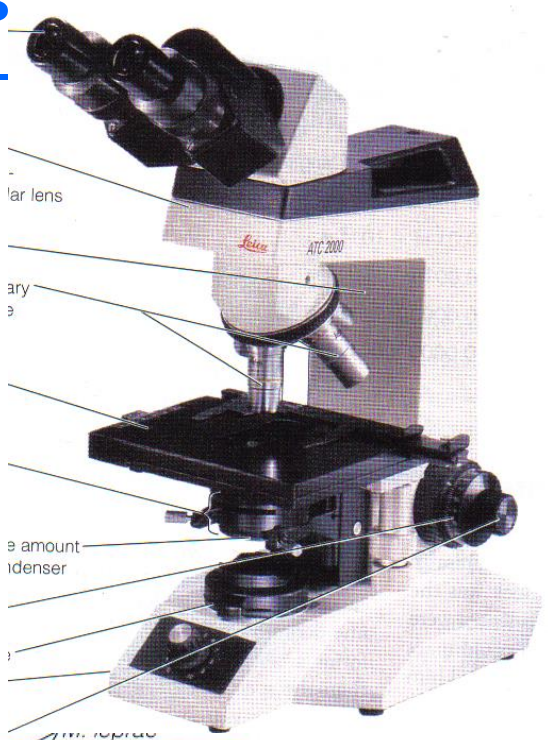
Chap 3 & 4 Microscopy & Cell Components

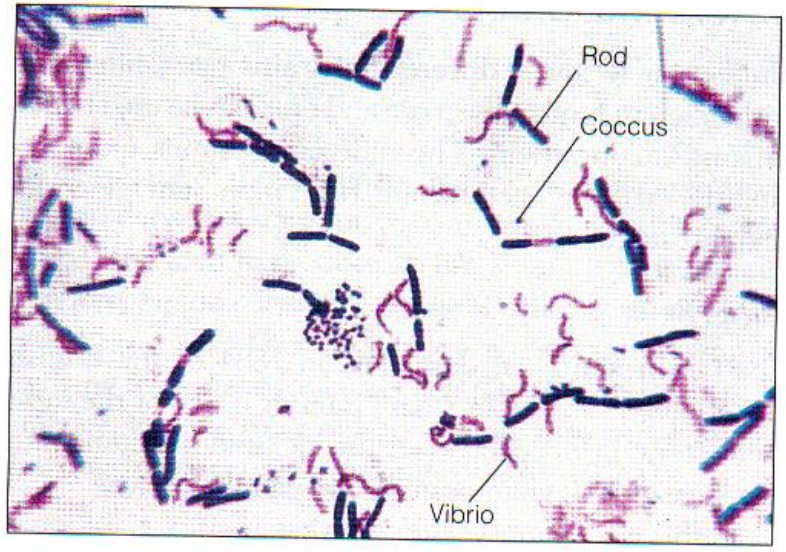
Prior Knowledge: Parts of the scope at the right? Type of scope? Proper care and use? Other types of scopes?

What are the 3 shapes of bacteria?

Preview of smears similar to what you will make:

Gram stain vs. acidfast stain; specificity, use & frequency.





(b)

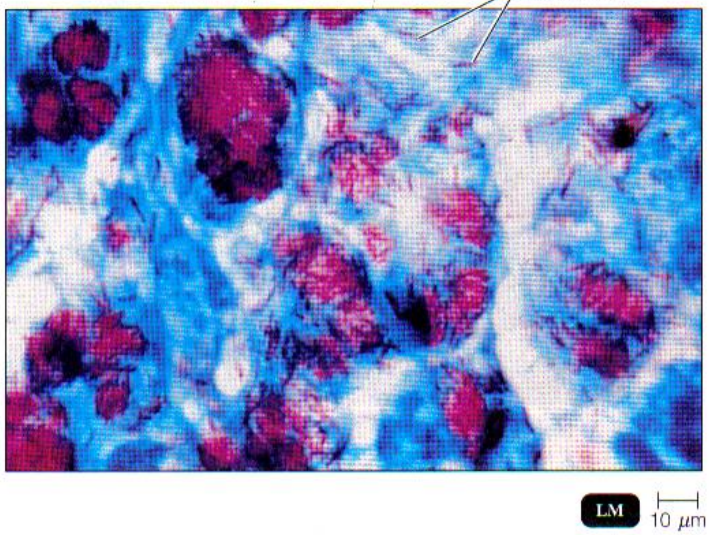


FIGURE 3.11 **Acid-fast bacteria.** The *Mycobacterium*

Objectives

1.White book: Read Chap 3 & p 77-98 & 108

2.Black book: Read Chap 3 & p75-96 & 106

Objectives:

1. List metric measurement units for microorganisms and convert to other metric units (m, mm, um, nm).

2. Identify parts & functions of the compound light microscope.

3. Define/calculate total magnification & resolution.

4. Compare, contrast, and identify uses (diseases/organisms) for brightfield, darkfield, fluorescent, electron-transmission, and electron-scanning microscopy.

5. Differentiate, compare, and explain the appearance and uses of each of the following: , fixing, acidic & basic dyes, simple, differential & special stains, capsule, endospore, acid-fast and flagella stains.

Objectives, cont'd

6. List specific chemicals that are used for each type of stain in the objective above, primary stain, mordant, decolorizer, counterstain.
7. Gram stain: list the steps, purpose, and the appearance of GP & GN cells after each step.
8. Identify the 3 basic shapes of bacteria and secondary arrangements.
9. Describe the structure & function of the glycocalyx, flagella (including arrangement), axial filaments, fimbriae, pili. Identify flagellar arrangements.
10. Compare & contrast the cell walls of GP bacteria, GN bacteria, archaea, mycoplasmas, and mycobacteria. (Including composition, antibiotic & chemical resistance, presence of toxins, staining reactions, effect of penicillin, lysozyme, etc.)

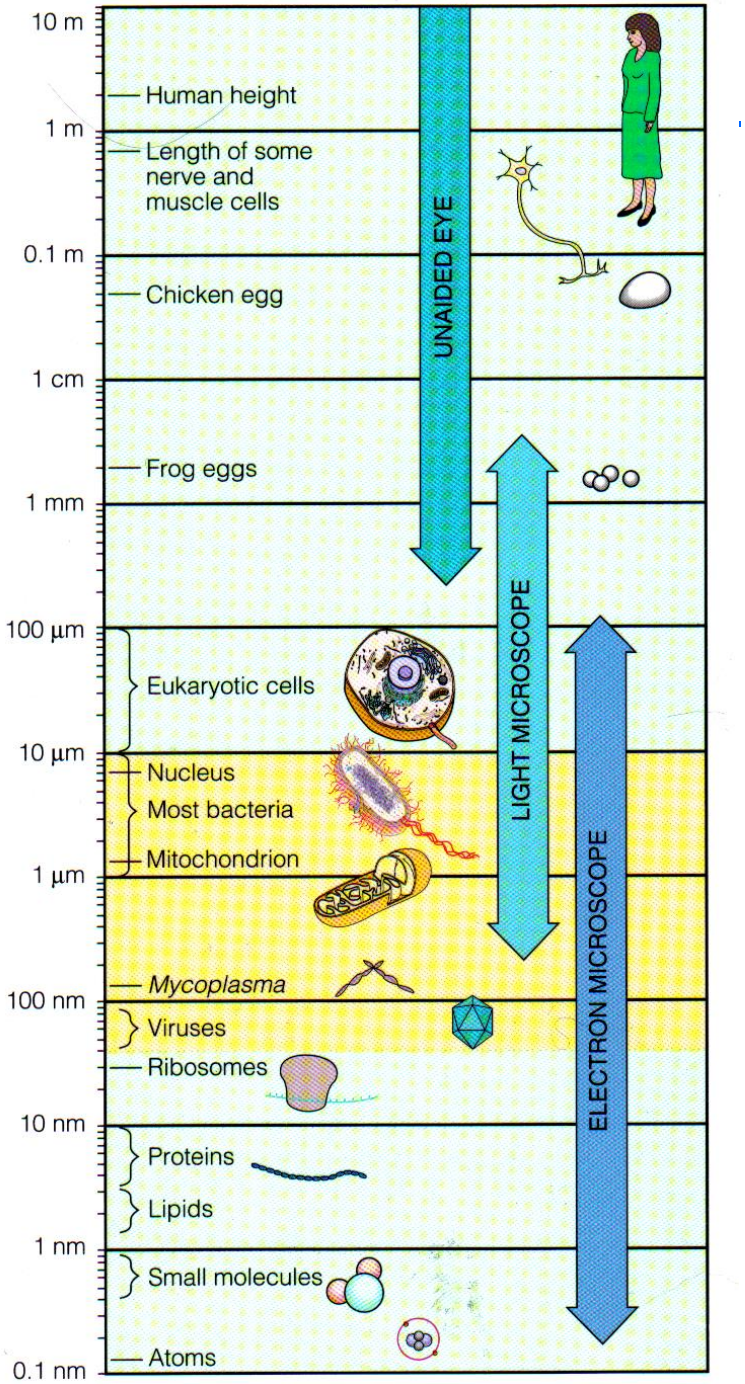
Objectives, Cont'd

11. Identify the functions of the cell/plasma membrane, chromatophores/thylakoids, nucleoid, ribosomes, endospores (including location), inclusions, plasmids.
12. Transport: passive (simple diffusion, osmosis, facilitated diffusion), active transport, hypertonic, hypotonic, isotonic, osmotic lysis, plasmolysis
13. Discuss several pieces of evidence that support the endosymbiotic theory of eukaryotic evolution.
14. Describe the overall structure and defining characteristics of prokaryotes, as compared to eukaryotes.
15. On given slides identify shape, arrangement, type of stain, gram reaction, endospore location, flagellar arrangement, presence/absence of a capsule.

Measurement Units & Terms

1. **Units**
   - A. **Micrometer (μm) = 10^{-6} m or 1/1,000,000 m or .000001m**
   - B. **Nanometer (nm) = 10^{-9} m**
     - i. **Example: Convert 21.5 nm to m**
       - **0.0000000215 m**
2. **Total Magnification Calculation: Ocular x Objective**
3. **Resolution: Distance apart needed to see 2 points as separate. (Ability to see fine detail)**

Fig 3.2 Resolution of eye & scopes



Resolution & Refractive Index

A. Resolving power = $\frac{\text{wavelength}}{\text{2 x N.A. numerical aperture}}$

N.A. depends on:

i. Refractive index of material between lens & slide.

ii. The angle of most divergent light ray

B. To improve resolution:

i. ⇓ wavelength (biggest impact)

ii. ⇑ N.A.: Use oil w/100x objective (similar to using "glasses" to improve resolution of eye)

iii. Altering position of CONDENSER (NOT amount of light)

C. Improve conditions but NOT resolution:

i. Contrast by staining

ii. Light adjustment

Fig 3.3 Refraction w/ & w/o Oil, p.59

Using oil does improve resolution, as it increases the numerical aperture, which will cause a better (smaller) resolving power number

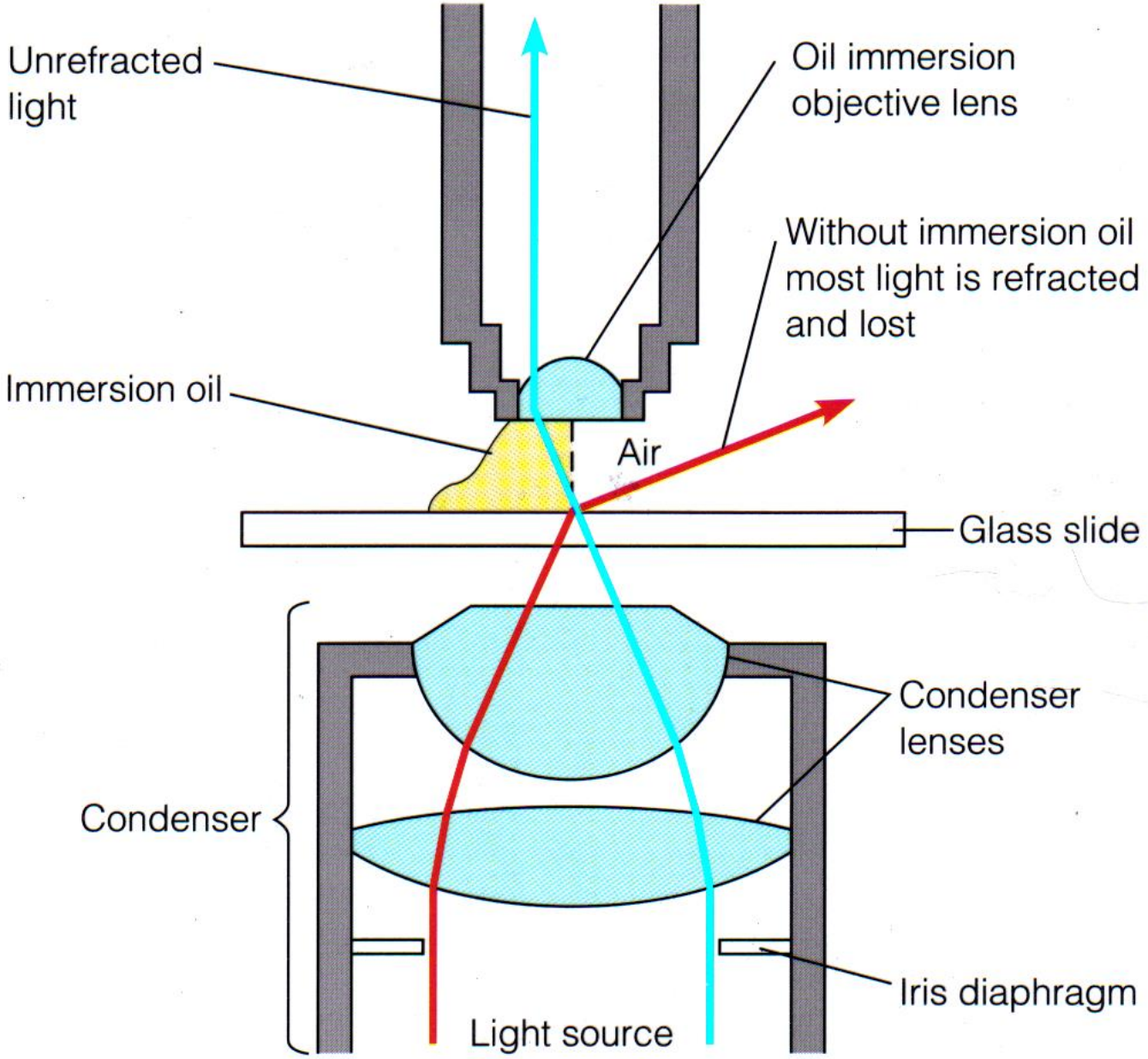


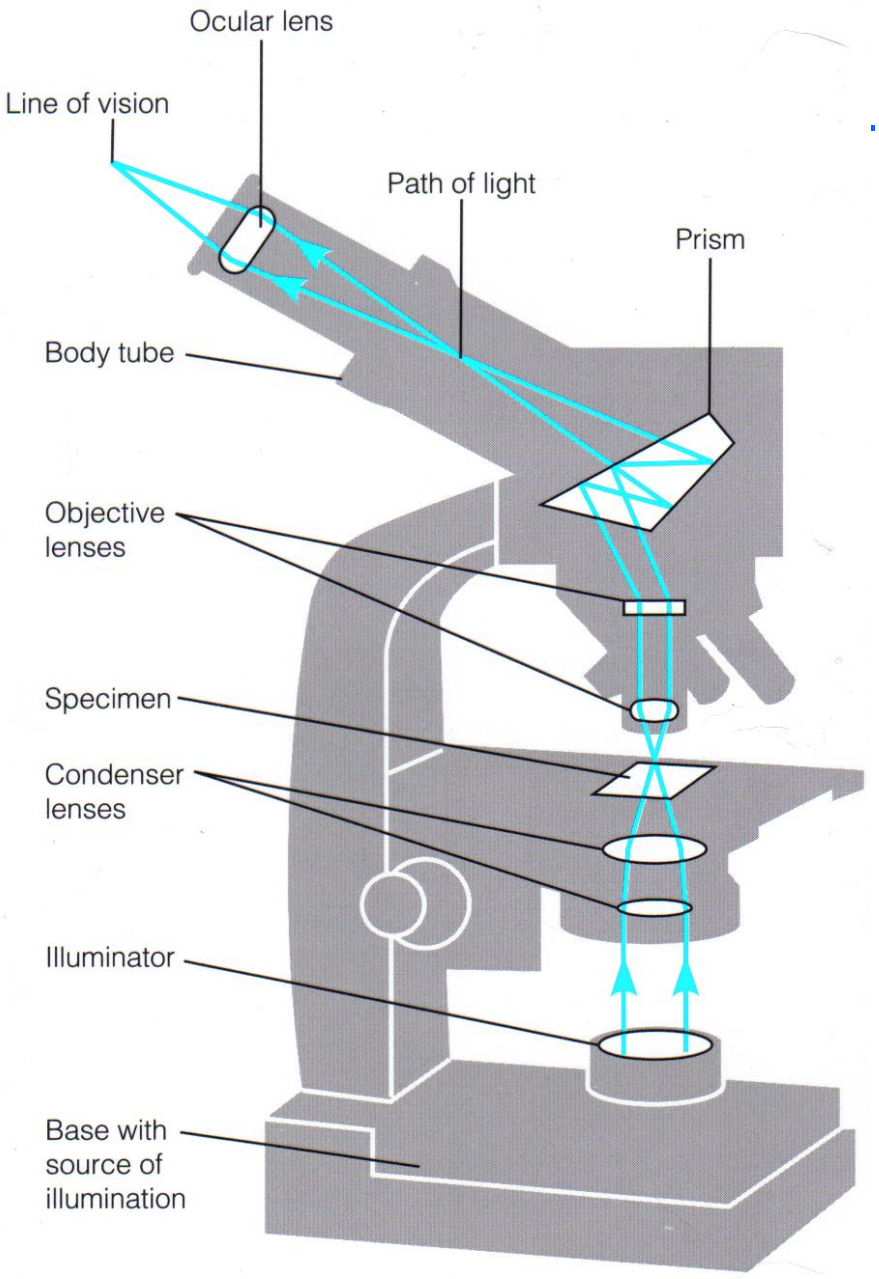
Fig 3.1b Compound Light Microscope

What does compound mean?

How does it apply to this scope?

Next slide will discuss 3 subtypes of light microscopy, using the same scope with different types of light.

In class, only Brightfield Light microscopy is used.

