# BIOL 4640 Mol Micro
# Lecture 18 10/7/26
# Cell Structures Part 4

- Assignment 11 – due on 10/7/26
- Assignment 12 due on 10/9/26
- Assignment 13 due on 10/9/26 – in class activity
- Assignment 14: In class Activity, Due on 10/12

# Unit 3. Features and Structures of Microbial Cells - C

A. Size and Shape of bacteria
B. Specialized External Structures of bacteria
C. Cell envelope of bacteria
D. Internal structures of bacteria
E. Cell Division in Bacteria
F. Viruses – Chapter 6
G. Atypical bacteria and viruses

Mostly Chapter 3 in book

# C. Cell Envelopes (rev)

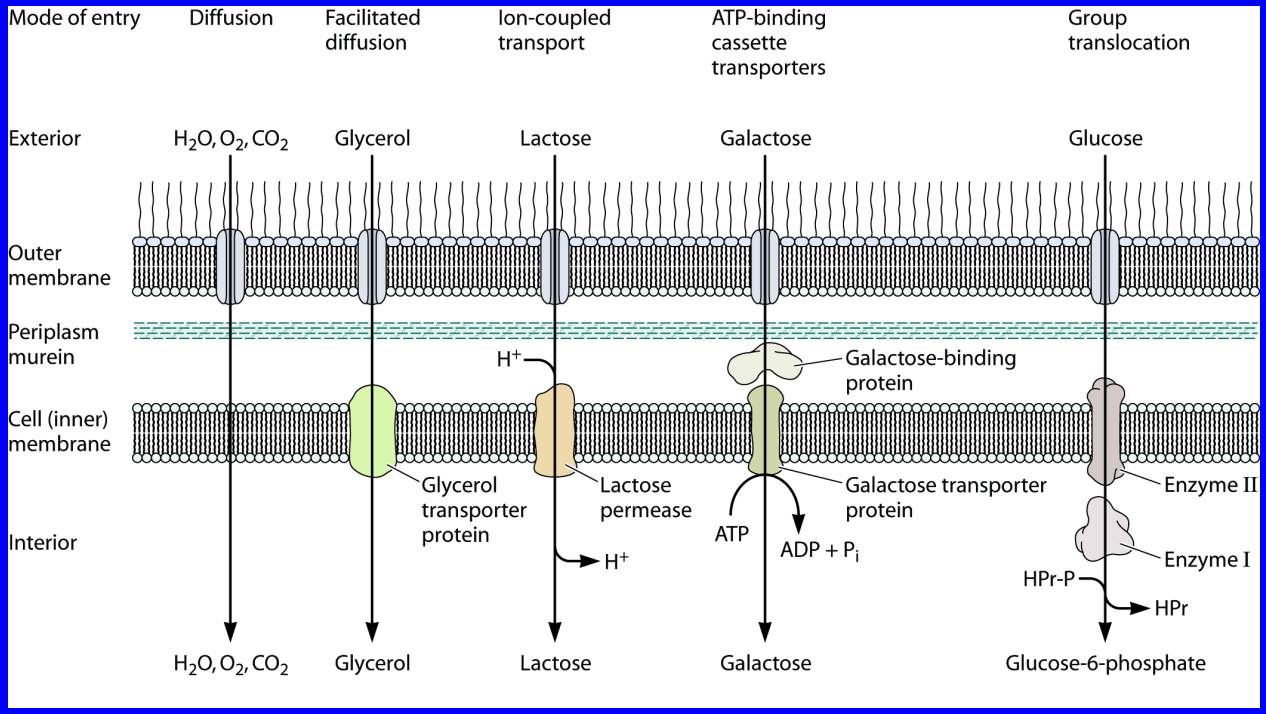
1. Major Styles/Configurations Observed
   a. Gram-positive (section 3.3)
   b. Mycobacterial (Gm+ actinomycete) - Acid-fast (section 3.3)
   c. Gram-negative (section 3.3)
   d. Mollicutes – *Mycoplasma* – no cell wall (pp. 734-735 6e)
   e. Archaeal (section 19.1)
2. Layers and Components
3. Peptidoglycan Synthesis and inhibition
4. Antibacterial Chemicals and resistance

# C. Cell Envelope of Bacteria

2. Layers and Components
   a. Glycocalyx – capsule
   b. S layer -
   c. Outer membrane
      - LPS and phospholipids
   d. Cell wall -
      - Peptidoglycan, Pseudopeptidoglycan, Teichoic acid, Mycolic acids, Arabinogalactan, Periplasm
   e. Periplasm
   f. Cell membrane
      - Phospholipids, Isoprenoids and hopanoids

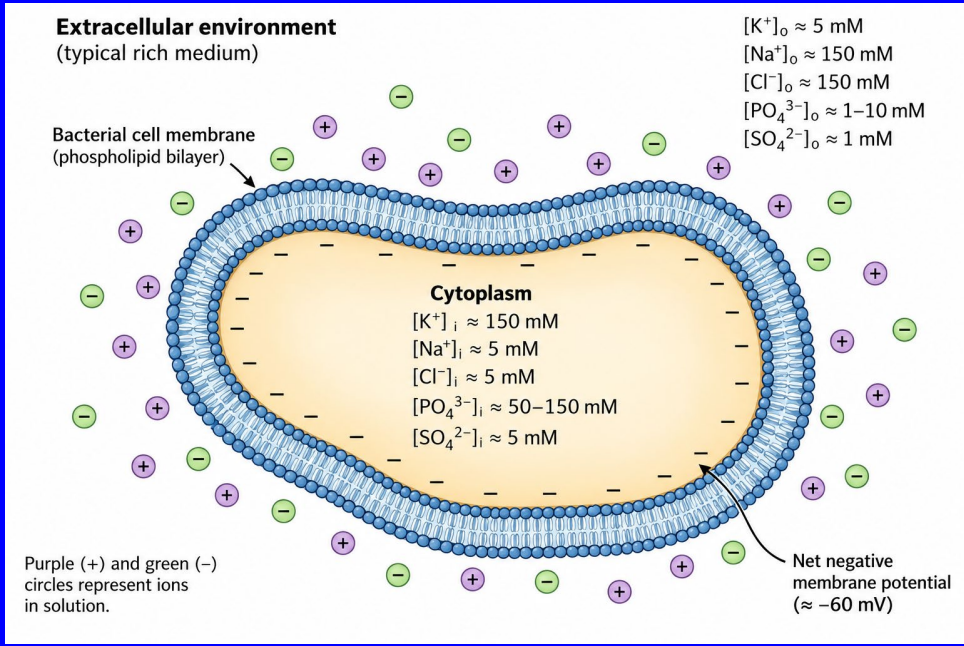
# f. Cell membrane

1) Components
2) Function
3) Transport across cell membrane
   - see sections 3.2 and 4.2



# 3) Transport across Cell Membranes

- Cells try to maintain concentration of certain ions and solutes inside the cell that differ from the outside environment




| ion conc. (mM) | sea water | *E. coli* | *S. cerevisiae* | mammalian cell (heart or RBC) | blood plasma |
|---|---|---|---|---|---|
| $K^+$ | ≈10 | 30-300 | 300 | 100 | 4 |
| $Na^+$ | ≈500 | 10 | 30 | 10 | 100-200 |
| $Mg^{2+}$ | ≈50 | 30-100 (bound); 0.01-1 (free) | 50 | 10 (bound) 0.5 (free) | 1 |
| $Ca^{2+}$ | ≈10 | 3 (bound);100 nM (free) | 2 (bound) | 10-100 nM (free) | 2 |
| $Cl^-$ | ≈500 | 10–200 media dependent | | 5-100 | 100 |

# 3) Transport across Membrane

Several mechanisms used (see sections 3.2 and 4.2)

a) Some passive (conc. gradient dependent)

b) Some active (requires energy)

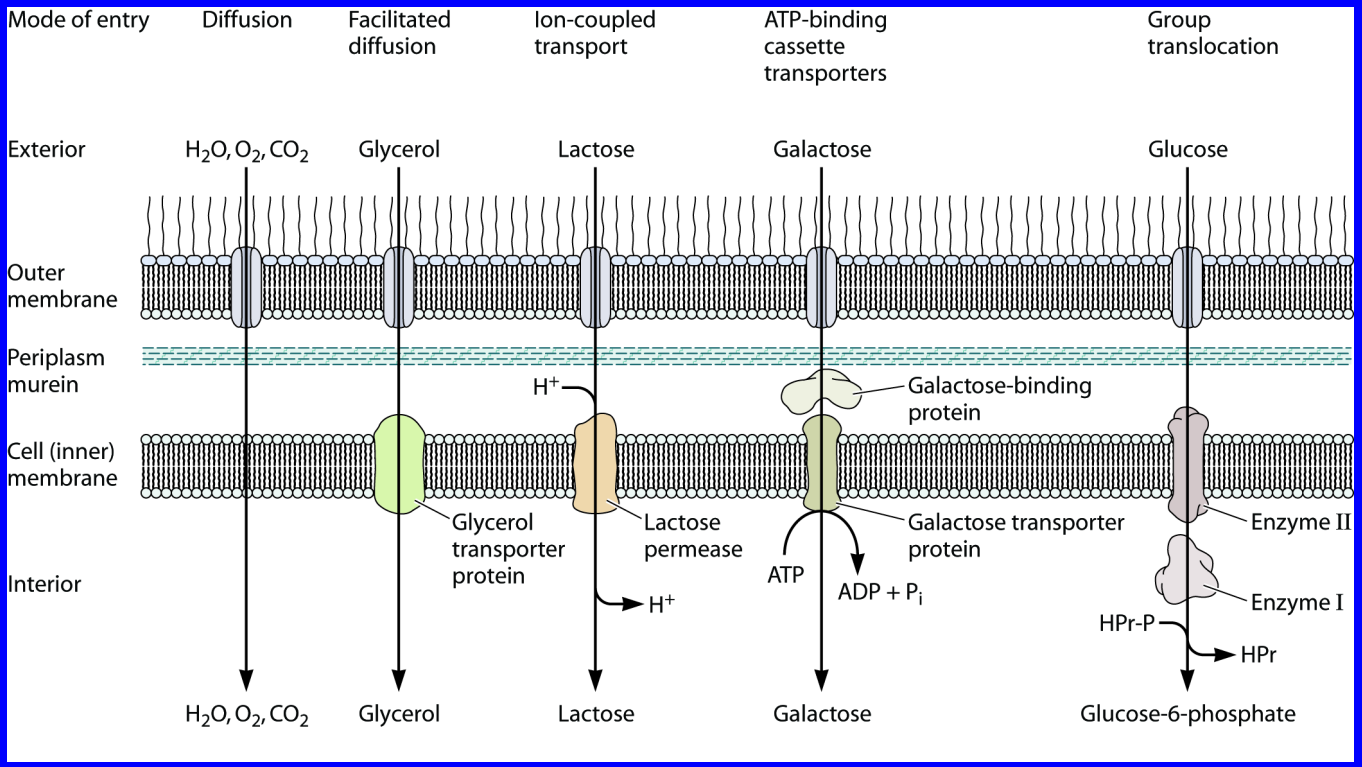



Figure 6.10
Microbe

# Assignment 14

1. What molecules are transported by each of the different mechanisms discussed?
2. How do the mechanisms of transport most frequently used for sugars differ from the mechanisms of transport most frequently used for ions

