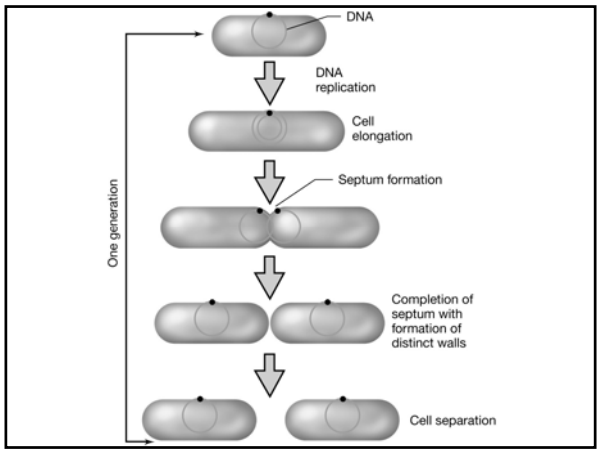
## The Process of Growth

- Metabolism required for growth, both anabolic and catabolic; ~2000 reactions!
- Usual Definition: **Increase in cell numbers**
  Other definitions possible – spores, UMC's, respiration, viable but nonculturable, morphology changes (life cycle)
- Divide via Binary Fission: 3 mechanisms involved!
  Cell Elongation – cell wall
  DNA Replication – rate limiting step
  Cell Division – septum formation



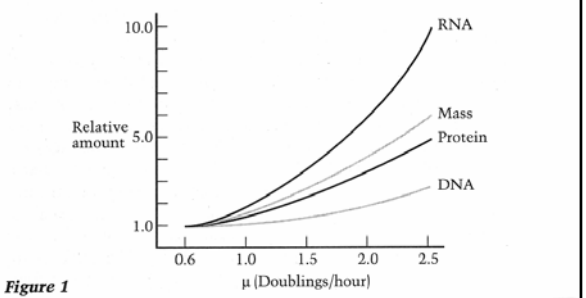




***Figure 1***

**Effect of nutrition-imposed growth rate on the composition of *Escherichia coli* B/r. All values are expressed in amounts per cell normalized to values at $\mu = 0.6$ (mass = $1.48 \times 10^{-13}$ g; protein = $1.00 \times 10^{-13}$ g; RNA = $2.0 \times 10^{-14}$ g; DNA = $6.3 \times 10^{-15}$ g). (Plotted from data in Bremer and Dennis, 1987.)**

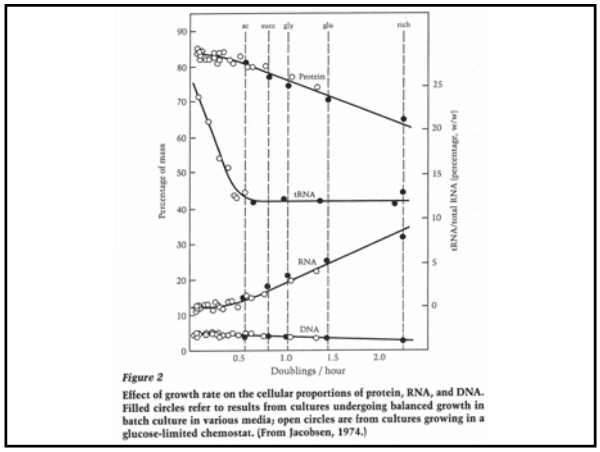
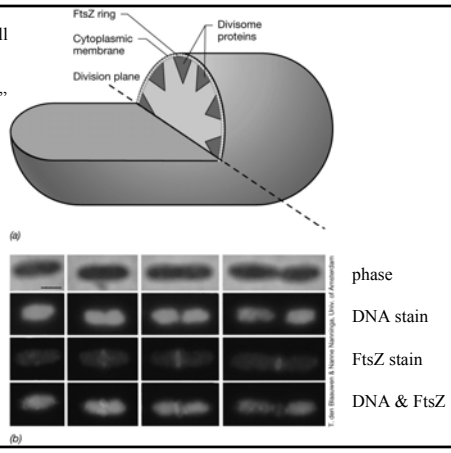



***Figure 2***

**Effect of growth rate on the cellular proportions of protein, RNA, and DNA. Filled circles refer to results from cultures undergoing balanced growth in batch culture in various media; open circles are from cultures growing in a glucose-limited chemostat. (From Jacobsen, 1974.)**

FtsZ ring & cell division

The "divisome"