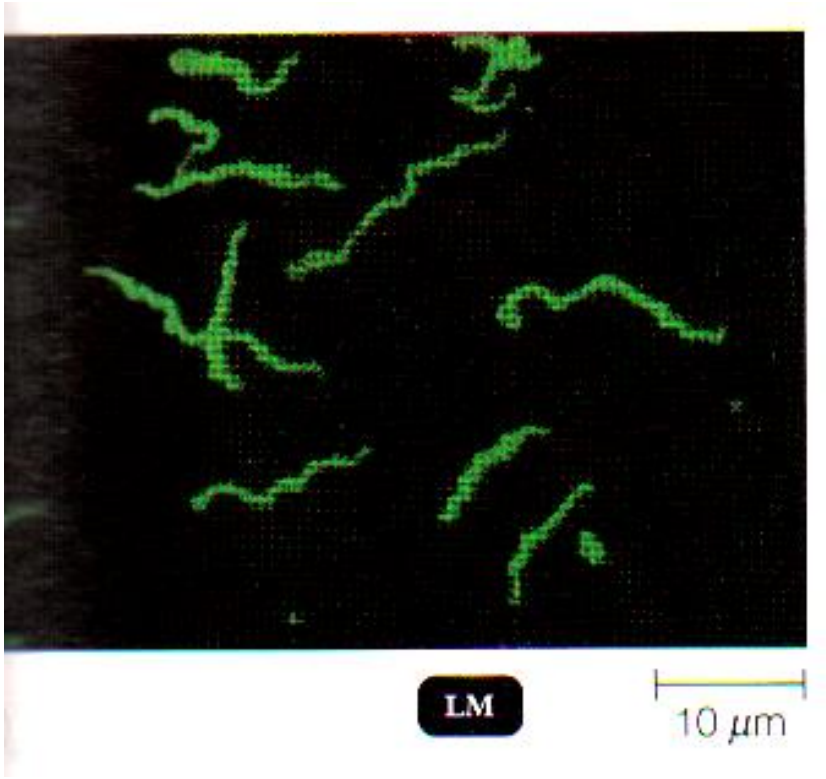


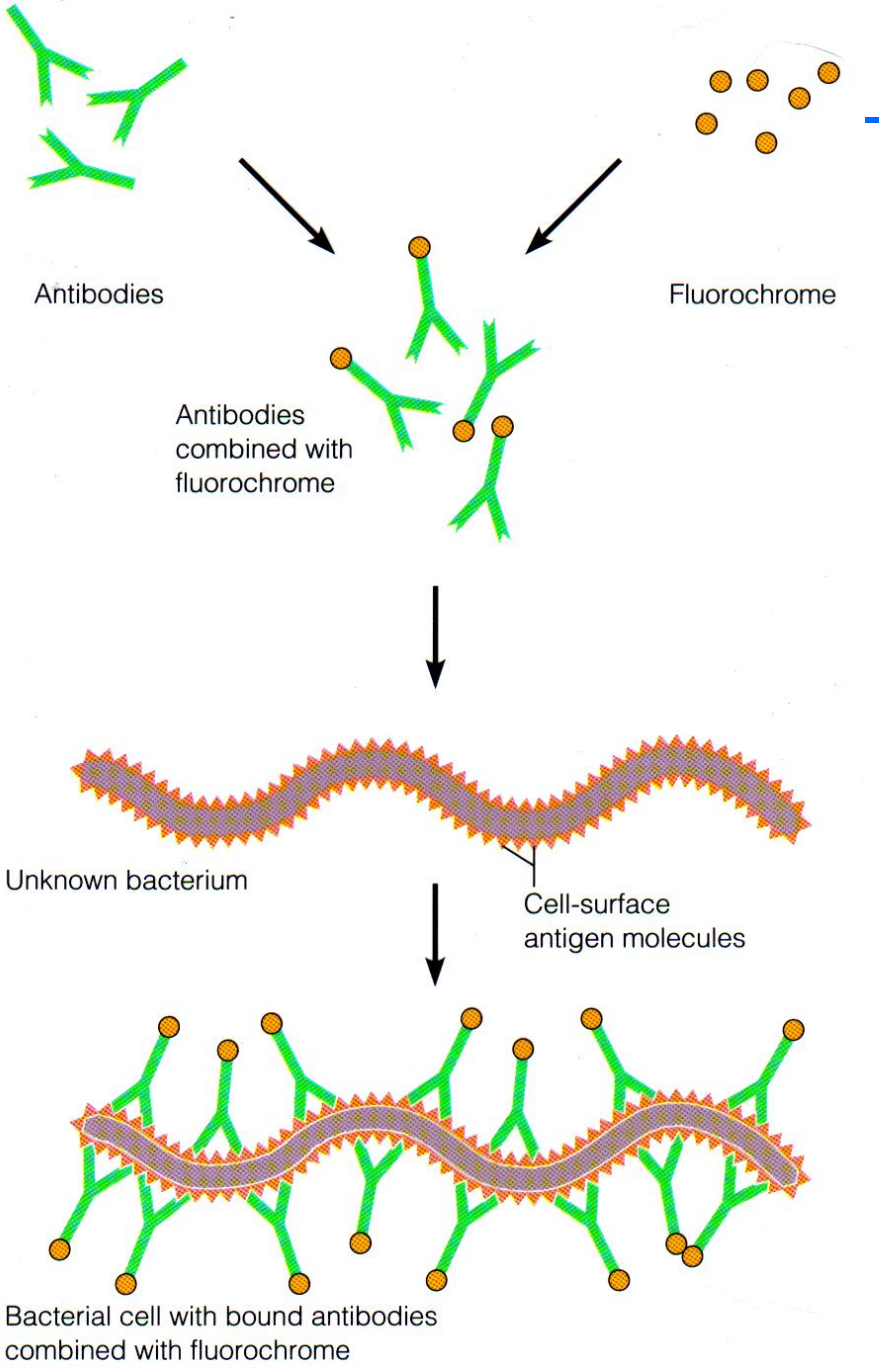
(b) The path of light (bottom to top)

Types of Scopes-3 subtypes of Light microscopes

Scope	Enhanced by	Advantages	Uses
Light, Brightfield: Background bright Visible light Res: 0.2um (200nm) Mag: 2000x	Stains Oil w/100x Diaphragm & light	Inexpensive Easy to use	Live specimens (unstained) Stained specimens Bacteria, protozoa
Light, Darkfield: Background dark & microbes clear Same	N/A	Easier to see unstained-(distinct borders) Smaller microbes	Live microbes: syphilis
Light, Fluorescent: Background dark & bright fluorescing microbes Same	Fluorescent-antibody dyes: Fluorescent dye on antibody to microbe antigen, microbe fluoresces	Rapid ID directly from specimen, w/o culture Detection of small # microbes compared to other light microscopy	When immediate diagnosis needed When cultures aren't avail, or take long TB, rabies, syphilis, anthrax

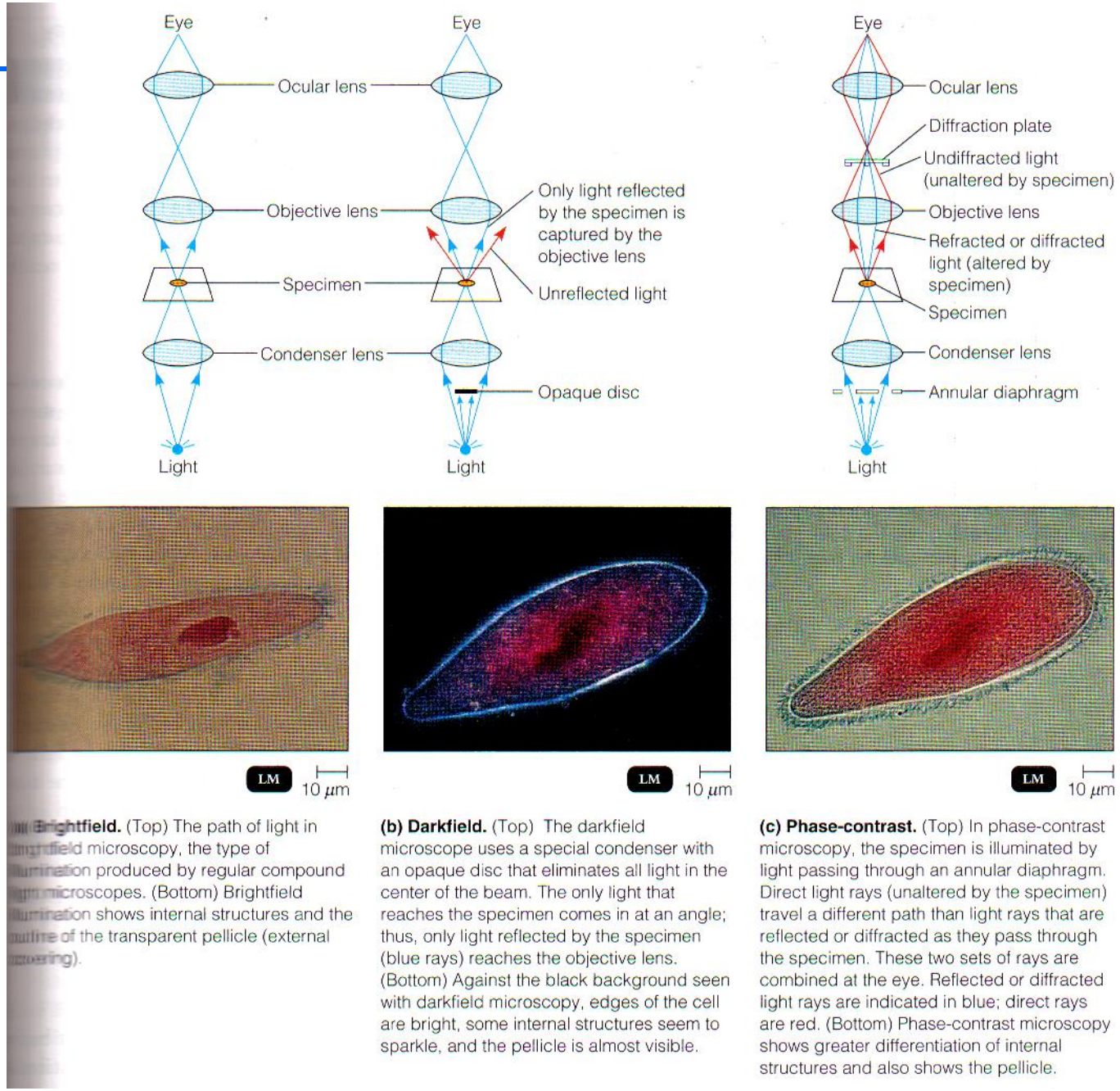
Fig 3.6 Immunofluorescent Staining Technique





Demo-Fluorescent marker drawings

Fig 3.4 p. 59 Micrographs comparing Bright-, Dark- & Phase-



Brightfield. (Top) The path of light in brightfield microscopy, the type of illumination produced by regular compound light microscopes. (Bottom) Brightfield illumination shows internal structures and the outline of the transparent pellicle (external covering).

(b) Darkfield. (Top) The darkfield microscope uses a special condenser with an opaque disc that eliminates all light in the center of the beam. The only light that reaches the specimen comes in at an angle; thus, only light reflected by the specimen (blue rays) reaches the objective lens. (Bottom) Against the black background seen with darkfield microscopy, edges of the cell are bright, some internal structures seem to sparkle, and the pellicle is almost visible.

(c) Phase-contrast. (Top) In phase-contrast microscopy, the specimen is illuminated by light passing through an annular diaphragm. Direct light rays (unaltered by the specimen) travel a different path than light rays that are reflected or diffracted as they pass through the specimen. These two sets of rays are combined at the eye. Reflected or diffracted light rays are indicated in blue; direct rays are red. (Bottom) Phase-contrast microscopy shows greater differentiation of internal structures and also shows the pellicle.

Scopes-Electron

Scope	Enhanced by	Advantages	Uses
Electron, Scanning Res: 20 nm AKA 0.02um AKA 0.000002mm Mag: 10,000x		3-D	Surfaces structures - eukaryote to virus
Electron, Transmission Res: 2.5nm AKA 0.0025 um Mag: 100,000x	Stain w/+ salt of heavy metal	Good res & mag DISADVANTAGE: Need THIN slice as e- can't penetrate All e- scopes- artifacts due to killing, & fixing under vacuum	Virus particles, bacterial flagella, internal cell structures, protein molecules

Both Electron Scopes – Why do they have better resolution?

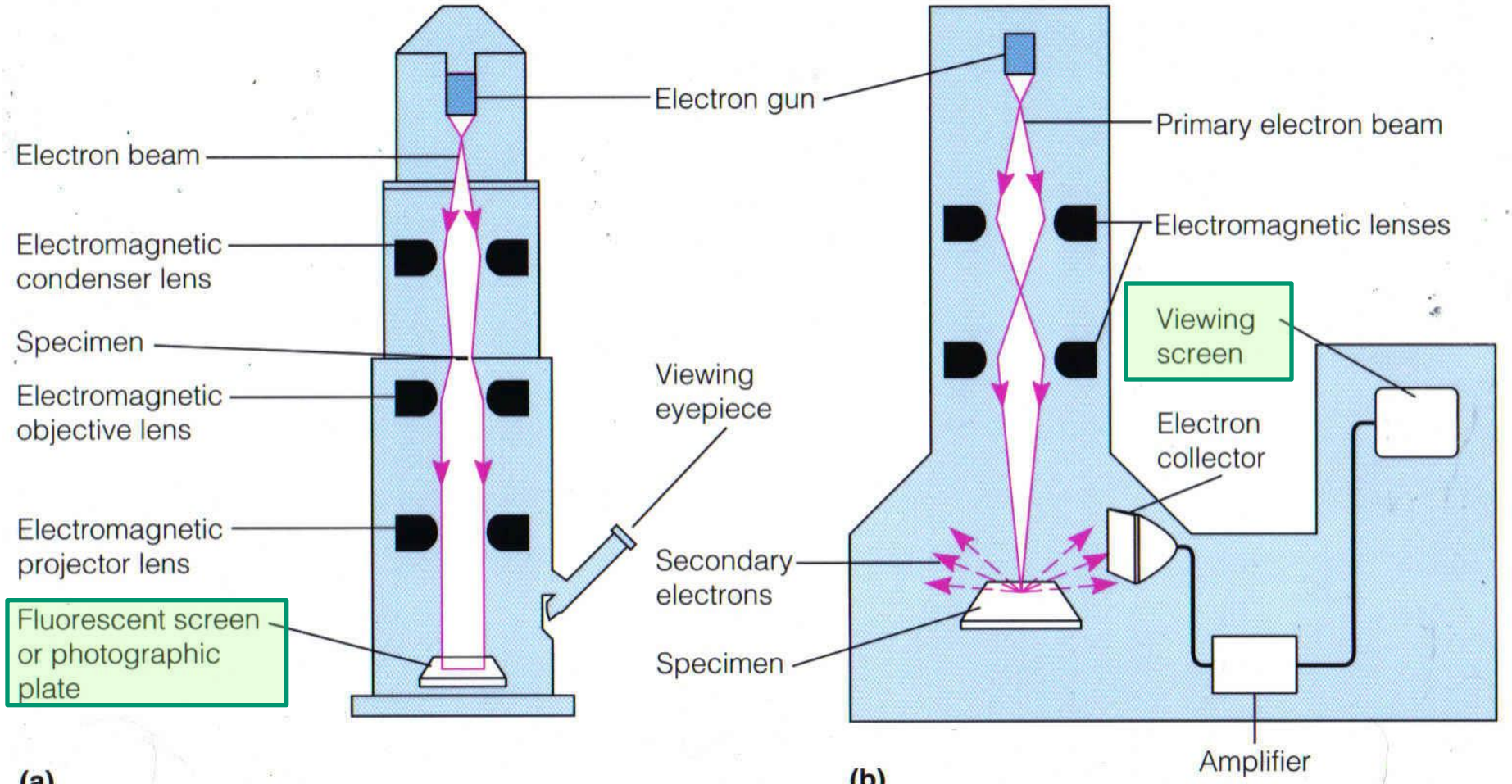
- e- wavelength is 100,000x shorter than the wavelength of light
- Note: Both always black & white. Color artificially

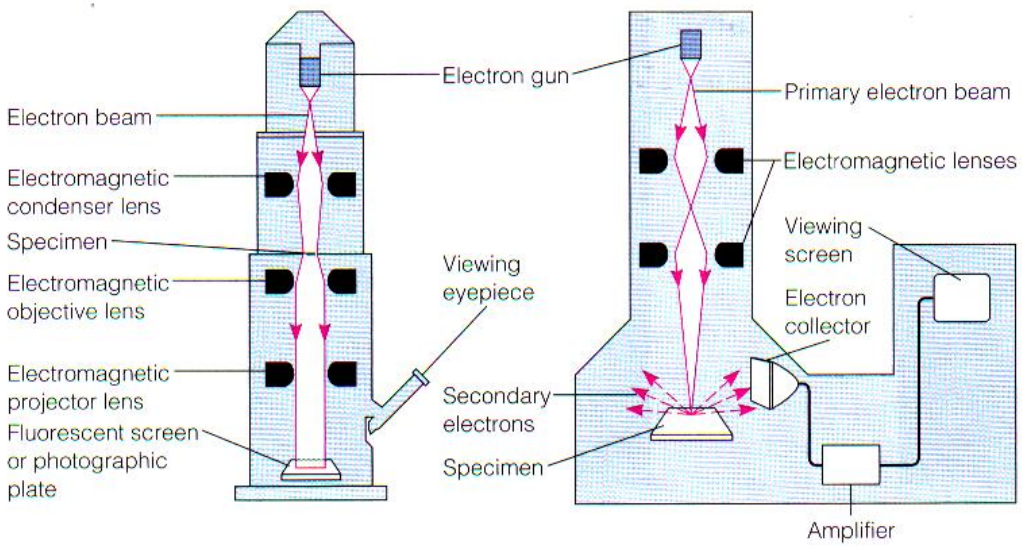
Fig 3.8 Diagrams of transmission & scanning electron

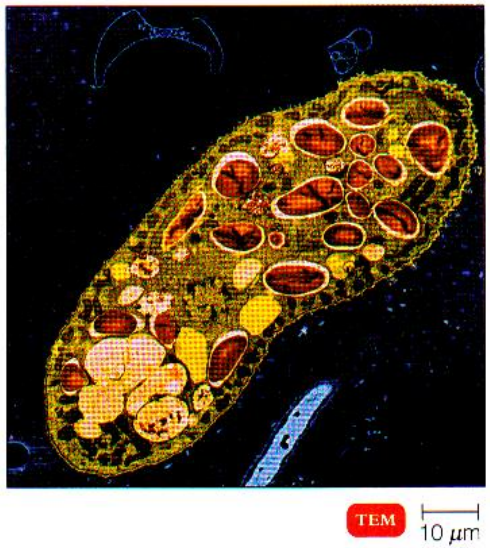
Which is which?

Note difference in specimen placement & path of electrons.

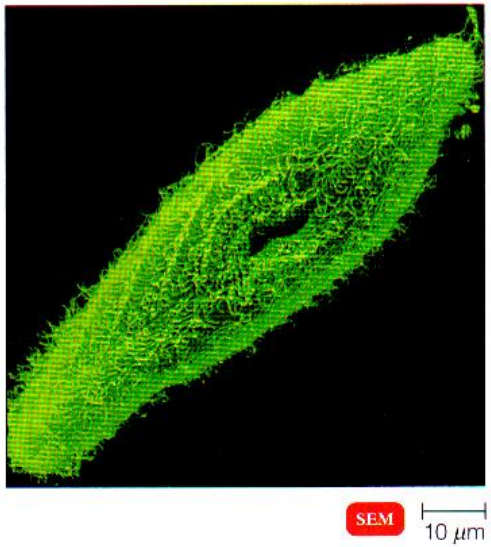
Do you look at the specimen directly, see actual object with eye? Explain.







(a) Transmission. (Top) In a transmission electron microscope, electrons pass through the specimen and are scattered. Magnetic lenses focus the image onto a fluorescent screen or photographic plate. (Bottom) This colorized transmission electron micrograph (TEM) shows a thin slice of a *Paramecium*. In this type of microscopy, the internal structures present in the slice can be seen.