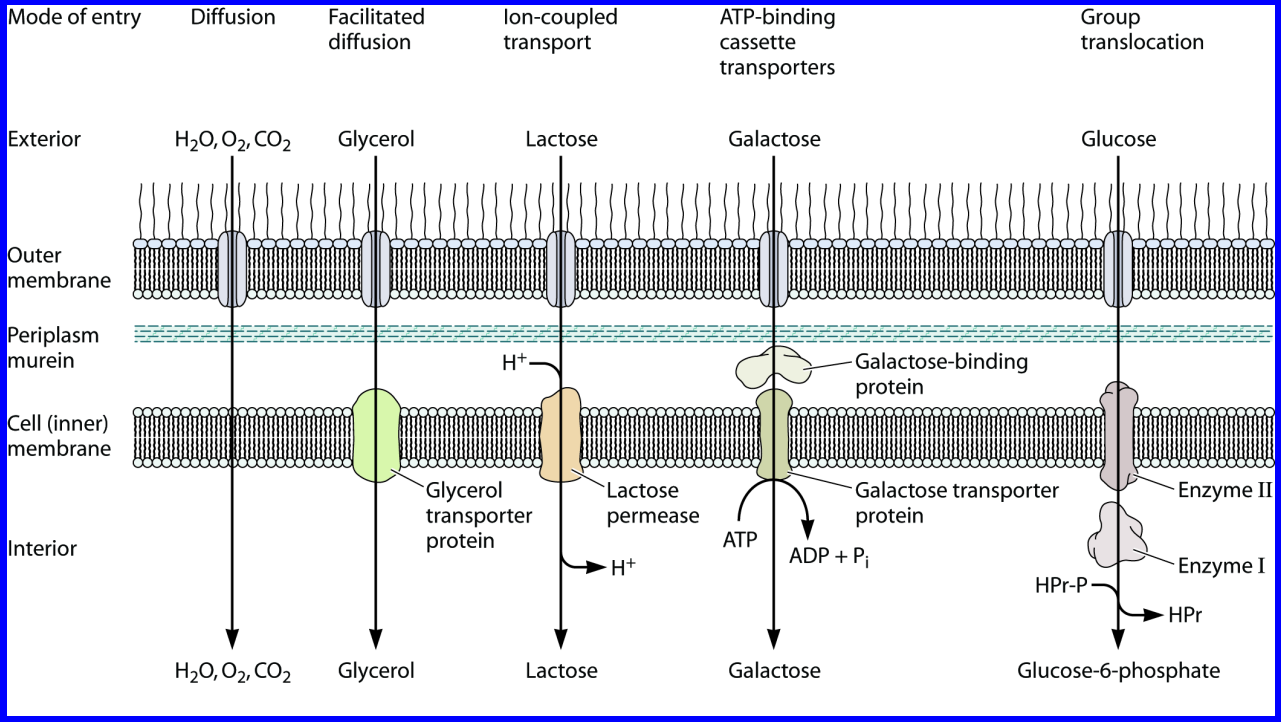
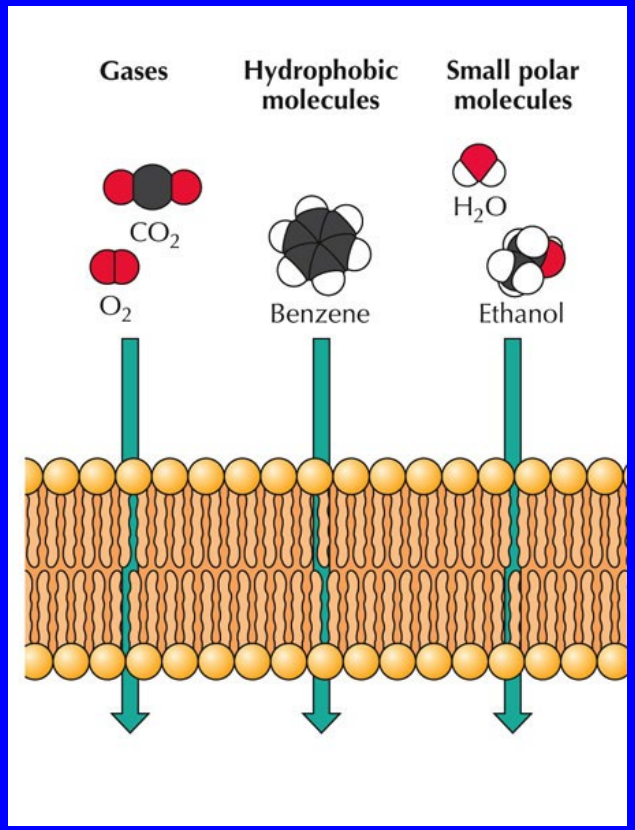


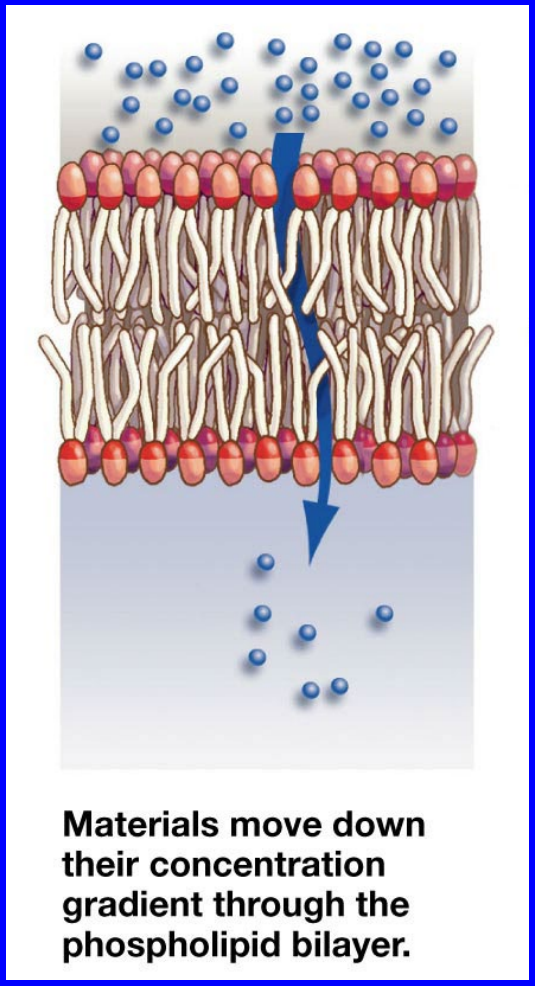

Figure 6.10
Microbe

# a). Passive Transport

1. Passive or simple diffusion – through bilayer
   a. Small uncharged molecules ($O_2$, $H_2O$) –
   b. Hydrophobic Molecules




Movement based on concentration gradient



# a). Passive Transport - 2

1. Passive or simple diffusion –
   c. Weak acids and bases
   - Can exist in two forms
     For an acid: uncharged acid (HA) or charged ($A^-$)
   - Uncharged form (HA) may be able to cross membrane
   - Once inside will deprotonate and be water soluble

Many drugs are weak acids and bases: aspirin, Prozac, penicillin, tetracycline

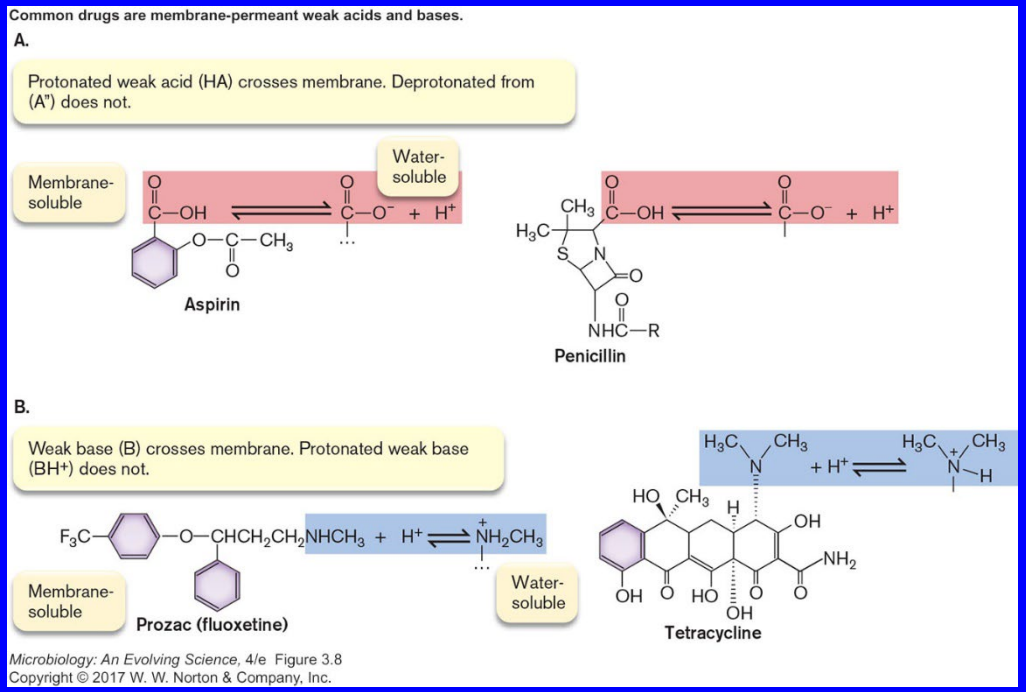


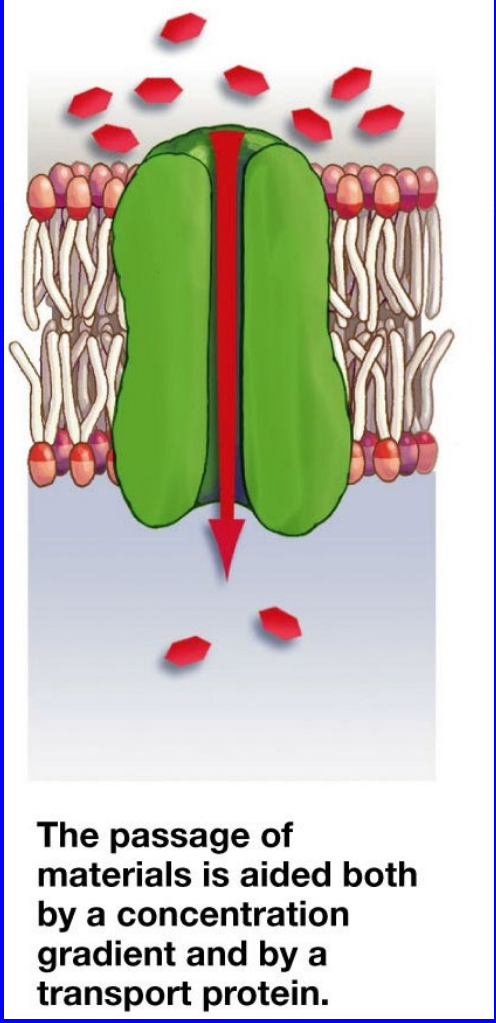

Figure 3.11
6th edition

# a). Passive Transport - 3

2. Facilitated diffusion
   - Go through membrane protein
   - Passive - No energy input
   - Flow determined by concentration and/or charge gradient (from high to low)

   Examples in bacteria

   a. Channel proteins - Outer membrane porins
   b. Carriers in inner/cell membrane in bacteria (rare – only a few)

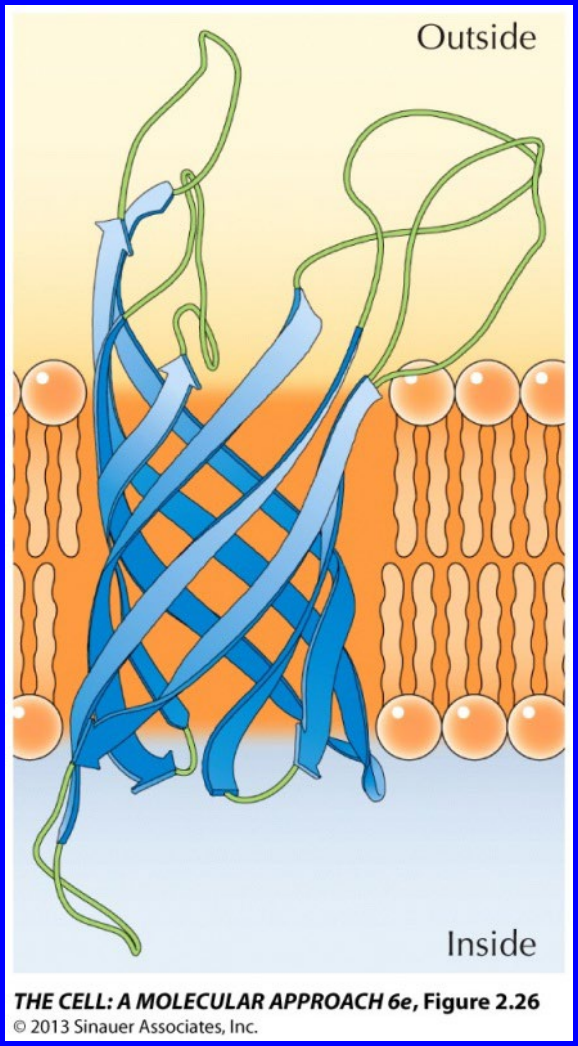


The passage of materials is aided both by a concentration gradient and by a transport protein.