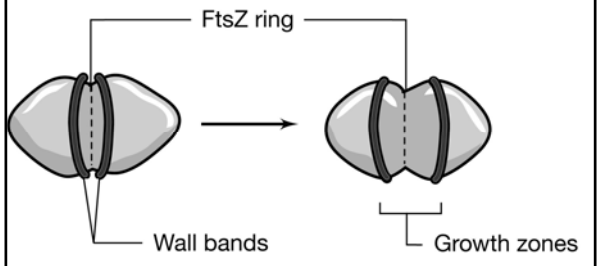

phase

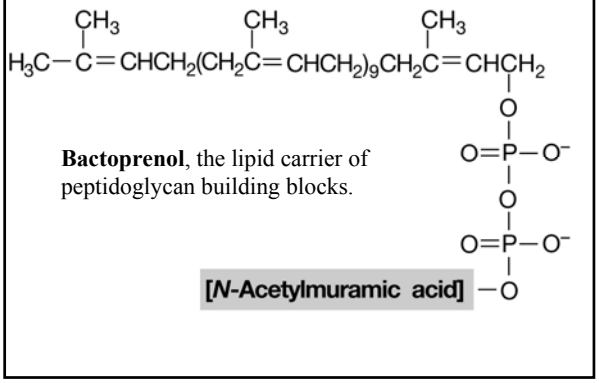
DNA stain

FtsZ stain

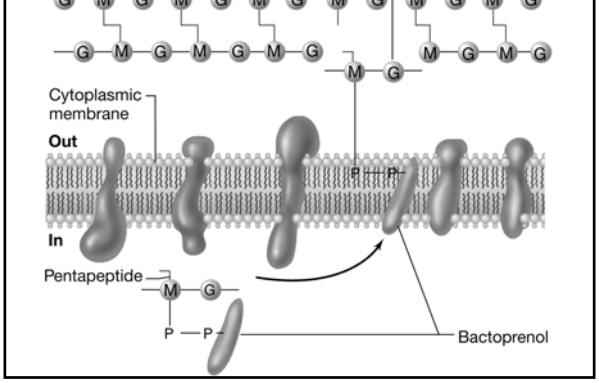
DNA & FtsZ

FtsZ similar to Tubulin
MreB similar to Actin

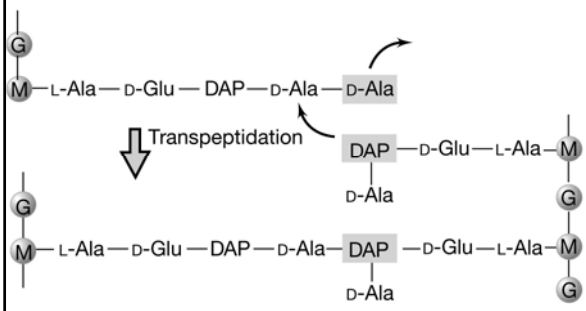





**Bactoprenol**, the lipid carrier of peptidoglycan building blocks.




**Penicillin** blocks this reaction



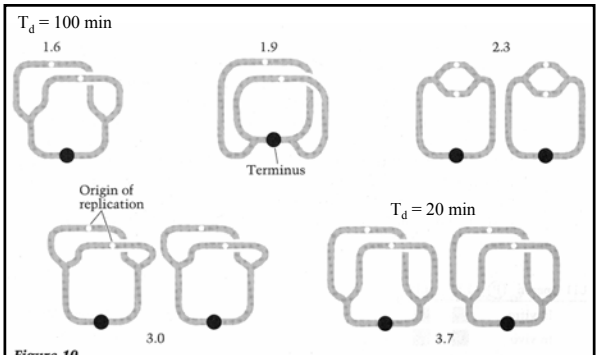




*Figure 10*

**Chromosome structure and equivalent DNA content of the average cell in culture of *E. coli* B/r growing at various rates. The numbers represent genome equivalents. (From Bremer and Dennis, 1987.)**

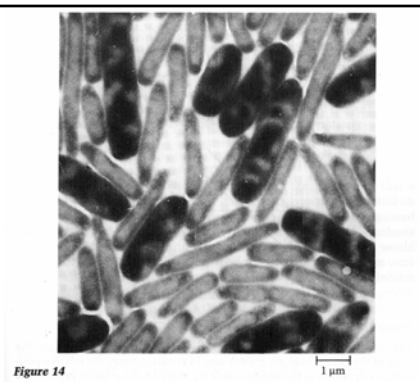



*Figure 14*

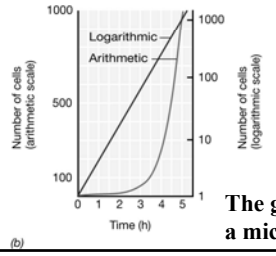
**Electron micrograph of a mixture of cells of *E. coli* B/r grown at different rates. The large cells grew with a doubling time of 22 minutes, the small ones with a doubling time of 72 minutes. (From Nanninga and Woldringh, 1985.)**

