Microscopy and Cell Structure

Chapter 3

3.3 Morphology of Prokaryotic Cells

Shapes

Arrangements

Prokaryotic Cell Shape & Arrangement

- Two types most common:
  - **Coccus**: spherical
  - **Bacillus**: cylindrical/Rod
    - Short rods sometimes called **coccobacillus**

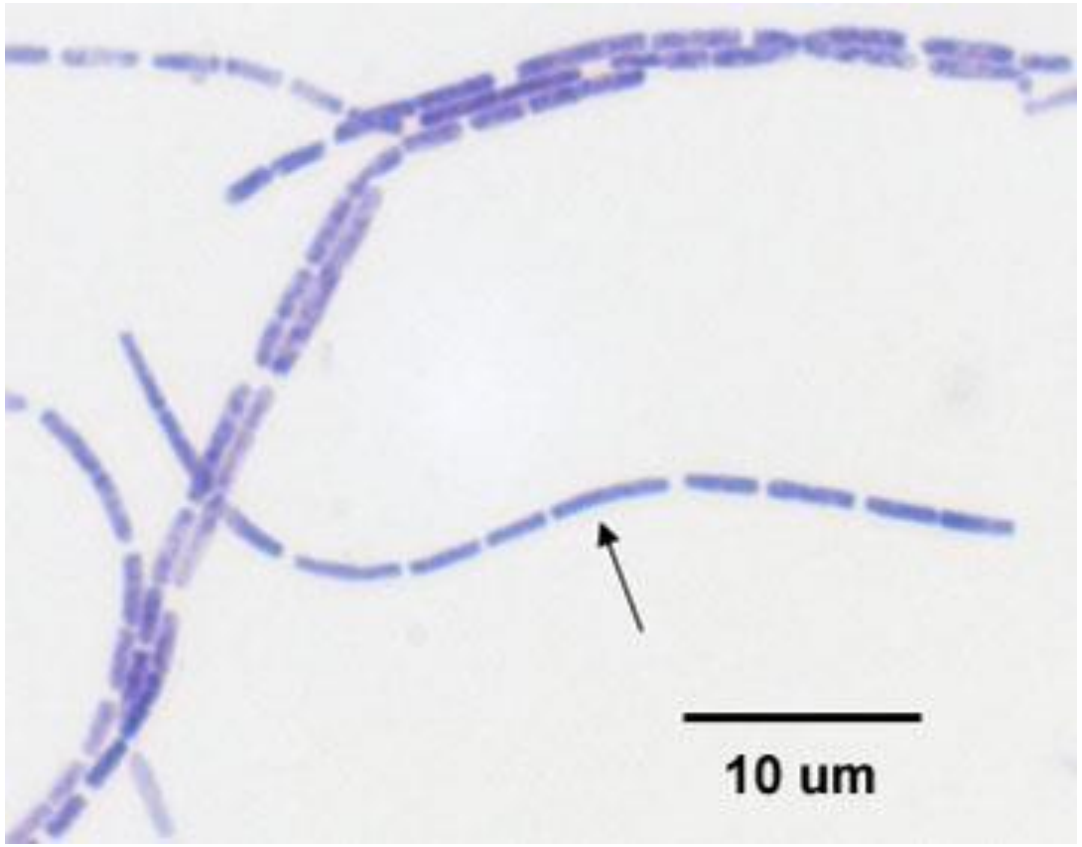
When bacteria reproduce, they often remain adhered to one another

Bacillus (pl. bacilli)

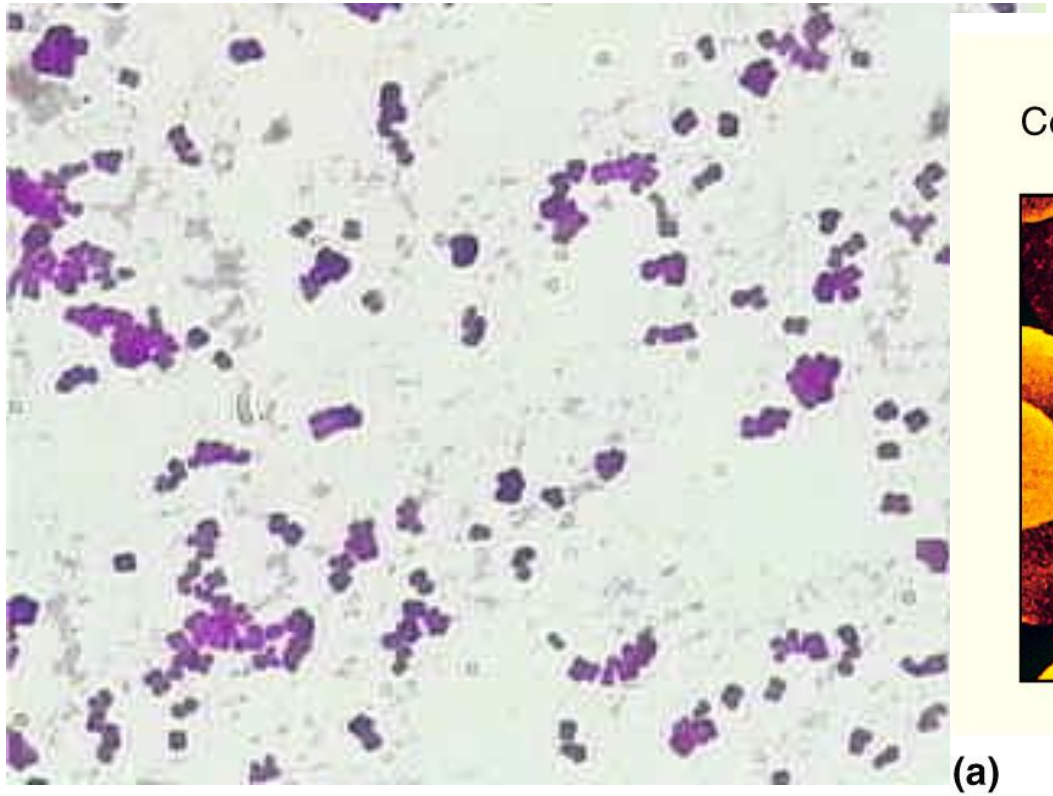
Rod (bacillus)

(b)

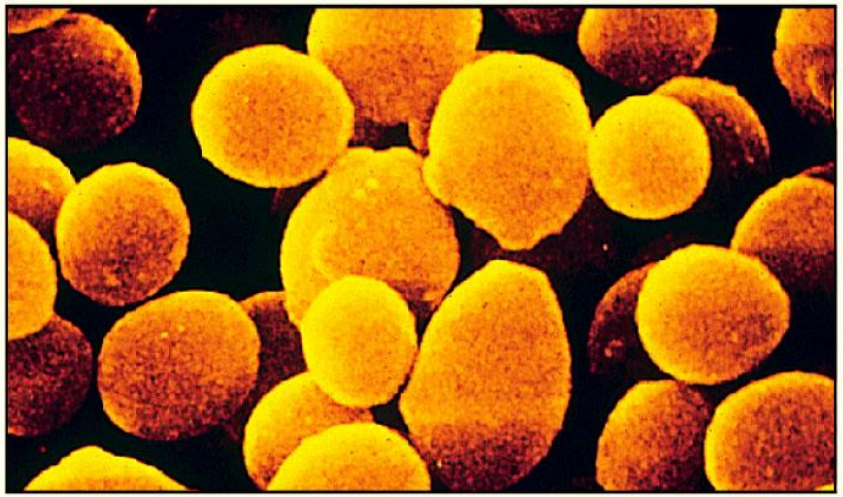
Streptobacillus



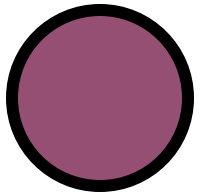
Coccus (pl. cocci)



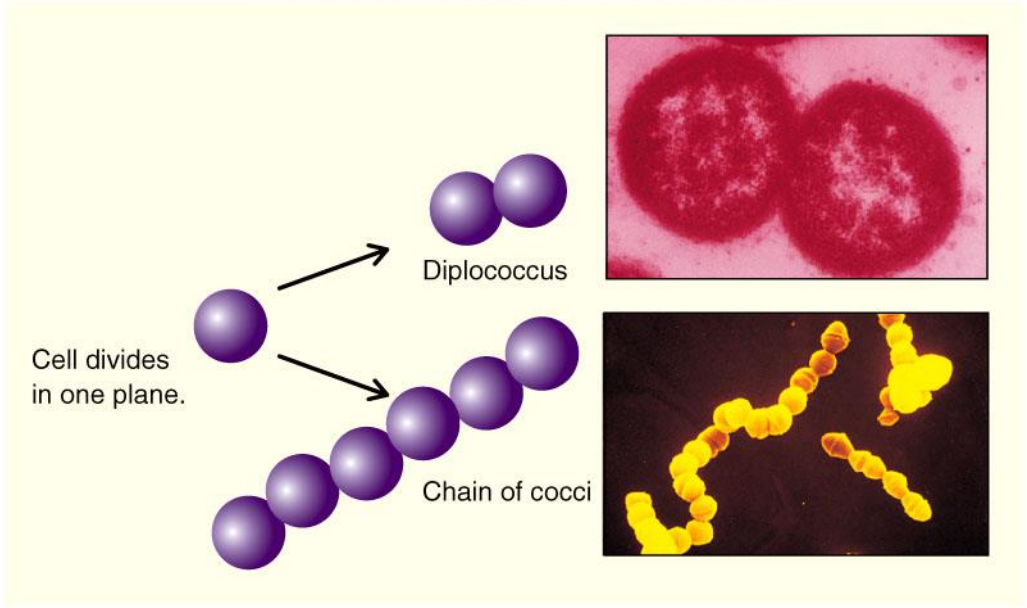
Coccus



(a)