# 2. Facilitated diffusion - a

a. Channel proteins

- Example: Outer membrane porins in Gram-negative bacteria
  - Some selective as far as type and size
  - Maximum size: 700 daltons (short chains of sugars and amino acids)




THE CELL: A MOLECULAR APPROACH 6e, Figure 2.26
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# a. Channel proteins

Porin channel proteins in *E. coli*

| Protein | Function |
|---|---|
| **OmpF** | Nonspecific channel for small molecules; receptor for phage T2 |
| **OmpC** | Nonspecific channel for small molecules; receptor for phage T4 |
| **OmpA** | Specific for peptides; receptor for mating pili |
| **LamB** | Porin for maltose and maltodextrin; receptor for phage λ |
| **PhoE** | Anion-selective channel |

# 2. Facilitated diffusion - b

b. Carrier proteins
- Binds to specific molecules and transport them across membrane
- Uncommon in bacteria –
- One example: glycerol transporter
  - Usually transport glycerol in, Conc of glycerol inside cell is very low as it is used up

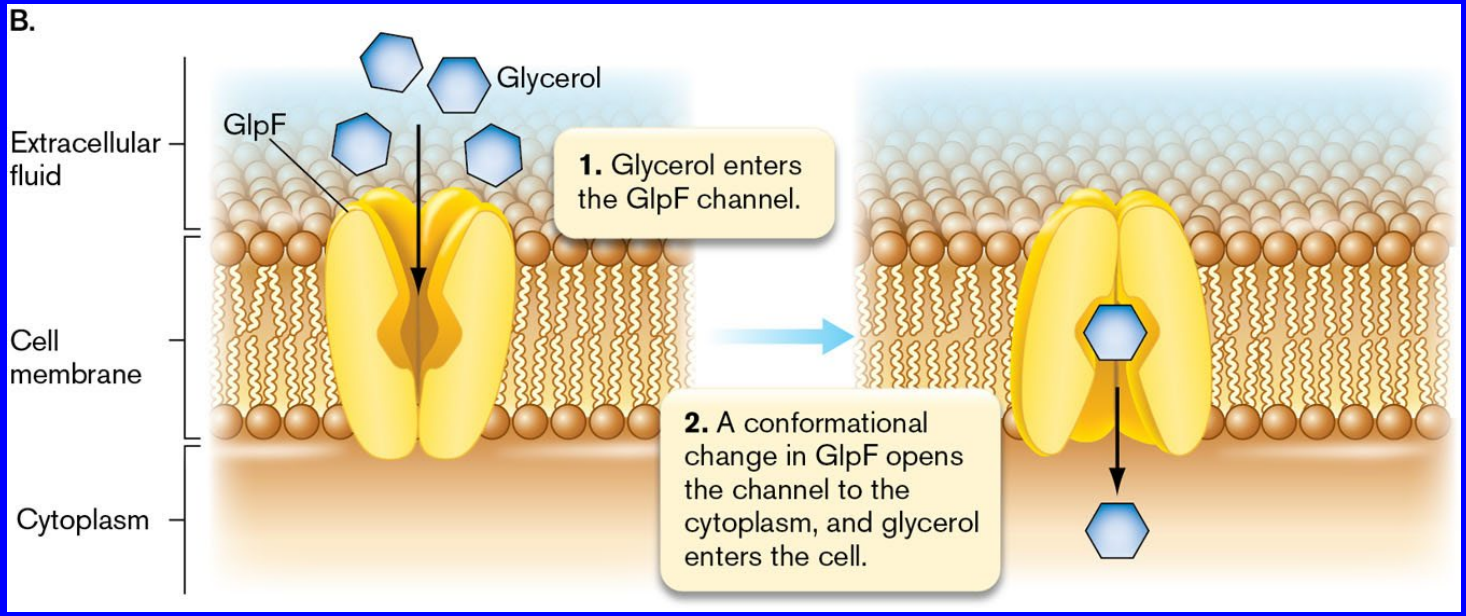
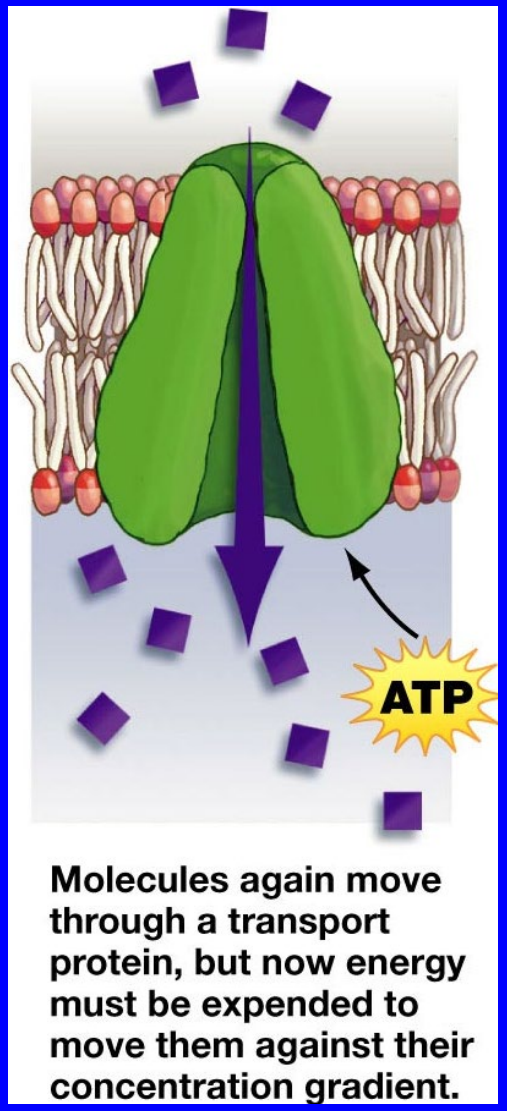



Figure 4.6 6th edition Glycerol transporter

# 3). Transport across Membrane

b. Active transport
   - Against concentration gradient
   - Requires energy to drive transport

1. Ion coupled – use ion gradient to drive transport
2. ATP driven – use energy from cleavage of high energy phosphate bond



# b. Active transport

1. Ion coupled transport
   – Transport an ion with concentration gradient to power transport of a molecule against conc. gradient

- Antiport – two liked charged ions, opposite directions
- Symport – Two ions in same dir.

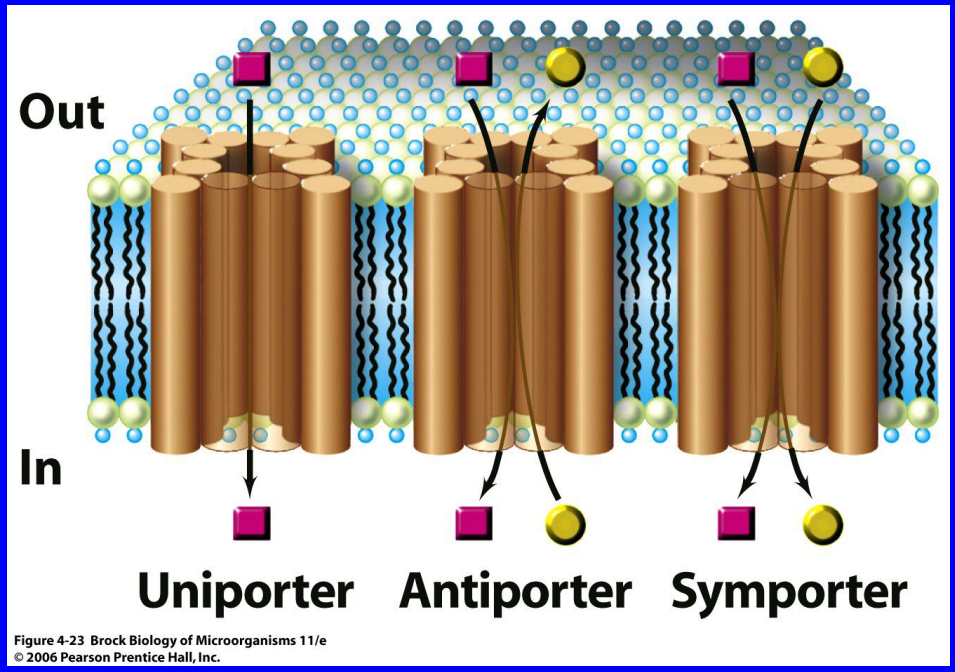


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# 1. Ion-coupled transport - 1

- Bacteria use $H^+$ and/or $Na^+$ ions to power transport
- Most use $H^+$ going in (proton gradient)
- Some use $Na^+$

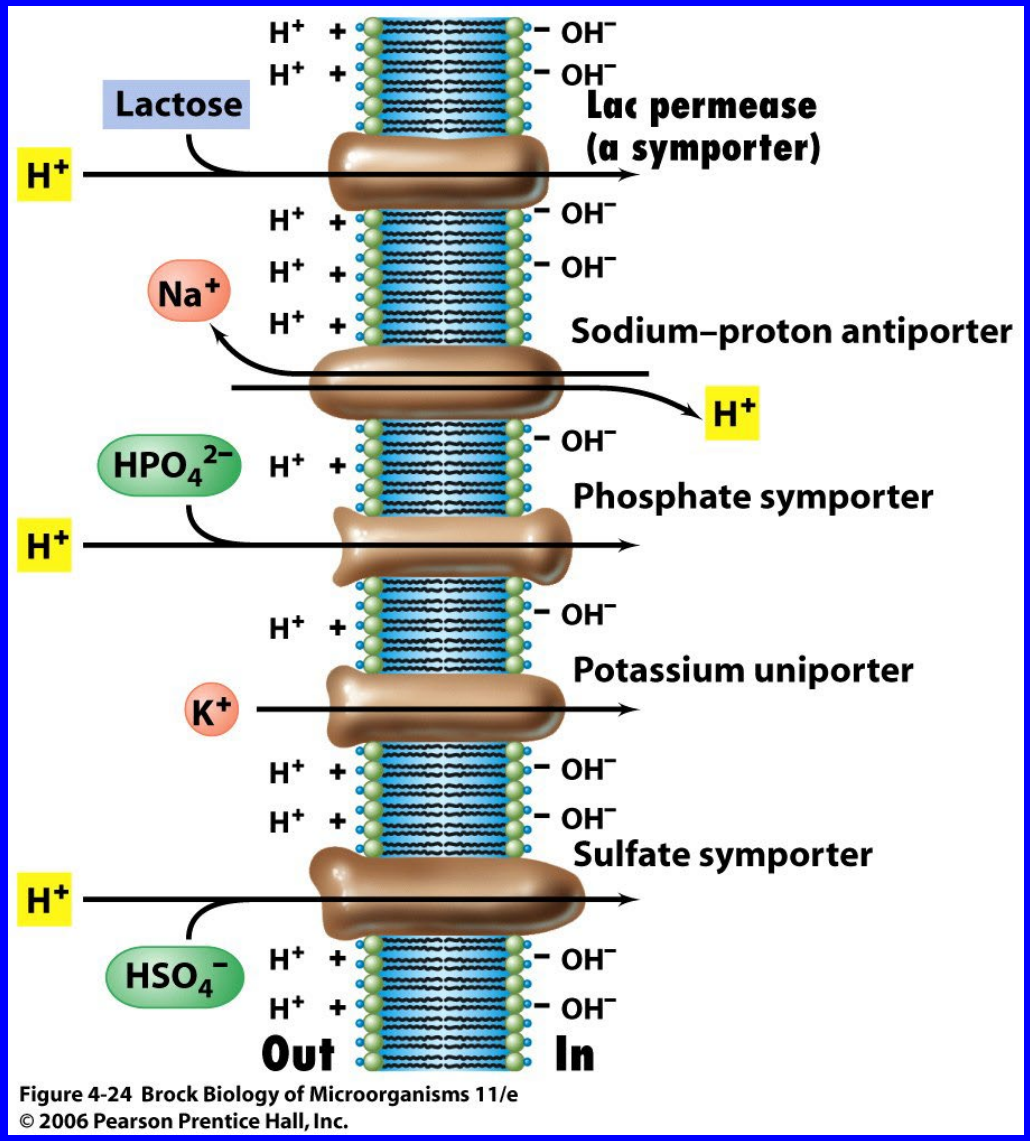



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# 1. Ion coupled Transport - 2

- Best known electrochemical gradients is with $H^+$ ions or protons that creates proton motive force across inner membrane of mitochondria and Gm neg bacteria

In bacteria, used for transport in addition to ATP synthesis

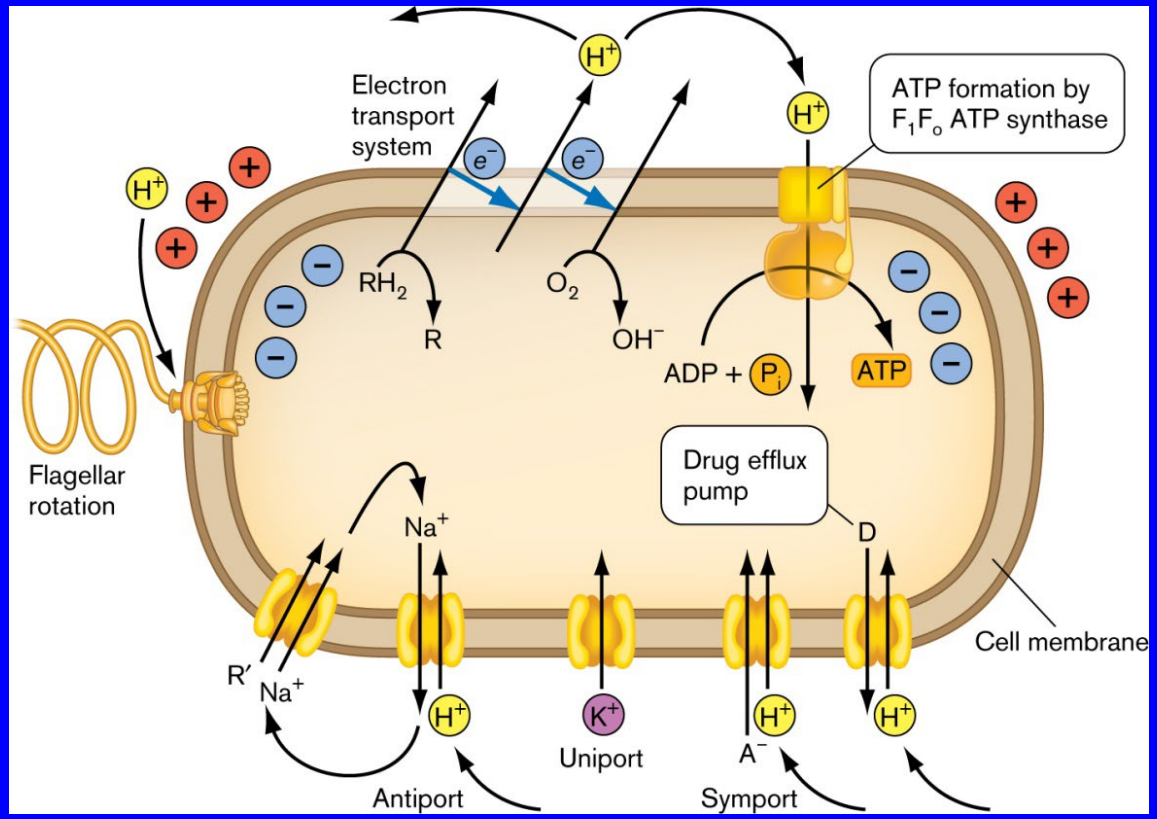



Figure 14.10
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# 1. Ion coupled Transport - 3

- Nebraska and other places have alkaline and brine lakes and marshes. Haloalkaliphilic bacteria use both $Na^+$ and $H^+$ gradients to power transport and other processes

What is powered by $Na^+$ ion transport?