## The Process of Growth

- Growth Rate: Time it takes to reproduce
  $t_{½} = \ln 2/\mu = 0.693/\mu = g$
- Phases of Growth in Batch culture
  Lag, Log, Stationary, Death
- Measurement of Growth
  Total cell counts
  Viable cell counts
  Turbidity

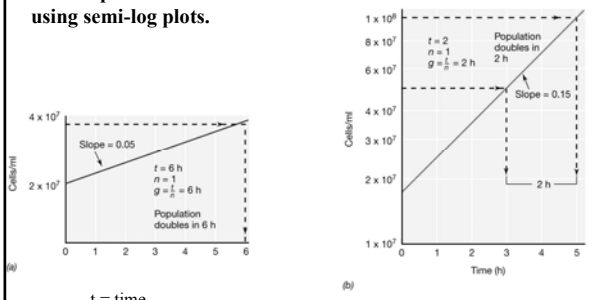
| Time (h) | Total number of cells | Time (h) | Total number of cells |
|---|---|---|---|
| 0 | 1 | 4 | 256 |
| 0.5 | 2 | 4.5 | 512 |
| 1 | 4 | 5 | 1,024 |
| 1.5 | 8 | 5.5 | 2,048 |
| 2 | 16 | 6 | 4,096 |
| 2.5 | 32 | . | . |
| 3 | 64 | . | . |
| 3.5 | 128 | 10 | 1,048,576 |

(a)




**The growth rate of a microbial culture**

**How to estimate the generation times of an exponential microbial culture using semi-log plots.**

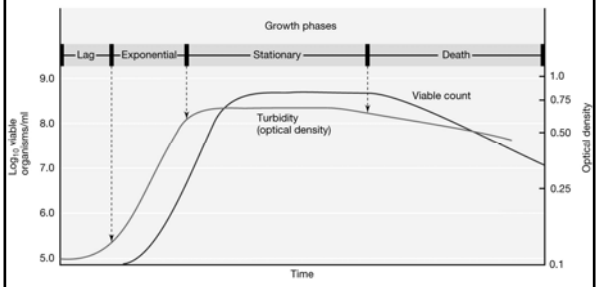



t = time
n = generations
g = generation time

**Table 6.1** Approximate generation times for several organisms growing in media optimal for growth

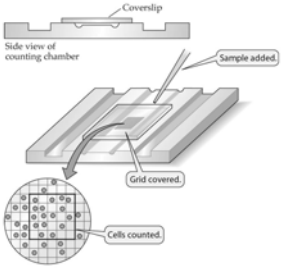
| Species | Generation Time |
|---|---|
| *Escherichia coli* | 20 min |
| *Bacillus subtilis* | 28 min |
| *Staphylococcus aureus* | 30 min |
| *Pseudomonas aeruginosa* | 35 min |
| *Thermus aquaticus* | 50 min |
| *Thermoproteus tenax* | 1 hr 40 min |
| *Rhodobacter sphaeroides* | 2 hr 20 min |
| *Sulfolobus acidocaldarius* | 4 hr |
| *Thermoleophilum album* | 6 hr |
| *Thermofilum pendens* | 10 hr |
| *Mycobacterium tuberculosis* | 13 hr 20 min |