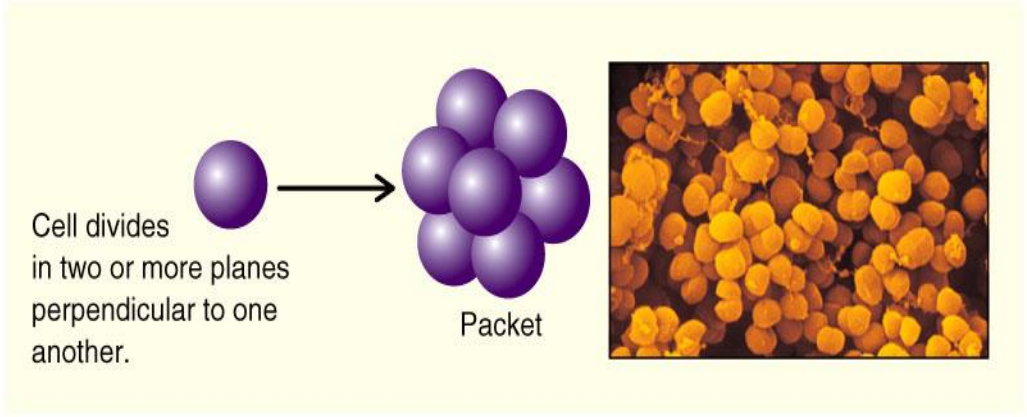


coccus



(a) Chains

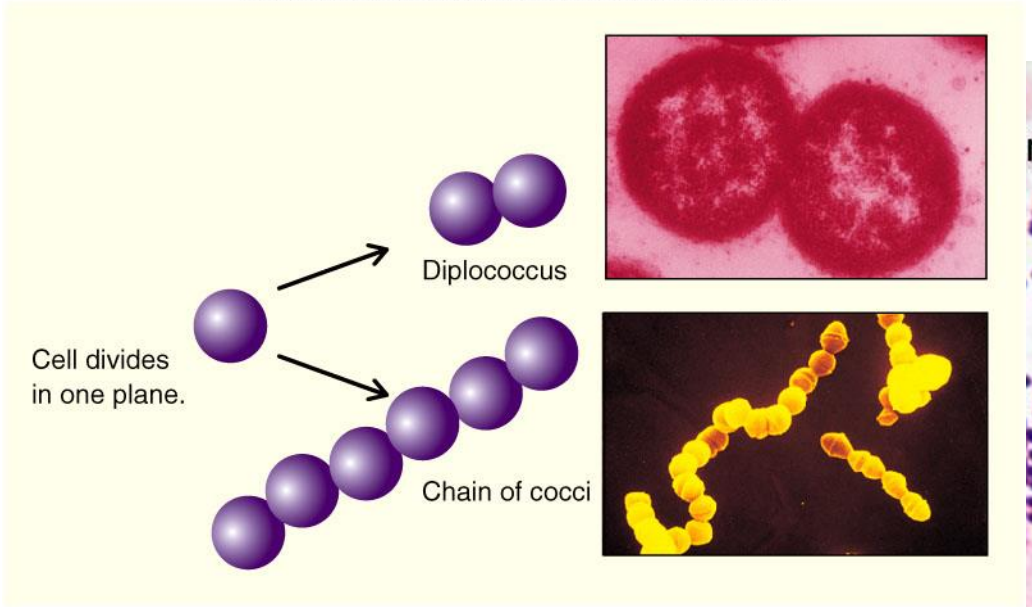
streptococci



(b) Packets

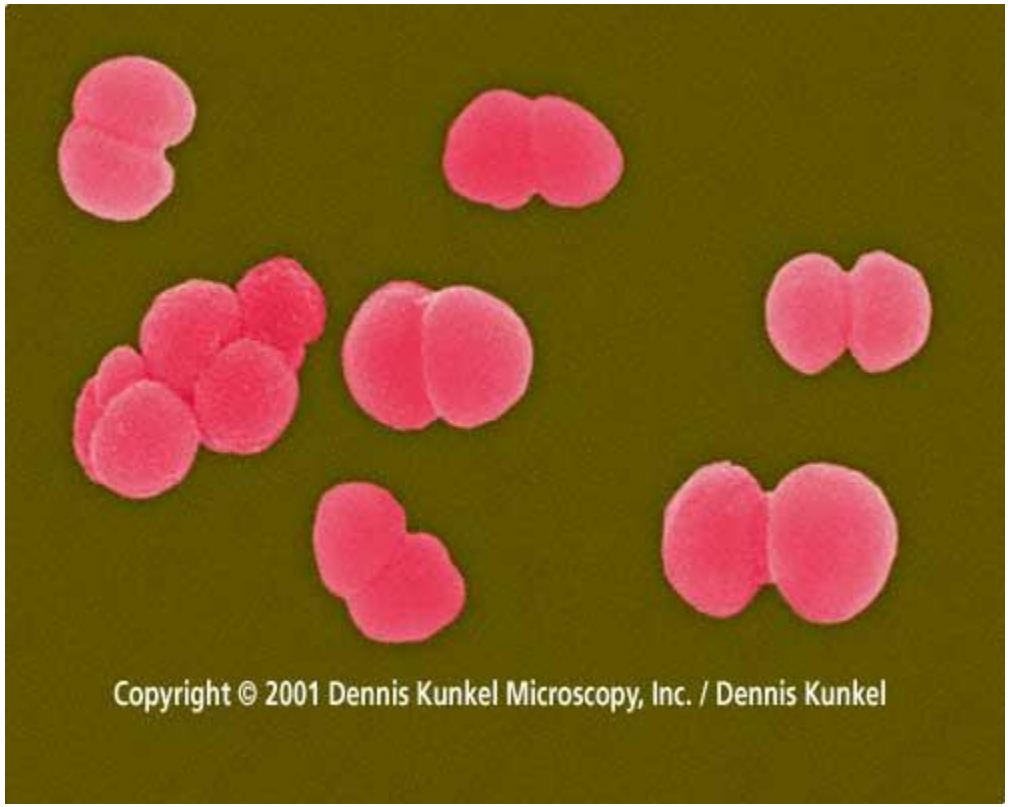
staphylococci

Streptococcus

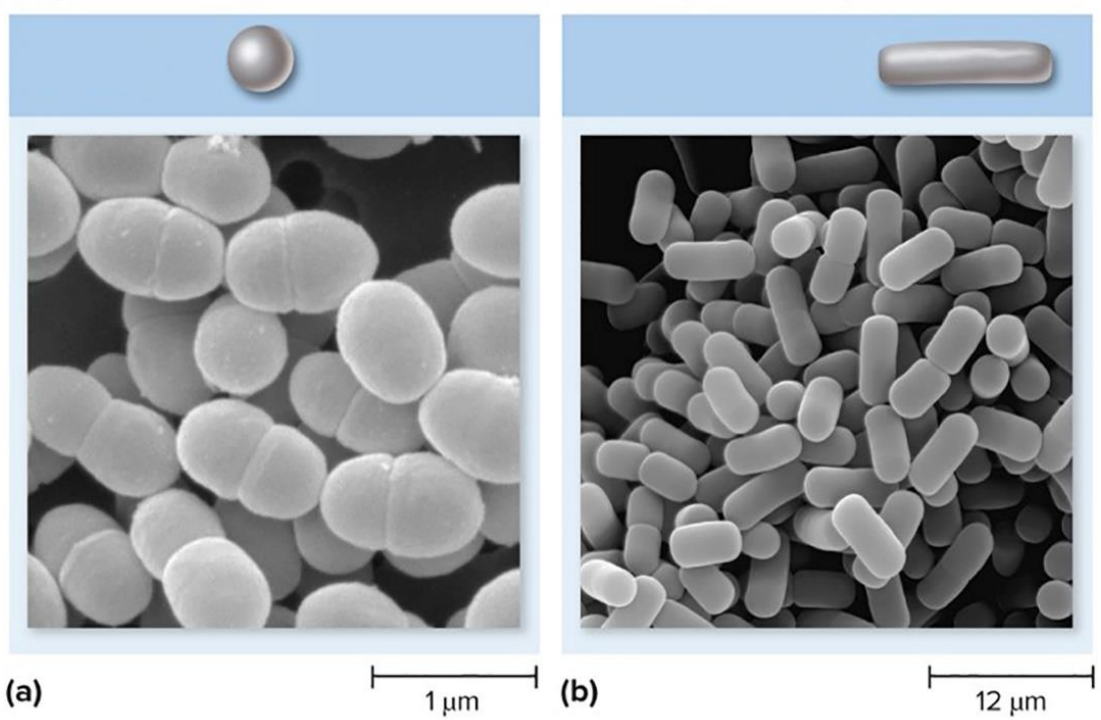


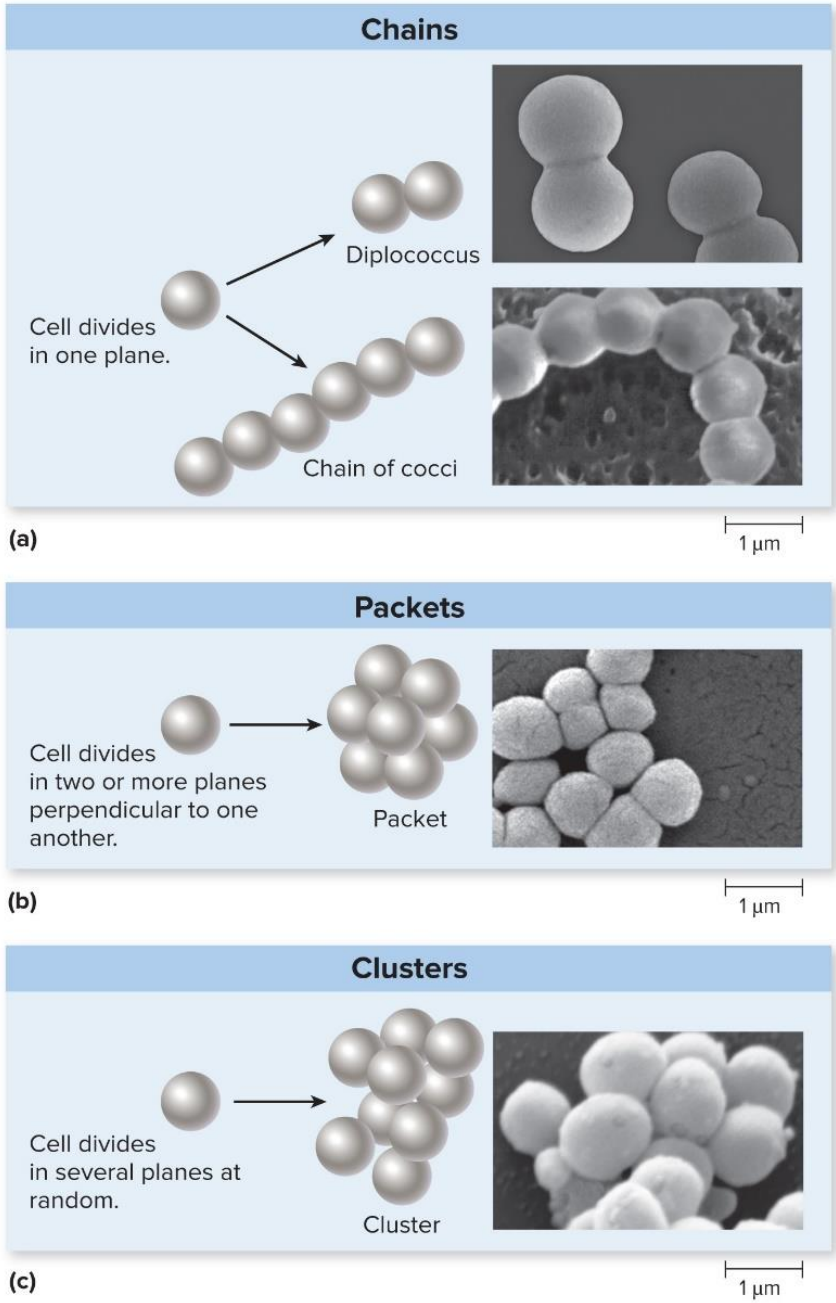
(a) Chains

Diplococcus



Neisseria gonorrhoeae

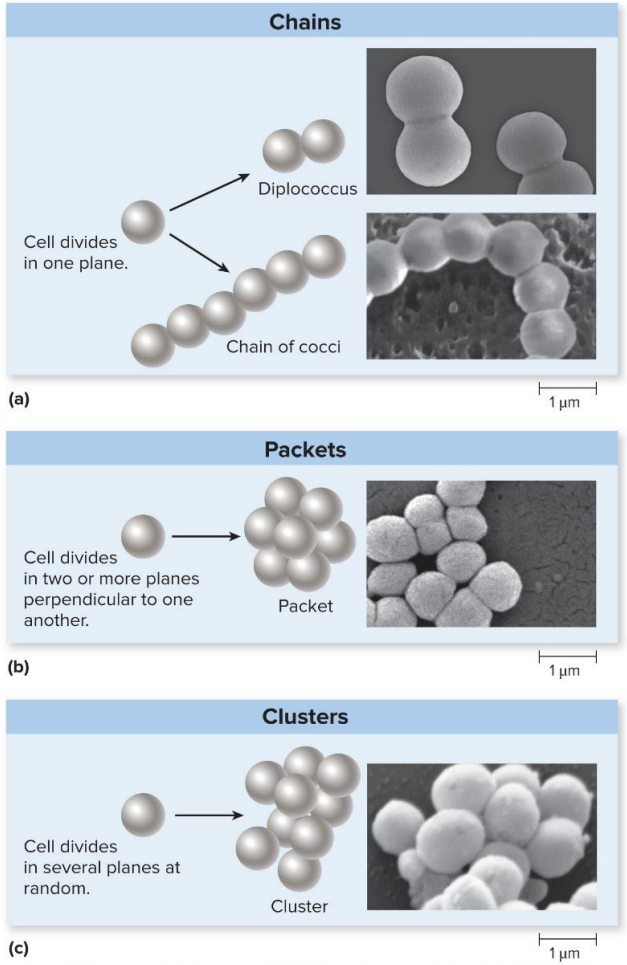

(a)
1 µm
(b)
12 µm

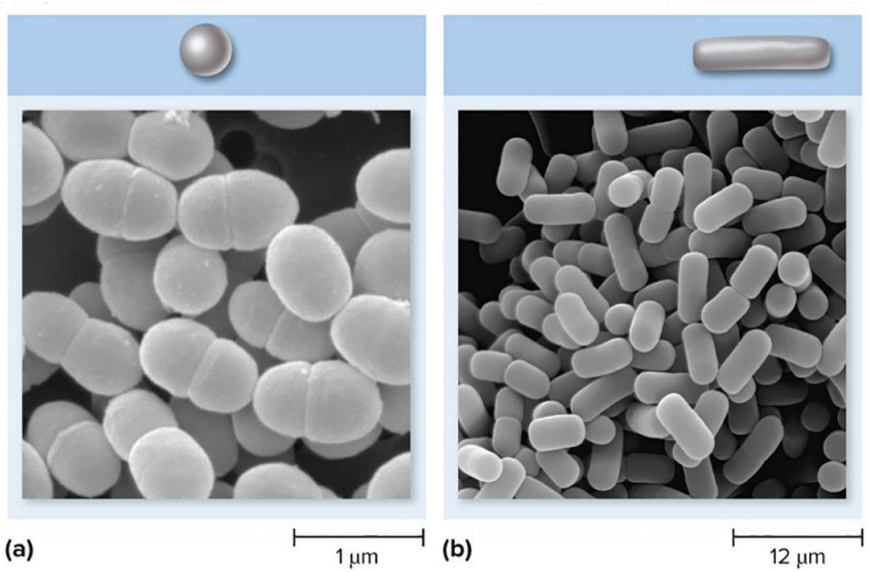

Chains
Diplococcus
Cell divides
in one plane.
Chain of cocci
(a)
1 µm
Packets
Cell divides
in two or more planes
perpendicular to one
another.
Packet
(b)
1 µm
Clusters
Cell divides
in several planes at
random.
Cluster
(c)
1 µm

Prokaryotic Cell Shape & Arrangement

- Two types most common:
  - Coccus: spherical
  - Bacillus: cylindrical/Rod
    - Short rods sometimes called coccobacillus

- Most prokaryotes divide by binary fission
  - Cells often stick together following division to form characteristic groupings





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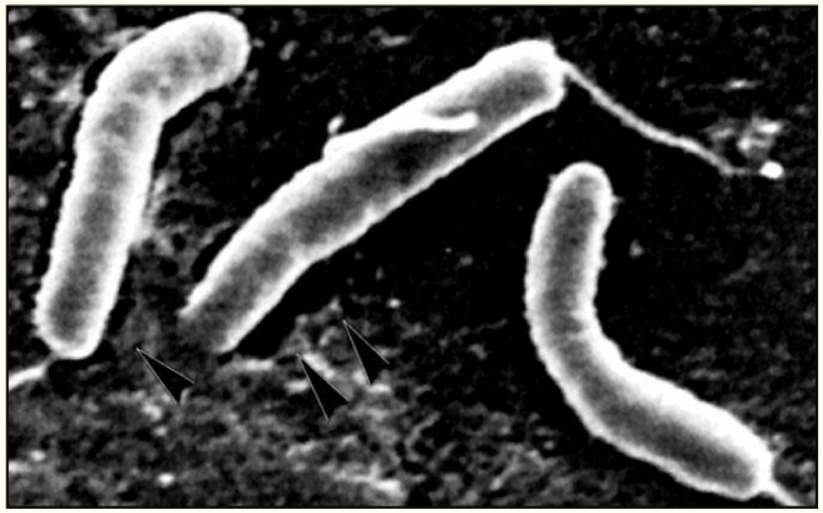
Spiral bacteria

- **Vibrio (pl. Vibrios)**
- **Spirillum (pl. Spirilla)**
- **Spirochete (pl. Spirochetes)**

Spiral bacteria

Vibrio

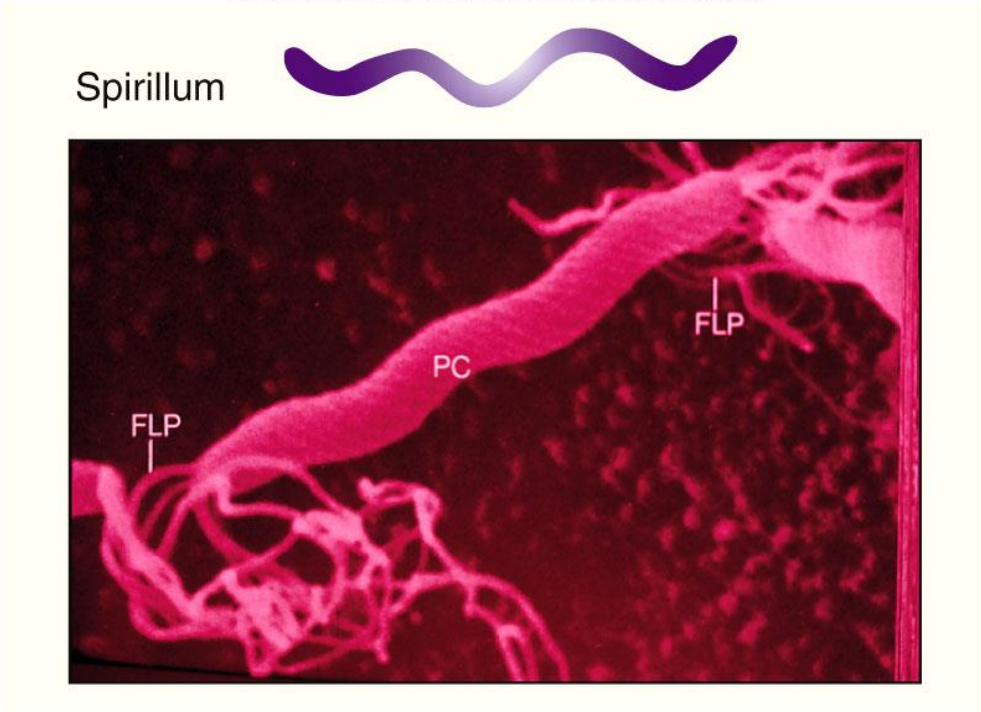
Vibrio



(d)

Spiral bacteria

Spirillum



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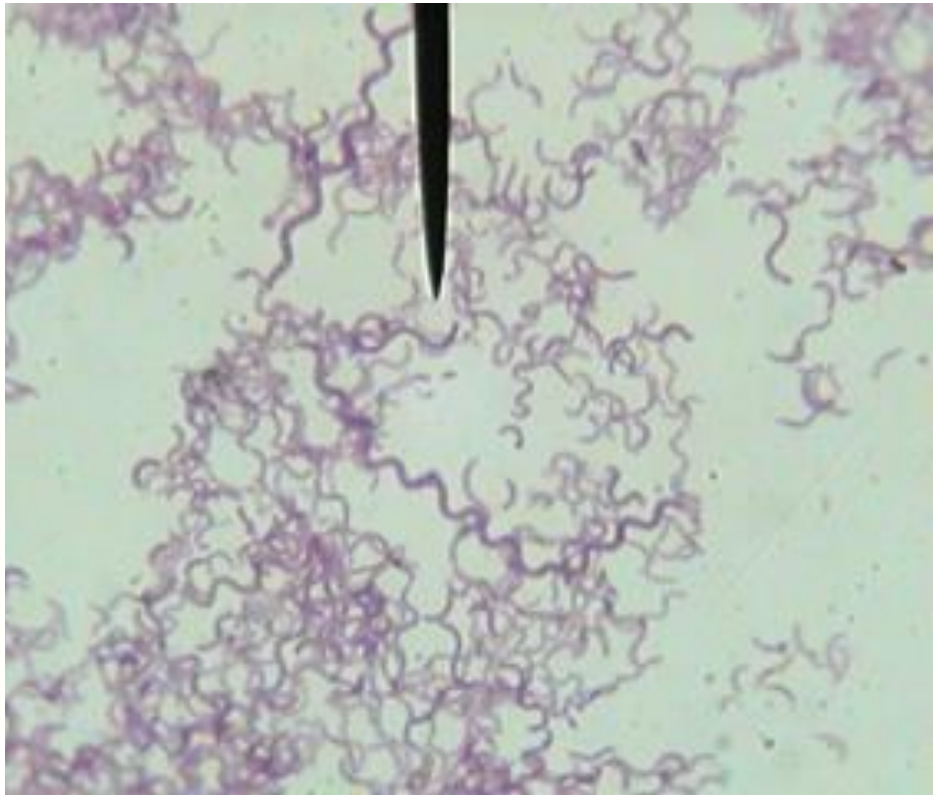
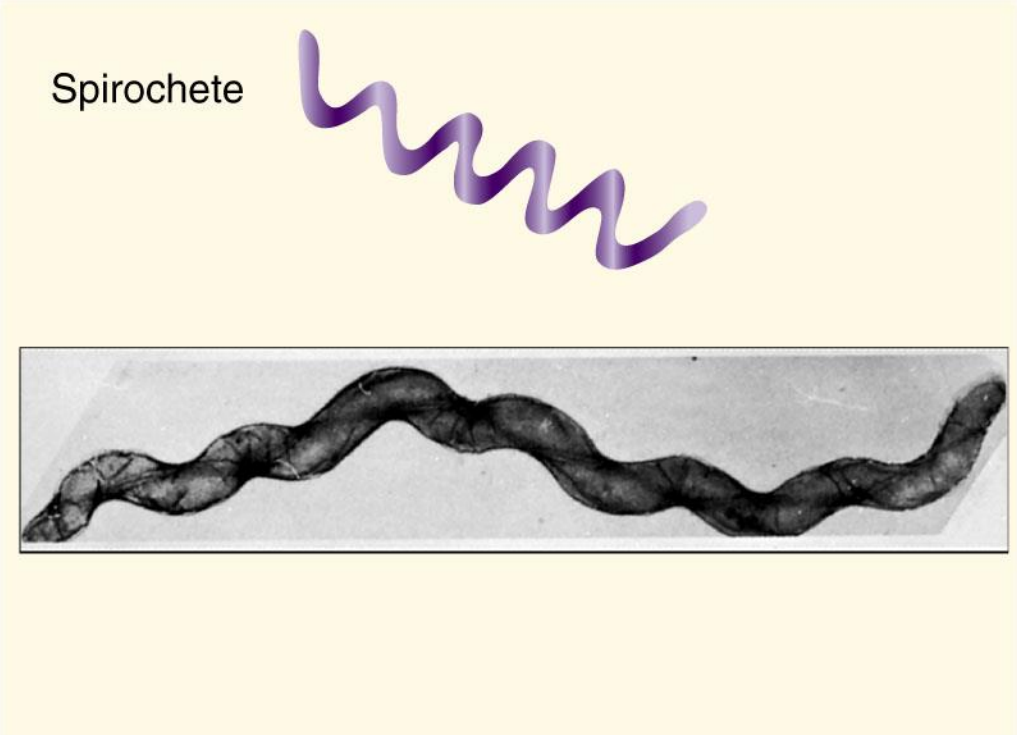
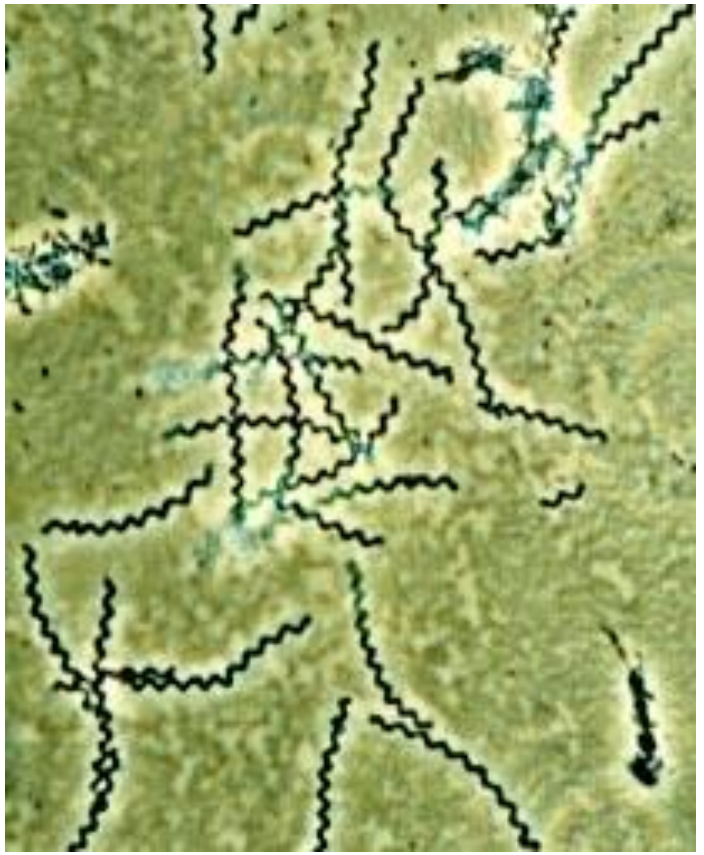