(b) Scanning. (Top) In a scanning electron microscope, primary electrons sweep across the specimen and knock electrons from its surface. These secondary electrons are picked up by a collector, amplified, and transmitted onto a viewing screen or photographic plate. (Bottom) In this colorized scanning electron micrograph (SEM), the surface structures of a *Paramecium* can be seen. Note the three-dimensional appearance of this cell, in contrast

Fig 3.8 Transmission vs. Scanning

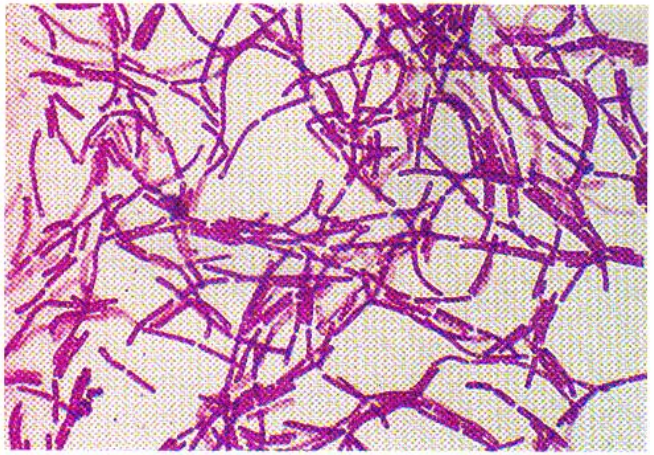
Stains-Slide Prep & Basic Stains

Slide Prep:

1. **Smear**
2. **Fix – attach to slide (won't wash off)**
   - A. **Alcohol or heat**
   - B. **Kills**
   - C. **Adheres to slide**
   - D. **HOPEFULLY-preserves w/ no artifacts (AKA minimal distortion)**

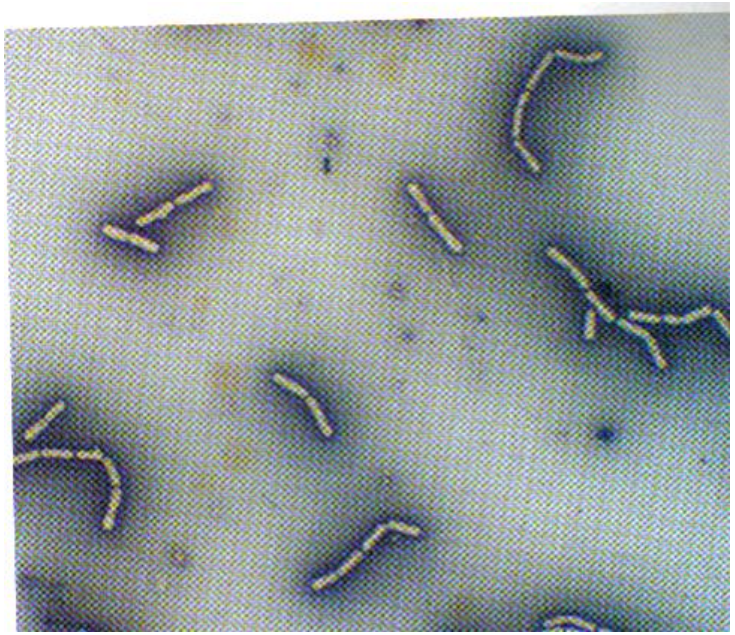
Staining

1. **Basic dye/Positive stain: Colored (+) ion of a salt**
   - A. **Attracted to (-) bacterial cell; stains cell**
   - B. **Crystal violet, methylene blue, safranin, malachite green**



Acidic Dye / Negative Stain

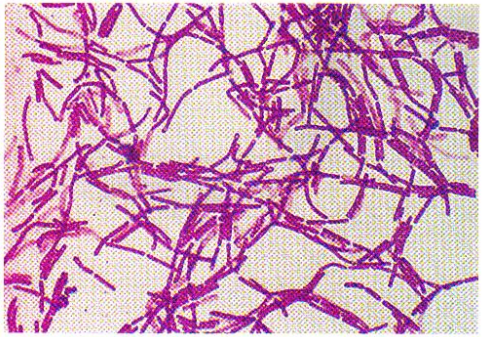
2. **Acidic dye / Negative stain: Colored (-) ion**
   - A. **Repelled & stains background**
   - B. **For cell shape & size, to detect capsules**
   - C. **Advantage: less distortion (no heat fixing & stain doesn't enter so accurate size & shape)**
     - **No heat fixing so DON'T rinse or might wash off**
   - D. **Examples: Acid fuchsin, nigrosin**



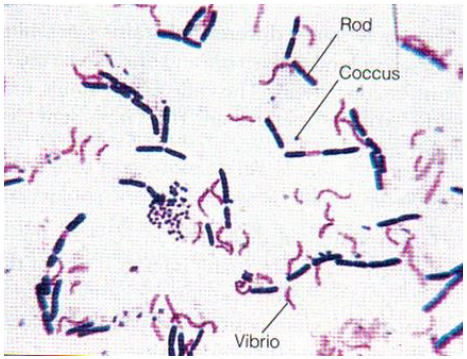
Mordant, Simple Stain, Differential Stain

3. **Mordant**: Substance used to cause more intense staining

   NOTE: This is not the stain that gives color, only helps the stain be more intense color



4. **Simple stain**: Single basic dye
   A. All microbes - same color
   B. Only for morphology (shape, arrangement, size)
5. **Differential Stain**: Use of >=2 stains to distinguish groups of bacteria
   A. Examples: gram stain, acid fast stain
   B. 1º = 1st stain applied. Then based on cell wall composition it is differentially removed, from some bacteria.
   C. 2º = 2nd stain applied & is taken up by cells that lost 1º stain - so that our eyes can see the cells.



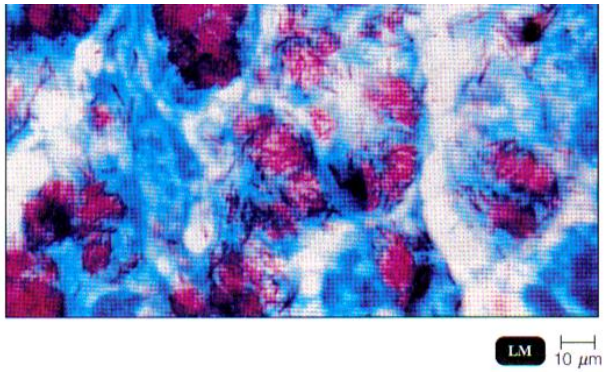


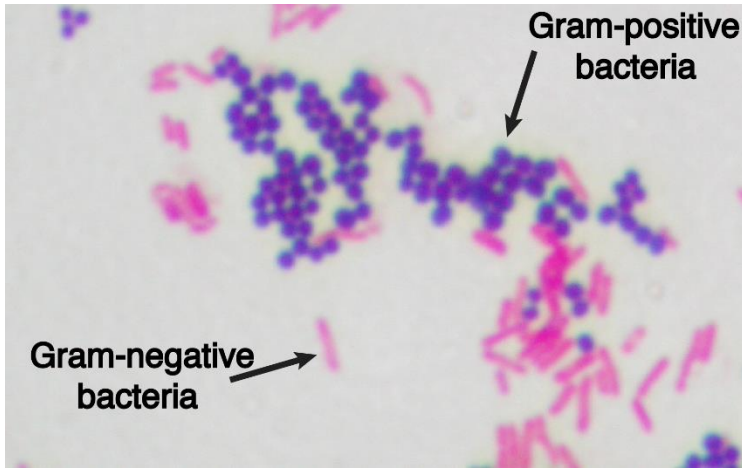
FIGURE 3.11 **Acid-fast bacteria.** The *Mycobacterium*

Gram Stain

6. **Gram Stain: Differential - due to cell wall differences**

   A. **GP = gram positive, purple, retain primary stain**
      - **Usually susceptible to penicillin**

   B. **GN = gram negative, red, loses primary stain & accepts counterstain**
      - **Resistant to penicillin**

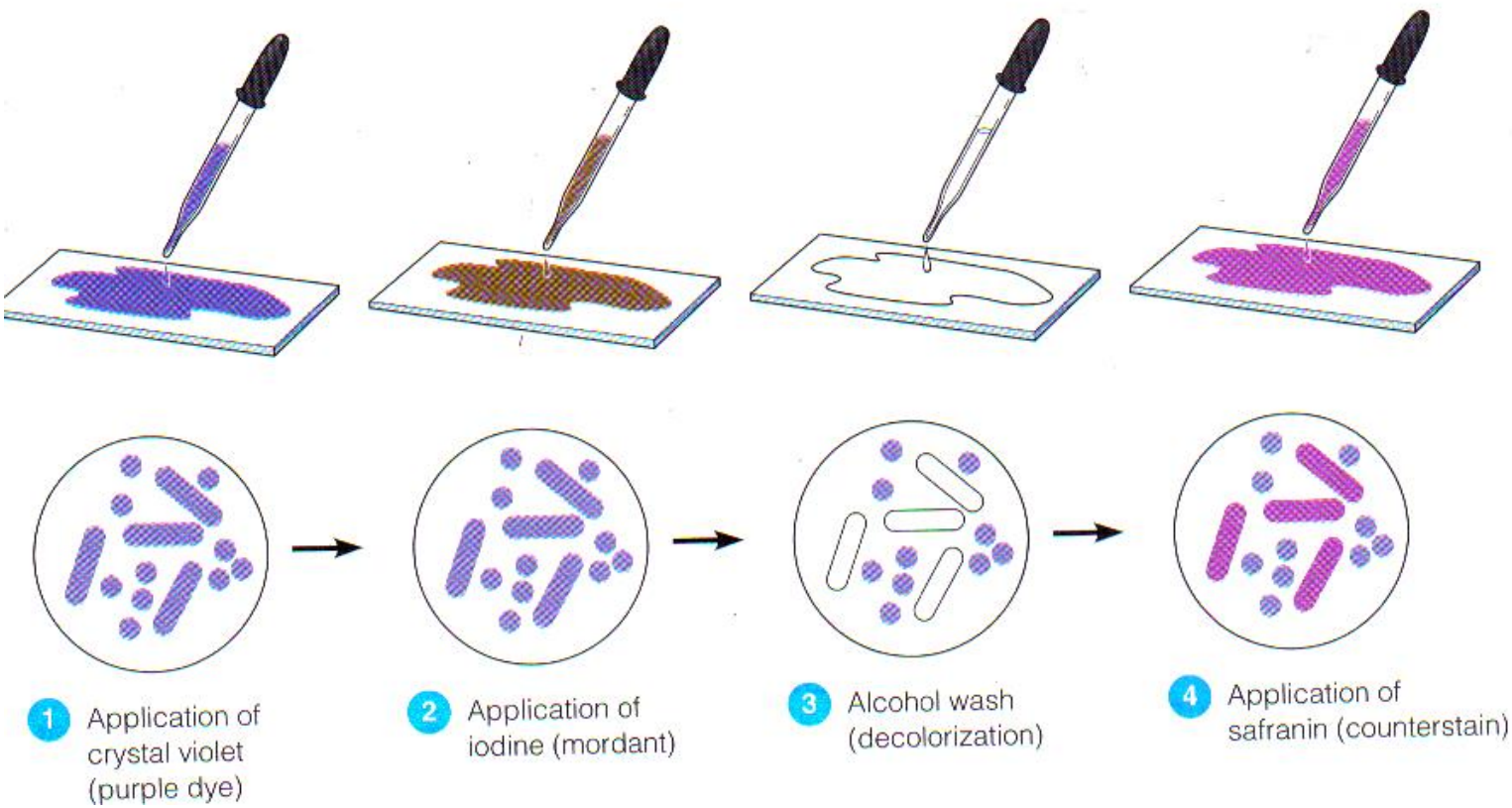


   C. **Staining problems**
      i. **Need young cultures**
      ii. **Decolorization timing is critical**
      iii. **Potential artifacts-structures/distortions that appear due to prep or staining procedures** **NOTE: this is potential problem w/all stains**

Most common stain in medical microbiology

Know procedure-steps, purpose of each step/stain, appearance of cells after each step, how cell wall causes differential staining (Chap 4)

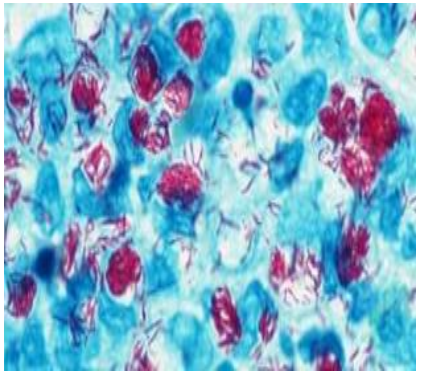
Gram Stain Diagram



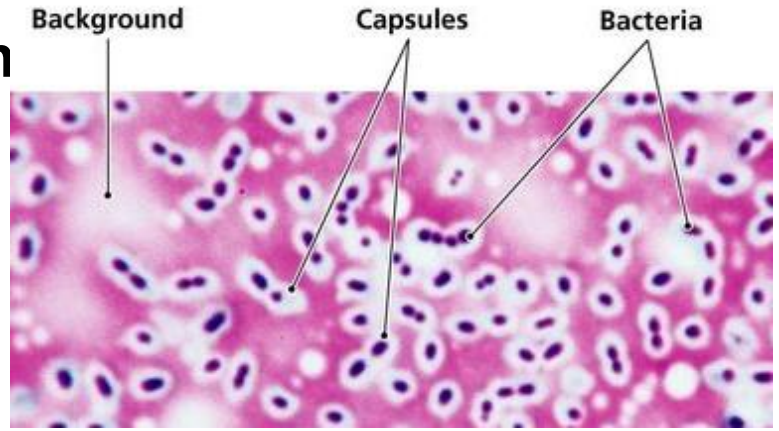
- Shapes above?
- GN or GP?
- Combine?
- GNR/GNB & GPC

Stains: Acid Fast & Capsule

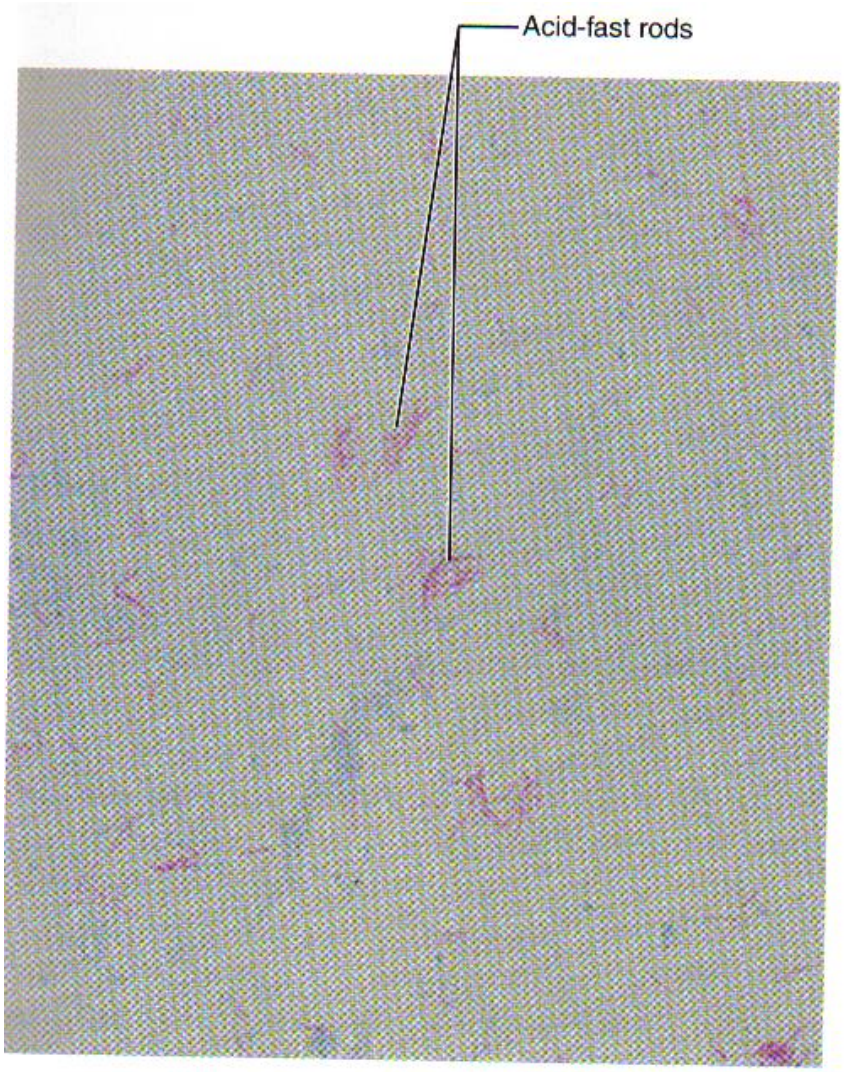
7. **Acid Fast Stain**
   - A. **Acid-fast positive = red (due to wax/lipid in cell wall)**
   - B. **Acid-fast neg = blue**
   - C. **ID *Mycobacterium* species, TB (tuberculosis)**

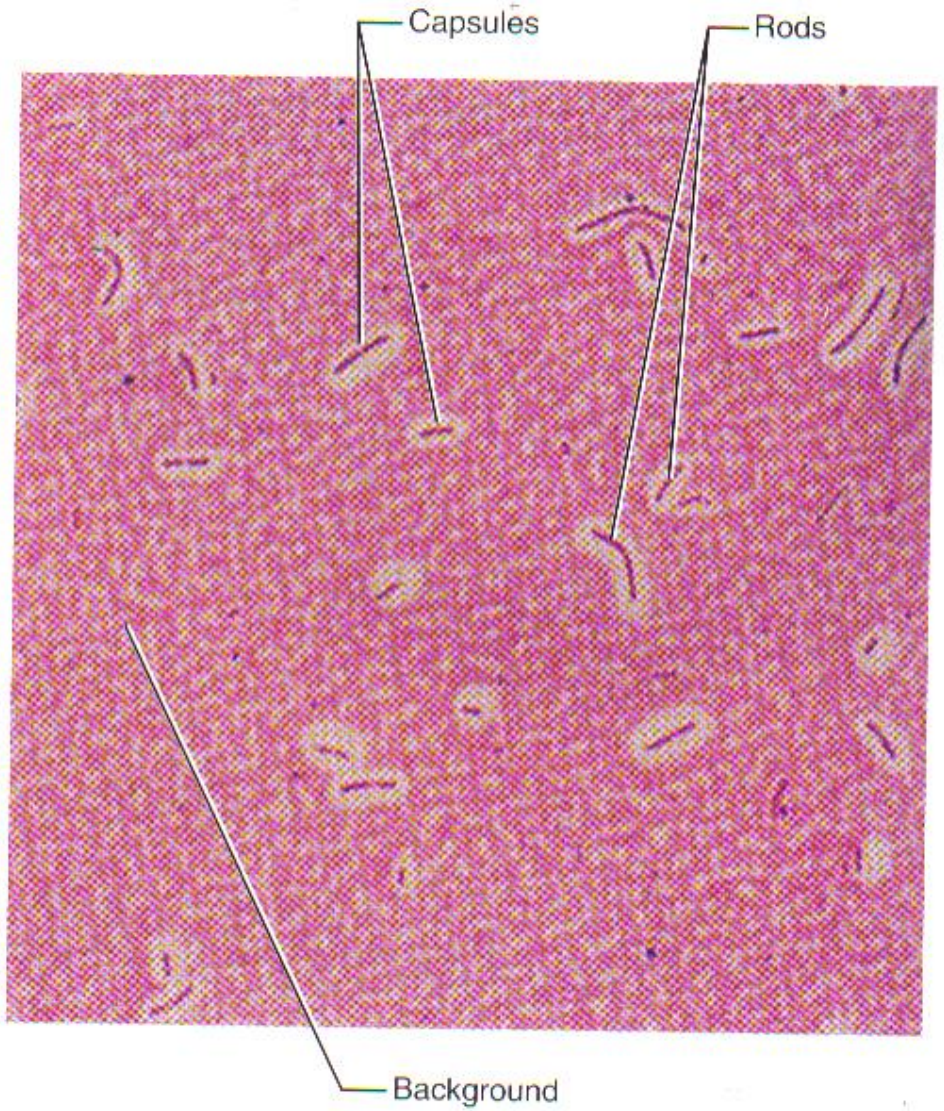


8. **Capsule Stain (w/ neg stain)**
   - A. **Capsule = gelatinous covering on**
   - B. **Variation w/2 stains:**
     - i. **Positive stains bacteria**
     - ii. **Negative stains background**
     - iii. **Clear "halo" of capsule left between the stains**
   - C. **Problems: capsule may wash off**