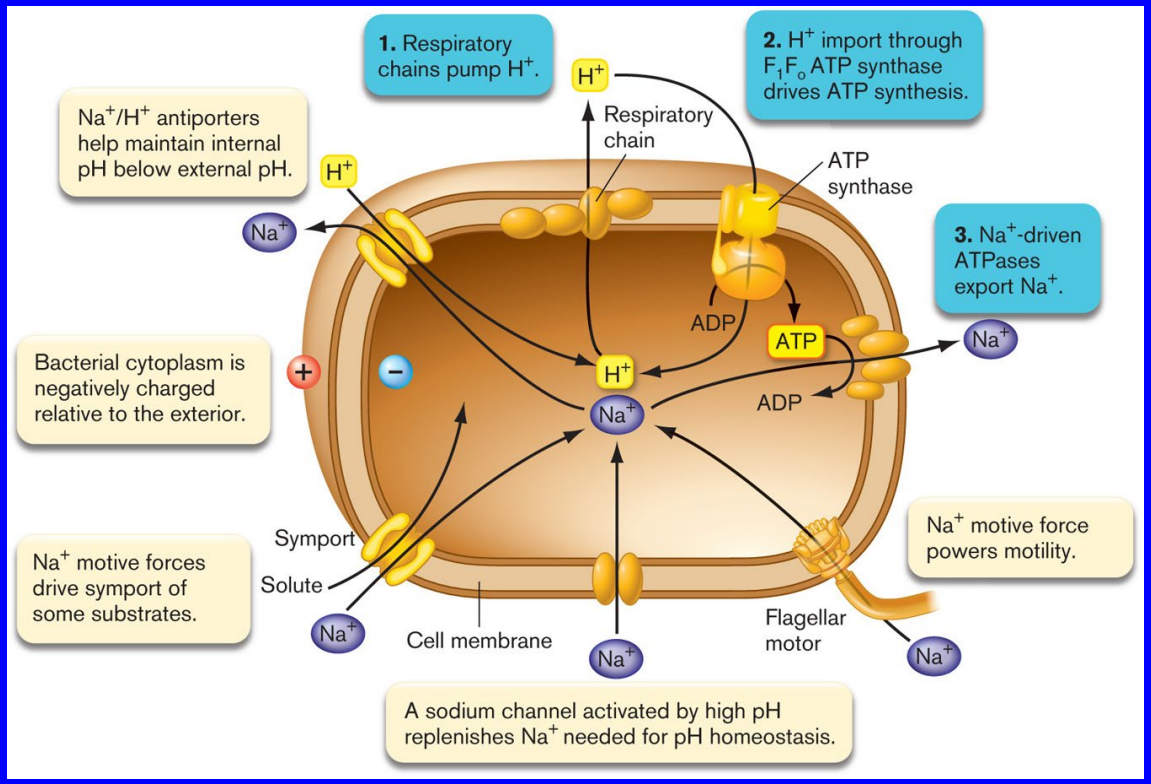
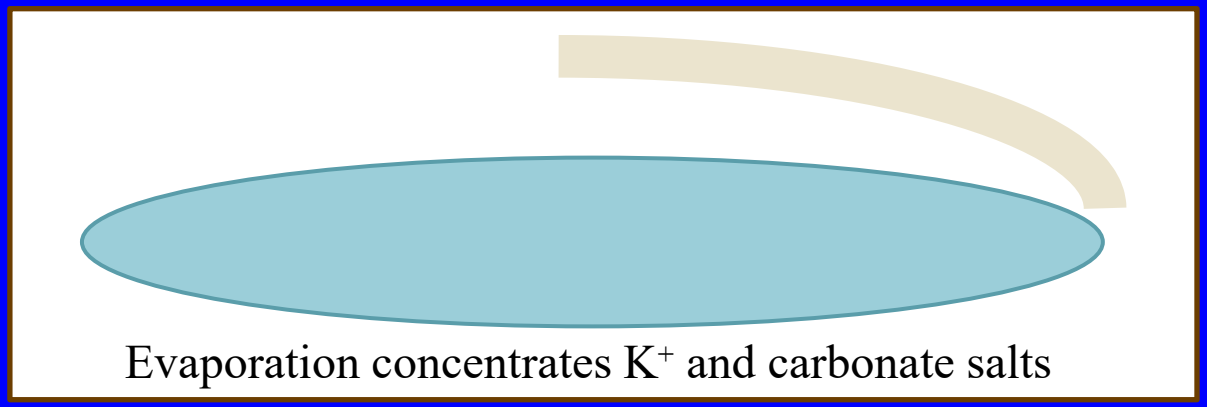



Figure 5.15
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# Western Sandhill Alkaline Lakes

**Where:** especially Garden and Sheridan Counties
**Chemistry:** closed/interdunal basins concentrate salts by evaporation; many lakes reach pH >10
**Unusual feature:** carbonate-rich water with unusually high potassium; white salt crusts can form along shorelines
**Microbiology:** selects for haloalkaliphiles and other polyextremophiles; some communities include chemolithoautotrophs



Evaporation concentrates $K^+$ and carbonate salts

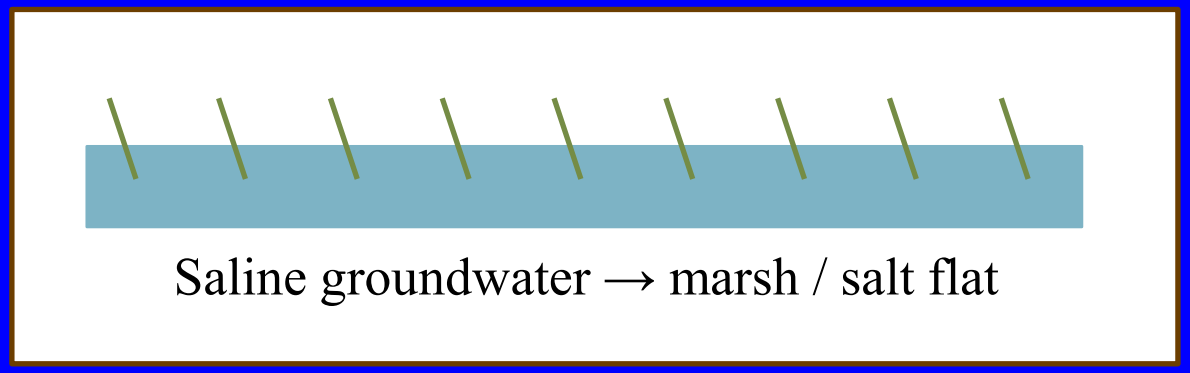
# Eastern saline wetlands ("salt marshes")

**Where:** Salt Creek floodplain and tributaries in Lancaster and southern Saunders Counties

**Source of salt:** saline groundwater moving through ancient marine salt-bearing formations

**Landscape:** saline marshes, ponds and salt flats; summer drying can leave visible salt crusts

**Ecology:** supports salt-tolerant plants and specialized microbial communities adapted to osmotic stress

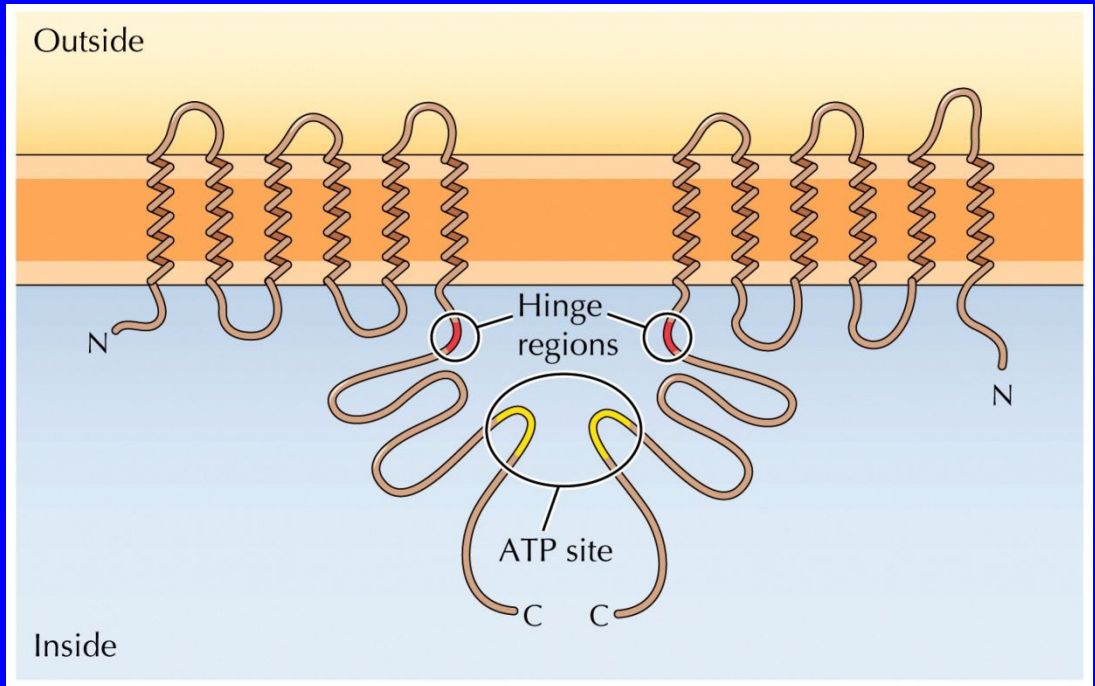


# b. Active transport

2. ATP driven transport
   a. ABC (ATP-binding cassette) family of transporters
      – Common in bacteria (*E. coli* has 78)
      – Formed by 2 or 4 proteins