Spiral bacteria

Spirochete



(f)



Pleomorphic

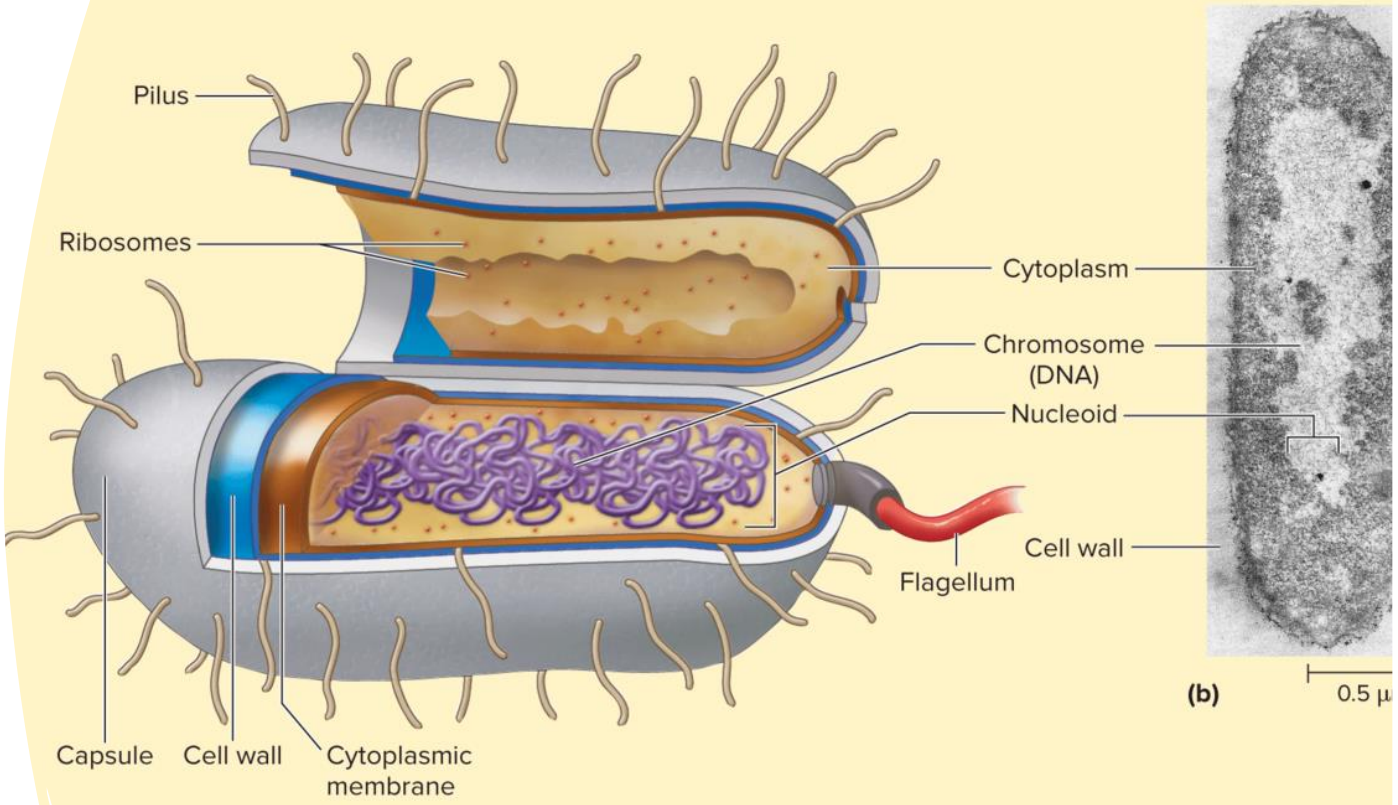
- **vary in their shape**
- **pleo: many**
- **morphic: shape**

Prokaryotic cells

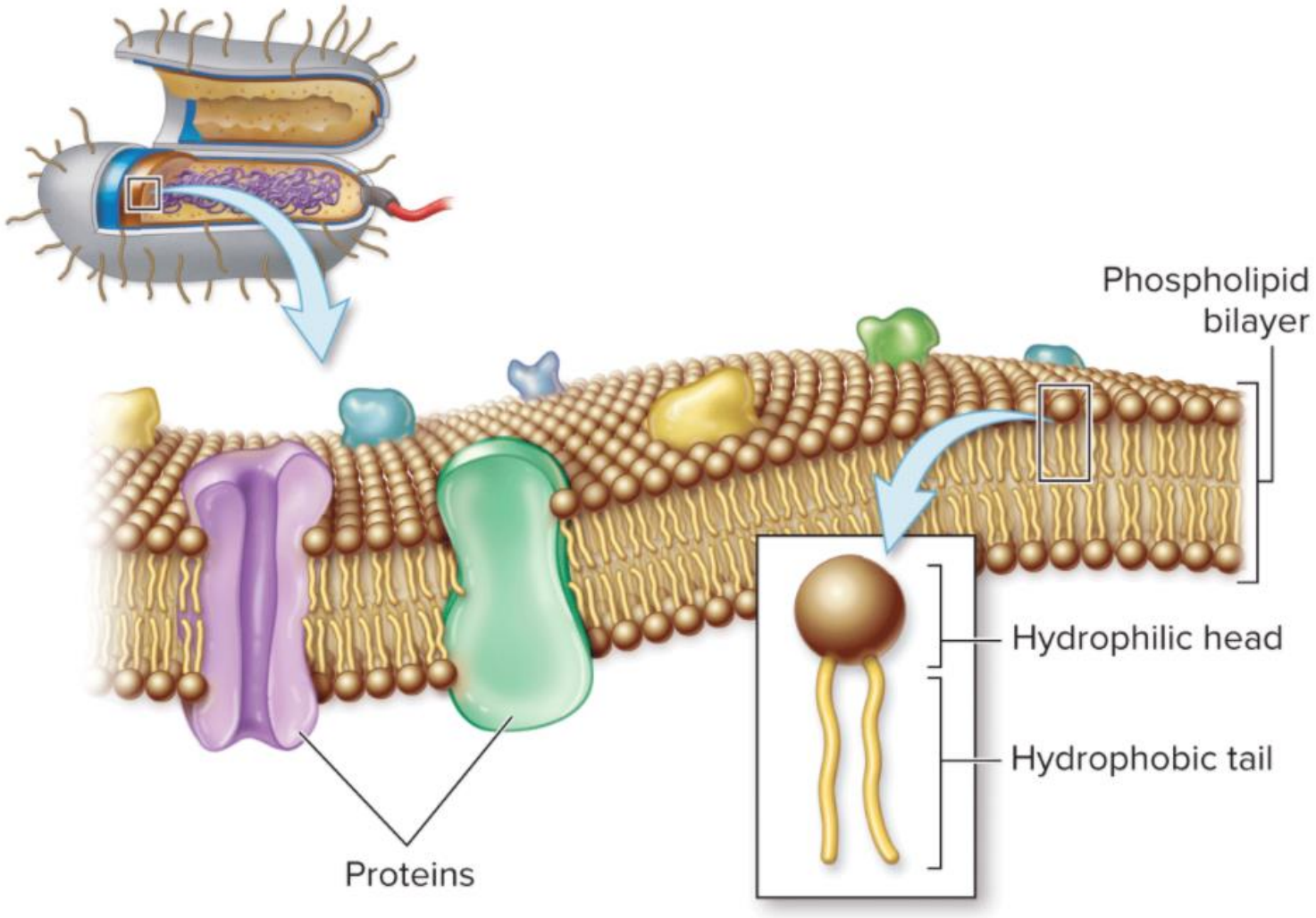
- Cell envelope
- Cytoplasm
- Nucleoid

Cell envelope of prokaryotic cells

- Cytoplasmic membrane
- Cell wall
- Capsule/Slime layer

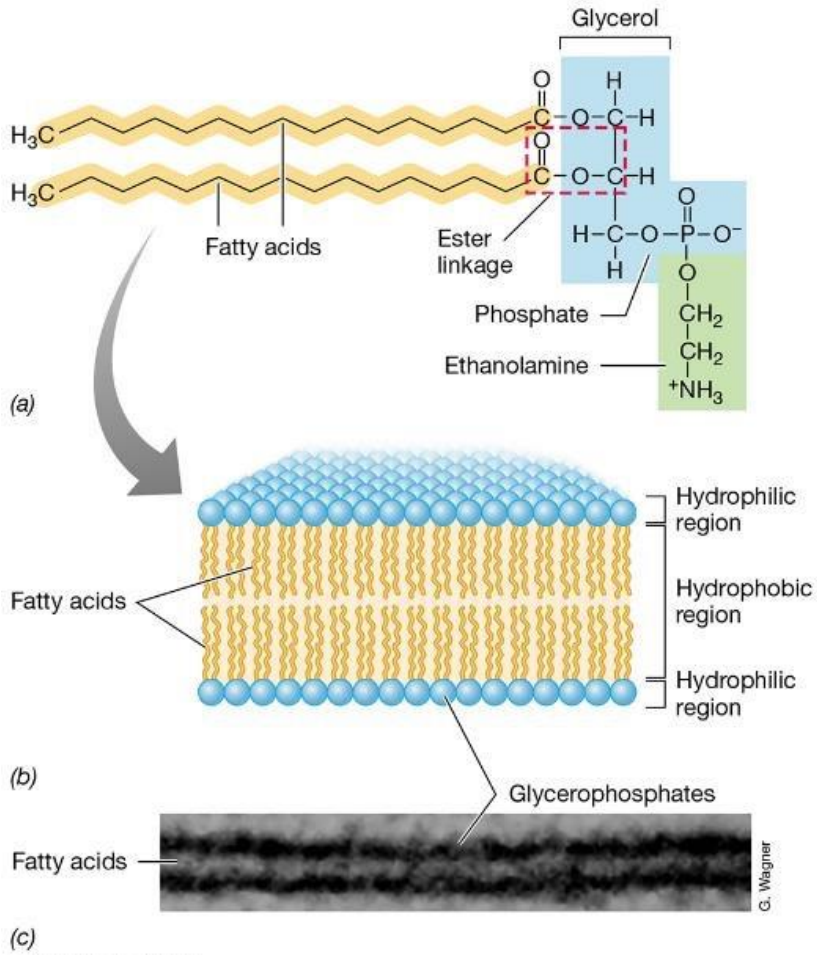


Cytoplasmic membrane or plasma membrane



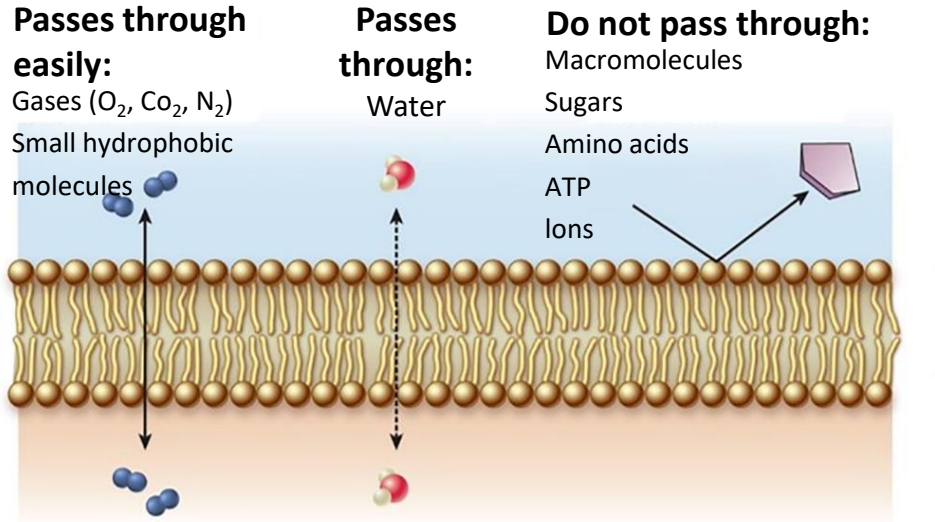
Cytoplasmic Membrane

- Lipid bilayer
- Hydrophobic, fatty acids
  - Middle of membrane
- Hydrophilic, Glycerol bonded to phosphate
  - Ester bond
  - Faces inside & out cell
- Integral & Peripheral proteins



Permeability of Cytoplasmic Membrane

- Cytoplasmic membrane is <u>selectively permeable</u>
  - O_2, CO_2, N_2, small hydrophobic molecules, and water pass freely
  - Some cells facilitate water passage with <u>aquaporins</u>
  - Other molecules must be moved across membrane via transport systems
- Cell wall prevents cell from bursting



3.5 The Cell Wall of Prokaryotic Cells

Cell Wall

- Maintain **cell shape**!!!
- **Protect** against **osmotic shock**
  - Hypotonic solution: cell will not lyse
  - Hypertonic solution: cell will not shrivel up

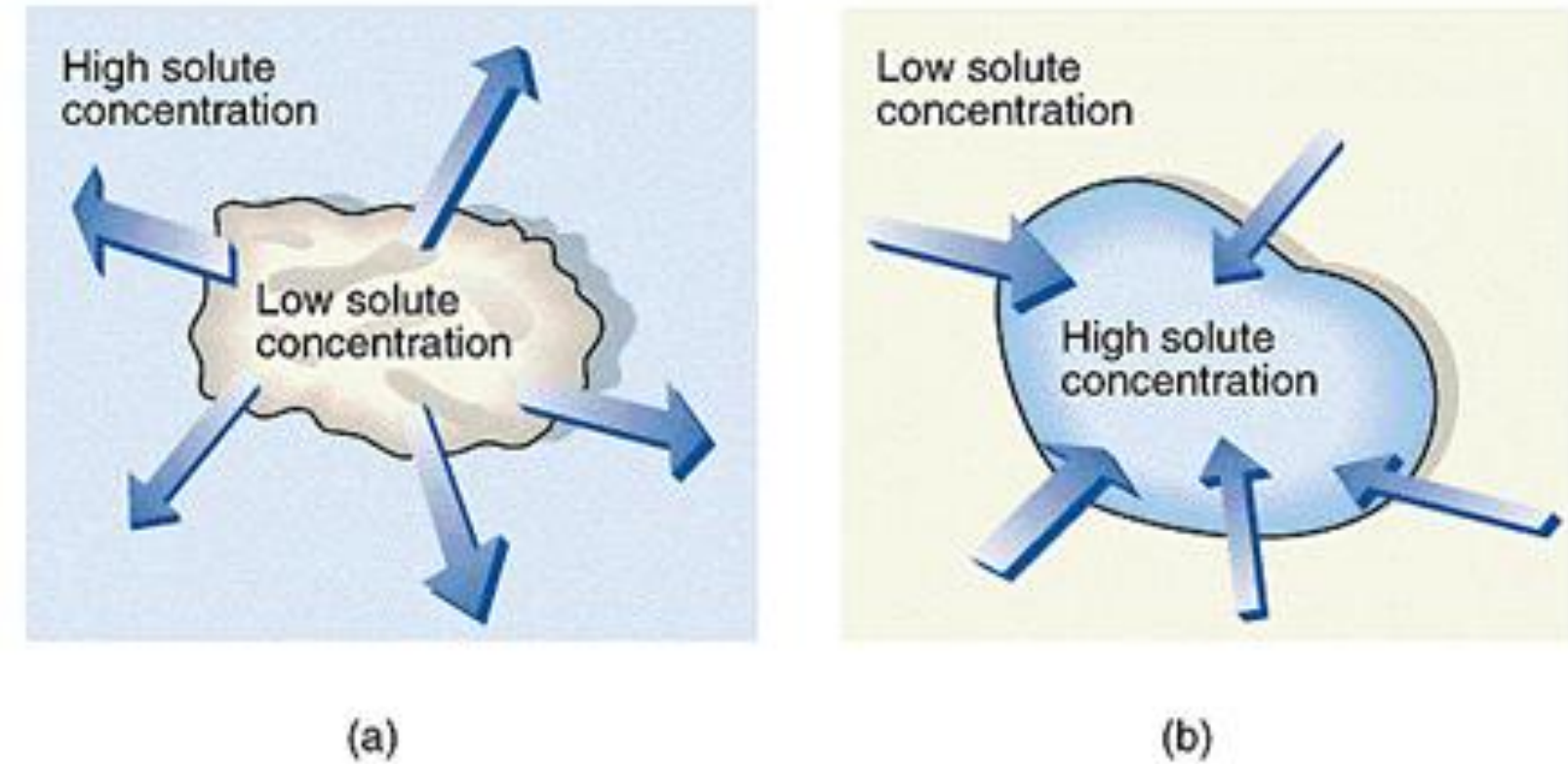
    Plasmolysis!!