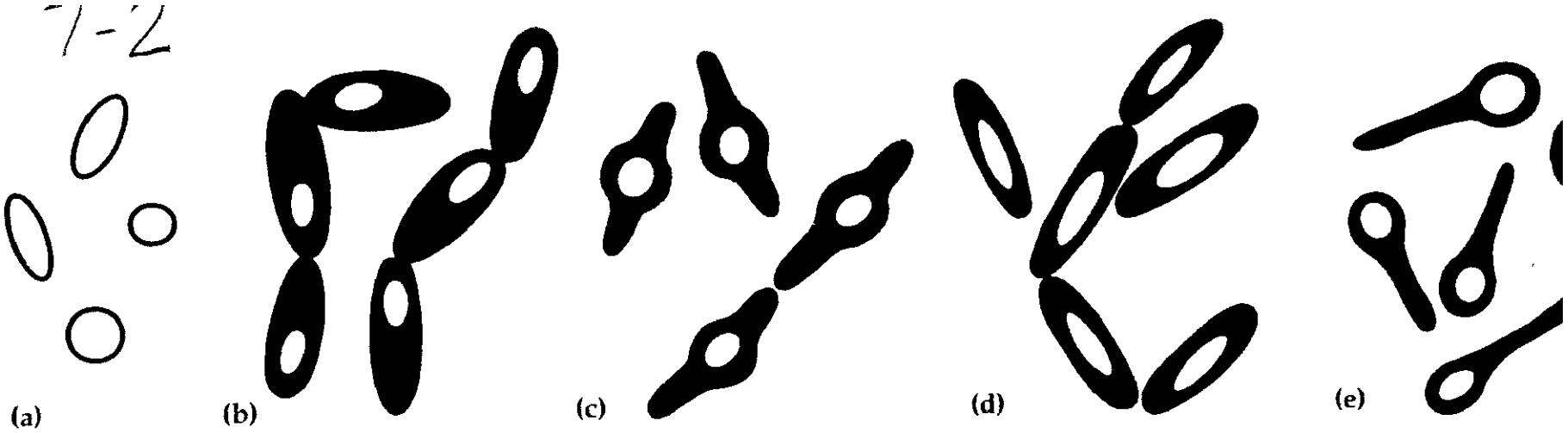
Pictures-Acid Fast & Capsule Stains



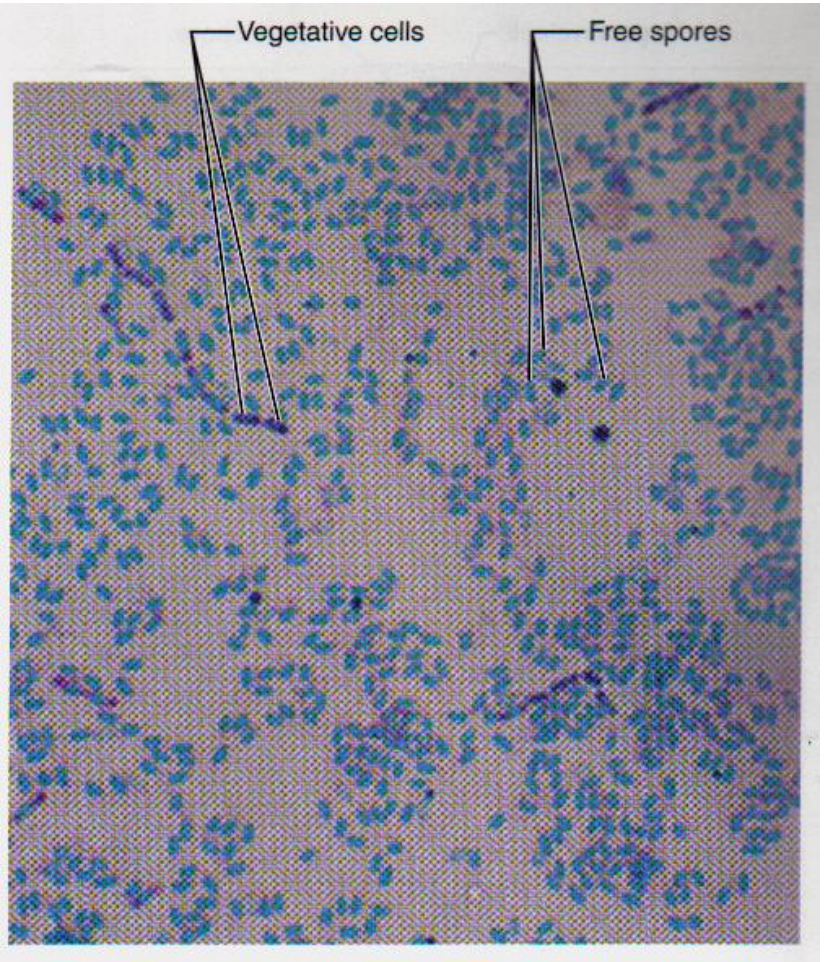


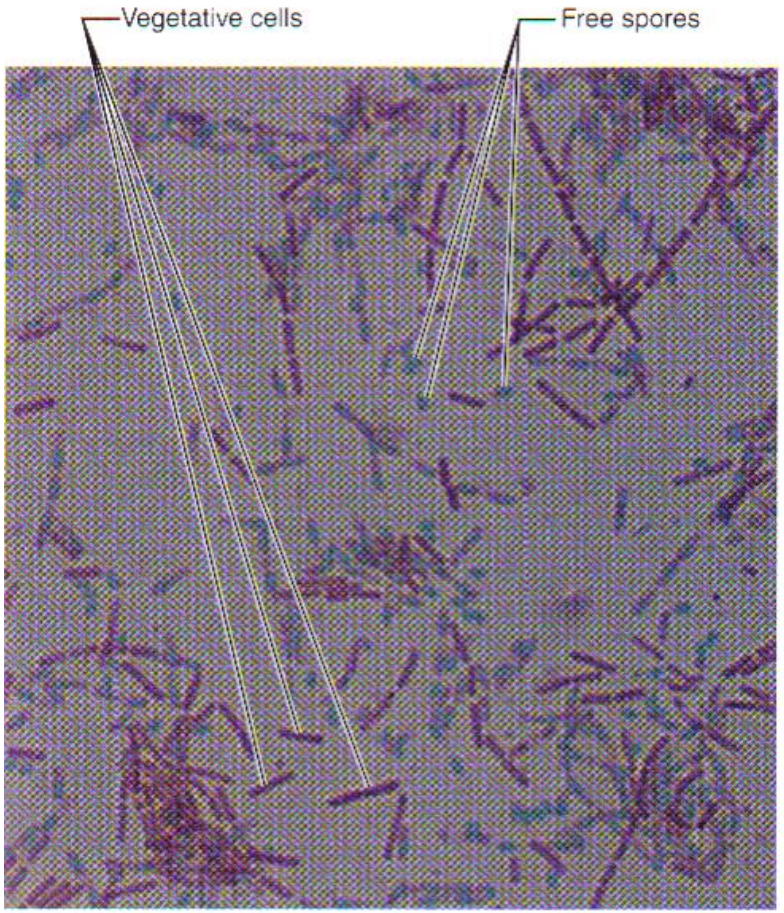
Stains: Endospore

9. **Endospore Stain**
   - A. **Endospore: Resistant, dormant, resting structure to protect microbe from adverse conditions**
     - i. **Position used to ID species; terminal, subterminal, central**
   - B. **Uses heat to force dye into stain-resistant endospores**



Stains; Endospore Pictures





1. Discuss vegetative vs. endospores. Free vs. still in cell.
2. Which of the 2 pictures above has been subjected to adverse conditions longer? Explain.

Stains; Flagella

10. **Flagella Stain**
    - A. **Flagella = whiplike structure for motility**
    - B. **# & arrangement used to ID bacteria**

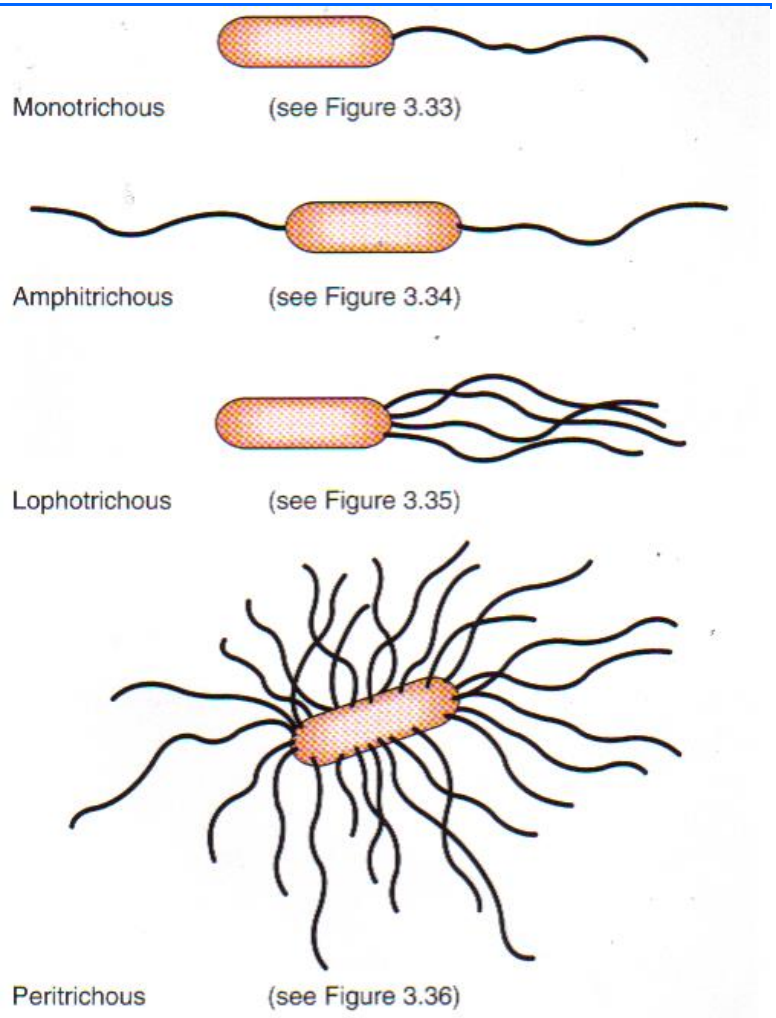


FIGURE 3.32 Flagella arrangements in bacteria. In *monotrichous* flagellation, a single flagellum is located at one end of the cell. In *amphitrichous* flagellation, a single flagellum is located at both ends of the cell. In *lophotrichous* flagellation, many flagella are grouped at one end of the cell. *Peritrichous* flagella are located all around the cell.

Chapter 4: Prokaryotic Cells

Prokaryote

1. Nucleoid region, no nucleus
2. Ribosomes, but no membranous organelles
3. 1 circular chromosome (DNA w/o histone)
4. Binary fission
5. Bacteria – cell wall peptidoglycan (AKA murein)
6. Archaea – no cell wall or pseudomurein

Fig 4.5a Prokaryotic Cell

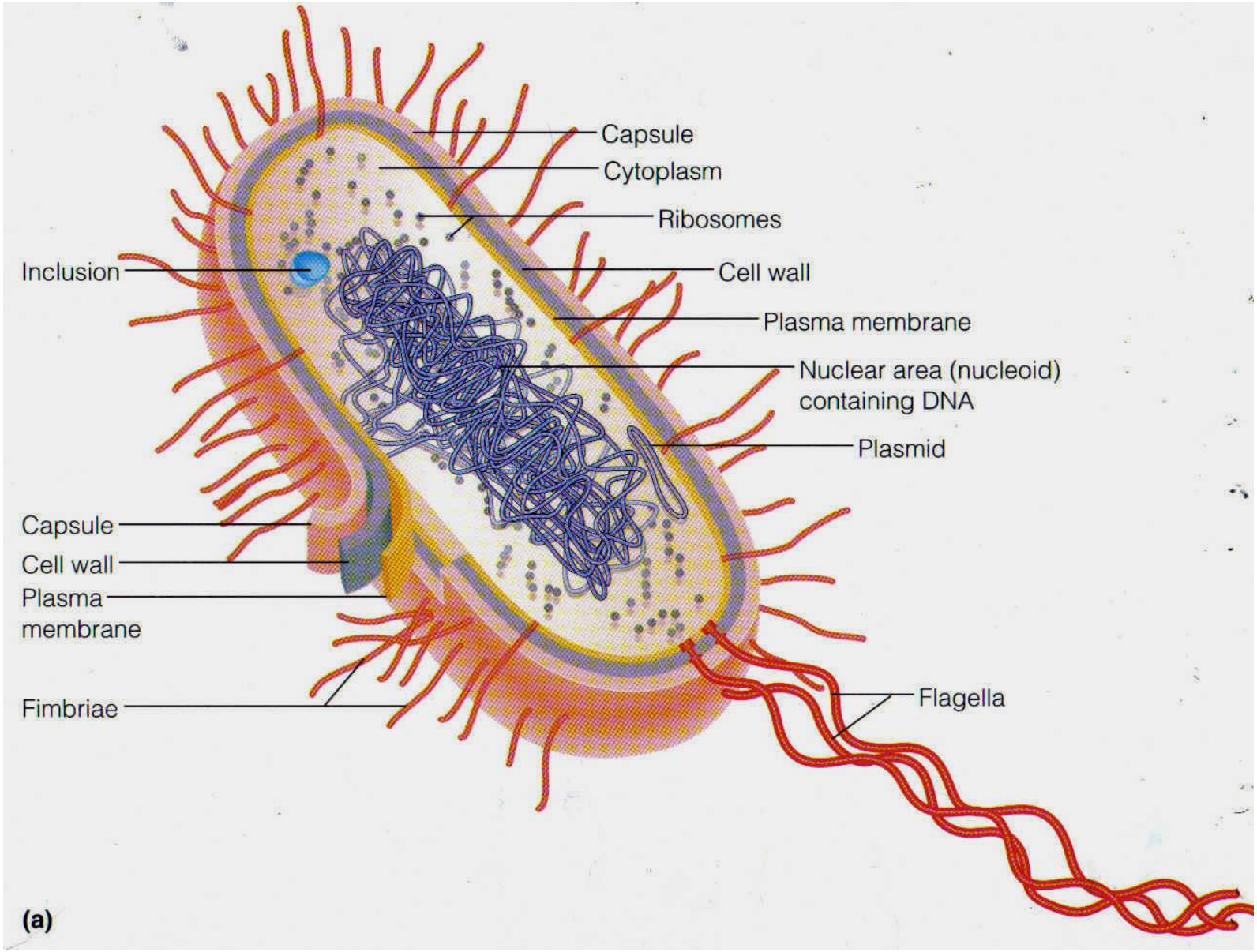
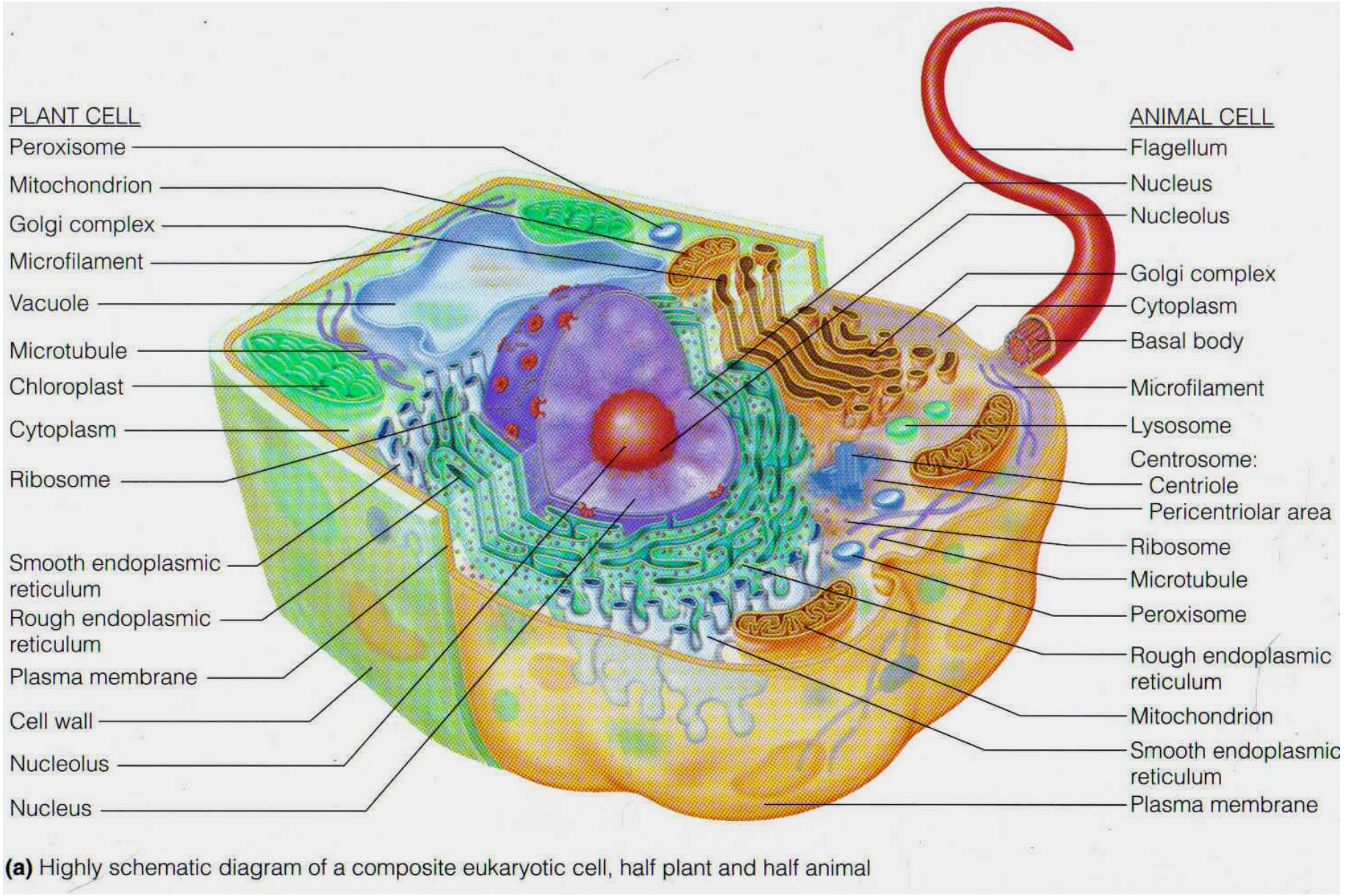
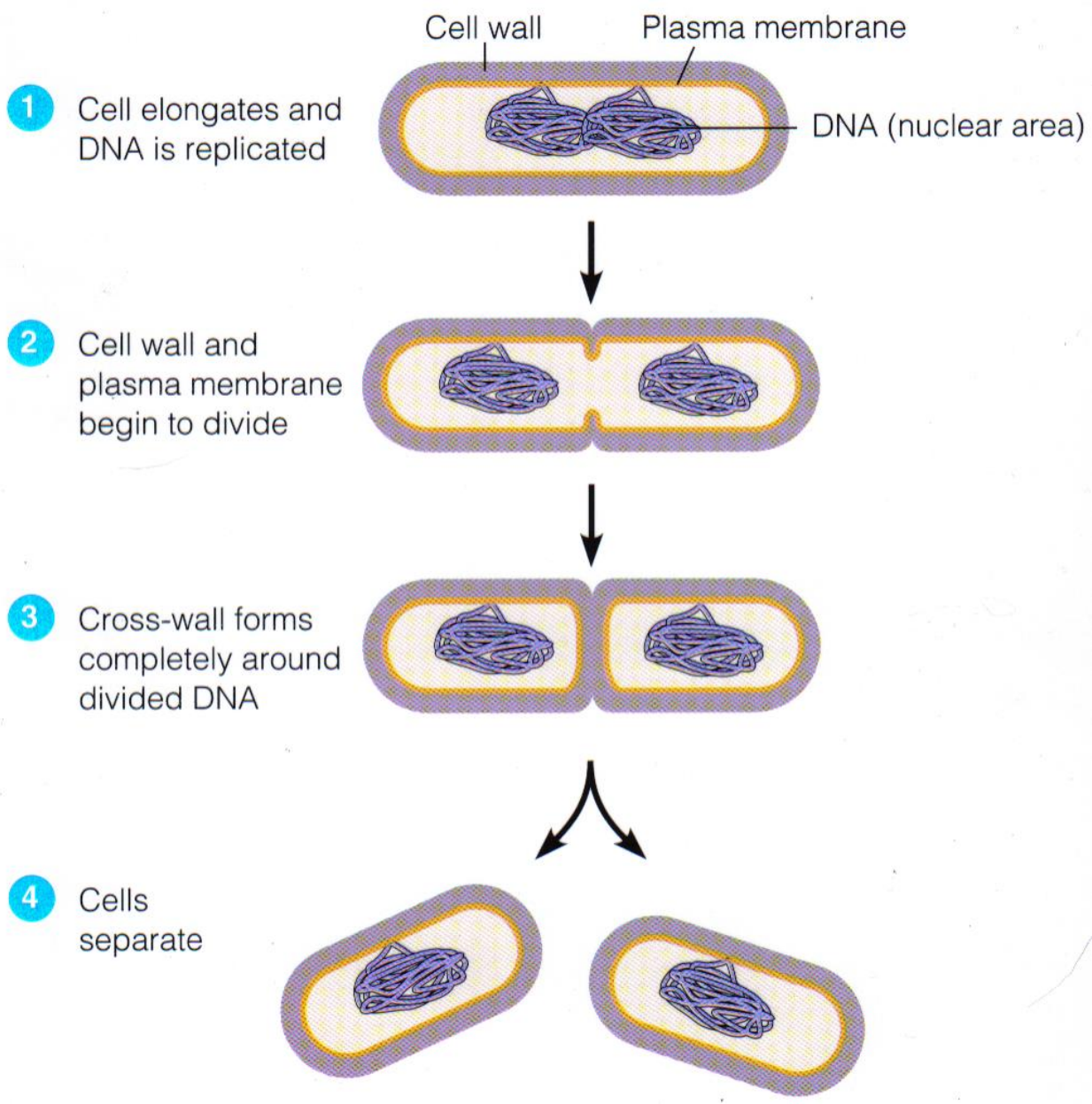