Two domains
1. Transmembrane
2. ATP binding

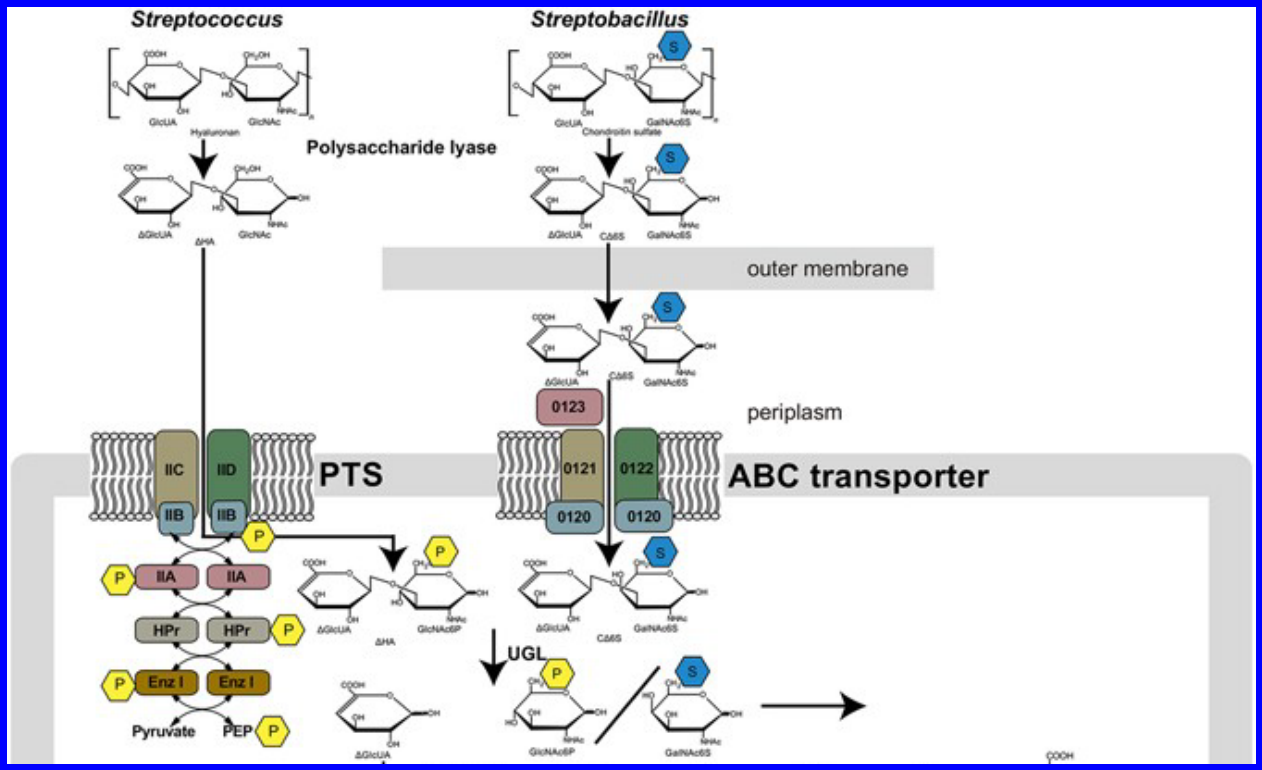
- Can be in same protein or separate




*THE CELL: A MOLECULAR APPROACH 6e*, Figure 13.32
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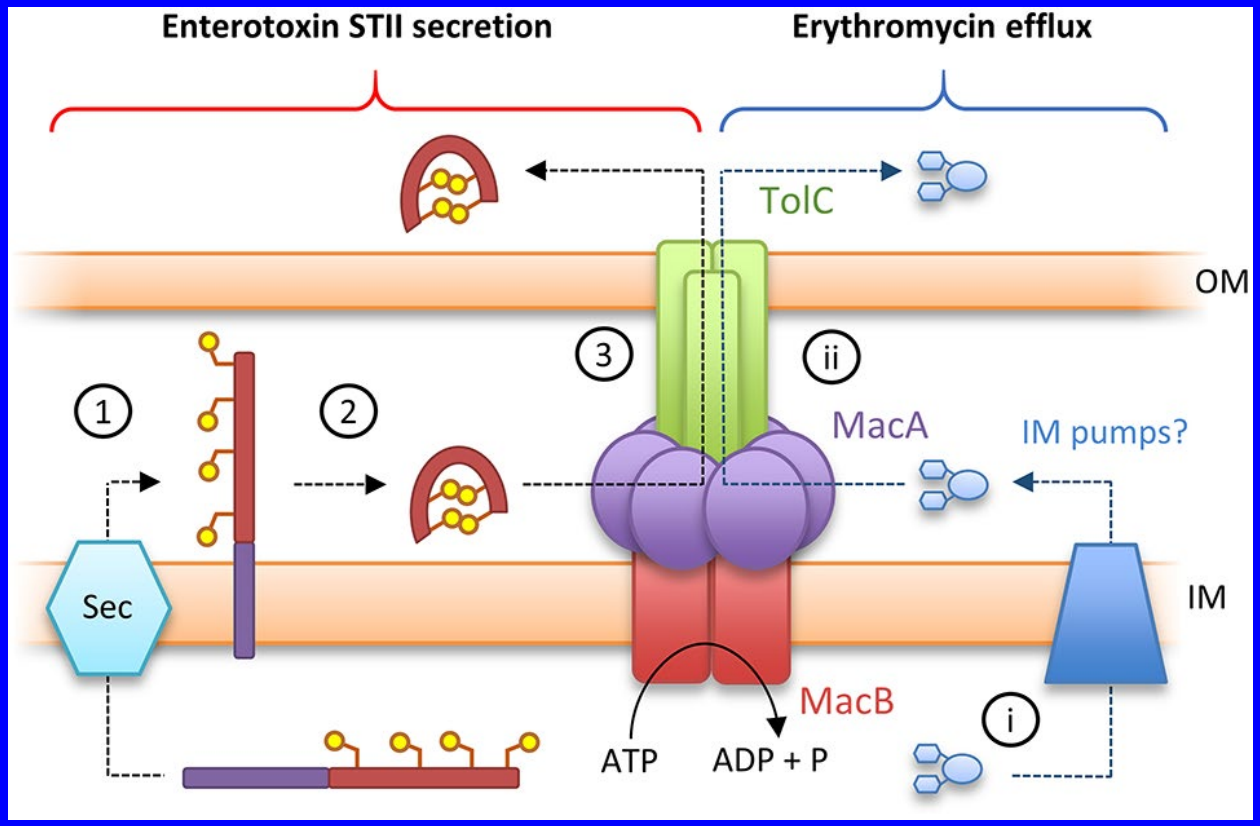
# a. ABC transporters - 2

1) Example of an ABC (ATP-binding cassette) Proteins
   - Transporter of Glycosaminoglycans (GAGs) disaccharides in *Streptobacillus*



# a. ABC transporters - 3

2) Some transport chemicals out such as some multidrug exporters



# a. ABC transporters - 4

3) May also have a solute binding protein in periplasm which bring solute to transporter

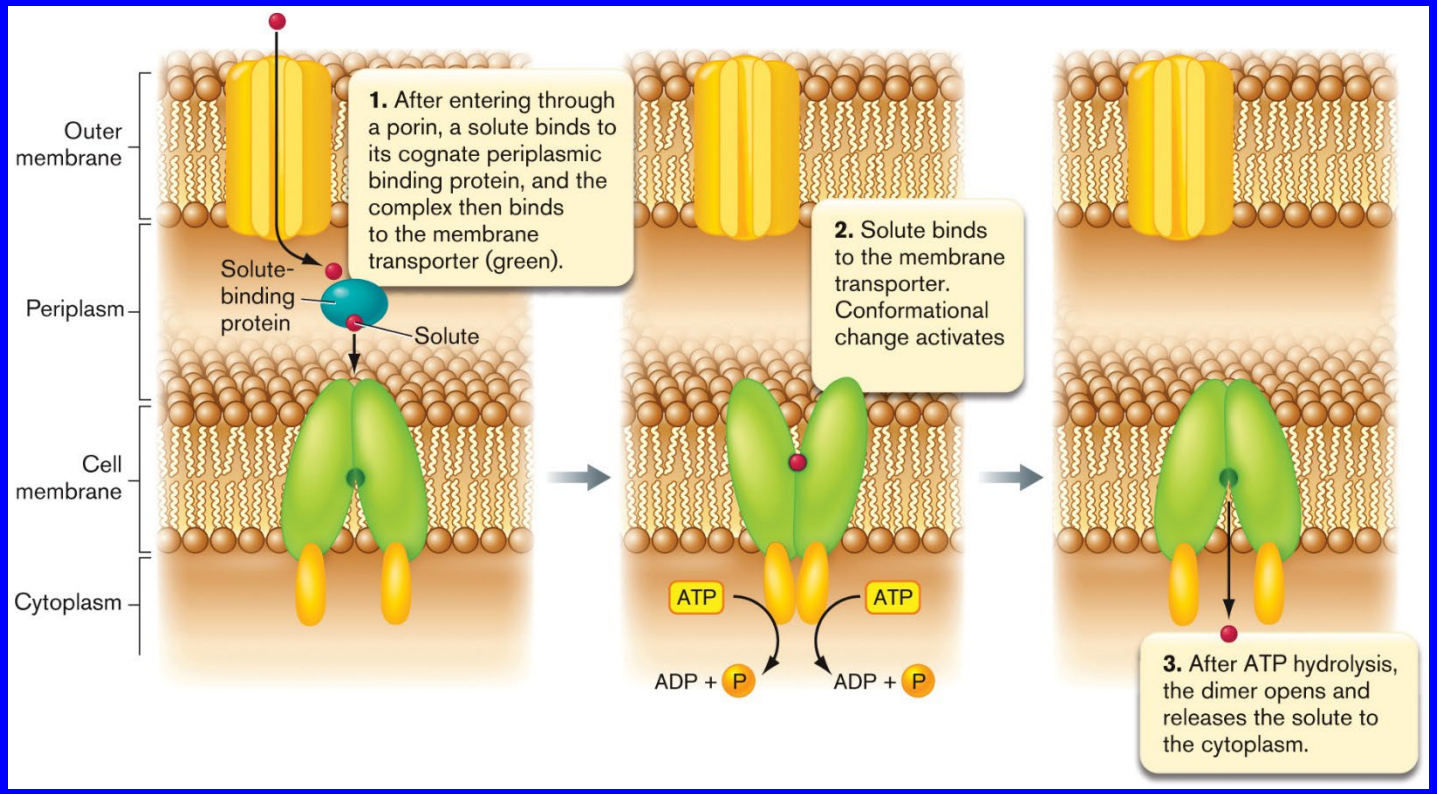



Figure 4.9
6th edition

# a. ABC transporters - 5

3) Siderophore mediated Transport Example

- Bacteria secrete siderophores which bind an iron ($Fe^{3+}$) ion
- Complex transported across cell membrane by ABC type transporter
- Inside cell, $Fe^{3+}$ ion released

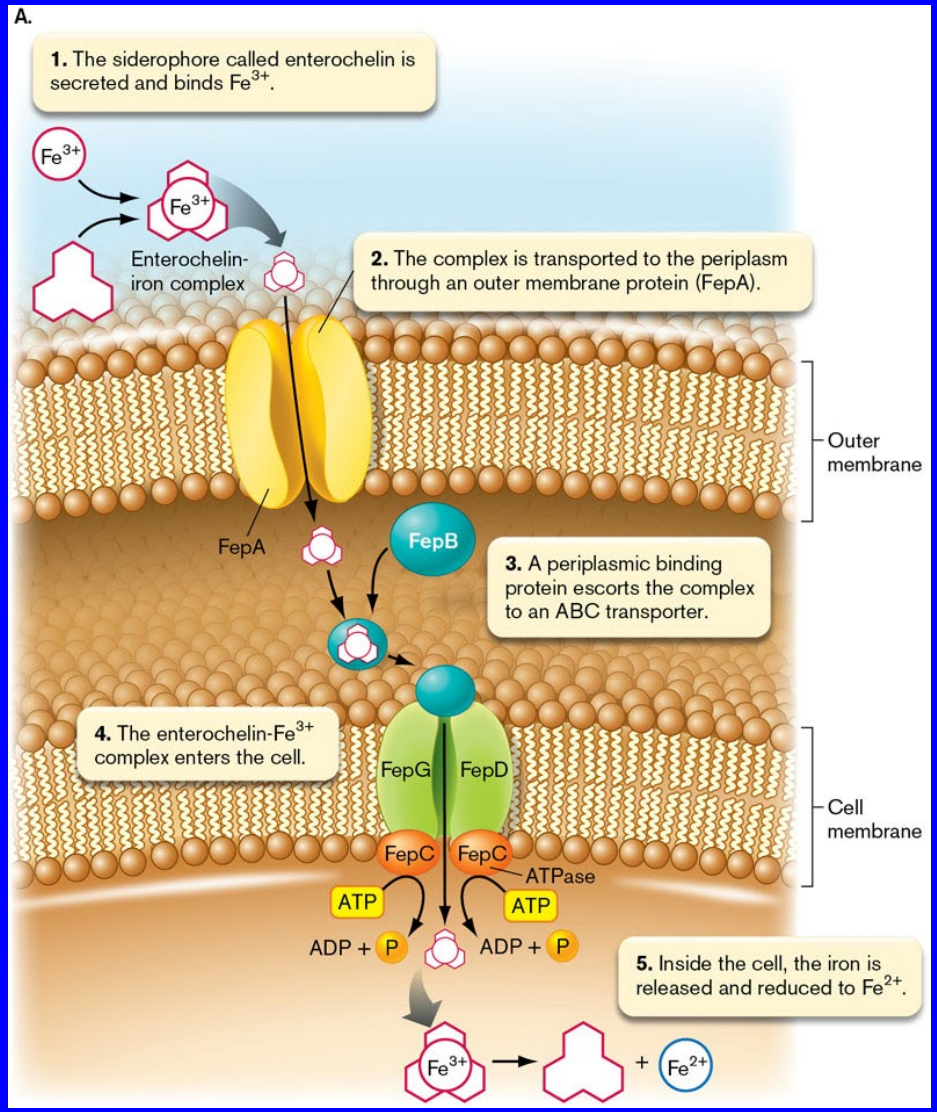



Figure 4.10
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# 2. ATP driven Transport - b

b. Group Translocation or Phosphotransfer system (PTS)

- Molecule transported in is modified during transport
- Used for transport of some sugars into cells
- A phosphate from PEP is added to the sugar as it is transported in
- Only found in bacteria and some Archaea

How can this system make transport of glucose be with the concentration gradient (concentration lower inside than outside)?