I. Differentiate between **species**

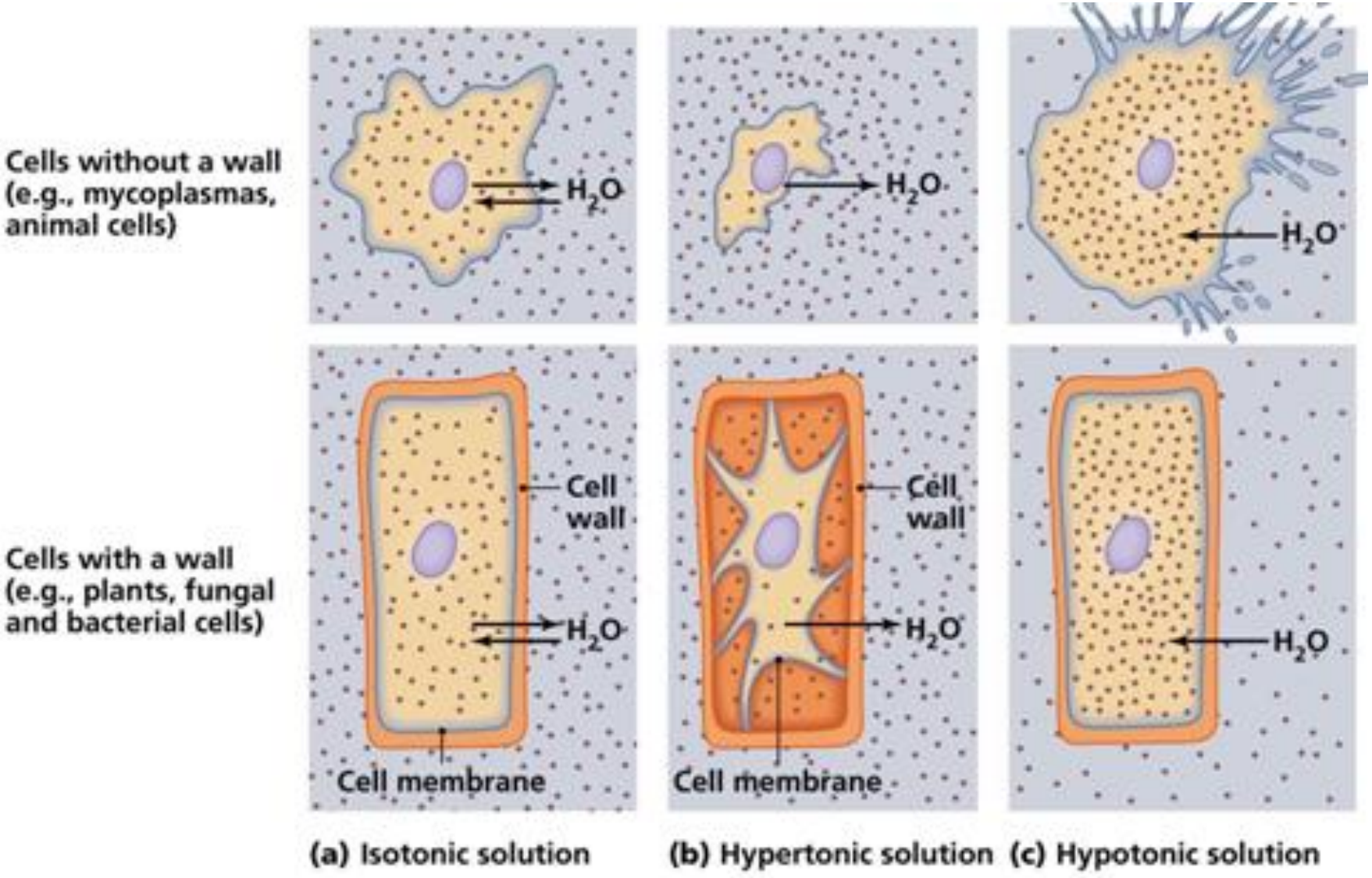
II. Affect **pathogenicity**

III. Anchor the flagella

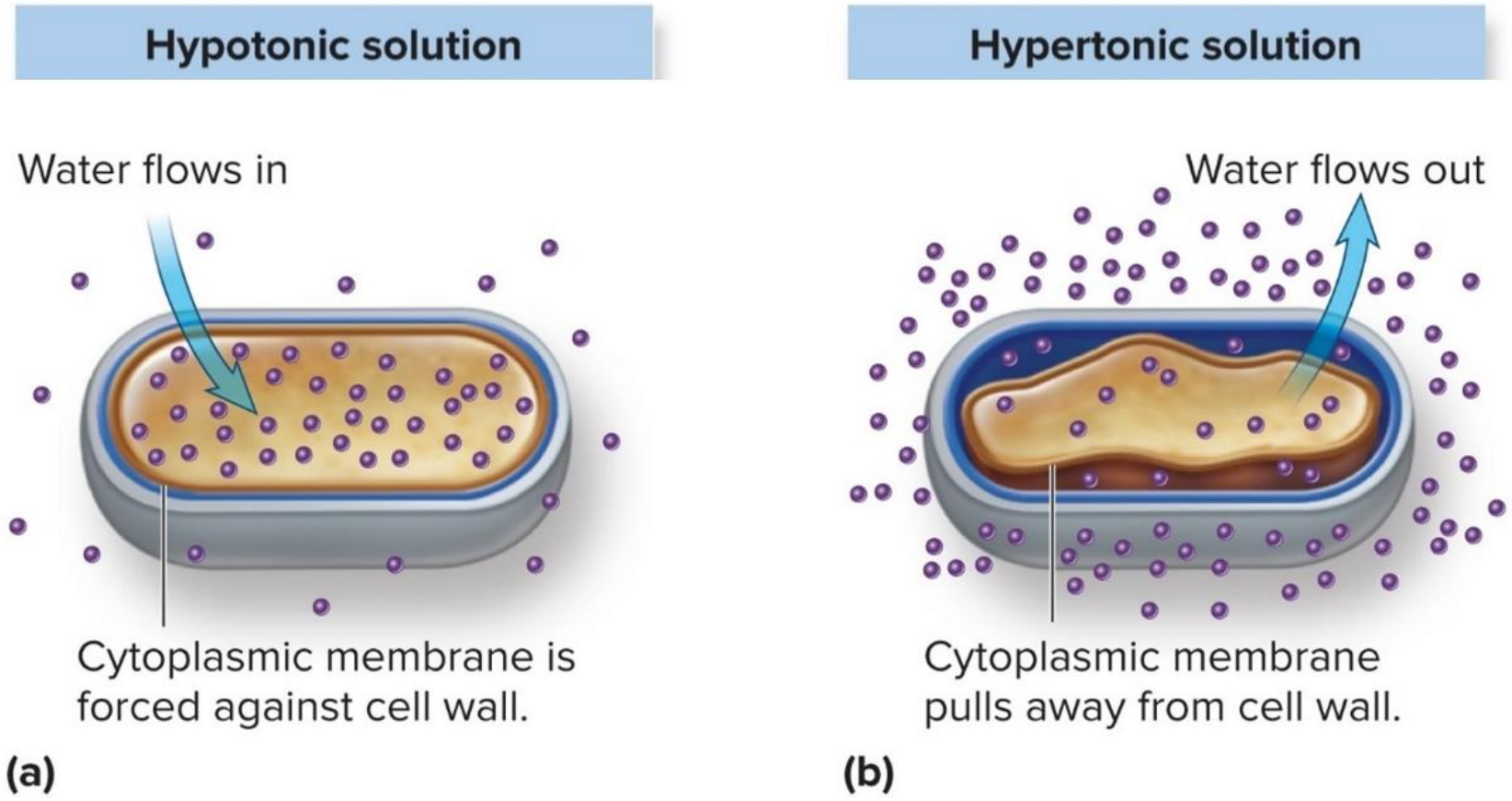
Osmosis


High solute concentration
Low solute concentration
(a)
Low solute concentration
High solute concentration
(b)

Osmosis


Cells without a wall
(e.g., mycoplasmas, animal cells)
Cells with a wall
(e.g., plants, fungal and bacterial cells)
H_2O
Cell wall
Cell membrane
(a) Isotonic solution
(b) Hypertonic solution
(c) Hypotonic solution

Osmosis

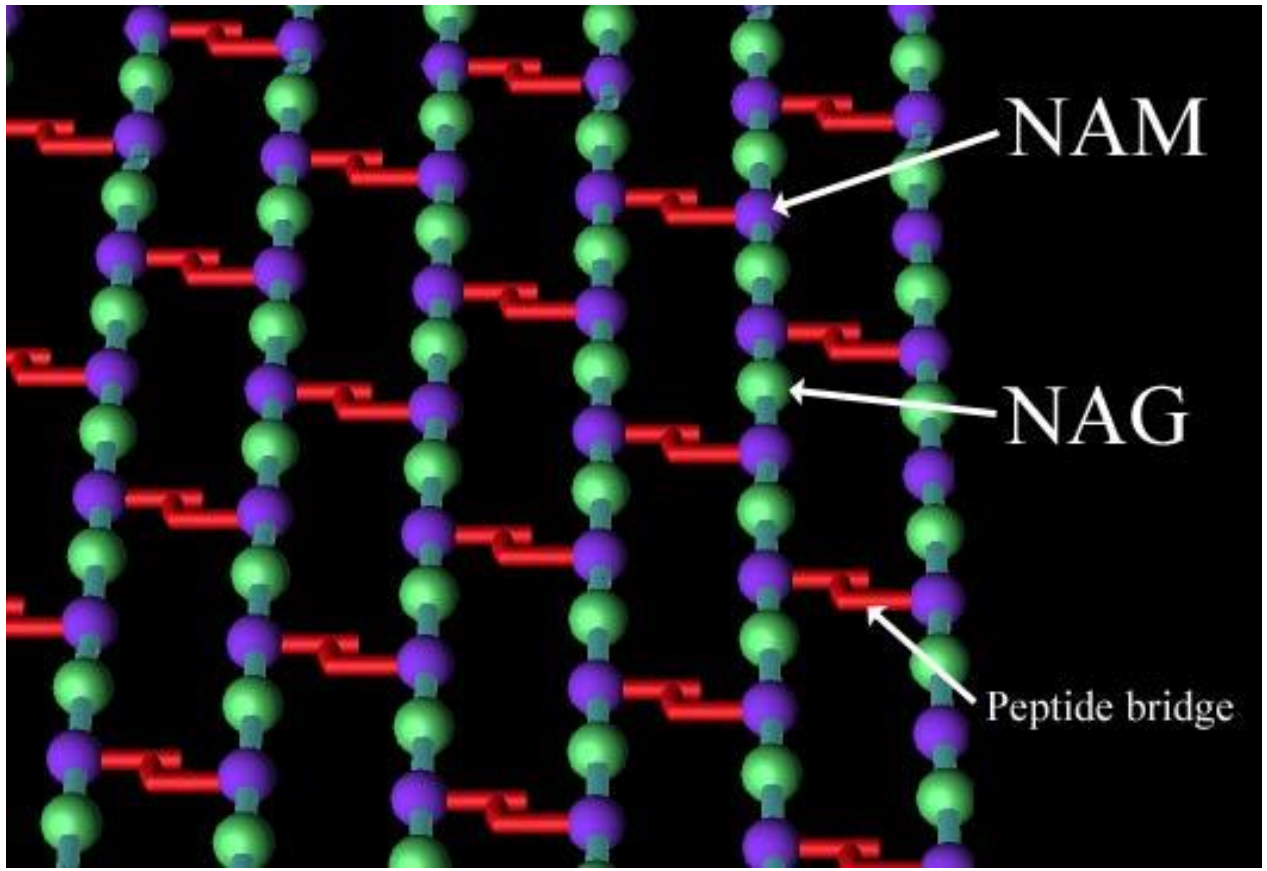

Hypotonic solution
Water flows in
Cytoplasmic membrane is forced against cell wall.
(a)
Hypertonic solution
Water flows out
Cytoplasmic membrane pulls away from cell wall.
(b)