Cryptic Growth

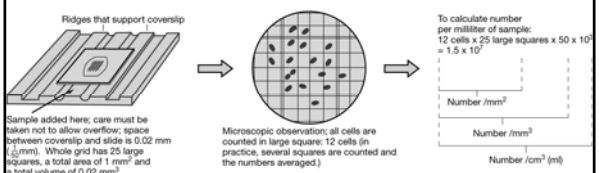



Typical growth curve for a bacterial population

Total Cell counts using the Petroff-Hausser Counter

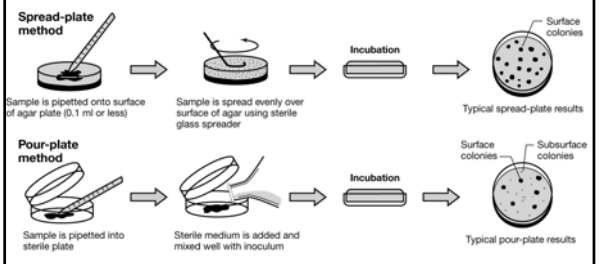



Total Cell counts using the Petroff-Hausser Counter

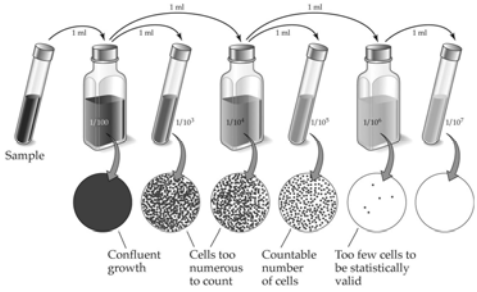


## Viable cell count methods

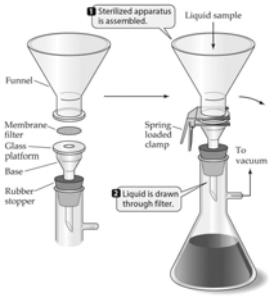
30-300 on standard Petri Dish



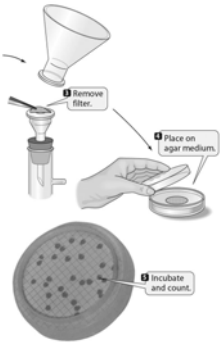

## Counting the number of viable cells by serial dilution and plate count



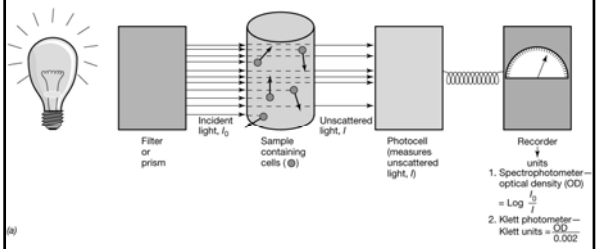

## (Part 1) Concentration of cells by membrane filtration



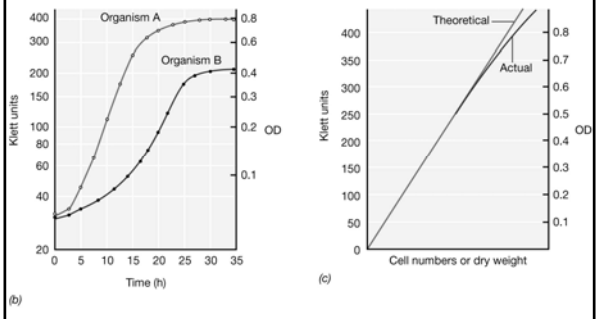

## (Part 2) Concentration of cells by membrane filtration




## Turbidity measurements of microbial growth




## Turbidity measurements of microbial growth

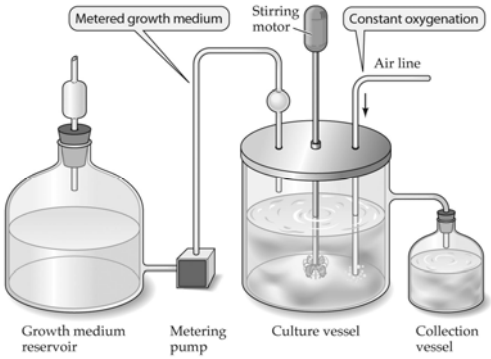


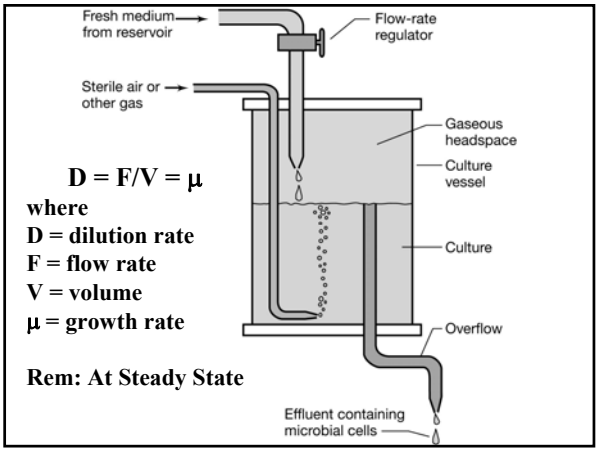
## The Process of Growth

- Continuous Culture: The wonders of the Chemostat
  - Steady State
  - Reproducible Physiology
  - Fine control