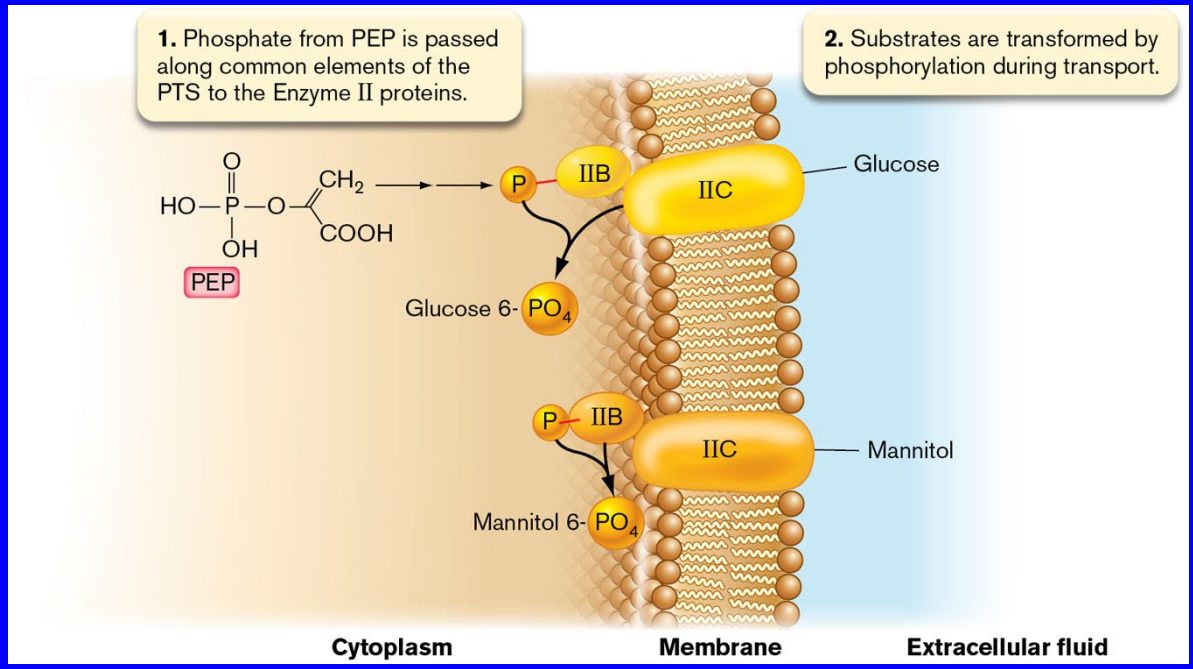



Figure 4.11
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# Assignment 14 - 2

1. What molecules are transported by each of the different mechanisms discussed?
2. How do the mechanisms of transport most frequently used for sugars differ from the mechanisms of transport most frequently used for ions

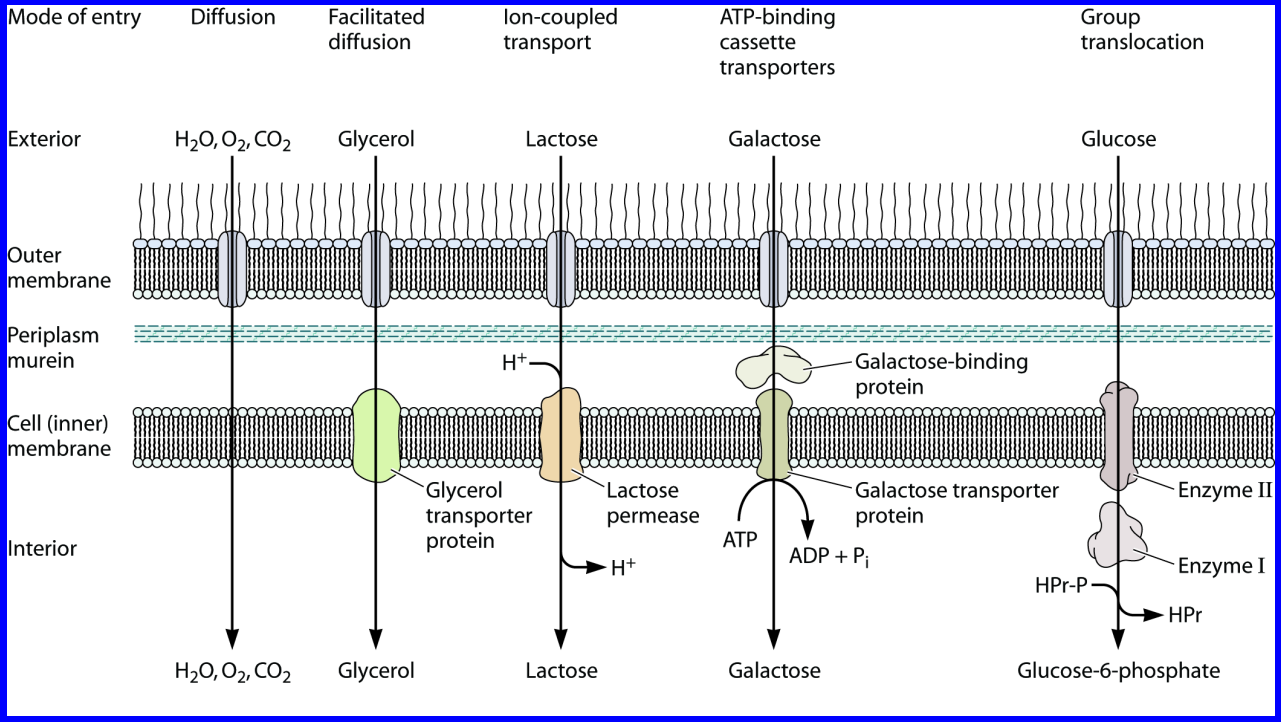



Figure 6.10
Microbe

# C. Cell Envelopes - 3

3. Peptidoglycan Synthesis and Antibiotics
   a. Peptidoglycan Synthesis is unique to bacteria
   b. Many antibacterial drugs (antibiotics) interfere with or inhibit peptidoglycan synthesis
      - Antibiotics inhibit different steps
   c. Some enzymes cleave peptidoglycan
      - Lysozyme – defensive enzyme of our body

# a. Peptidoglycan Synthesis

- New peptidoglycan needs to be synthesized for both cell growth/division and repair/replacement
- Have to maintain integrity of layer while cutting links between existing peptidoglycan and new peptidoglycan

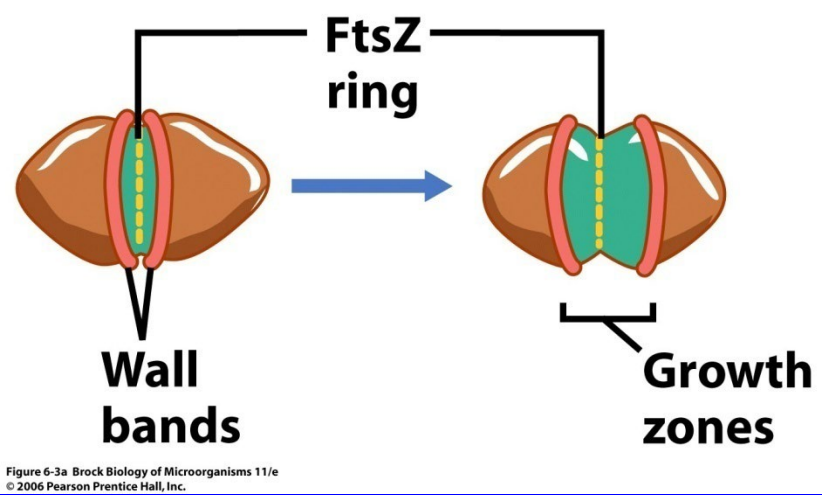


Figure 6-3a Brock Biology of Microorganisms 11/e
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Green is new peptidoglycan

# a. Peptidoglycan Synthesis -2

- Site where new peptidoglycan synthesis occurs varies
  - Can occur in middle, one end or dispersed

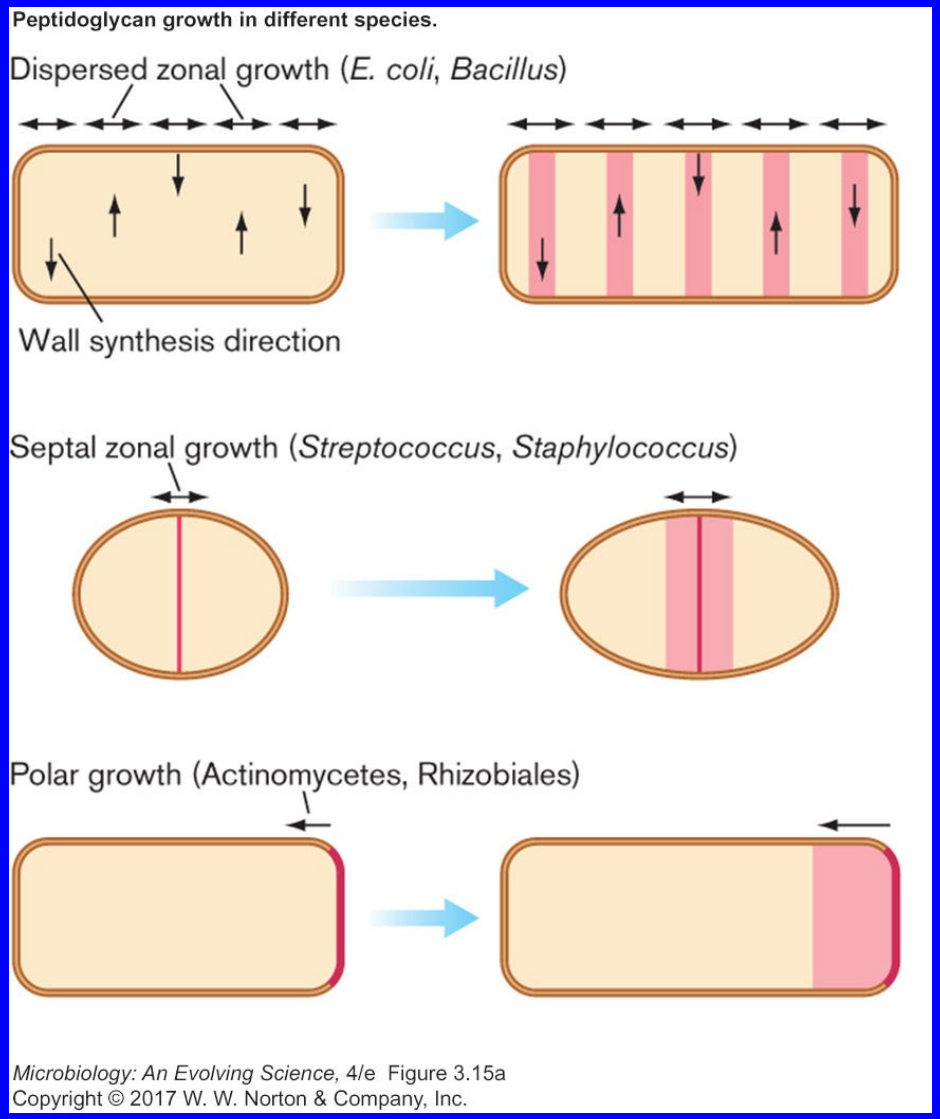



Figure 3.14A
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# a. Peptidoglycan Synthesis - 3

Overview

1. Synthesize two sugar units (NAG and NAM) with peptide off NAM
2. Add 2 sugar unit to an existing chain
3. Then form crosslink to peptide in a different chain