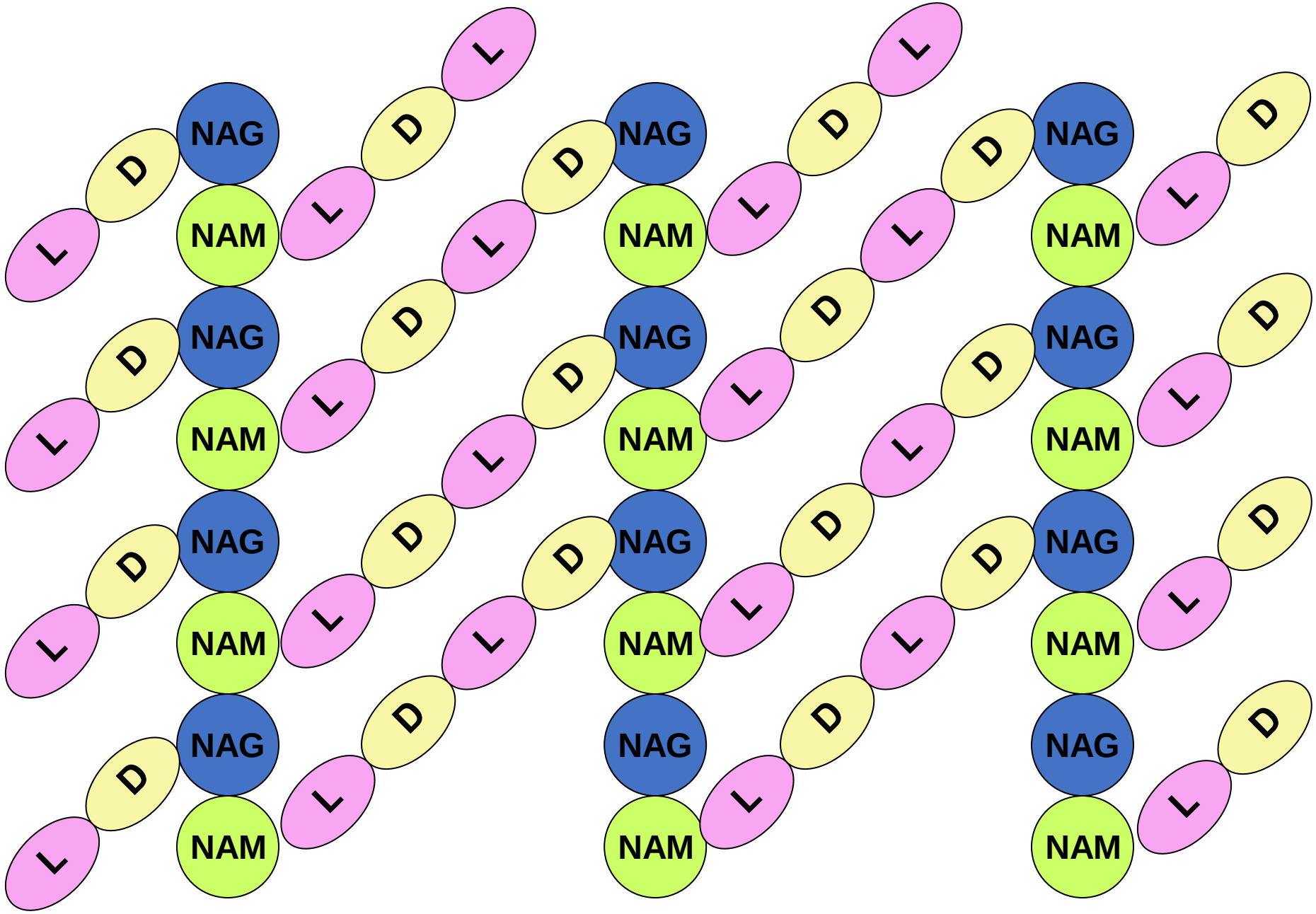
Bacterial Cell Walls - Peptidoglycan

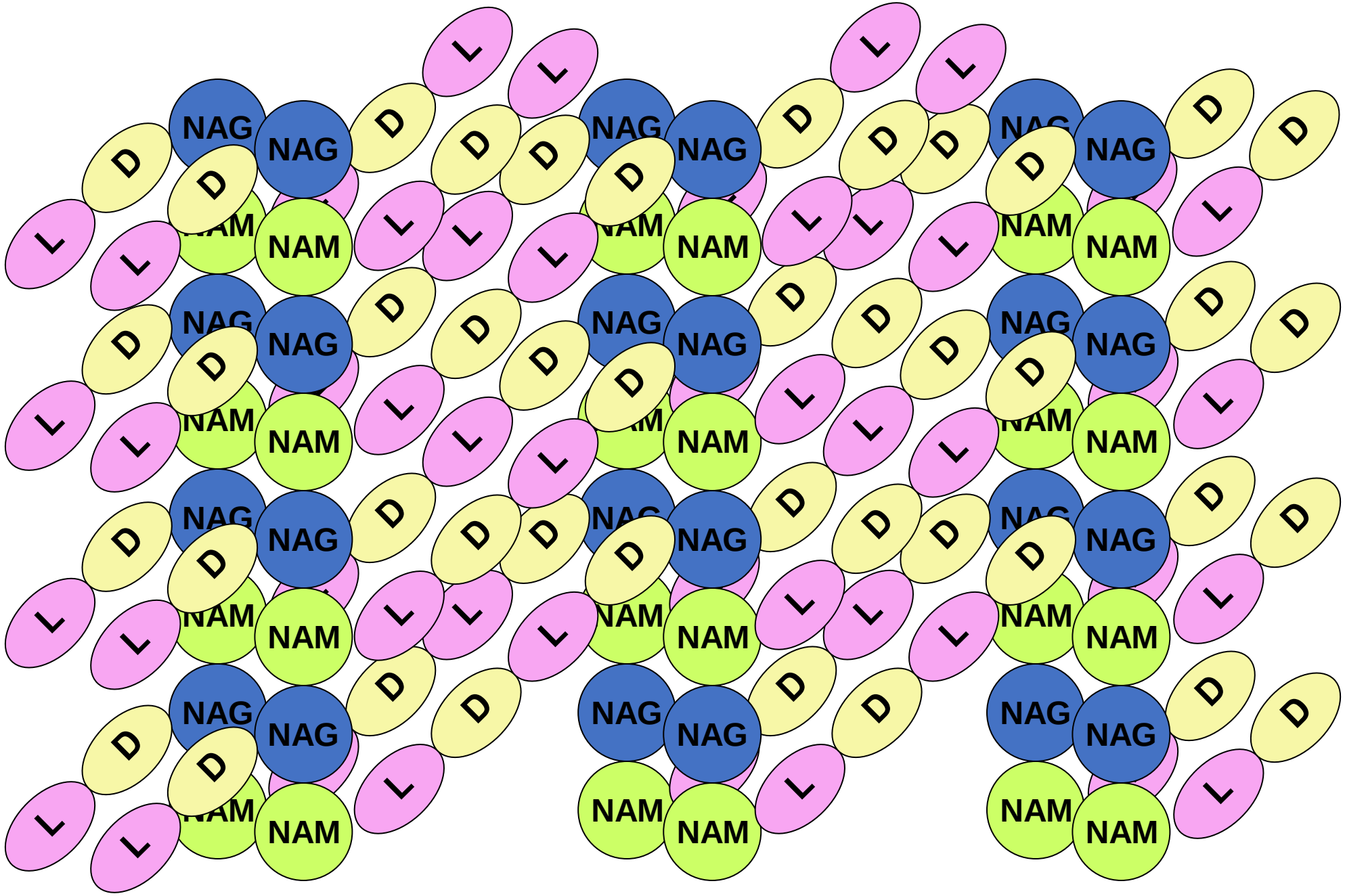
- Major component of **bacterial cell wall**!!

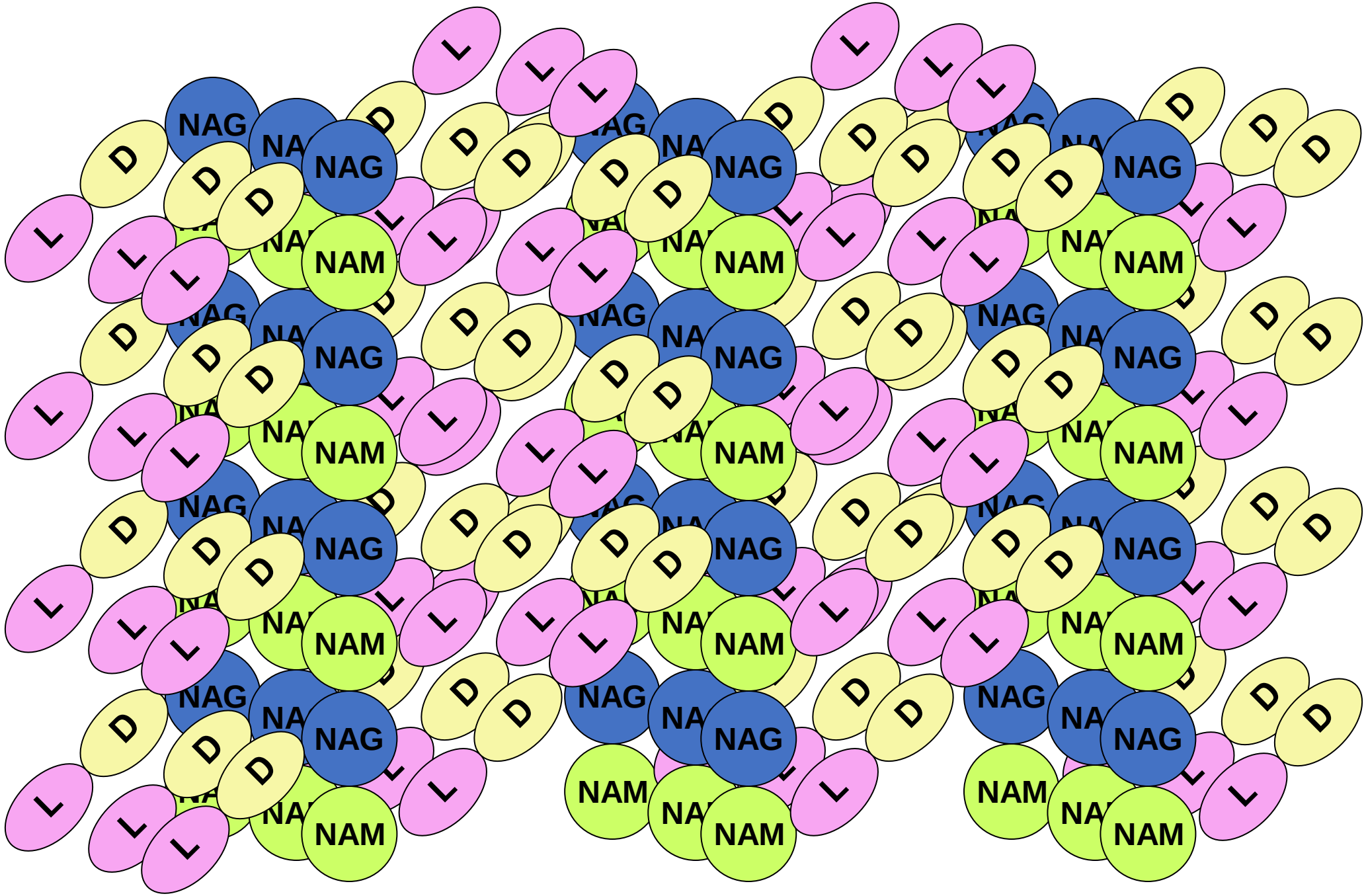
- Composed of repeating sugars(disaccharides)
  - **NAG** = N-Acetyl glucosamine
  - **NAM**= N-Acetylmuramic acid

- The peptidoglycan chains are held together by a short protein composed of only **4 amino acids**


NAM
NAG
Peptide bridge


NAG
NAM
D
L

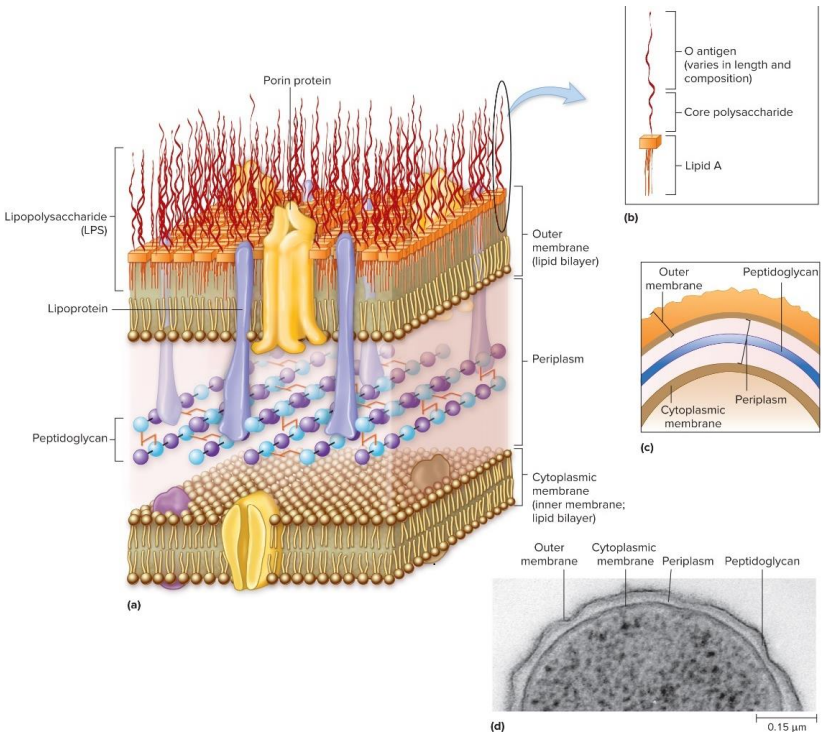

NAG
NAM
D
L


NAG
NAM
D
L

Bacterial Cell Walls - Peptidoglycan

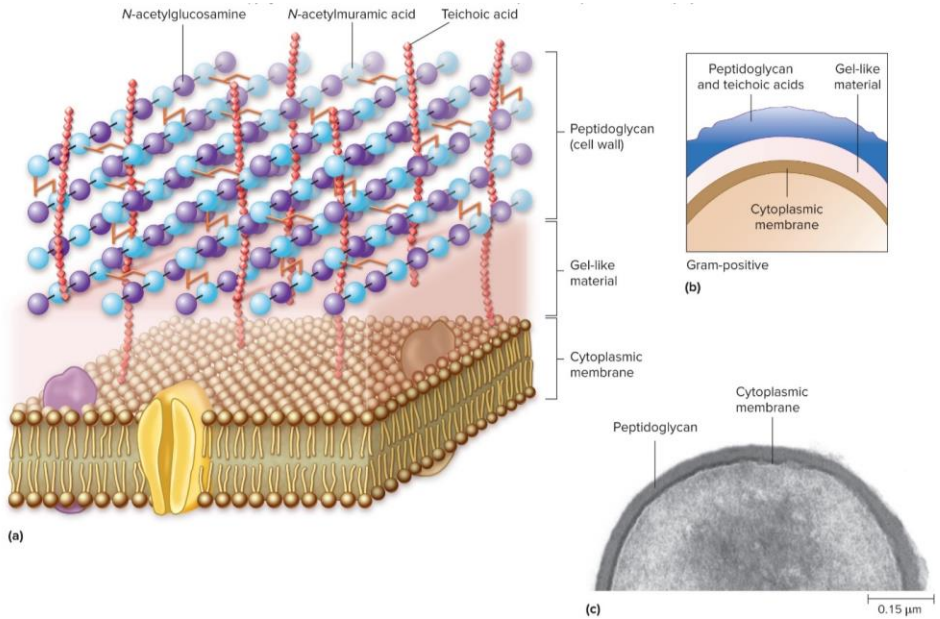
Gram Negative

- at least two layers: LPS and peptidoglycan

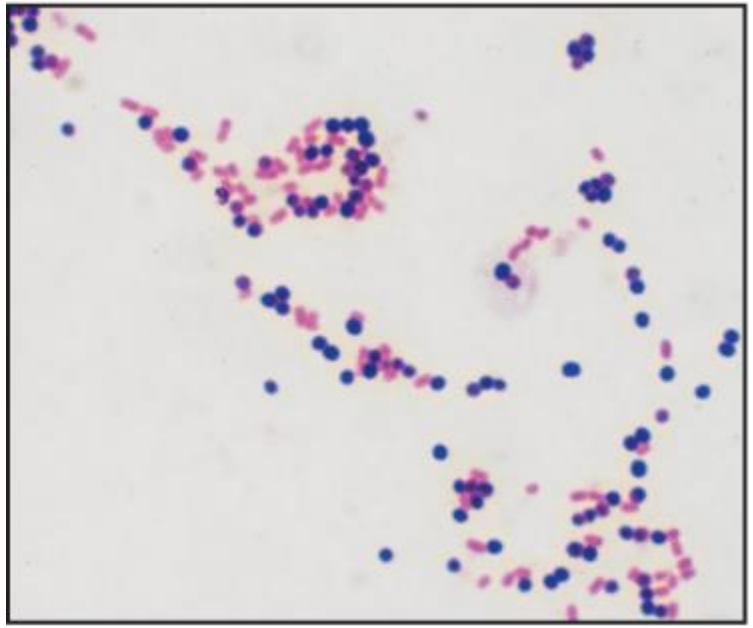


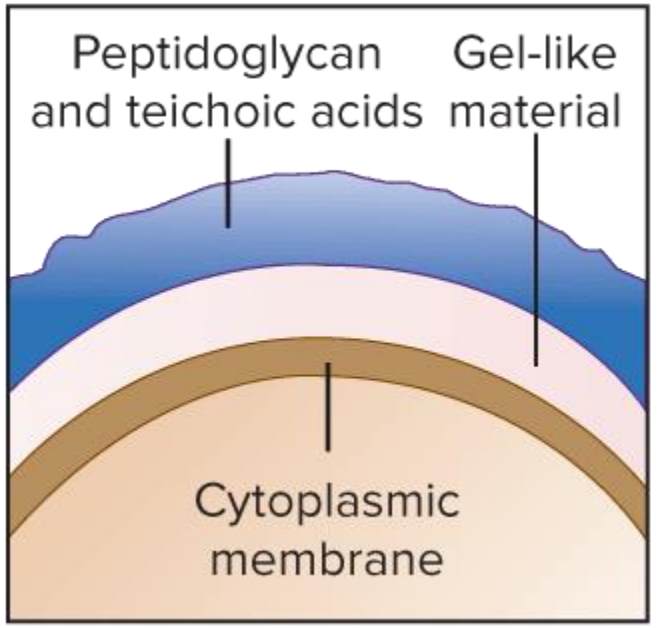
Gram Positive

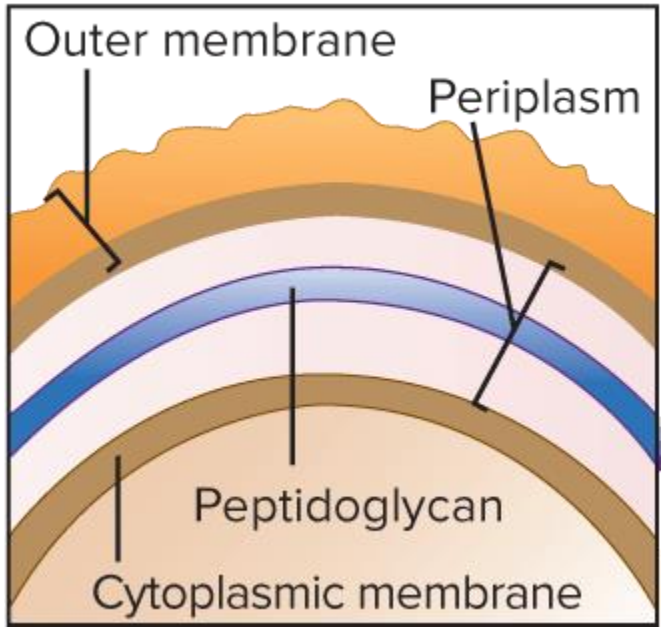
- thicker, primarily one layer of peptidoglycan



3.5 The Cell Wall of Prokaryotic Cells







Cell Wall

Gram positive bacteria

vs.

Gram negative bacteria

Why is this important?

- **Penicillin** (and other antibiotics) interfere with the peptide cross-linking!!
- Differences in cell walls are the basis for the **gram stain**

Gram Negative Cell Wall

- **Periplasmic Space**
- High Concentration of LPS (Lipopolysaccharides) in Outer Membrane

LPS (Lipopolysaccharides)

Lipid A component is TOXIC

Other things about the Gram (-) outer membrane!!!

- The outer membrane of gram (-) bacteria acts as a protective barrier. It is a major permeability barrier.

- Prevents many drugs like penicillin from getting to the thin peptidoglycan layer.

- Gram (-) bacteria are less sensitive to many medications.

Effects of Penicillin and Lysozyme

- **Penicillin**
  - Interferes with the **synthesis of peptide linkages** between the peptidoglycan chains
  - More effective against gram positive cells
  - **Most Gram Negative** are protected by outer membrane (EXCEPTIONS TO THIS!!)
- **Lysozyme**
  - Found in saliva, tears, mucus
  - Breaks the bond between NAG-NAM
  - Only affects gram positives!!!