Fig 4.21a: Eukaryotic Cell for Comparison



(a) Highly schematic diagram of a composite eukaryotic cell, half plant and half animal

Fig 6.11a Binary Fission

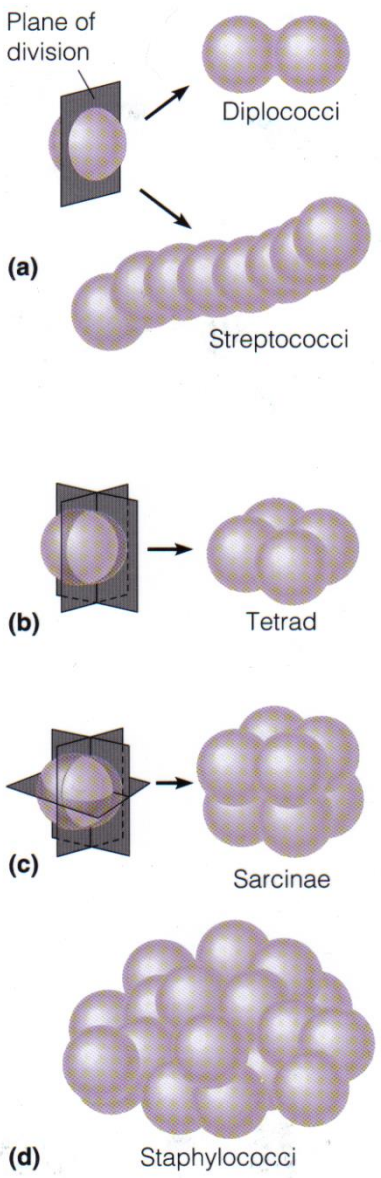


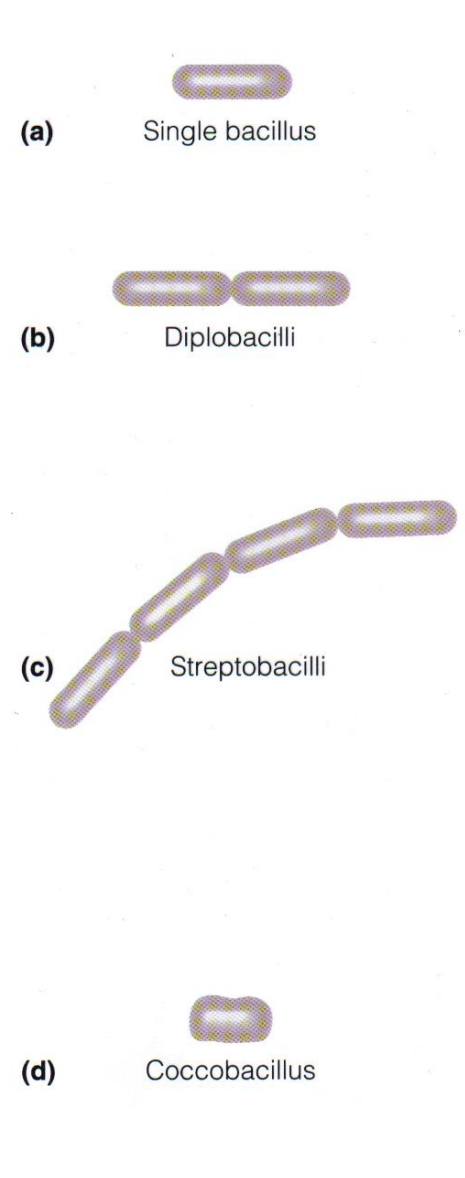
Arrangement

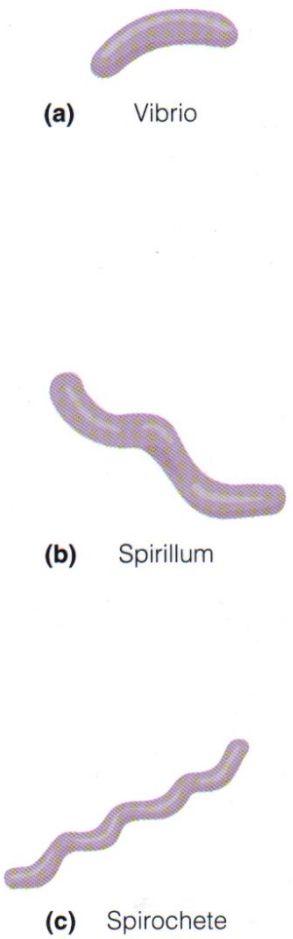
- Size of bacteria
- 0.2 – 8 um vs.
- resolution of light microscope?

Arrangement Review:

- Shape (Morphology)?
- Arrangements?







Cell Wall - Bacteria

Bacterial Cell Wall

1. **Function:**
   - A. Maintains shape
   - B. Prevents rupturing due to osmotic pressure
2. **Clinical importance**
   - A. Site of antibiotic activity
   - B. Differentiate bacteria, ie. GN vs. GP
3. **Made of: Peptidoglycan**

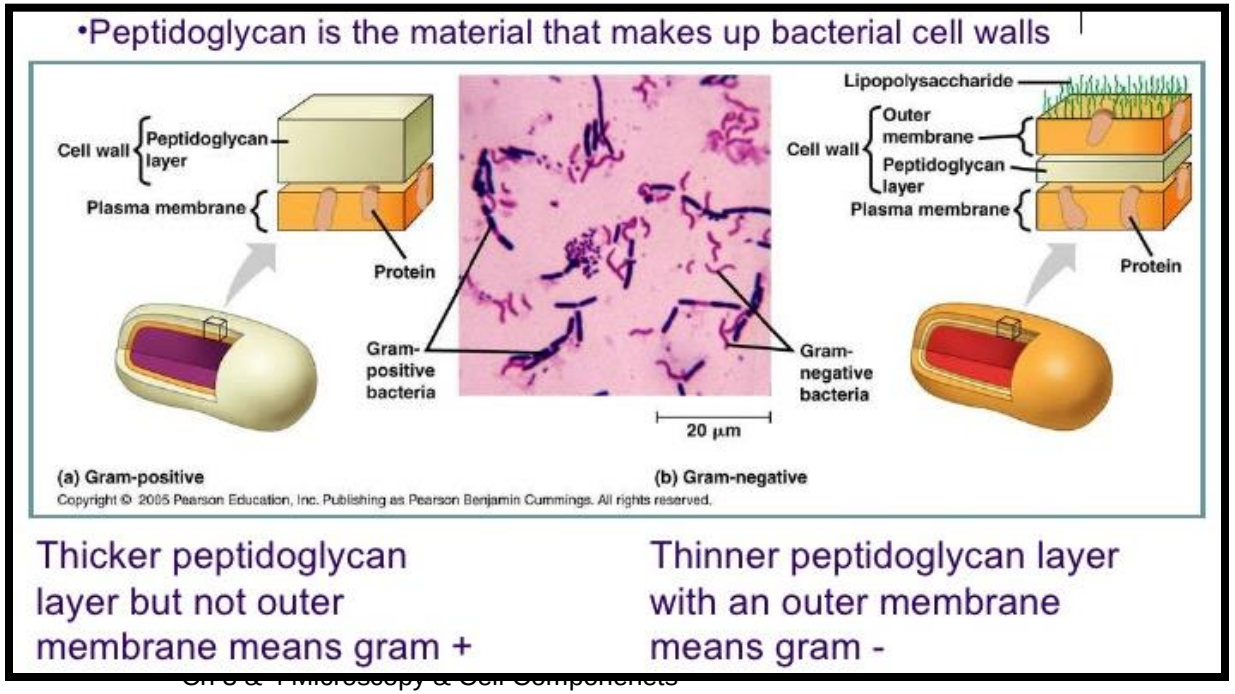
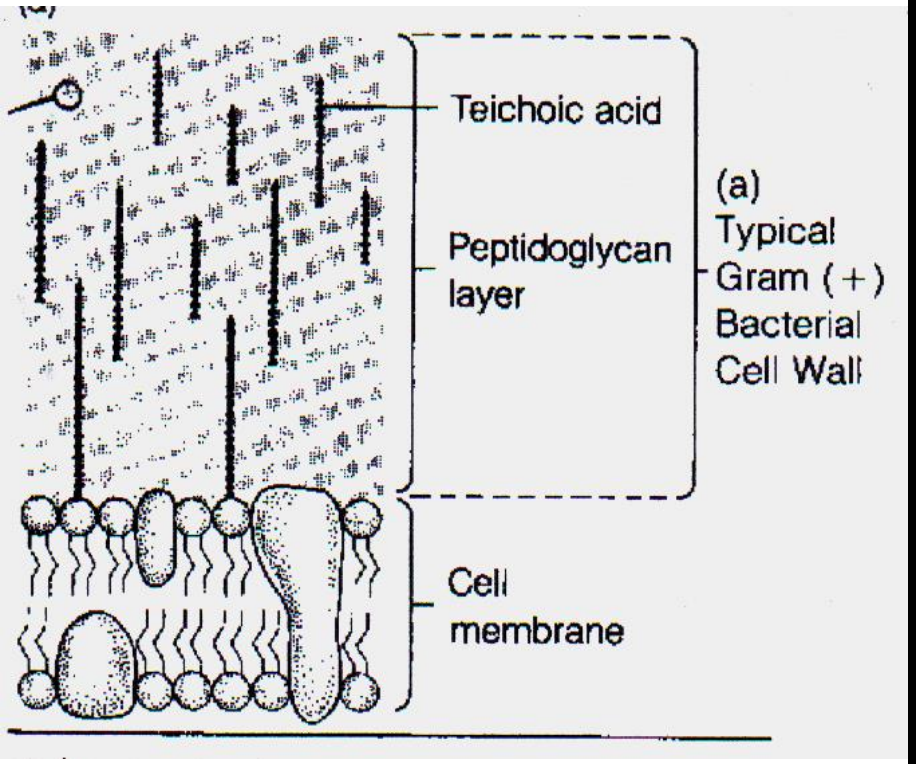


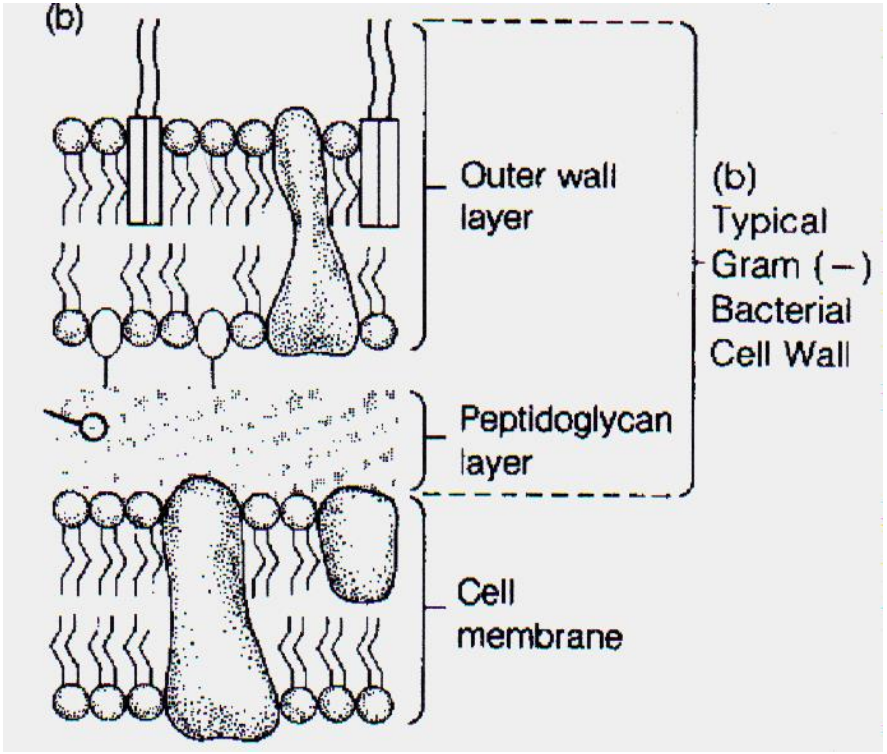
Diagram –Cell Wall Diagrams

Outside Cell



Inside Cell

Outside Cell



Inside Cell

Table – GP vs. GN Cell Wall Characteristics

GP Wall	GN Wall
1. THICK peptidoglycan	1. Thin
2. Contains teichoic acids	2. None
3. None	3. OUTER Wall Membrane A. Evades phago, barrier to penicillin & enzymes B. Contains porins C. LPS – endotoxin ⇒ fever & shock
4. None	4. Periplasm- space between outer & plasma membrane (where peptidoglycan is) A. Contains degradative enzymes

Fig 4.12 GP Cell Wall

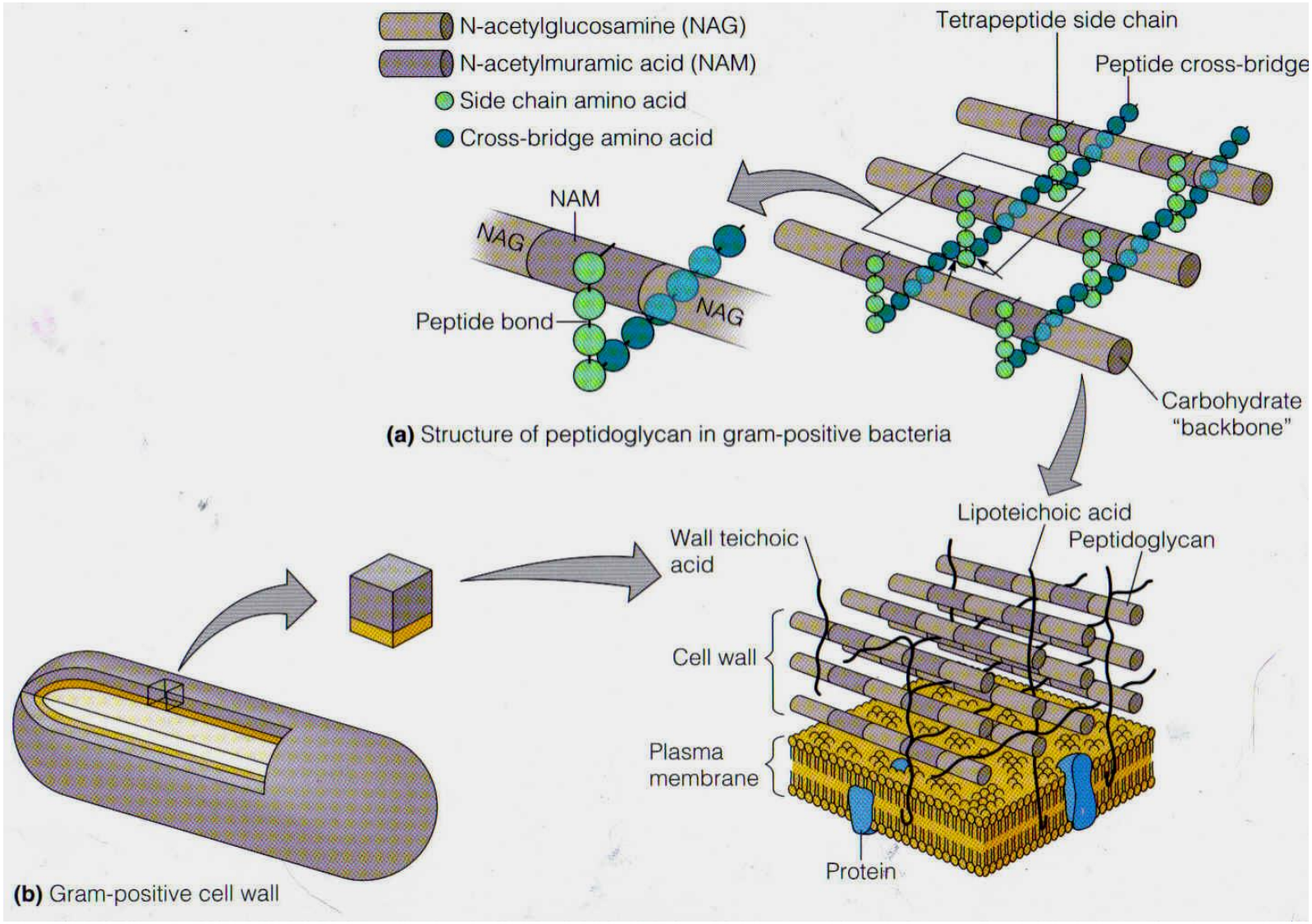
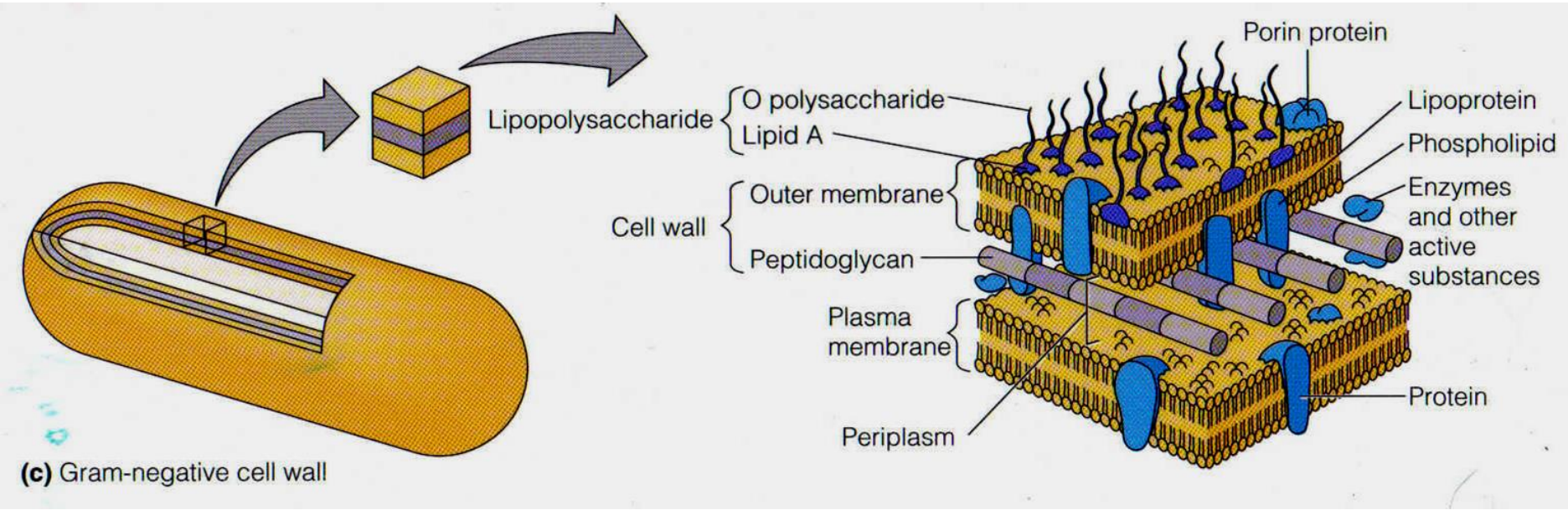