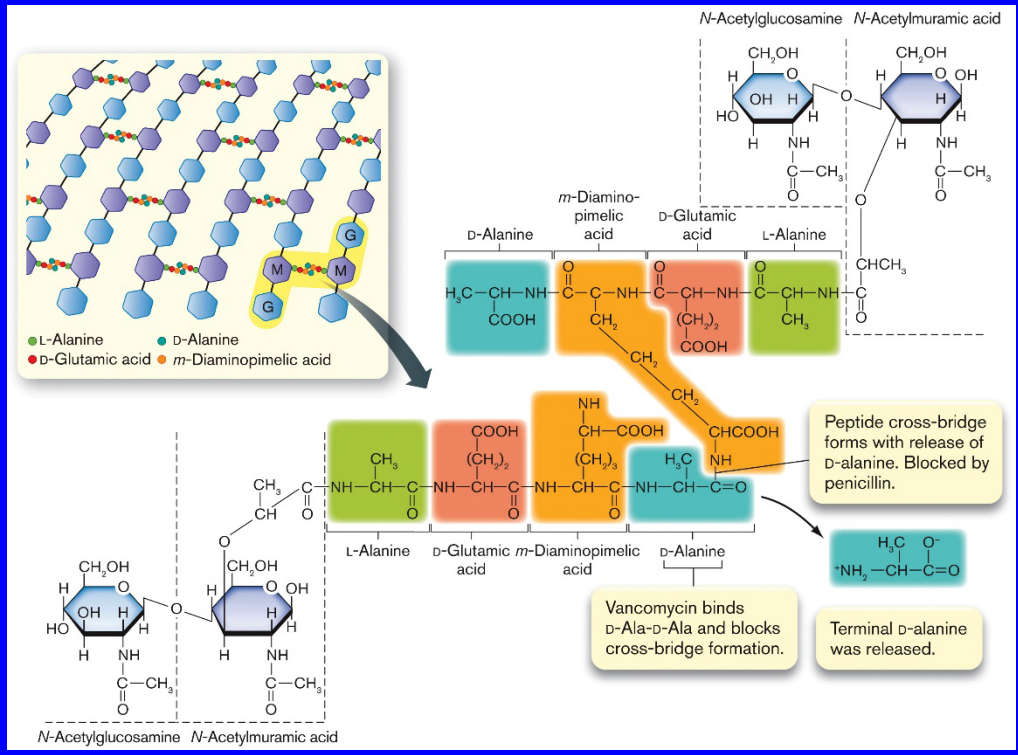
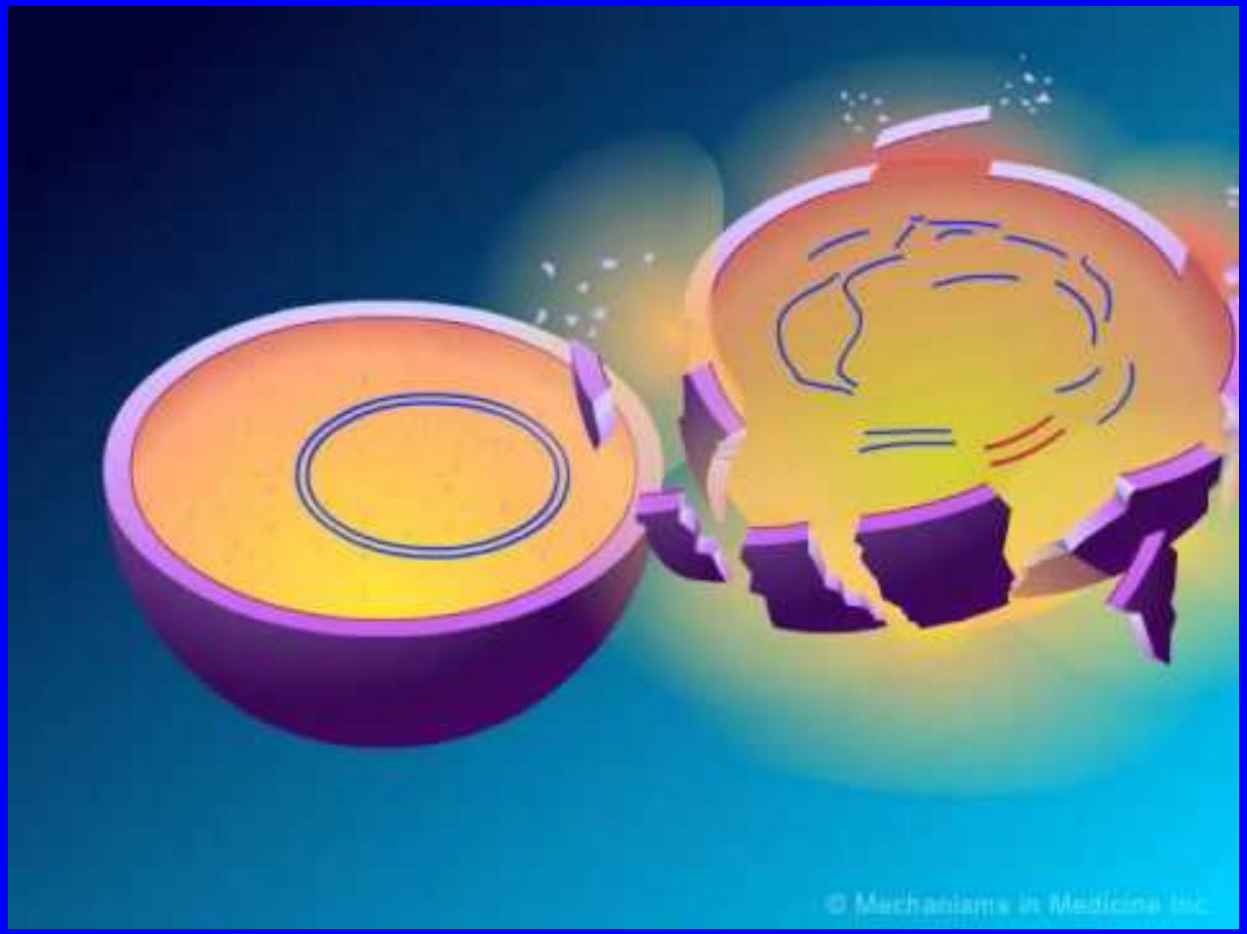



Figure 3.12b
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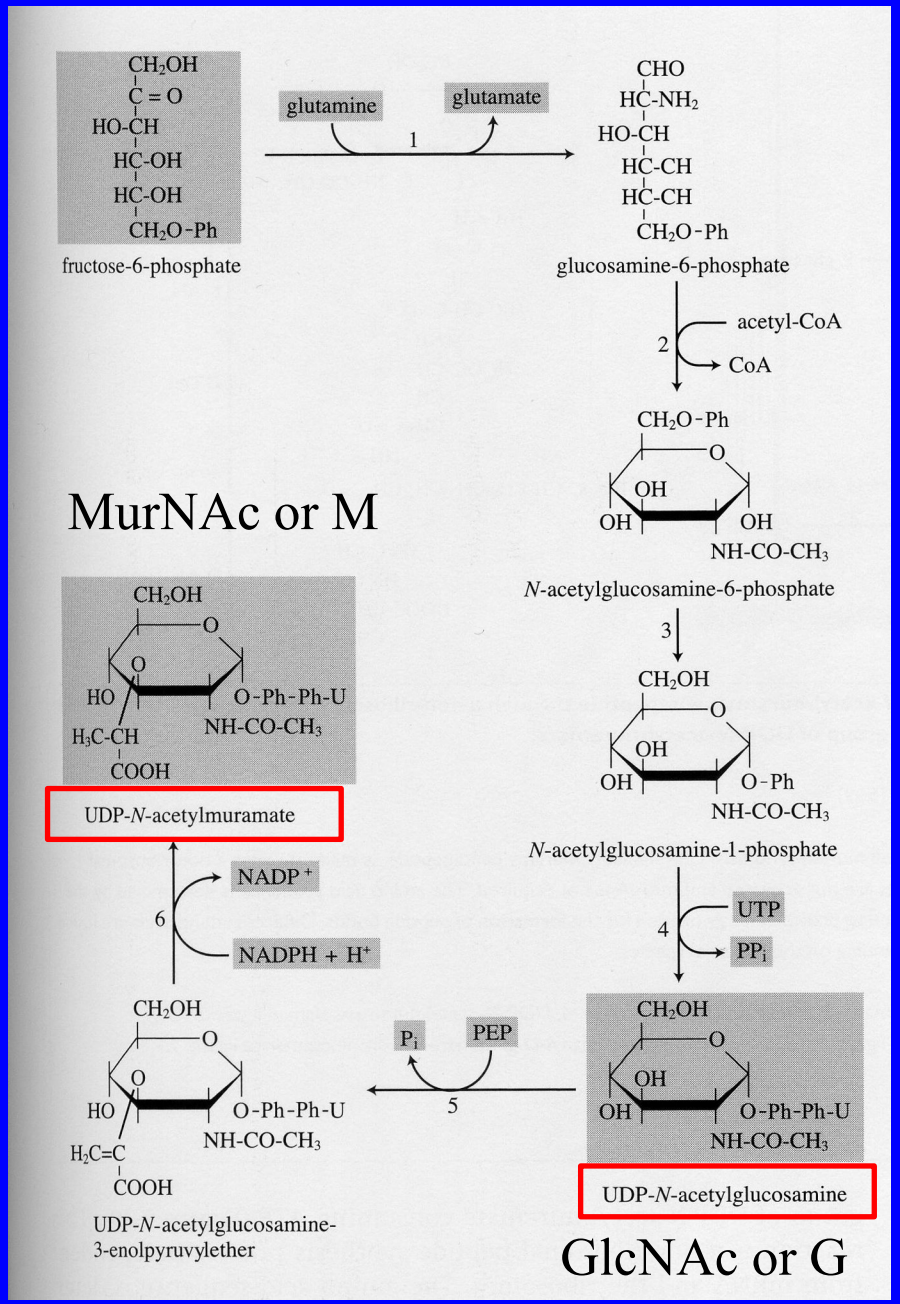
# a. Peptidoglycan Synthesis - 4

Video ß-Lactams: Mechanisms of Action and Resistance: 0:01-2:30 covers Peptidoglycan synthesis



# a) Peptidoglycan Synthesis

1. Make sugar precursors (NAM and NAG)
   a. Both sugar precursors made from Fructose-6-P in cytosol
   b. Sugars initially attached to UDP



# a) Peptidoglycan Synthesis

This is in a Gram-positive bacteria. Similar in Gm neg. but some steps occur in periplasm between membranes.

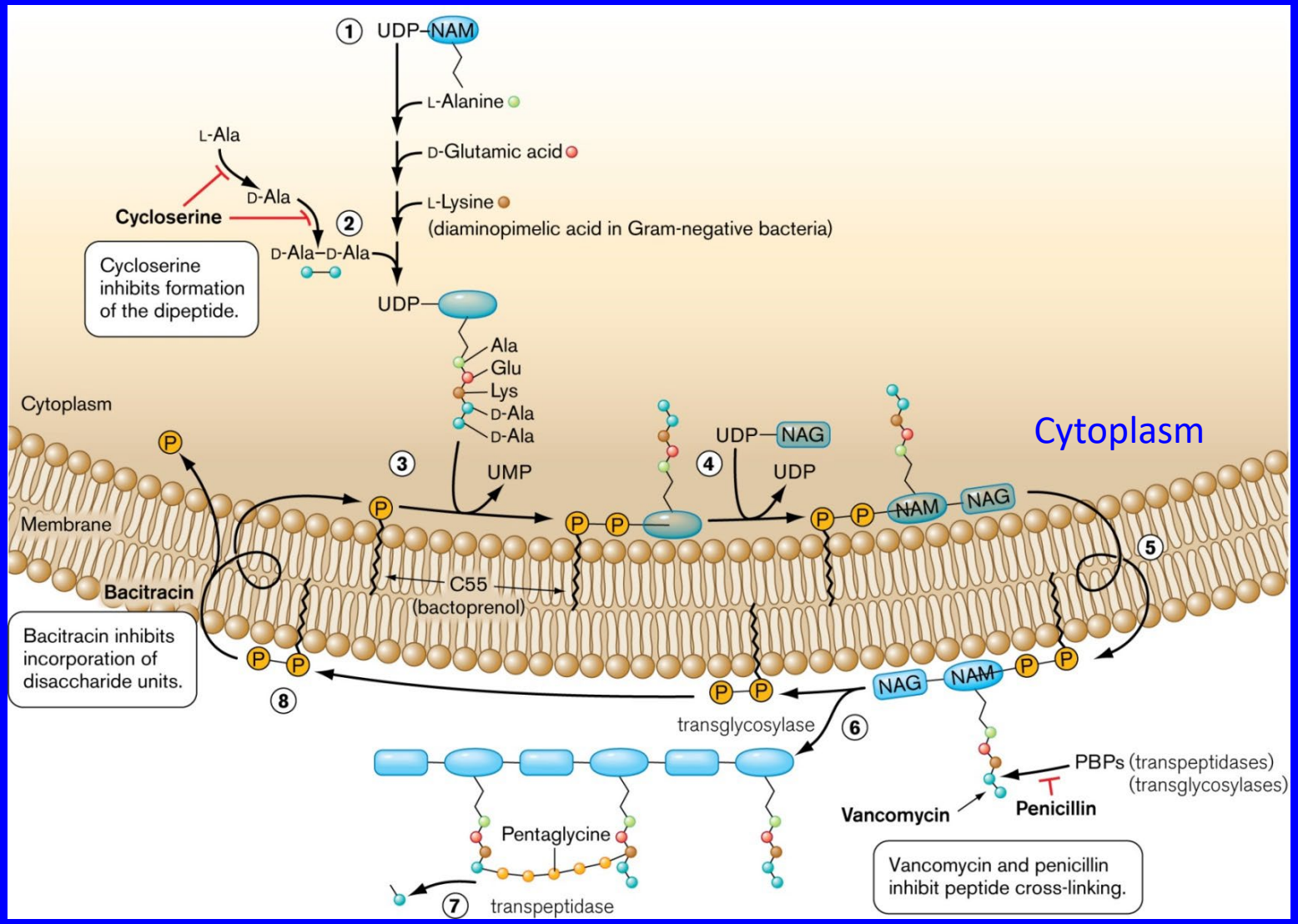



Figure 27.7 of 3rd edition

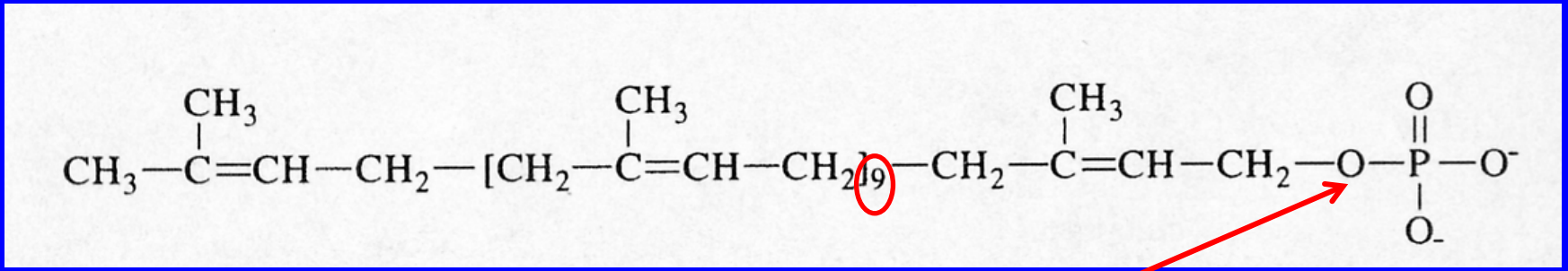
# a) Peptidoglycan Synthesis

2. Add 3 amino acids to UDP-NAM to begin peptide
3. Add dipeptide of 2 D-alanine amino acids to peptide
4. Transfer NAM-pentapeptide to bactoprenol (C55) in cell membrane
5. Link NAG to NAM
6. Flip 2 sugar unit to other side of bilayer
7. Transglycosylation - link sugars to a glycan chain
8. Transpeptidase – form cross link
9. Regenerate bactoprenol-P by cleaving off a phosphate group