	Gram-Positive	Gram-Negative
Color of Gram-Stained Cell	Purple	Pink
Representative Genera	*Bacillus, Staphylococcus, Streptococcus*	*Escherichia, Neisseria, Pseudomonas*
Distinguishing Structures/Components		
Peptidoglycan	Thick layer	Thin layer
Teichoic acids	Present	Absent
Outer membrane	Absent	Present
Lipopolysaccharide (endotoxin)	Absent	Present
Porin proteins	Absent (unnecessary because there is no outer membrane)	Present; allow molecules to pass through outer membrane
General Characteristics		
Sensitivity to penicillin	Generally more susceptible (with notable exceptions)	Generally less susceptible (with notable exceptions)
Sensitivity to lysozyme	Yes	No

Gram Stain

Flood smear with primary stain

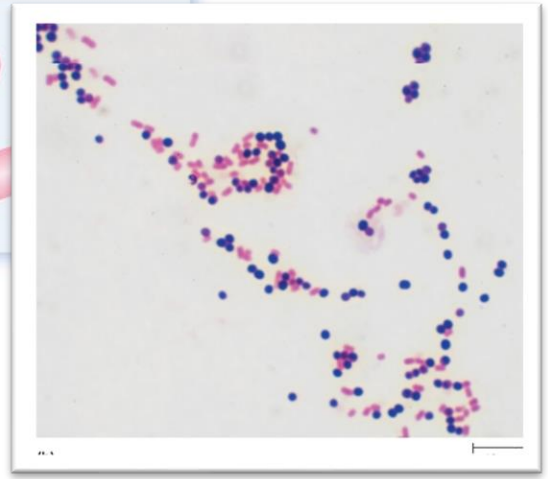
Rinse and flood with iodine, mordant that stabilizes the dye in the cell

Rinse and briefly add alcohol a decolorizing agent, to remove dye complex from Gram-negative cells

Rinse and flood smear with counterstain that adds a different color to Gram-negative cells

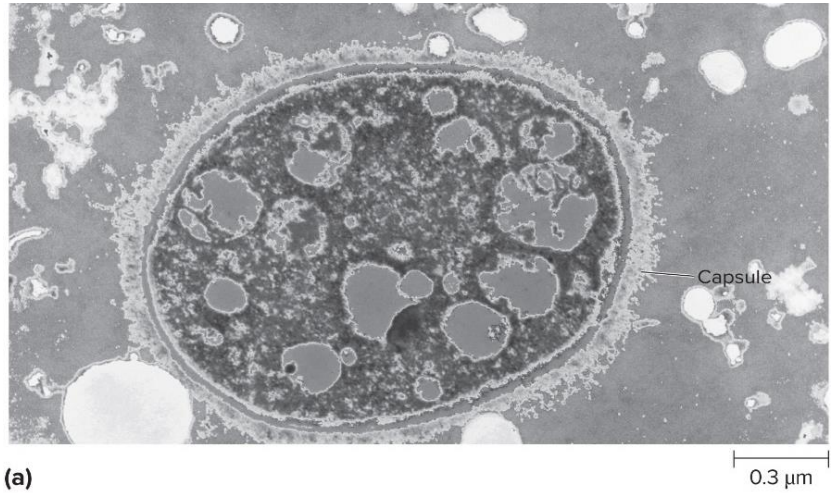
Steps in Staining	State of Bacteria	Appearance
1 Crystal violet (primary stain)	Cells stain purple.	
2 Iodine (mordant)	Cells remain purple.	
3 Alcohol (decolorizer)	Gram-positive cells remain purple; Gram-negative cells become colorless.	
4 Safranin (counterstain)	Gram-positive cells remain purple; Gram-negative cells appear pink.	

(a)

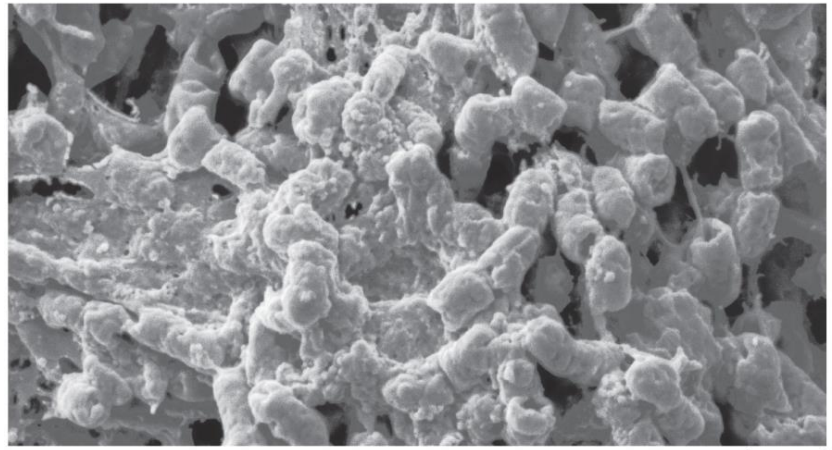


Bacterial Cell Surface - Glycocalyx

- **Gel-like layer** outside the cell wall that either protects the cell or allows it to attach to a surface
- **Capsule**
  - Composed of organized repeating units of organic chemicals
  - Firmly attached to cell surface
  - May prevent bacteria from being recognized by host
- **Slime layer**
  - Loosely attached to cell surface
  - Water-soluble
  - Sticky layer allows prokaryotes to attach to surfaces



(a)



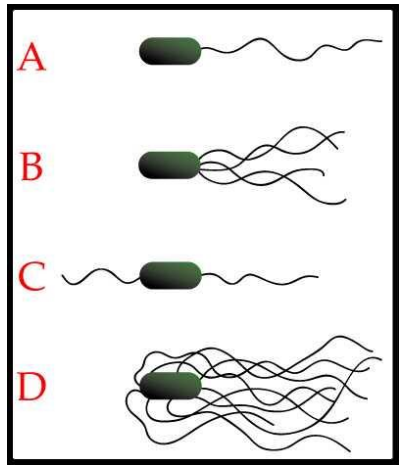
(b)

Cell attachments

Flagella (flagellum)

Fimbriae (fimbria)/ Pili (pilus)

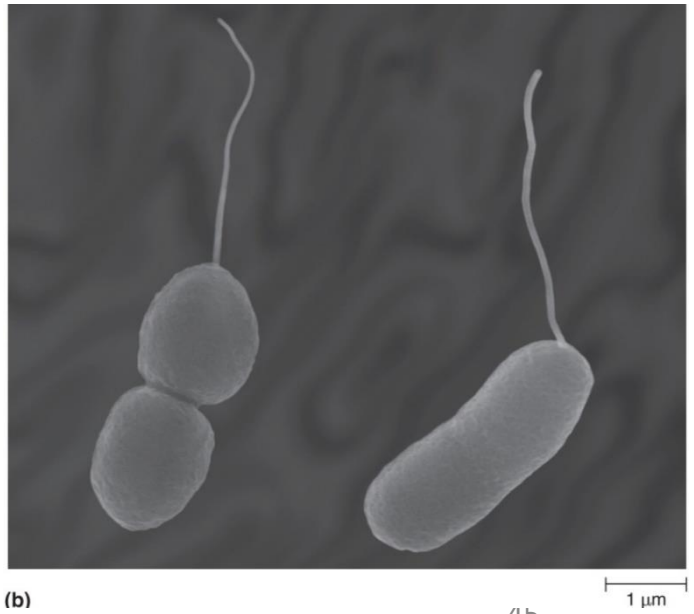
Flagella & Archaella



- Structure that assists in swimming in Bacteria and Archaea
  - long, thin appendages (15–20 nm wide)
  - Archaella are half the diameter of bacterial flagella (10–13 nm)
- arrangements:
  - **monotrichous/polar:** one flagella
  - **Amphitrichous:** two or more
  - **Lophotrichous:** one at both ends
  - **Peritrichous:** all around the periphery

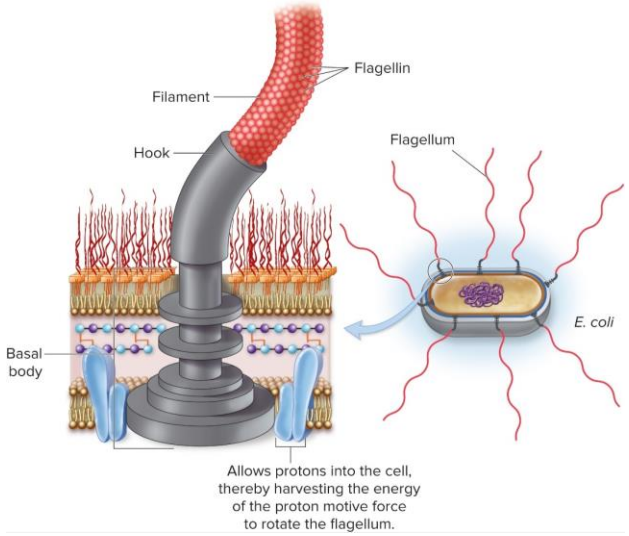
Peritrichous

Monotrichous (polar)



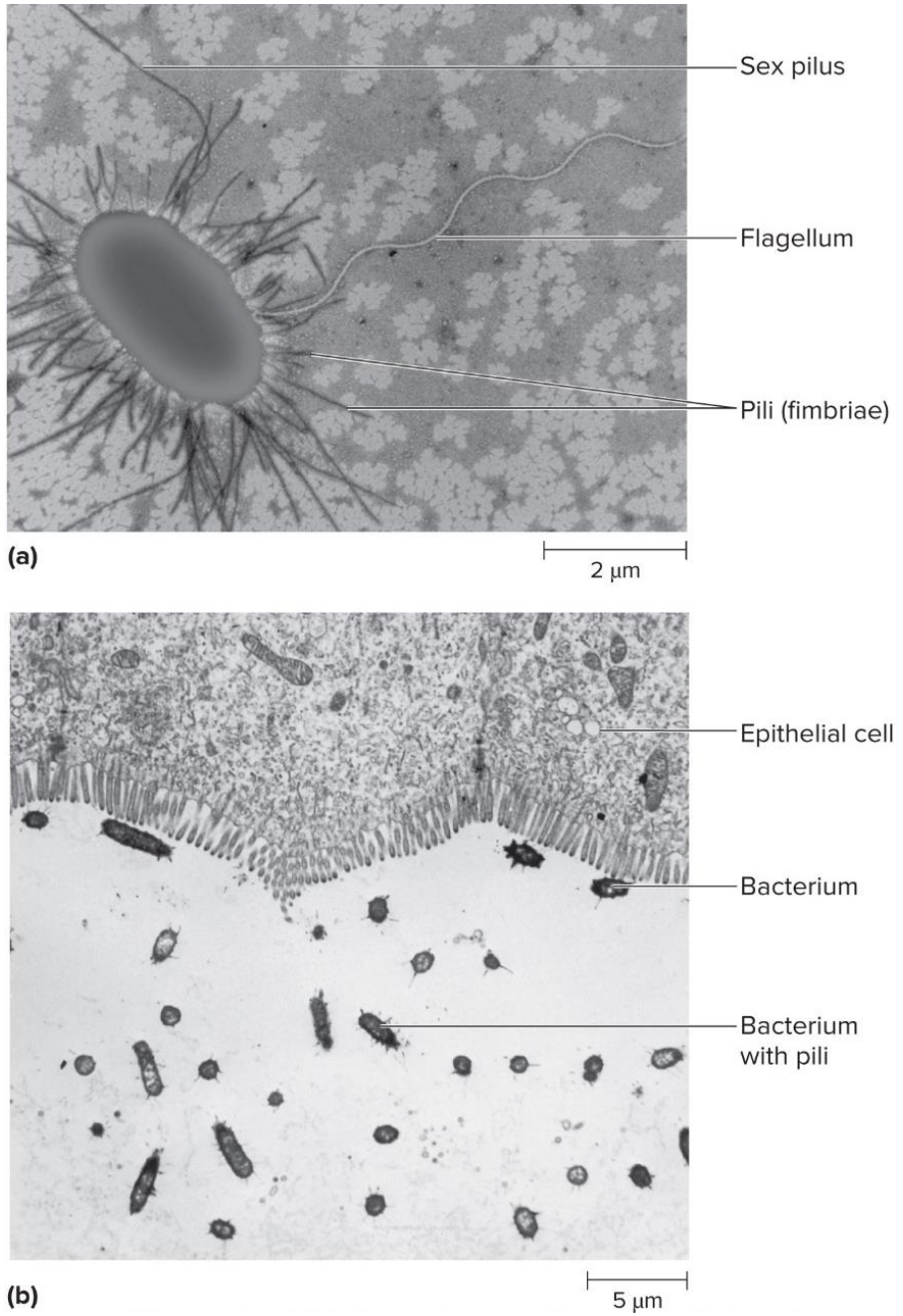
Flagellum composed of 3 Parts

- **Filament**
  - length of flagella
  - composed of protein flagellin
- **Hook-base**
  - to which flagella is attached
- **Basal body**
  - provides for spinning movement. Axel and Wheel movement



Pili

- Pili (singular: pilus) are shorter and thinner than flagella
- Types that allow surface attachment also called fimbriae
- *Twitching* motility and *gliding* motility involve pili
- Sex pilus used to join bacteria for a type of DNA transfer



- a: ©Dennis Kunkel/SPL/Science Source; b: Source: Harley W. Moon/U.S. Department of Agriculture

Gliding Motility

- Bacteria only; no Archaea
- Slower and smoother than swimming
- Movement typically occurs away from colony.
- Requires surface contact
- proteins)

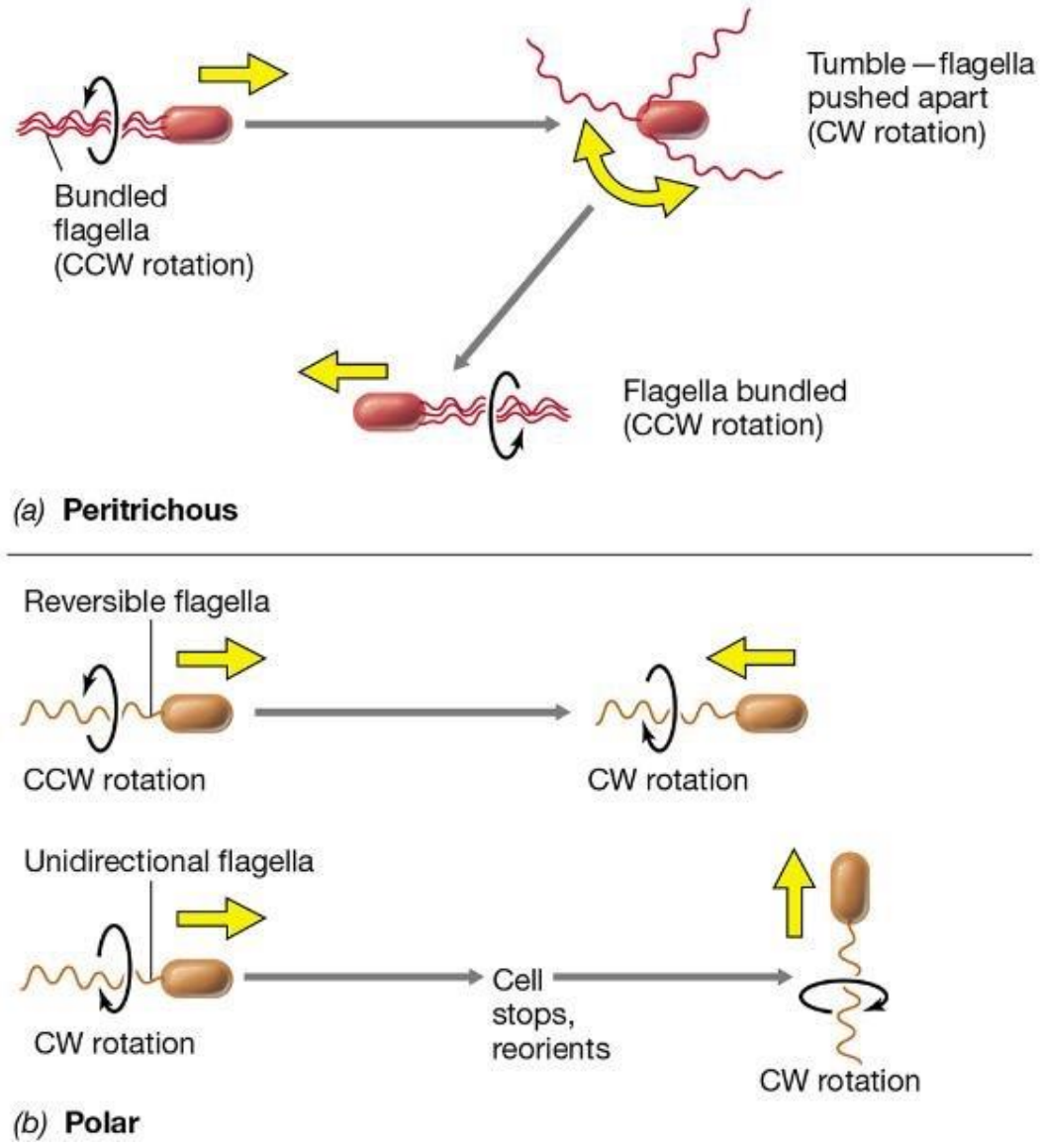
Mycoplasma gliding
https://youtu.be/RiBfycQ0eUM

Directional Movement

Taxis: directed movement in response to chemical or physical gradients

- chemotaxis: response to chemicals
- phototaxis: response to light
- aerotaxis: response to oxygen
- osmotaxis: response to ionic strength
- hydrotaxis: response to water
- magnetotaxis:

Movement

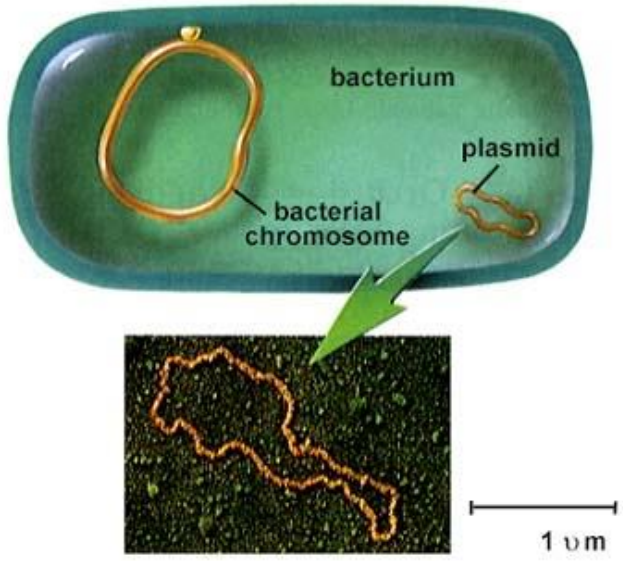


3.7 Internal Components of Prokaryotic Cells

- Inclusions & Vesicles
- Endospores
- Chromosome
- Ribosomes

Chromosome and **Plasmids**

- **Chromosome:** a single, circular double-stranded DNA molecule
- The **Nucleoid:** a gel-like region
- **Plasmids:** extra circular chromosomal DNA
  - usually encode for:
    - Antibiotic resistance

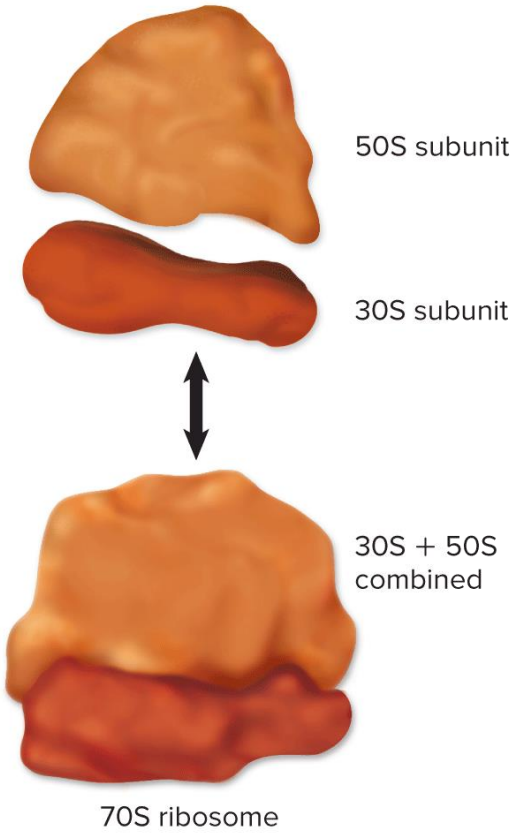


Ribosomes

- Translate mRNA into Proteins
  - **70S** In **Bacteria**

  Vs

  - 80S In Eukaryotic Cells



Svedberg units

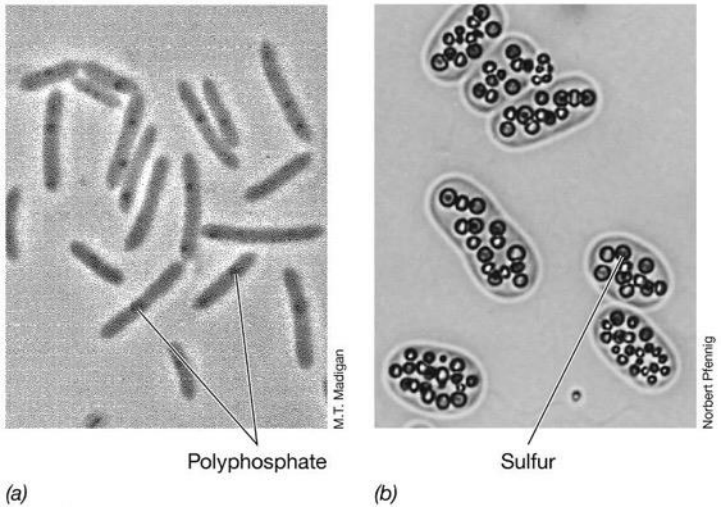
- a term that reflects the relative size and density of the molecules.
- In that term, the “S” (for Svedberg) is a unit used to indicate how fast particles settle when spun at very high speeds in an ultracentrifuge
- The faster a particle moves toward the bottom, the higher the S value and the greater the size and density.

Inclusions & Vesicles

Many types of storage granules
(starch, phosphate, lipid, sulfur)

Inclusions & Vesicles

- Inclusions function as energy reserves, carbon reservoirs, and/or have special functions.



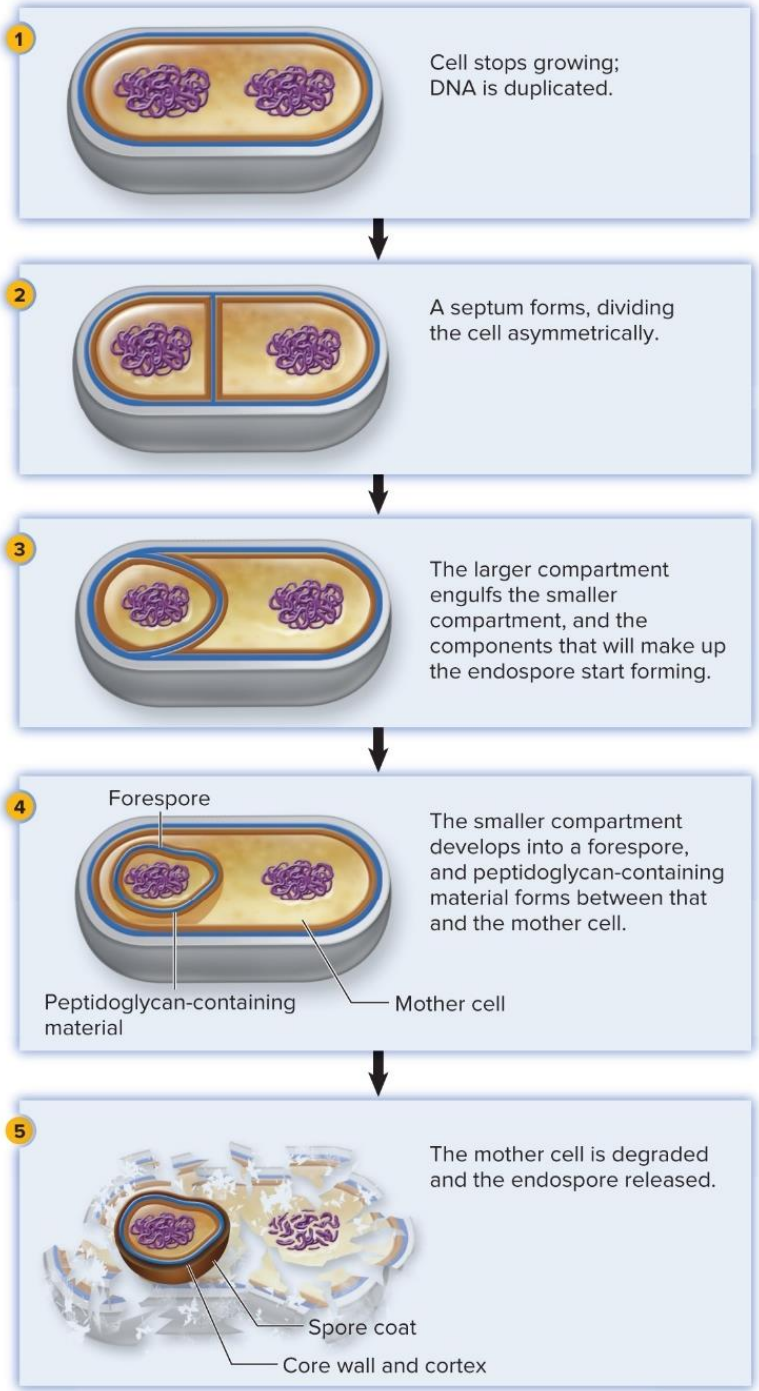
- Gas Vesicles confer buoyancy in planktonic cells
  - Impermeable to water and solutes

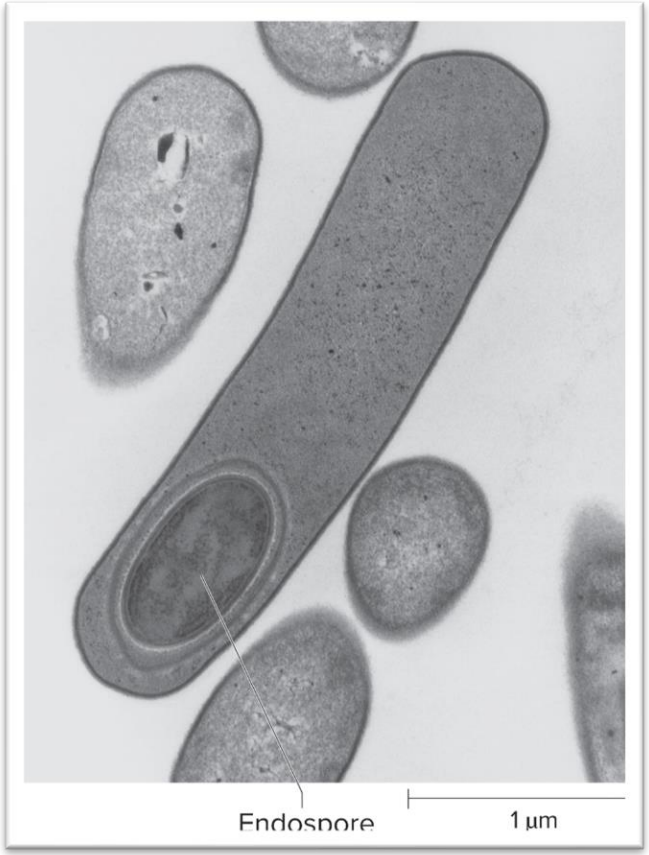
Metachromatic granules

- known as Volutin
- They are composed of polyphosphate, RNA & proteins
- Their main **function** is to supply phosphate for nucleic acid synthesis, cell division, energy metabolism and as a source of phosphorous for nutrition

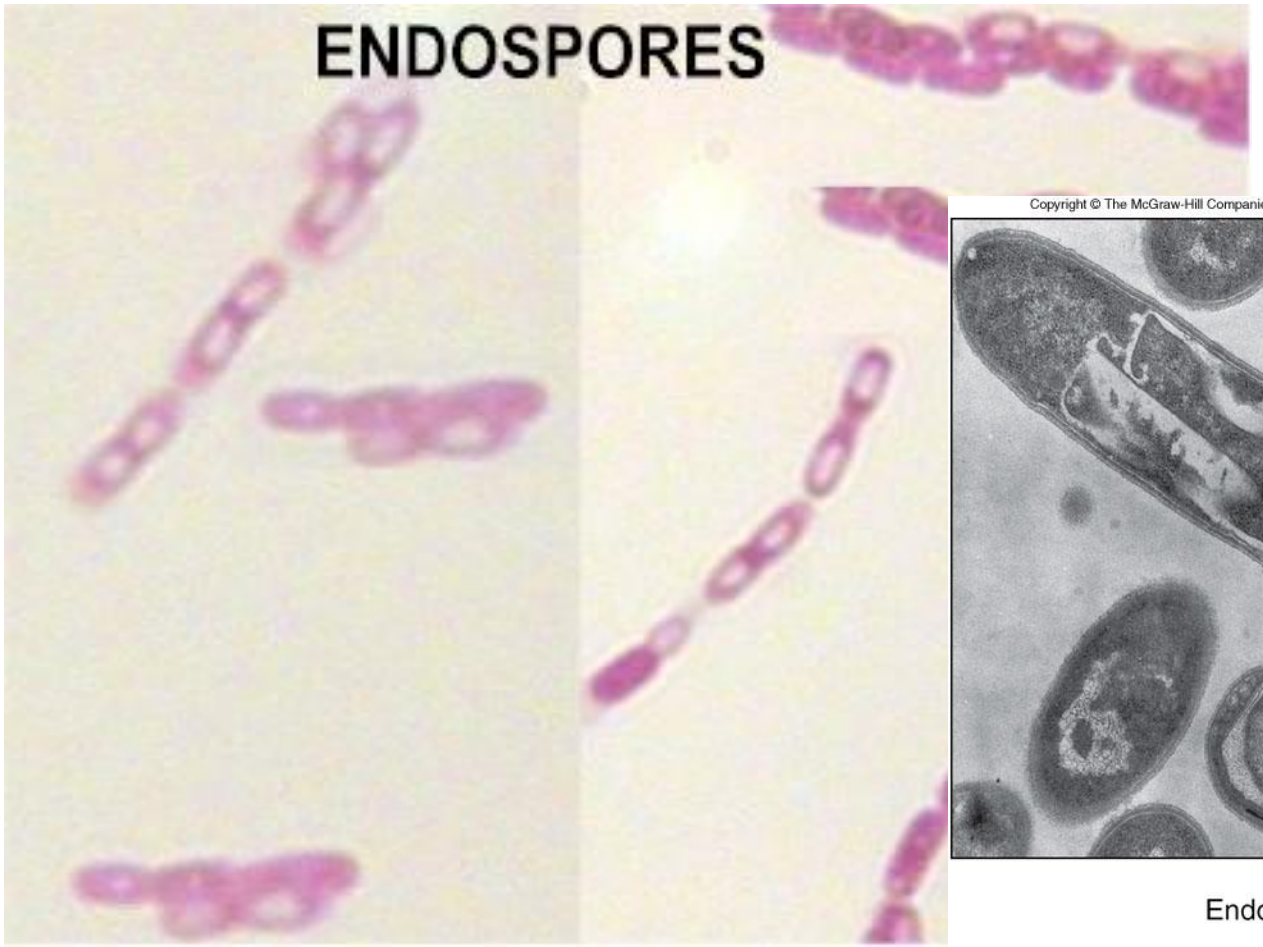
Endospores

- Survival structures to endure unfavorable growth conditions
- Ideal for dispersal via wind, water, or animal gut
- Present only in some gram-positive bacteria, (*e.g.*, *Bacillus* and *Clostridium*)





Endospores


ENDOSPORES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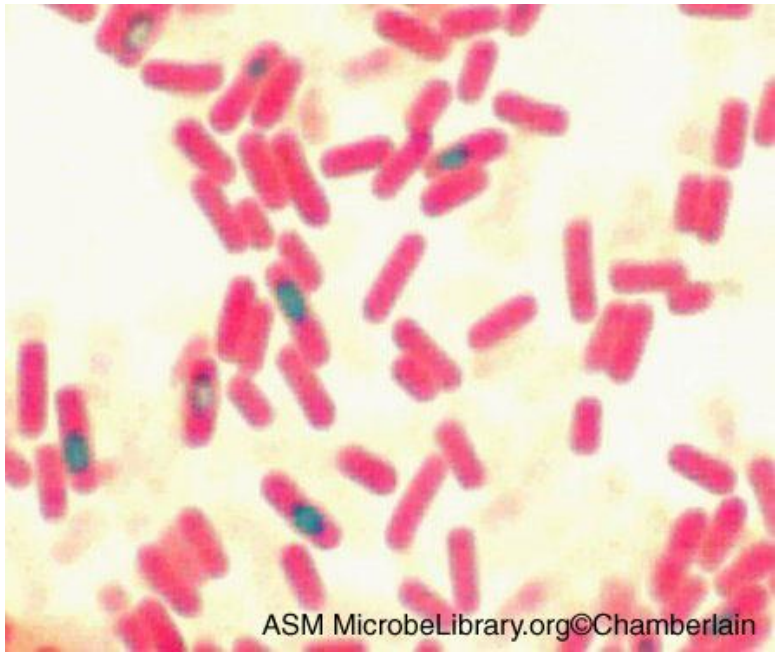
Endospore
1 µm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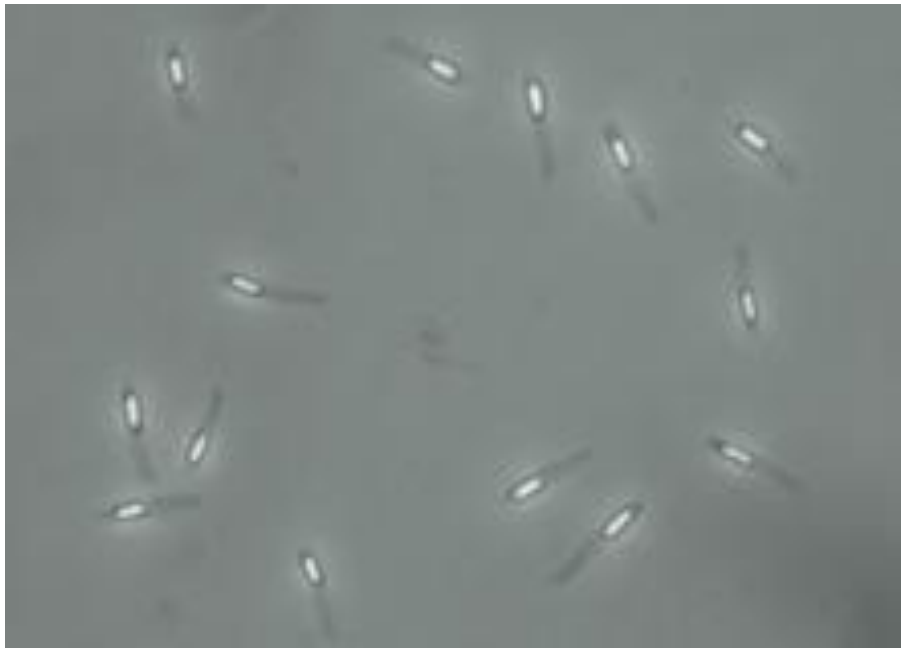