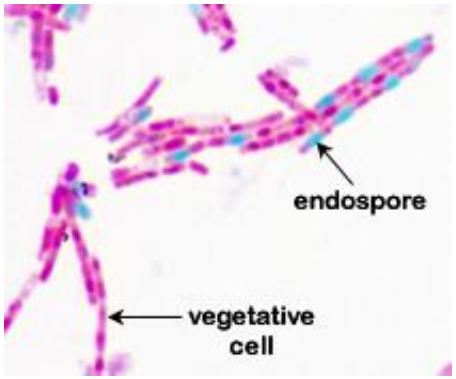
Fig 4.12 GN Cell Wall

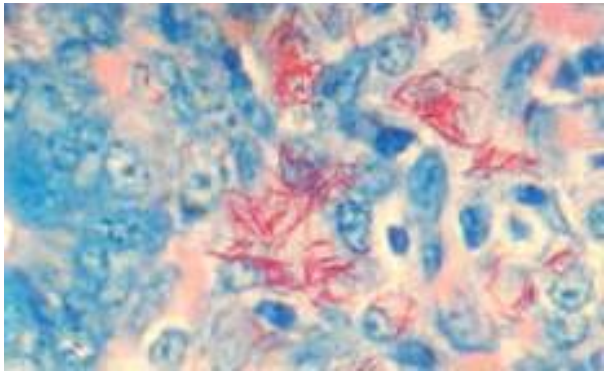


Gram Stain & the Cell Wall

Cell Wall & gram stain

1. **Iodine = mordant. Forms large crystals w/crystal violet**
2. **Alcohol**
   A. **GP-dehydrates & ⇑ density of thick wall-crystals can't leave**
   B. **GN-dissolves outer membrane & dehydration leaves hole in thin peptido-crystals exit & cell colorless**
3. **GP falsely stain GN when cell wall damaged due to age, heat damage during fixing, decolorizing with alcohol too lone**
4. **The following 2 stains should only be done IF the gram stains= shows GPR/GPB:**
   A. **Endospore stain: Bacillus & Clostridium**
   B. **Acid Fast stain: Mycobacterium (TB)**

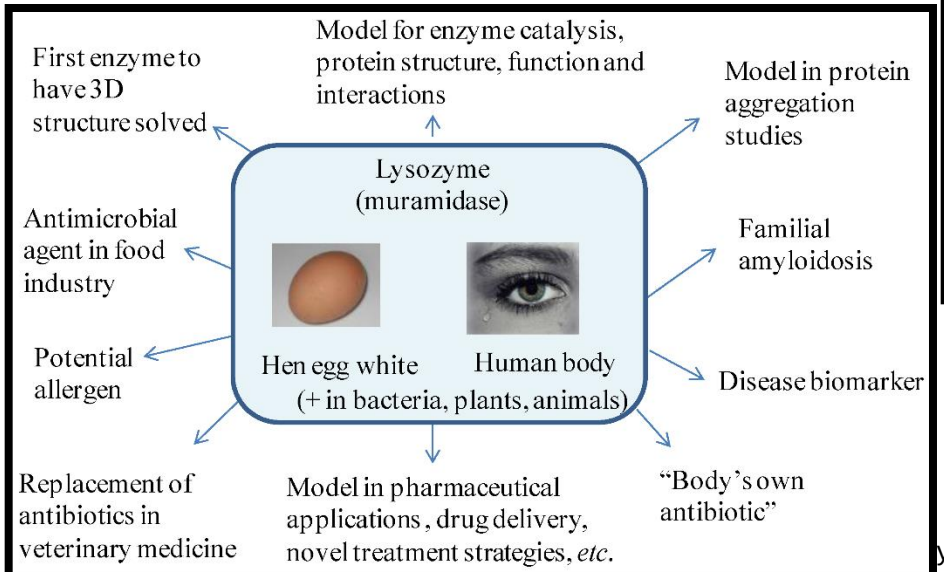


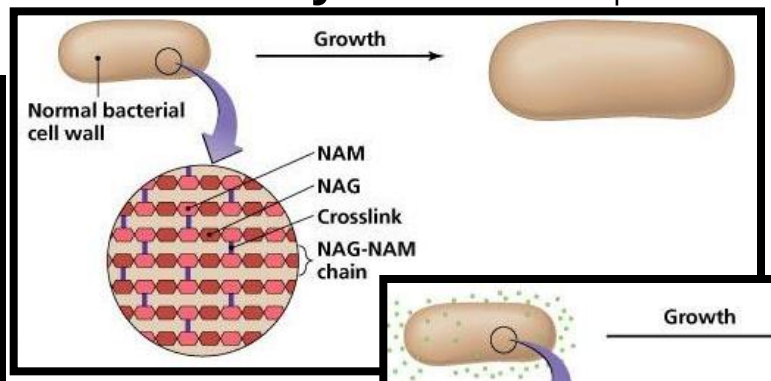


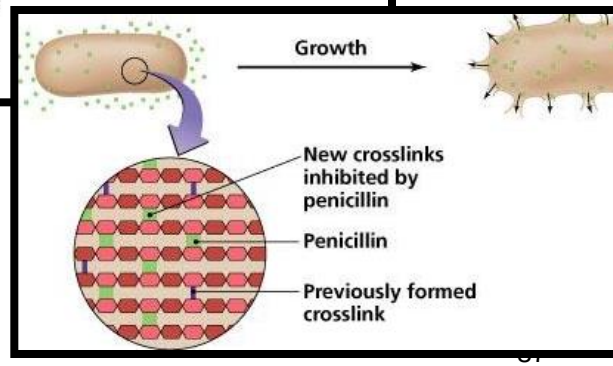
Chemicals & the Cell Wall

Chemical Effects on Cell Wall

1. Lysozyme: Digestive enzyme in saliva, tears, mucus
   A. Most effective on GP - hydrolyzes peptido bonds
2. Penicillin – How it works:
3. Affects GP cells ONLY
   A. Interferes w/peptide links in the peptidoglycan in GP cell wall
   B. Causes the cell wall to be weak
   C. When water enters cell through osmosis, the weak cell wall bursts & the cell undergoes osmotic lysis due to ↑ osmotic pressure







Gram Stain Diagram-Again

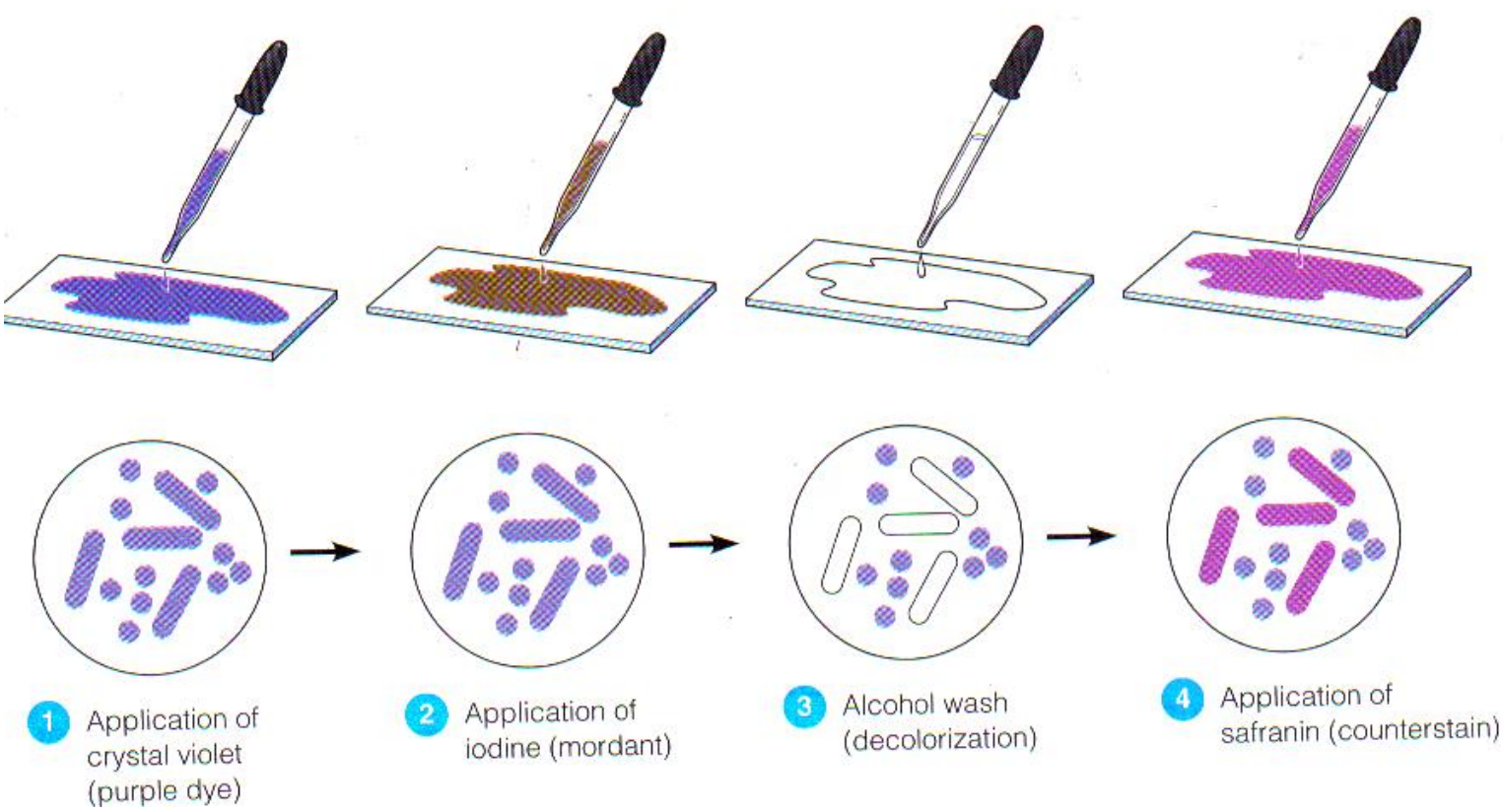
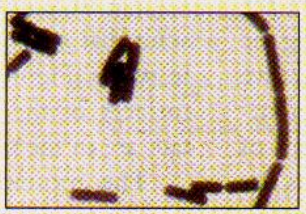


Table 4.1 Comparison of GP & GN bacteria

Characteristic	Gram-Positive	Gram-Negative
	 LM 2 μm	 LM 2 μm
Gram reaction	Retain crystal violet dye and stain dark violet or purple	Can be decolorized to accept counterstain (safranin) and stain red
Peptidoglycan layer	Thick (multilayered)	Thin (single-layered)
Teichoic acids	Present in many	Absent
Periplasmic space	Absent	Present
Outer membrane	Absent	Present
Lipopolysaccharide (LPS) content	Virtually none	High
Lipid and lipoprotein content	Low (acid-fast bacteria have lipids linked to peptidoglycan)	High (due to presence of outer membrane)
Flagellar structure	2 rings in basal body	4 rings in basal body
Toxins produced	Primarily exotoxins	Primarily endotoxins
Resistance to physical disruption	High	Low
Cell wall disruption by lysozyme	High	Low (requires pretreatment to destabilize outer membrane)
Susceptibility to penicillin and sulfonamide	High	Low
Susceptibility to streptomycin, chloramphenicol, and tetracycline	Low	High
Inhibition by basic dyes	High	Low
Susceptibility to anionic detergents	High	Low
Resistance to sodium azide	High	Low
Resistance to drying	High	Low

Atypical Cell Walls

Atypical Cell Walls

1. **Mycobacteria- High wax in wall**
   - **A. GP**
   - **B. AFB stain for TB**
2. **Archea; No cell wall or pseudomurein**

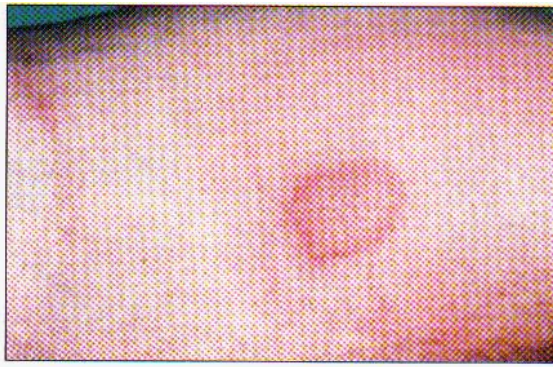
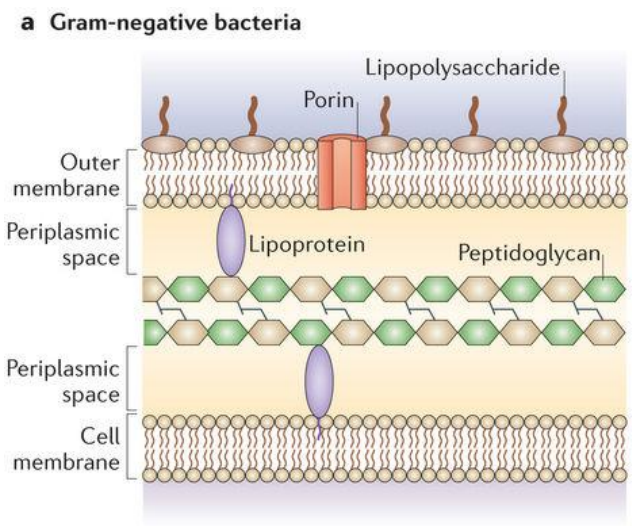
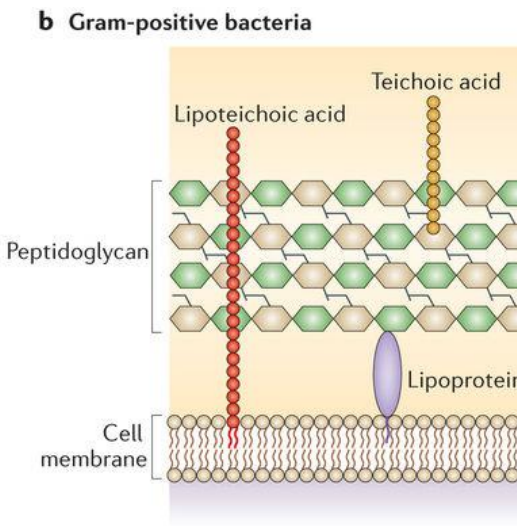
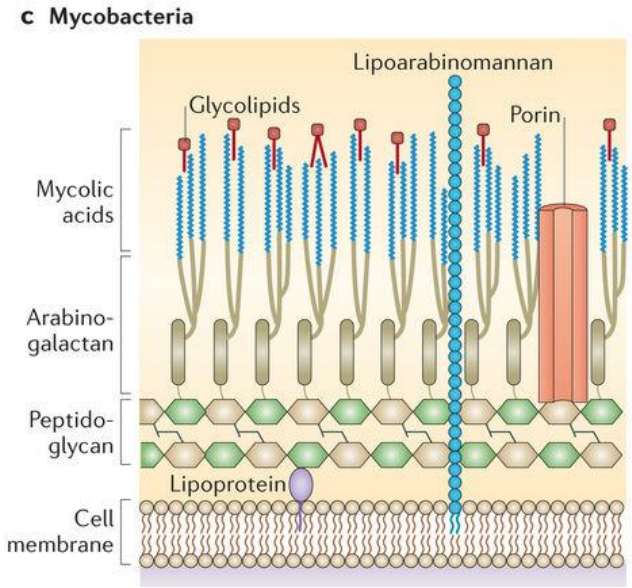
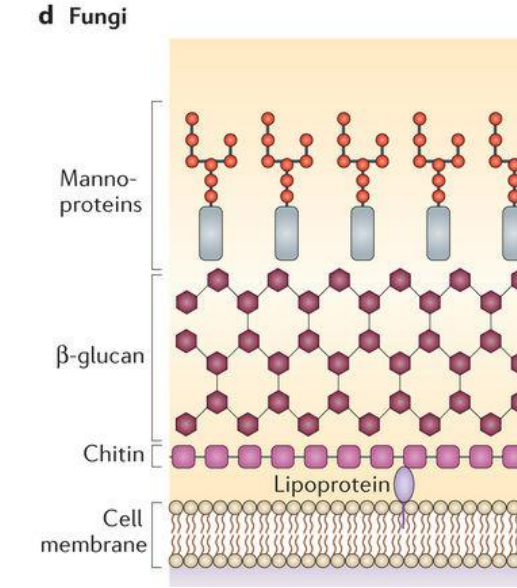


FIGURE 24.11 The tuberculin skin test on arm.





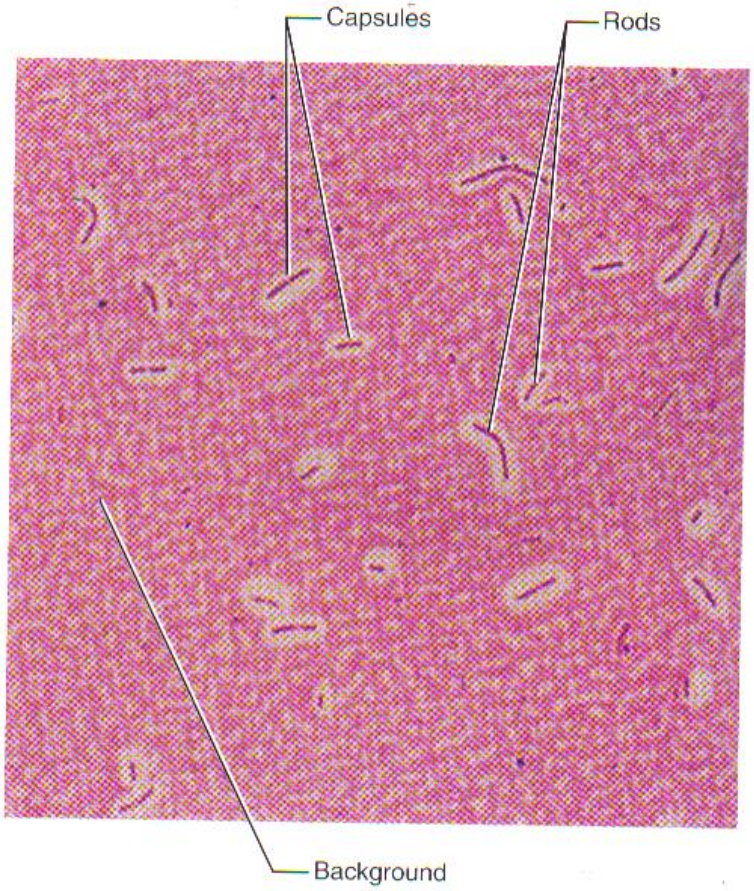




Structures External to Cell Wall

External Structures

1. **Glycocalyx/Capsule:**
   - A. **EPS (Extracellular polysaccharide & polypeptide polymer**
   - B. **⇑ virulence; evade phagocytosis, adherence, dehydration protection**
   - C. **Negative Stain, but uses 2 dyes**
     - i. **Basic stains organism**
     - ii. **Acidic stains background**
     - iii. **Halo between the stains is the capsule**



External Filamentous Structures

2. Table:

Flagella	Axial Filaments	Fimbrae	Pili
Movement	Movement (cork-screw)	Adherence	Transfer DNA "sex pili"
Bacilli & spiral	Spirochetes	GN	GN
Made of flagellin		pilin	pilin
Monotrichous - 1 at a pole Amphitrichous-tuft each pole Lophotrichous->=2 at 1 pole Peritrichous-all around	Spiralled around cell within outer sheath	Few to 100s	0-1/cell

Bacterial Conjugation (Sex Pilli)

https://www.youtube.com/watch?v=O-EdX4MaMFE

Flagella Diagrams & Photos

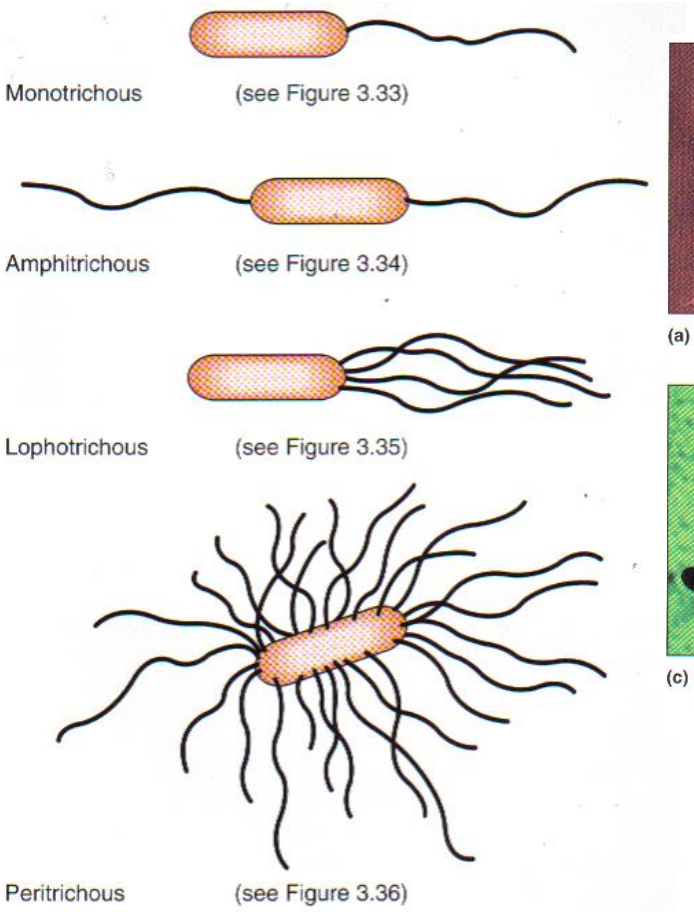


FIGURE 3.32 Flagella arrangements in bacteria. In *monotrichous* flagellation, a single flagellum is located at one end of the cell. In *amphitrichous* flagellation, a single flagellum is located at both ends of the cell. In *lophotrichous* flagellation, many flagella are grouped at one end of the cell. *Peritrichous* flagella are located all around the cell.