- Transglycosylation and transpeptidase done by penicillin binding proteins (PBPs)

# a). Peptidoglycan Synthesis

4. Sugars transferred to bactoprenol
   - Bactoprenol or undecaprenyl is large isoprenoid phosphate with 55 carbons ($C_{55}$)
   - Bactoprenol helps to carry peptiodoglycan units, LPS and teichoic acid through the cell membrane
   - Transmembrane protein MurJ (flippase) – flips sugars to other side




   - Sugar is attached via phosphate

# a) Peptidoglycan Synthesis - 5

7. Transglycosylation reaction
   - Linking newly synthesized two sugar units to existing peptidoglycan chain

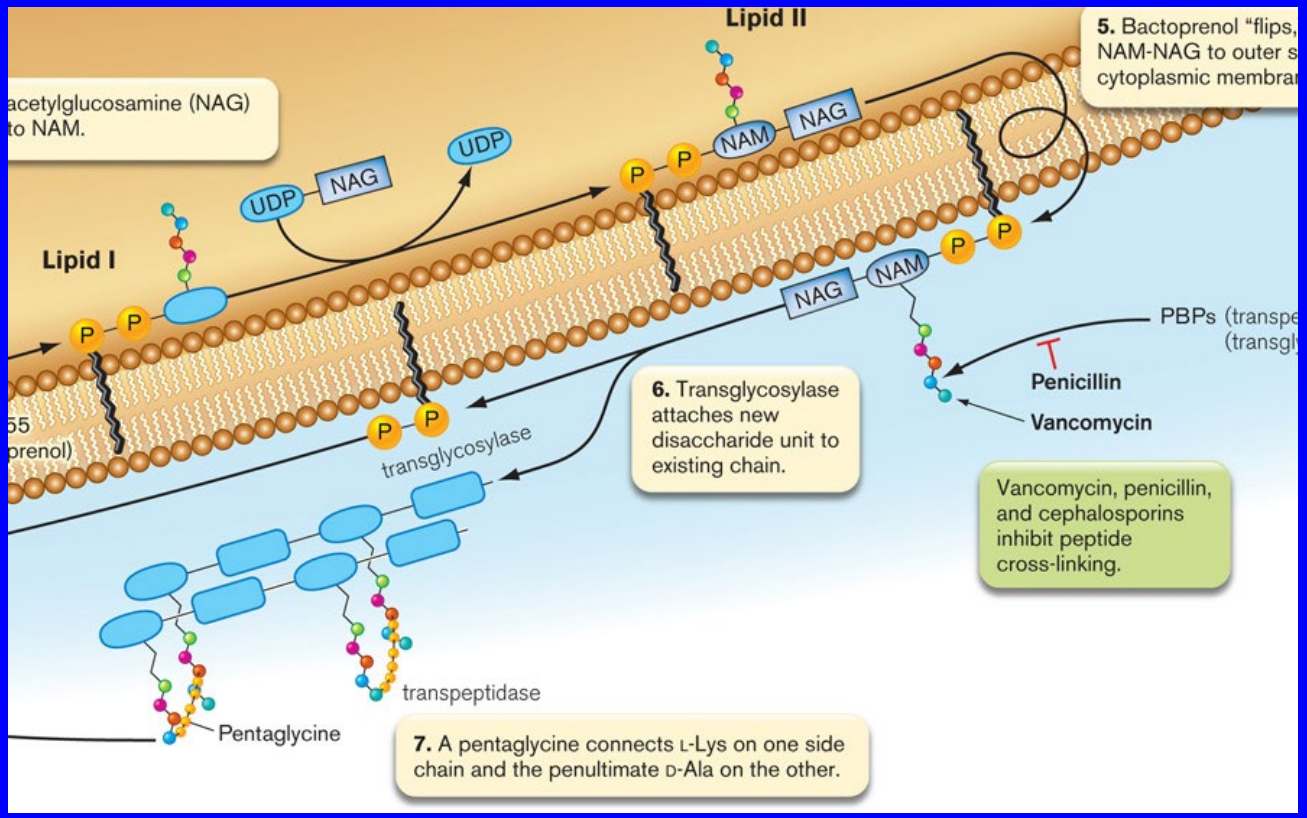



Figure 27.8B
6th ediition

# a) Peptidoglycan Synthesis - 6

8. Transpeptidation reaction
   - Crosslinking of peptides
   - In Gm negative, DAP in 3rd position attacks *D*-Ala in 4th position and displaces *D*-Ala in 5th position
   - Done by penicillin binding proteins

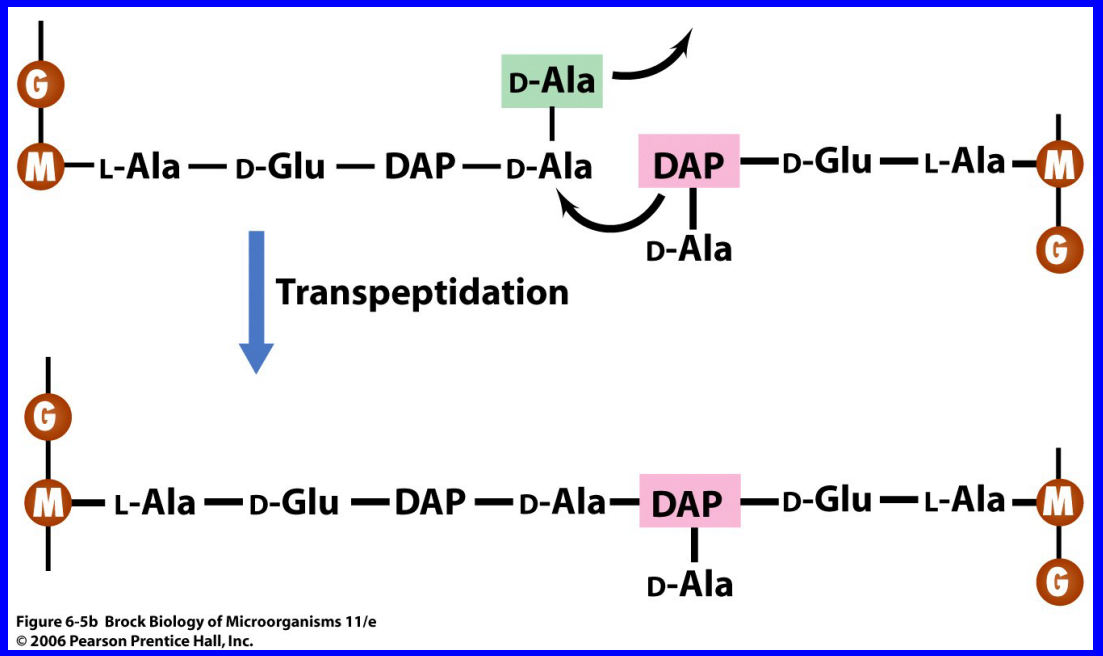



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# a) Peptidoglycan Synthesis - 7

- Penicillin binding proteins (PBB)
  - Part of complex involved in synthesis of peptidoglycan
  - Do transglycosylation and transpeptidation reactions

Fig 2.7 White Physiology and Biochem. of Prokaryotes

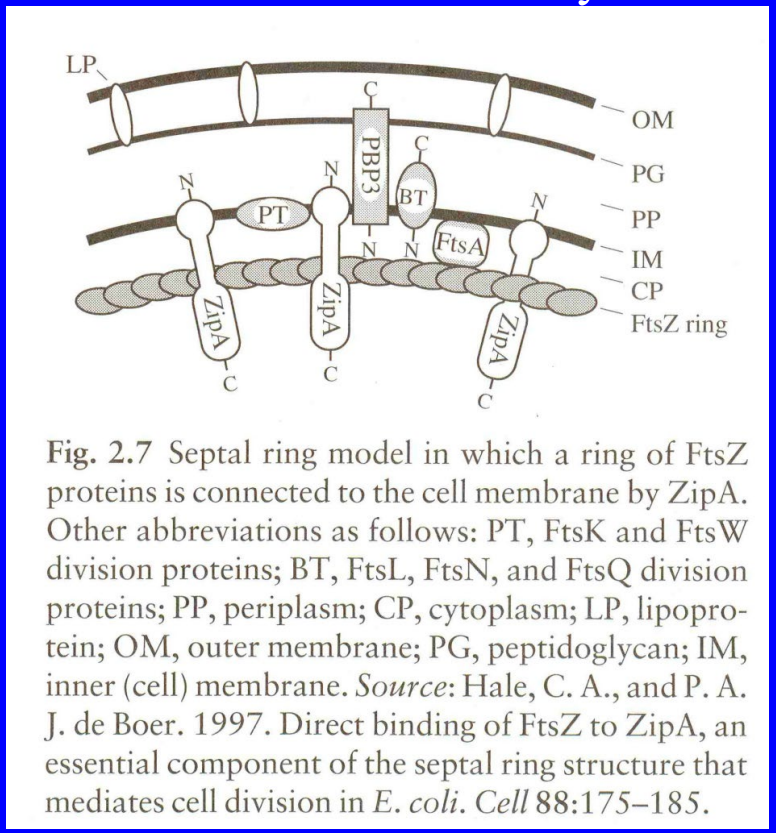



**Fig. 2.7** Septal ring model in which a ring of FtsZ proteins is connected to the cell membrane by ZipA. Other abbreviations as follows: PT, FtsK and FtsW division proteins; BT, FtsL, FtsN, and FtsQ division proteins; PP, periplasm; CP, cytoplasm; LP, lipoprotein; OM, outer membrane; PG, peptidoglycan; IM, inner (cell) membrane. *Source*: Hale, C. A., and P. A. J. de Boer. 1997. Direct binding of FtsZ to ZipA, an essential component of the septal ring structure that mediates cell division in *E. coli*. *Cell* 88:175–185.

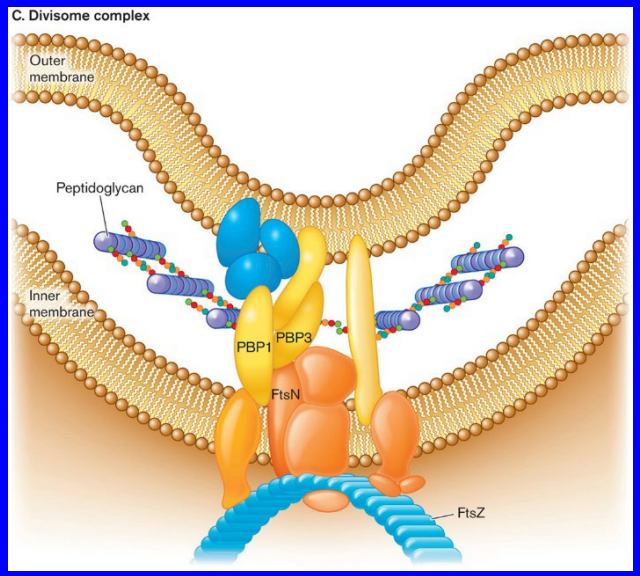



Fig 3.25C in 6th edition

# 3) Peptidoglycan Synthesis and Antibiotics

b) Antibiotics that interfere with peptidoglycan synthesis

1. Penicillin (β-lactam) family -
2. Cephalosporins -
3. Vancomycin –
4. Bacitracin –
5. D-cycloserine –

c). Enzymes that cleave peptidoglycan

1. Lysozyme
2. Autolysin – produced by bacteria

- Determine mechanism of action for Assignment 12

# End of Material covered in Lecture 18