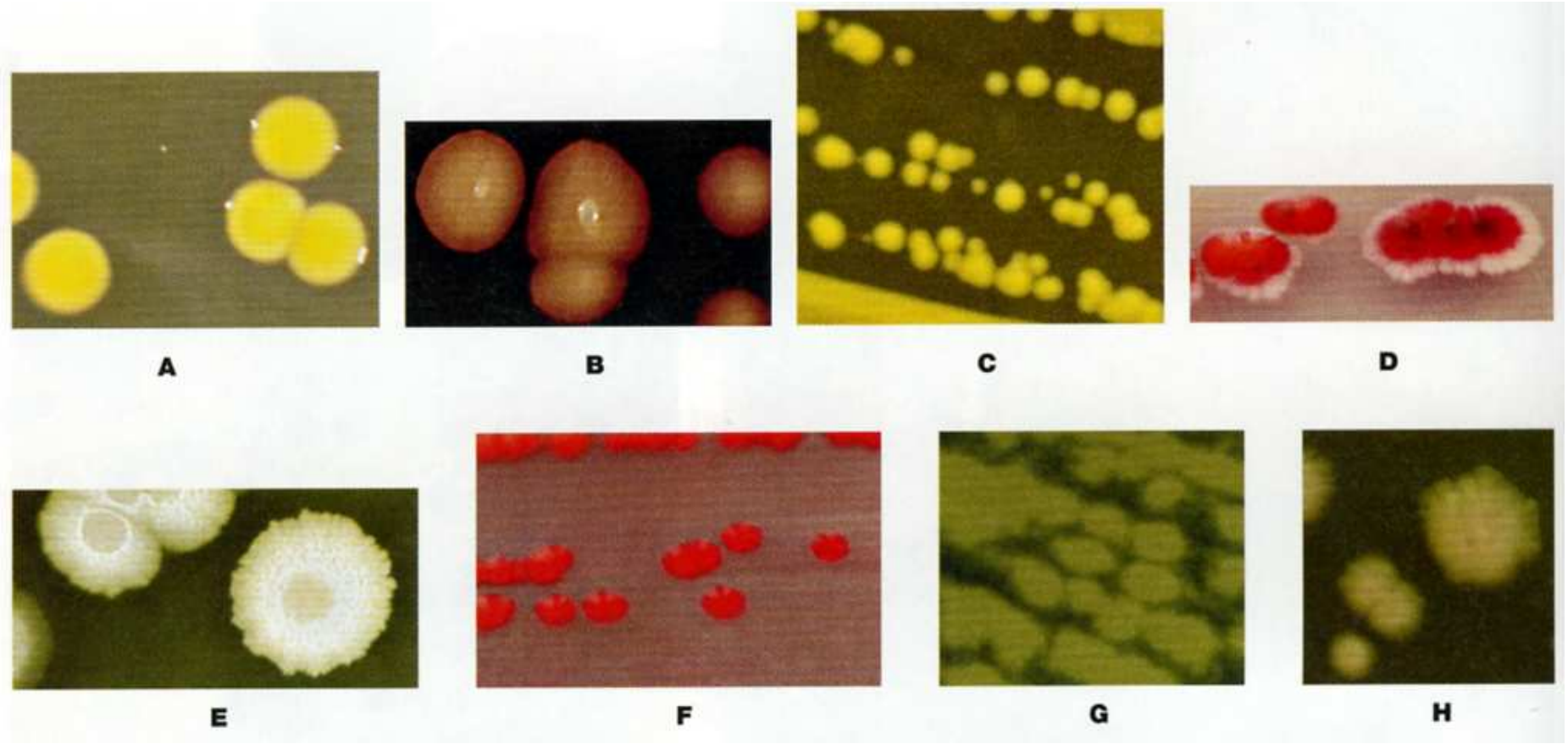
Streaking a Plate for Isolation




S. epidermidis
E. aerogenes


S. epidermidis
E. coli

Aspects of bacterial colony morphology



Aspects of bacterial colony morphology