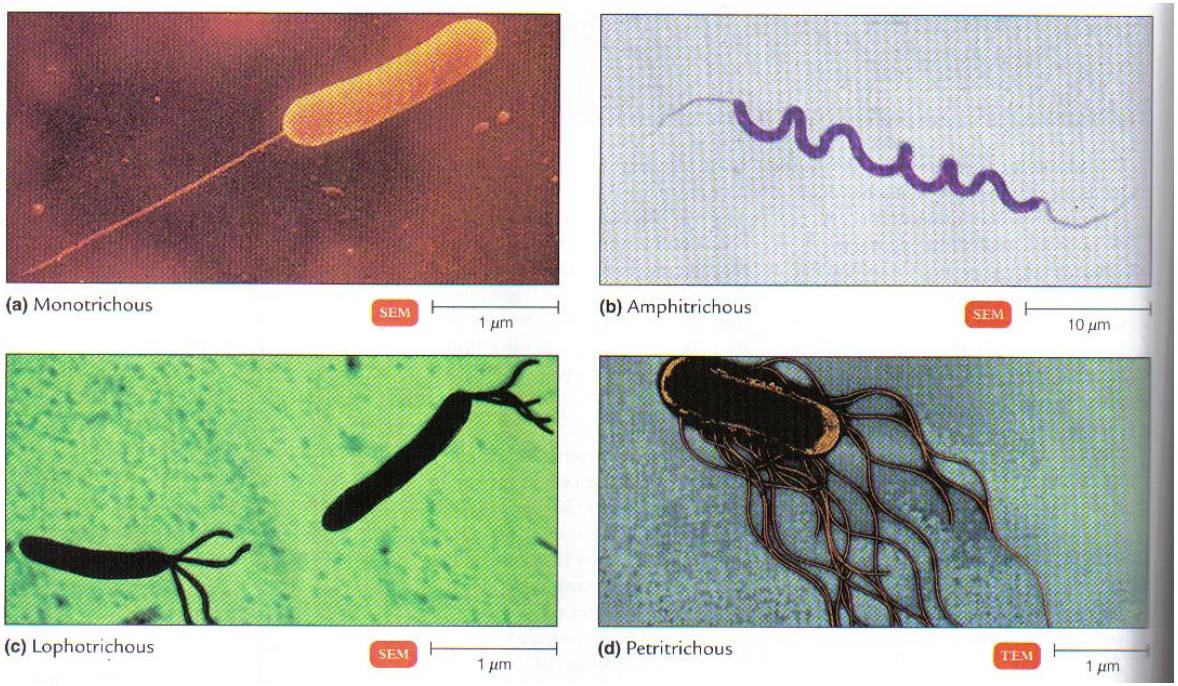
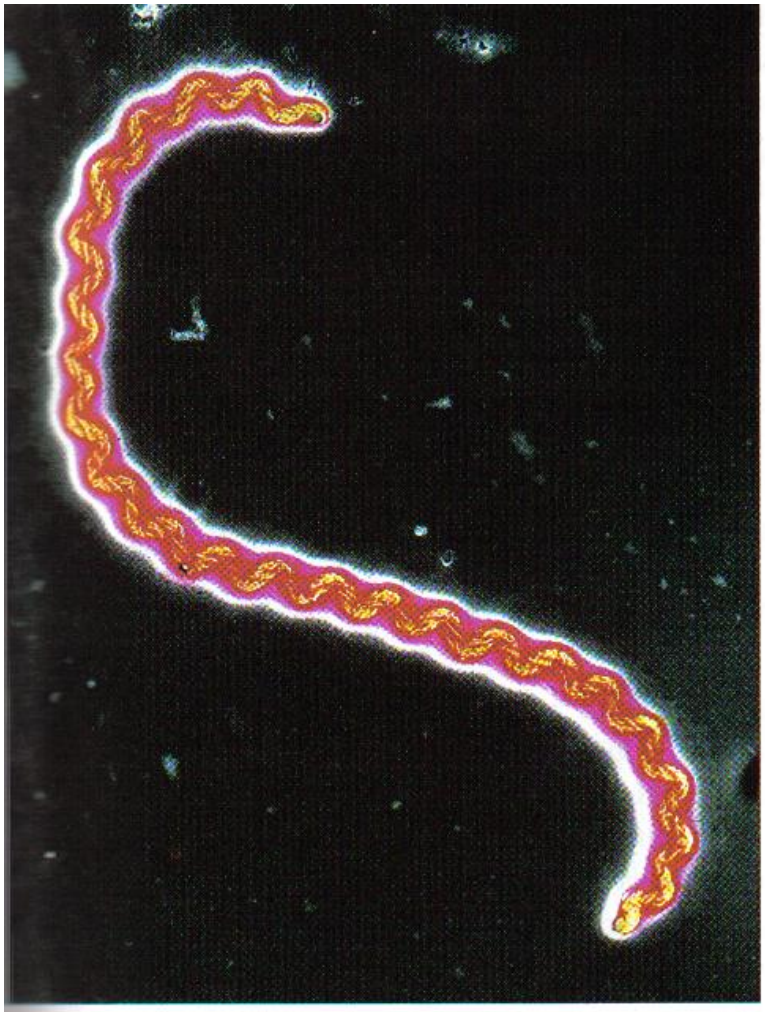


Diagram-Axial Filament



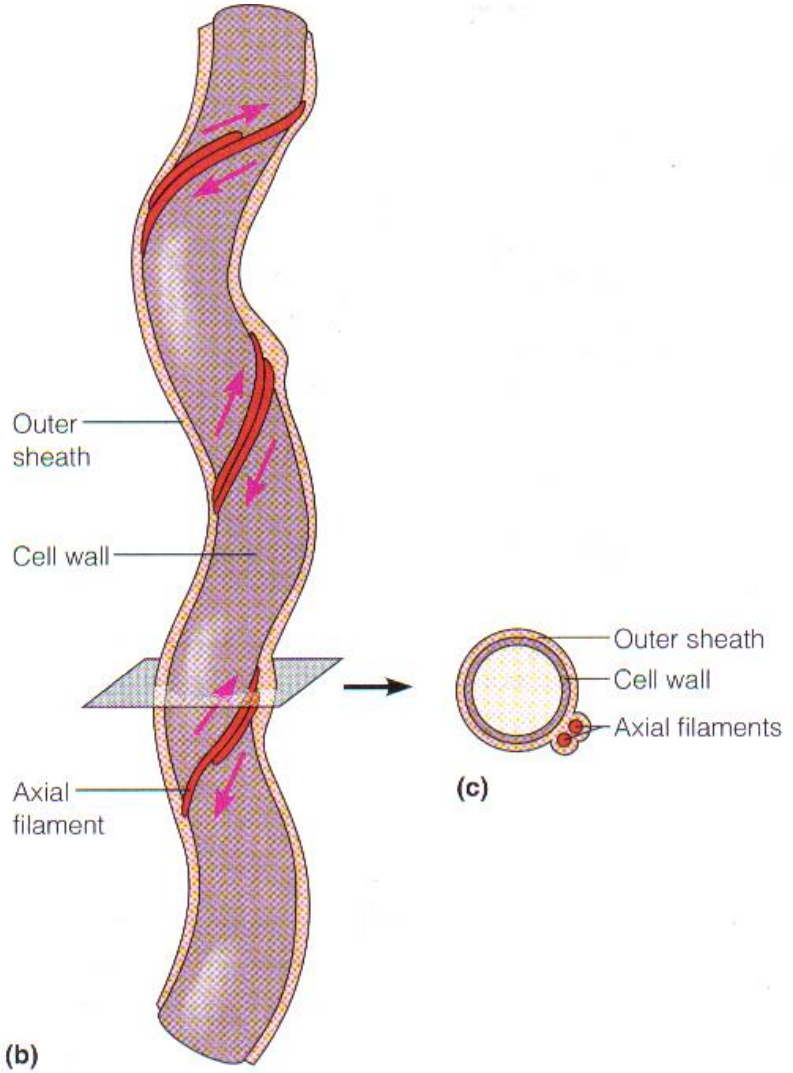
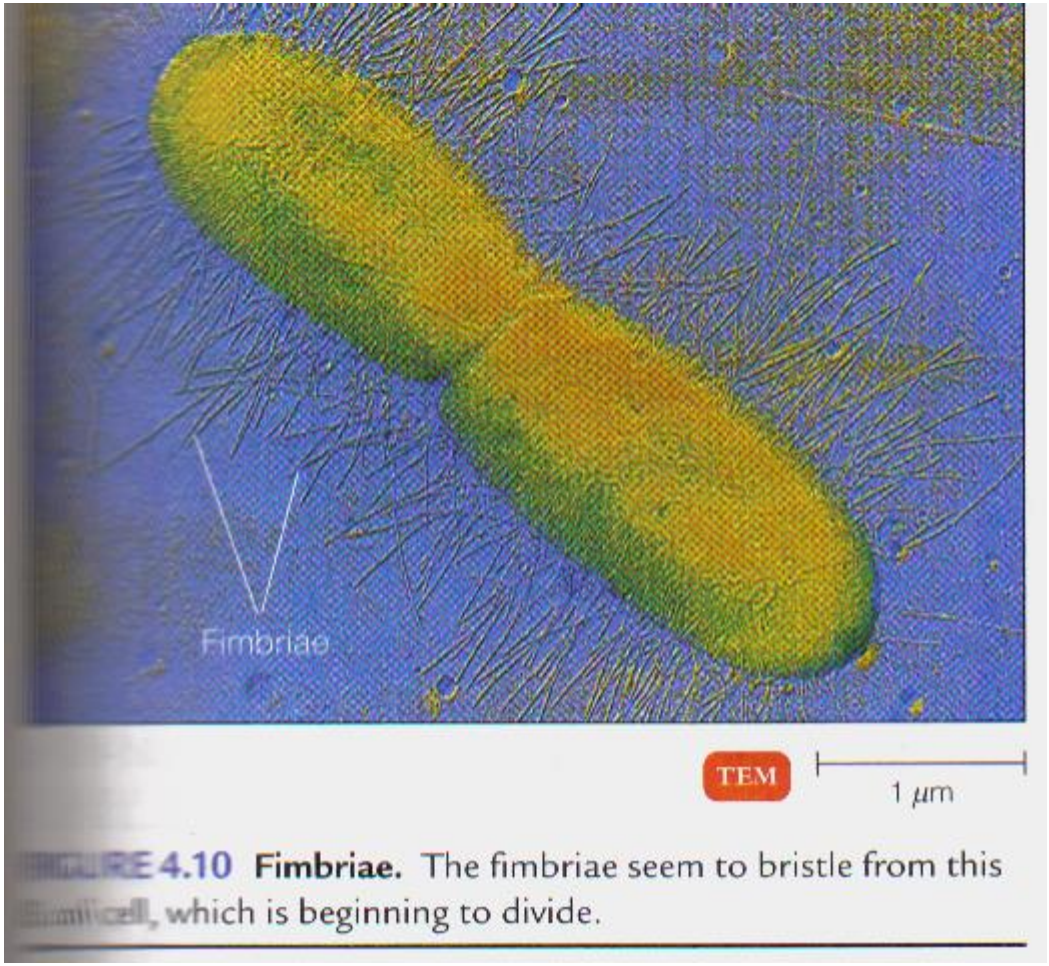


FIGURE 4.9 Axial filaments. **(a)** A photomicrograph of the spirochete *Leptospira*, showing an axial filament. **(b)** A diagram of axial filaments wrapping around part of a spirochete. **(c)** A cross-sectional diagram of the spirochete, showing the position of axial filaments.

Photo-Fimbriae



4.10 Fimbriae. The fimbriae seem to bristle from this cell, which is beginning to divide.

Fig 8.26 Bacterial Conjugation

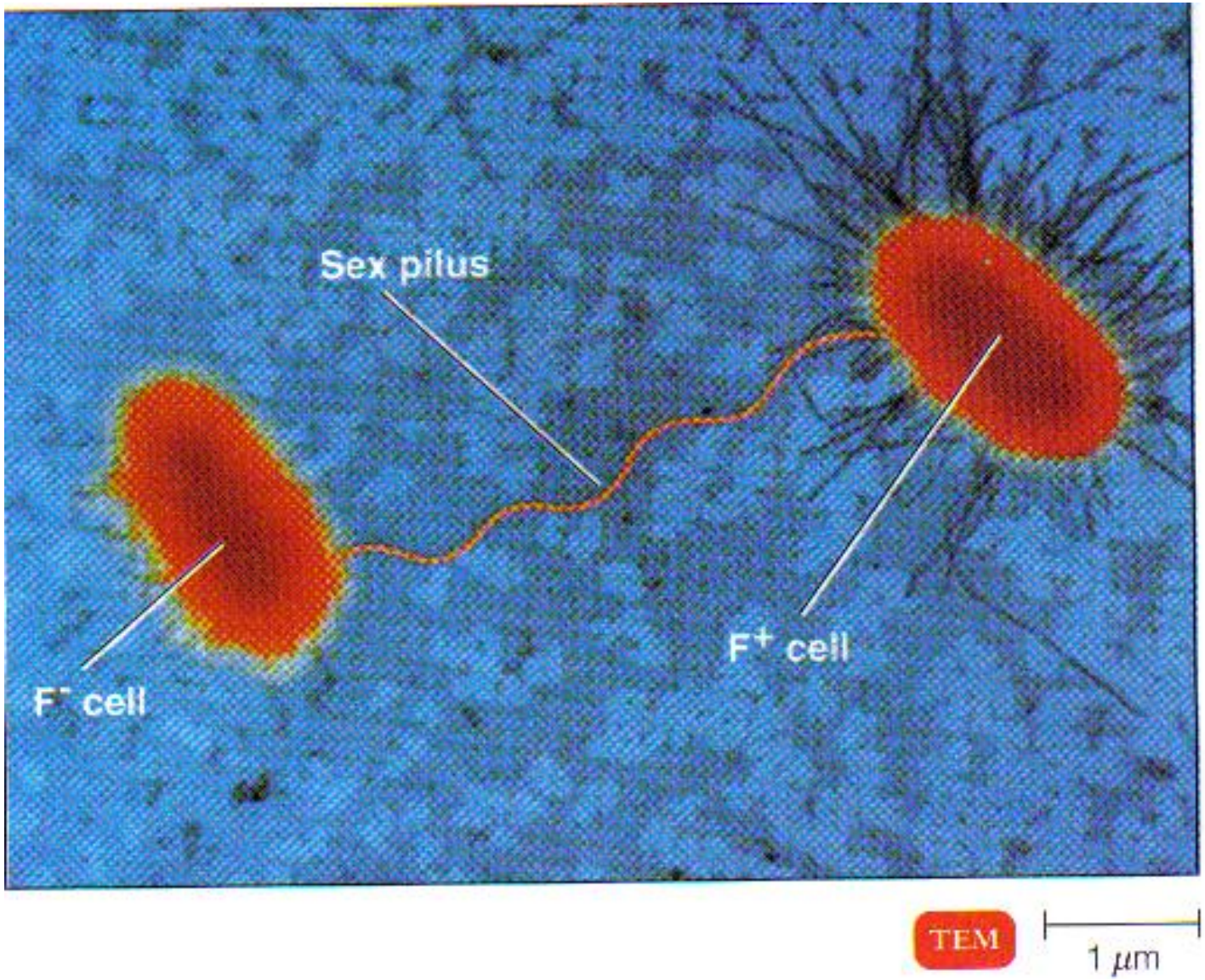
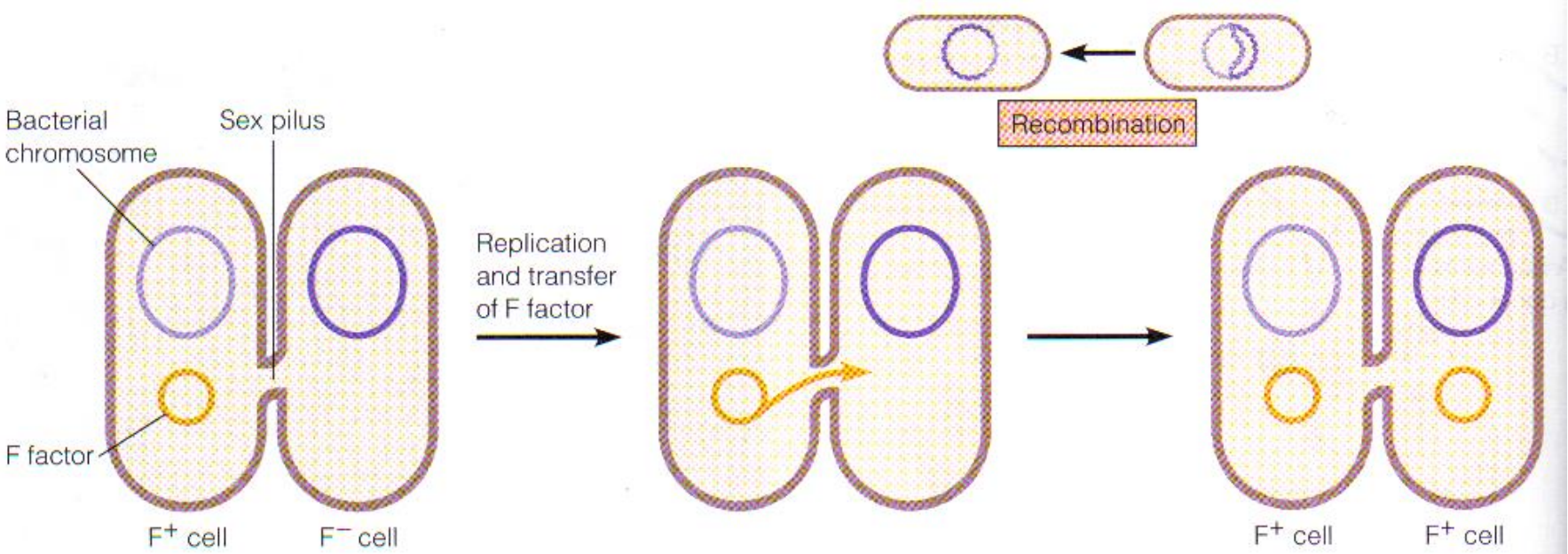


Fig 8.27 Conjugation in E. coli

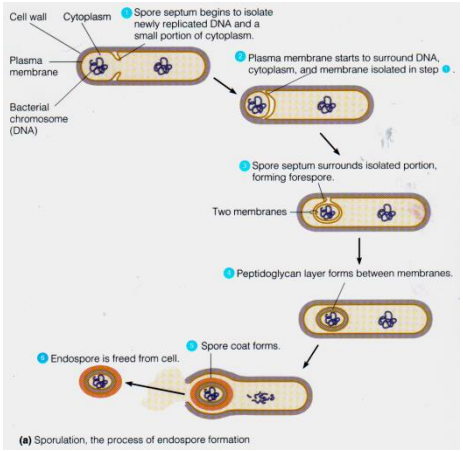


(a) When an F factor (a plasmid) is transferred from a donor (F^+) to a recipient (F^-), the F^- cell is converted into an F^+ cell.