  Key parameters – Ks, μmax, Yield

  Closed systems vs. Open systems vs. Nature!
  (Batch) (Chemostat)

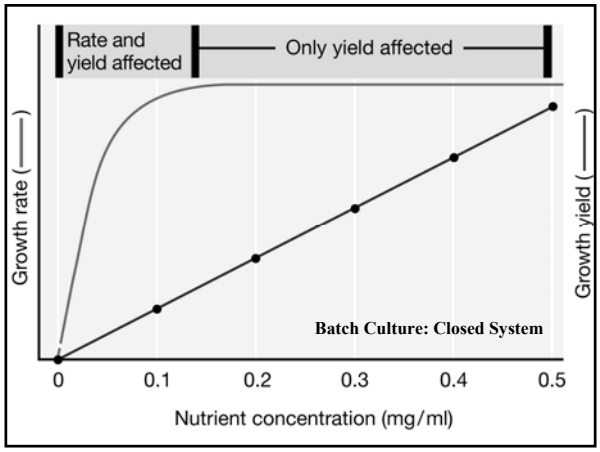


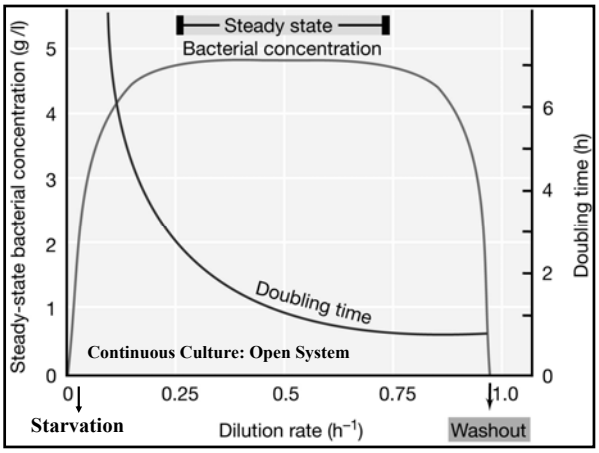




**D = F/V = μ**

**where**
**D = dilution rate**
**F = flow rate**
**V = volume**
**μ = growth rate**

**Rem: At Steady State**



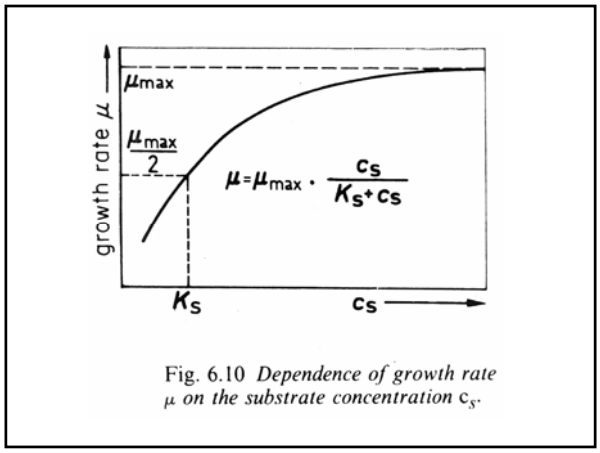




Fig. 6.10 *Dependence of growth rate μ on the substrate concentration* $c_s$.

Steady-state relationship between substrate concentration and output of bacterial mass

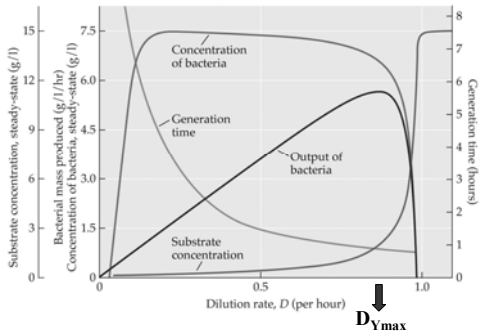



**Table 6.2** Growth yields of anaerobic bacteria utilizing glucose as the energy source

| | Mol ATP/Mol Glucose | $y_{max}$ (g of cell/mol Glucose) | $y_{ATP}$ (g of cell/mol ATP) |
|---|---|---|---|
| *Lactobacillus delbrueckii*[a] | 2 | 21 | 10.5 |
| *Enterococcus faecalis*[a] | 2 | 20 | 10 |
| *Zymomonas mobilis*[b] | 1 | 9 | 9 |

[a]Homolactic fermentation, Embden–Meyerhof pathway (see Chapter 10).
[b]Alcoholic fermentation, Entner–Doudoroff pathway (see Chapter 10).