A sporofied bacterium

can **germinate** back to the **vegetative state**

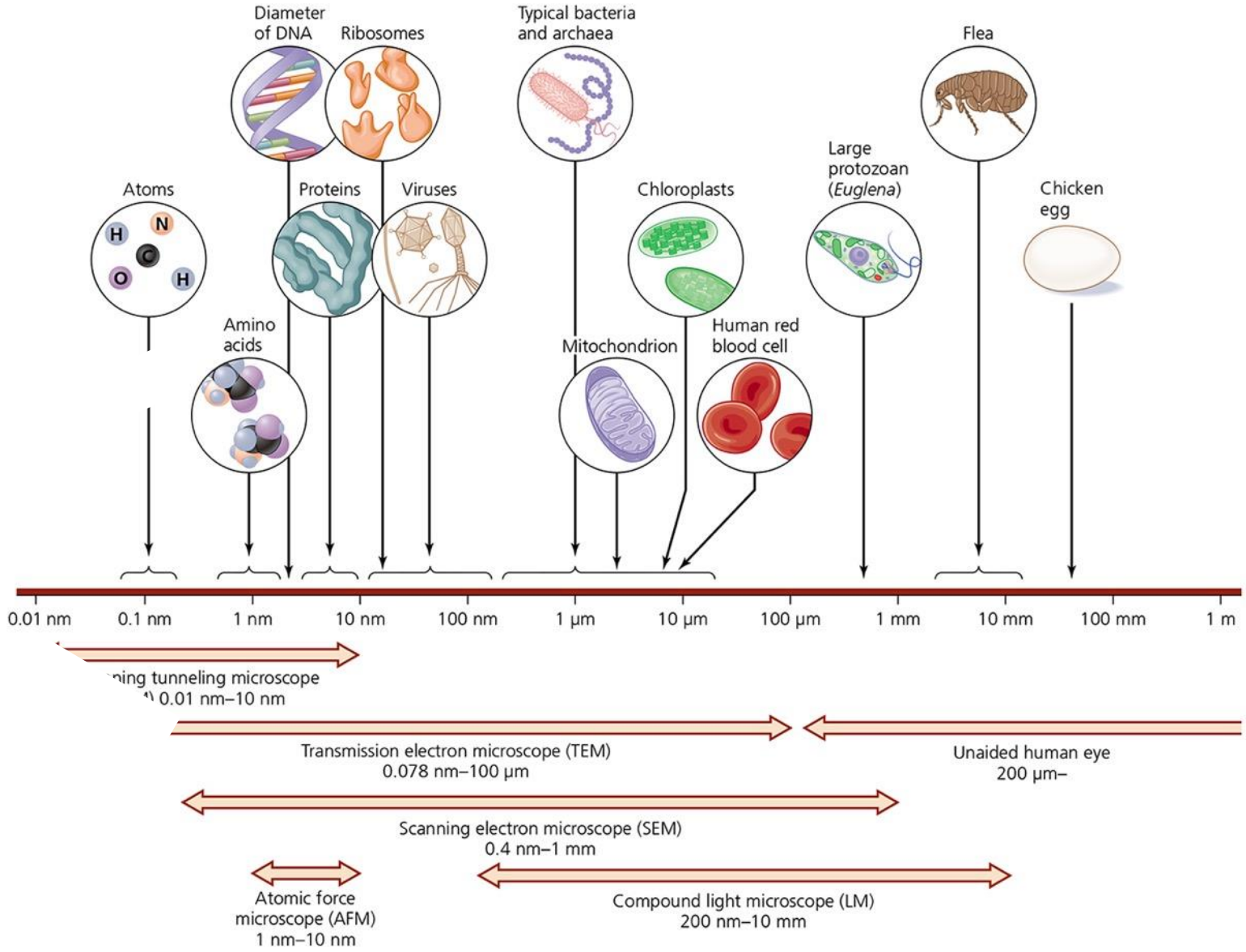
Microscopy

- Microscopy reveals two fundamental cell types:
- **Prokaryotic** cells (*Bacteria*, *Archaea*)
  - Smaller size gives high surface area to low volume
    - Facilitates rapid uptake of nutrients, excretion of wastes
    - Allows rapid growth
  - Disadvantages include vulnerability to threats including predators, parasites, and competitors
- **Eukaryotic** cells (*Eukarya*)
  - Larger, more complex, many cellular processes take place in membrane-bound compartments
  - Defined by the presence of a nucleus

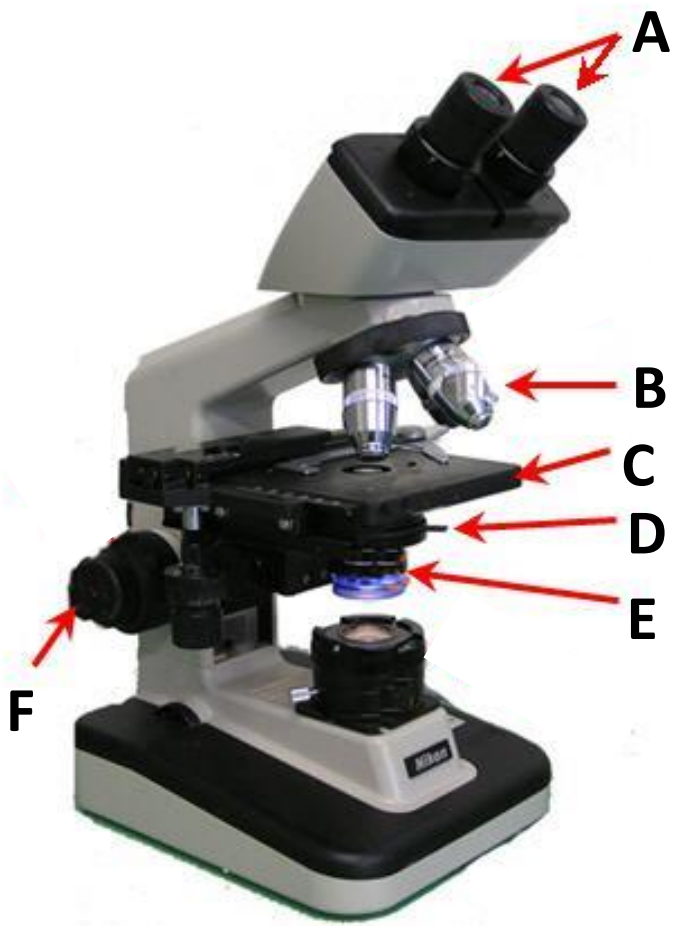
Relative sizes

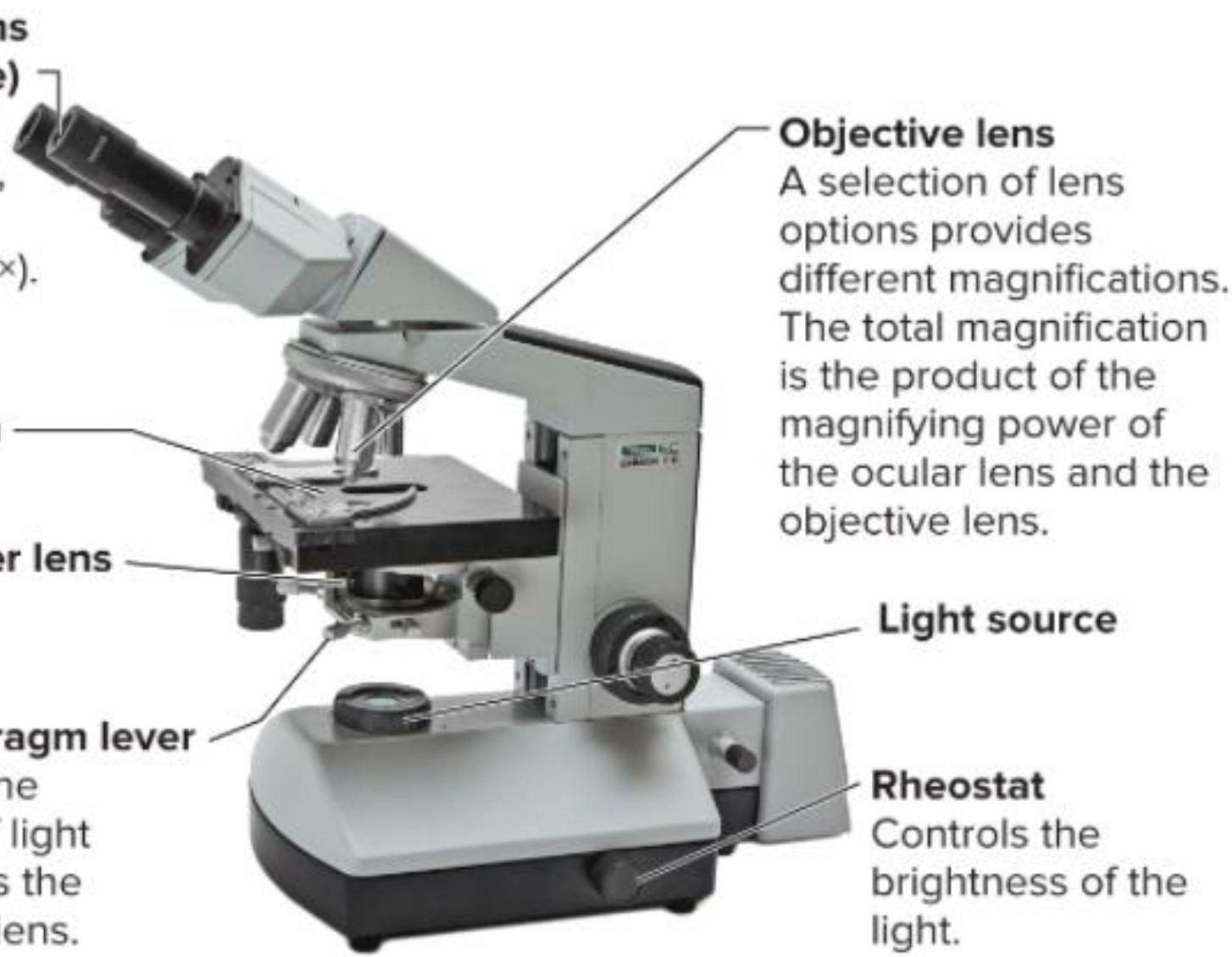
- 1 meter (m)= 1000000000, 10^9 nanometer
  - 1 **nanometer**(**nm**) = 10^{-9} meters
- 1 meter= 1000000, 10^6 micrometer
  - 1 **micrometer(µm)** =10^{-6} meters
- 1 micrometer =1000, 10^3 nanometer
  - 1 **nanometer** = 10^{-3} µm
- Angstrom (A): one ten –billionth of a meter

Macro → Micro



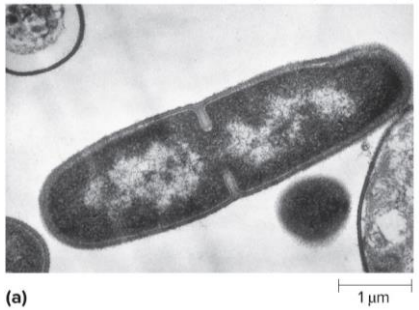
Macro → Micro



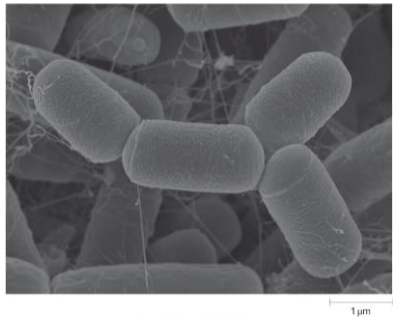

Ocular lens (eye piece)
Magnifies the image, usually 10-fold (10×).
Objective lens
A selection of lens options provides different magnifications. The total magnification is the product of the magnifying power of the ocular lens and the objective lens.
Specimen stage
Condenser lens
Focuses the light.
Light source
Iris diaphragm lever
Controls the amount of light that enters the objective lens.
Rheostat
Controls the brightness of the light.

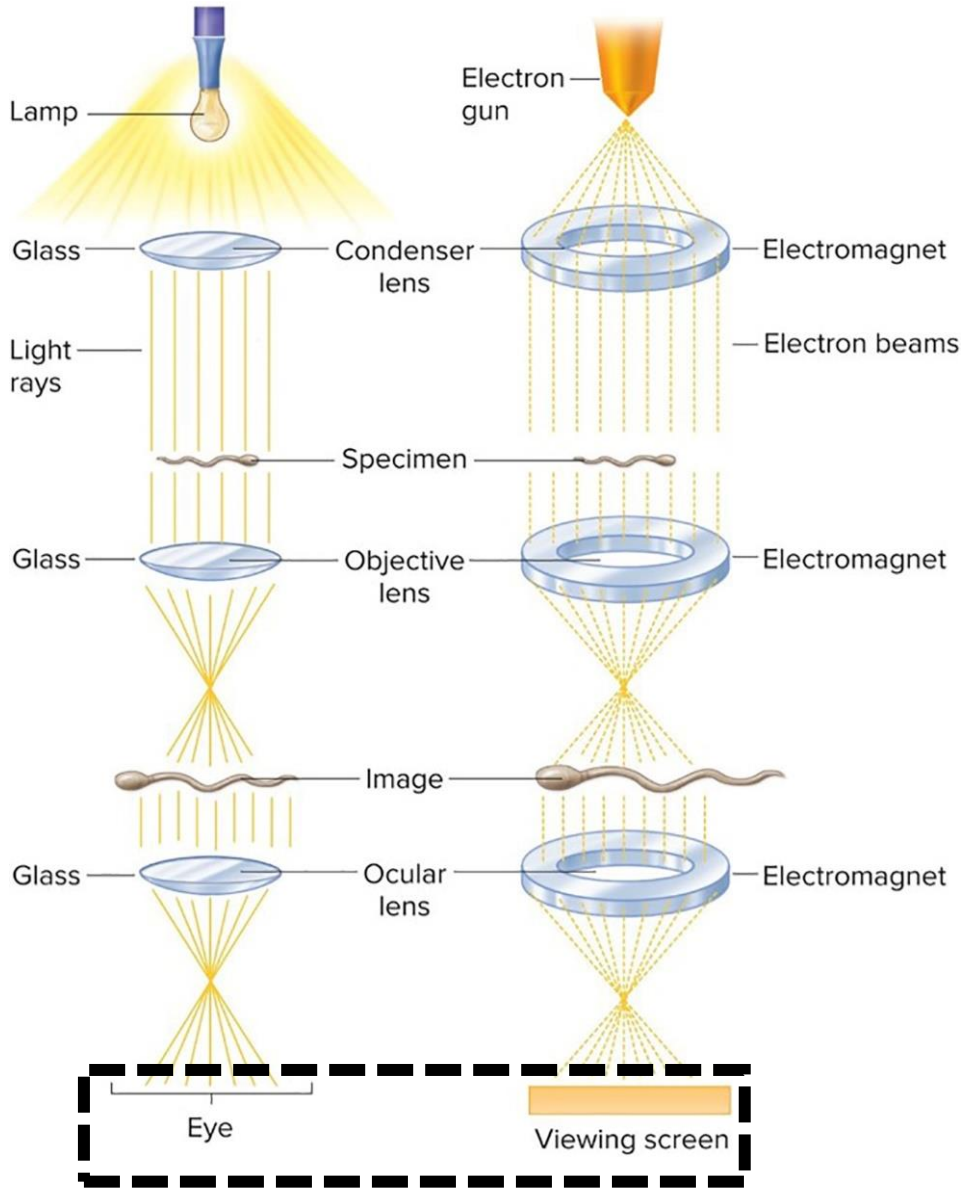
Electron microscope

- **Transmission** EM (TEM): Beam of electrons passes through specimen or scatters
  - Depends on density of region: dark areas are dense
  - Thin-sectioning used to view internal details, but process can distort cells



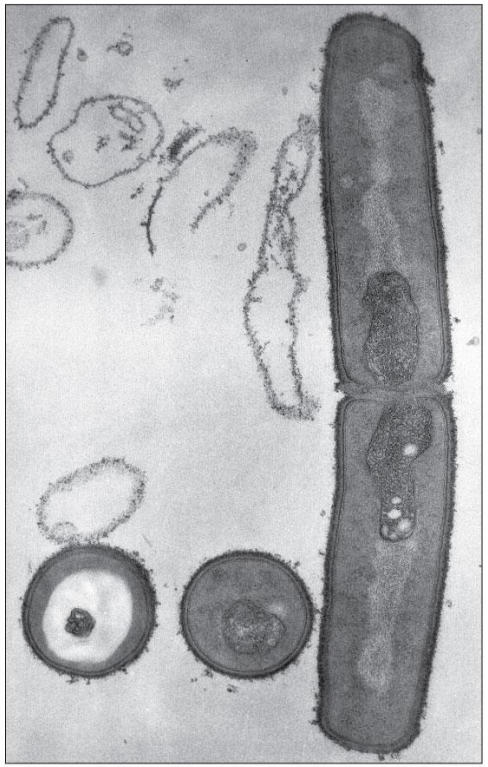
- **Scanning** EM (SEM): beam of electrons scans over surface of specimen
  - Used to observe surface details
  - Surface coated with thin film of metal




Lamp
Electron gun
Glass
Condenser lens
Electromagnet
Light rays
Electron beams
Specimen
Glass
Objective lens
Electromagnet
Image
Glass
Ocular lens
Electromagnet
Eye
Viewing screen

Transmission Electron Microscopy

To see structures inside of cells



(c)

Scanning Electron Microscopy

- **Surface Structures**
- **Produces a 3D effect**