Endospores

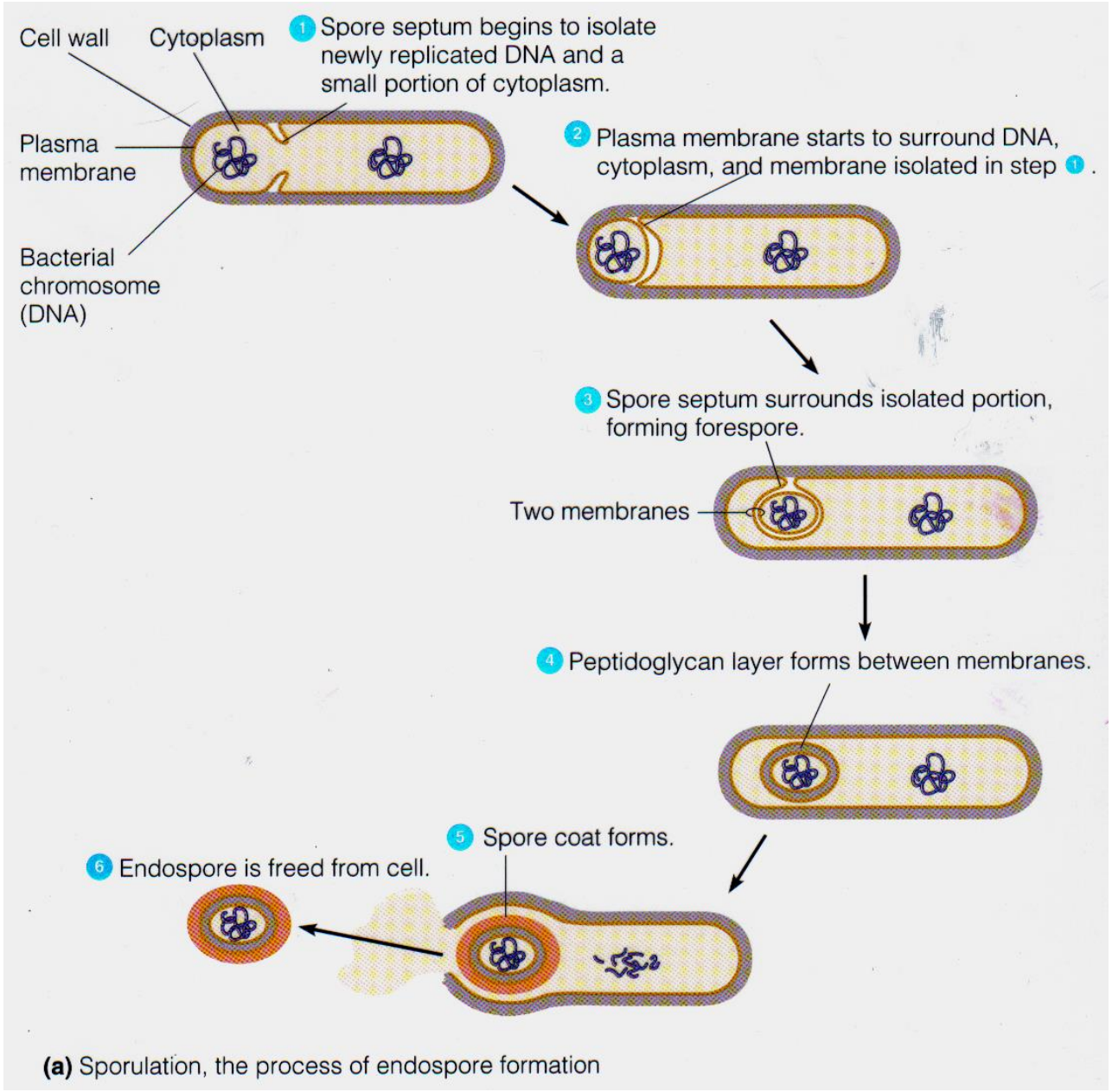
Structure Internal to Cell Wall

1. **Endospores: Resistant "resting" structures to survive adverse conditions**



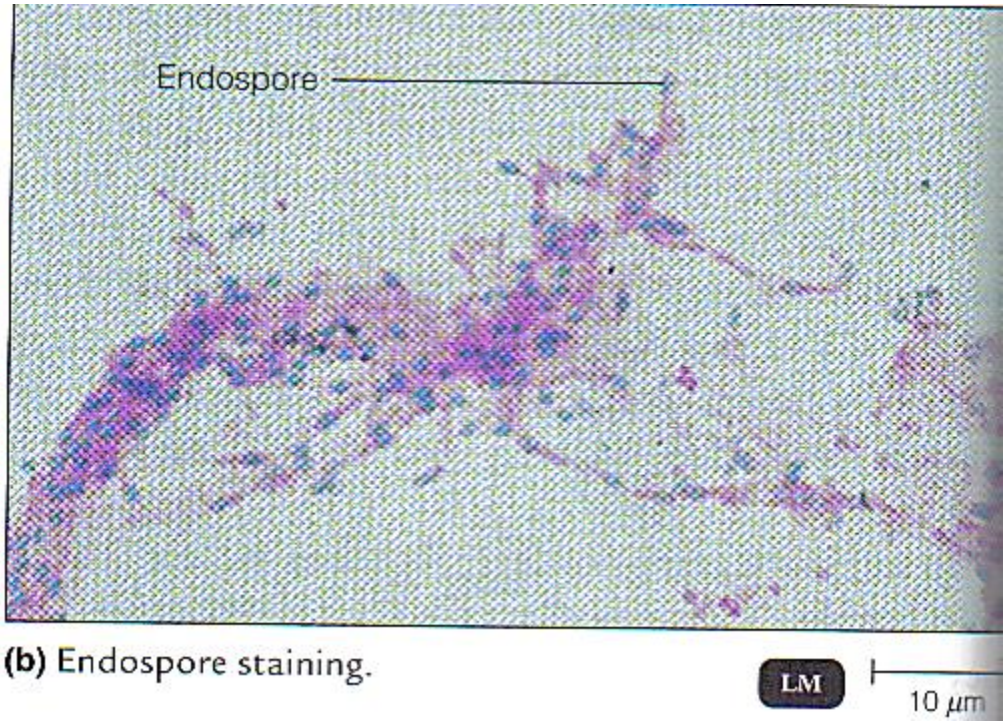
   A. **Only 2 genera, both GPR: Bacillus & Clostridium**
   B. **Sporulation / Sporogenesis: when bad conditions**
   C. **Germination – return to vegetative state**
   D. **NOT reproduction**
   E. **Location: Terminal, ~~subterminal~~, central**
   F. **Survive dehydration, heat, chemicals (antibiotics, disinfectants), improper canning (toxins ⇒ food poisoning) , etc**
   G. **Stains:**
      i. **Gram- appear clear**
      ii. **Endospore Stain:**
         - **Primary: basic stain w/heat forces stain into endospore**
         - **Rinse: removes stain from rest of slide/organism**
         - **Counterstain: basic stain colors rest of organism**

Fig 4.20a Endospore Formation

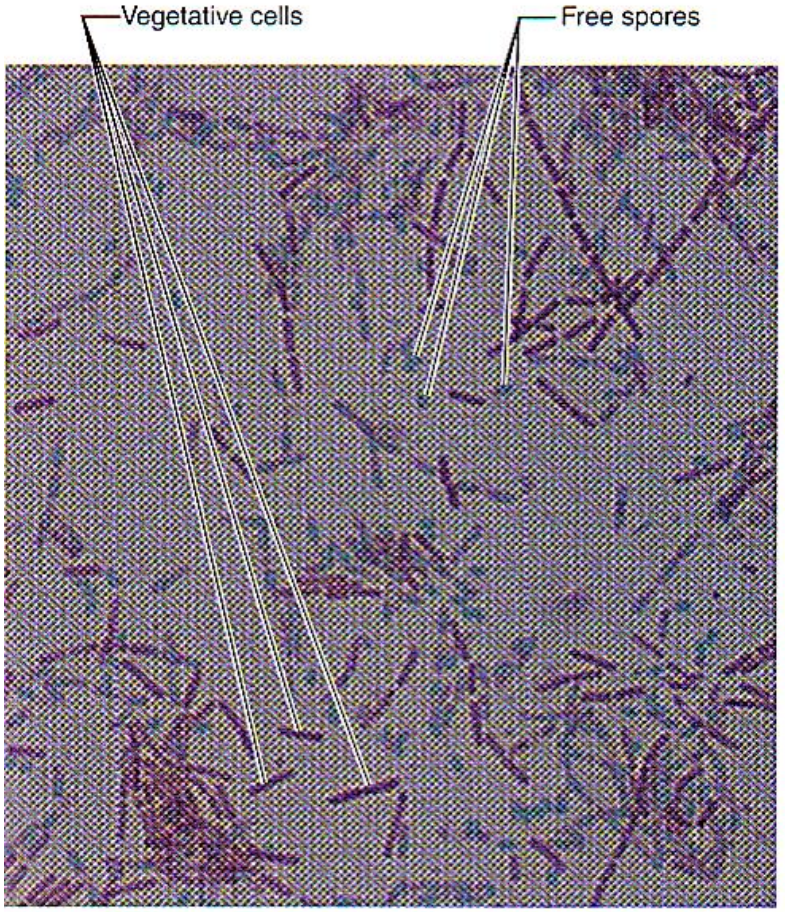


(a) Sporulation, the process of endospore formation

Endospore Stain Pictures



(b) Endospore staining.



Endospore stain

https://www.youtube.com/watch?v=ZlsPakEQeX0

Plasma/Cytoplasmic Membrane

2. **Plasma Membrane**
   - A. **Bilayer of phospholipids w/ proteins**
   - B. **Selectively permeable**
   - C. **Controls what gets in & out of cell**

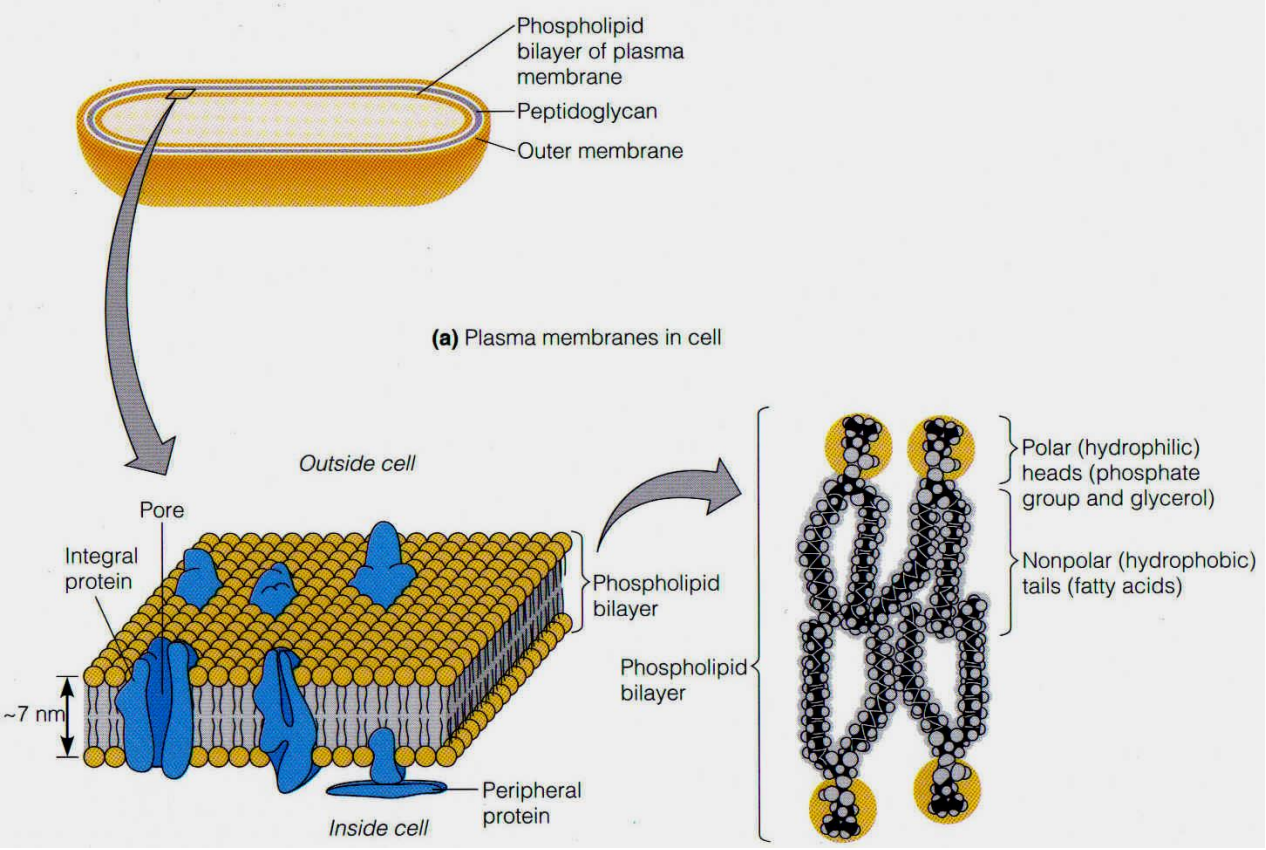


Fig 2.11 Phospholipids

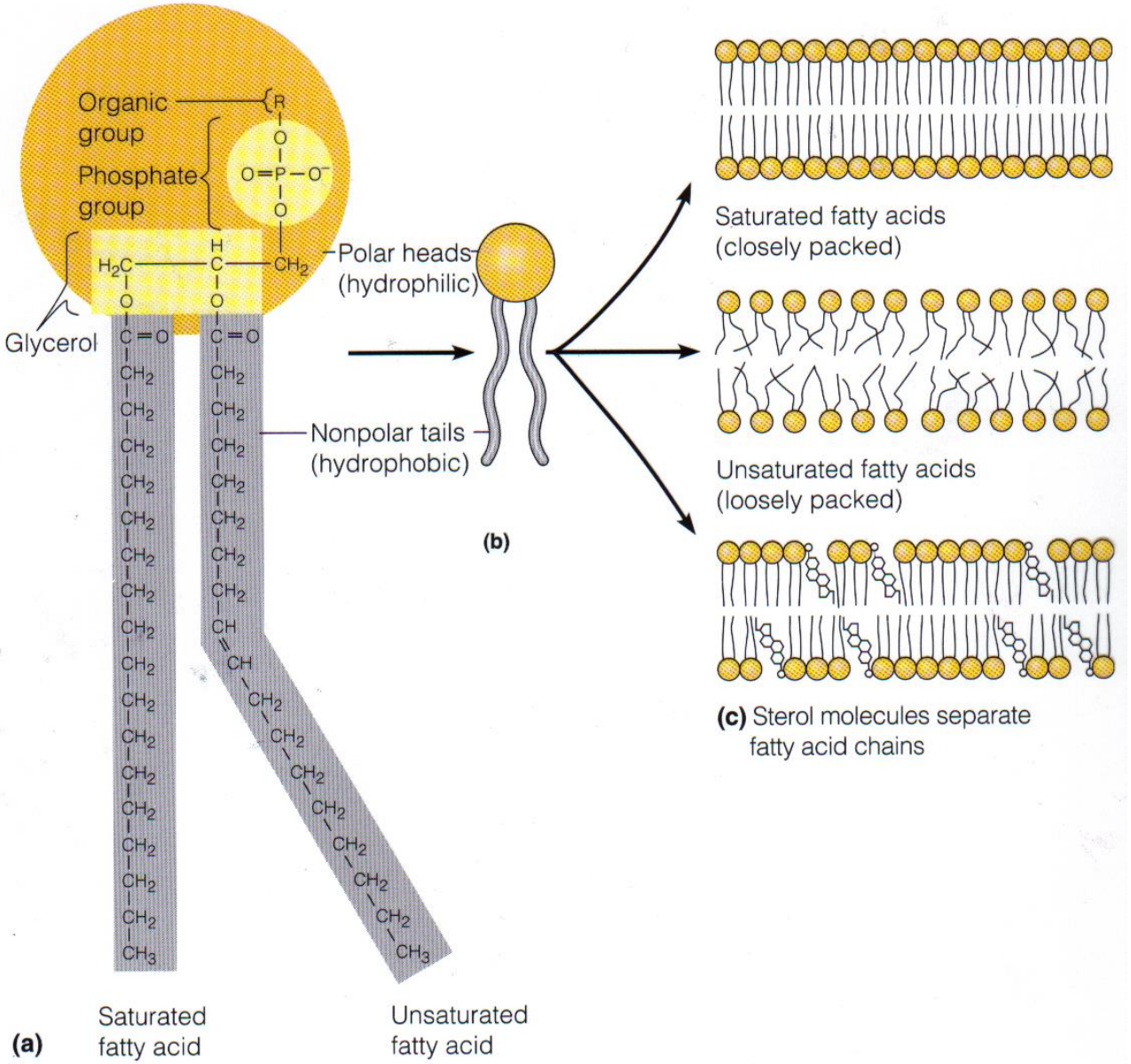


Fig 4.16 Diffusion

D. Diffusion: Passive transport from high to low [ ]

   i. Simple diffusion

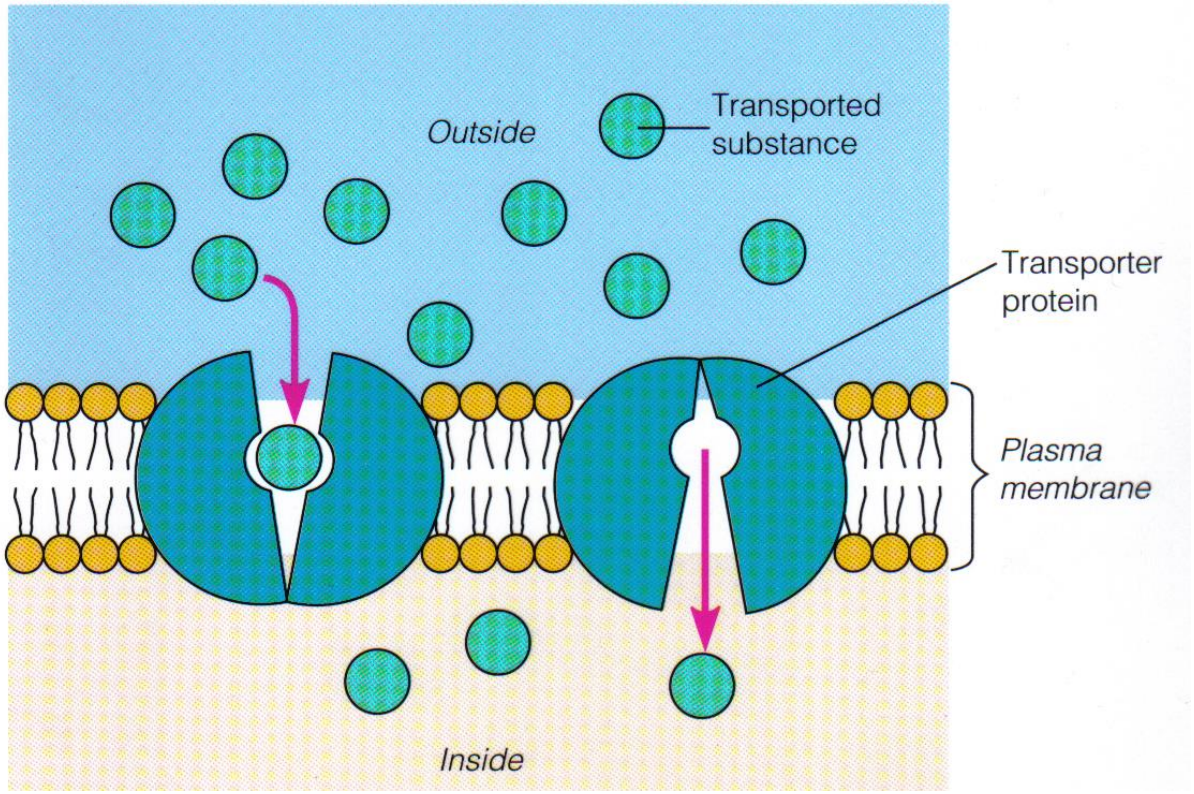
   ii. Facilitated diffusion

   iii. Osmosis

E. Active Transport: From low to high [ ], requires ATP & protein

Diagram on the right:

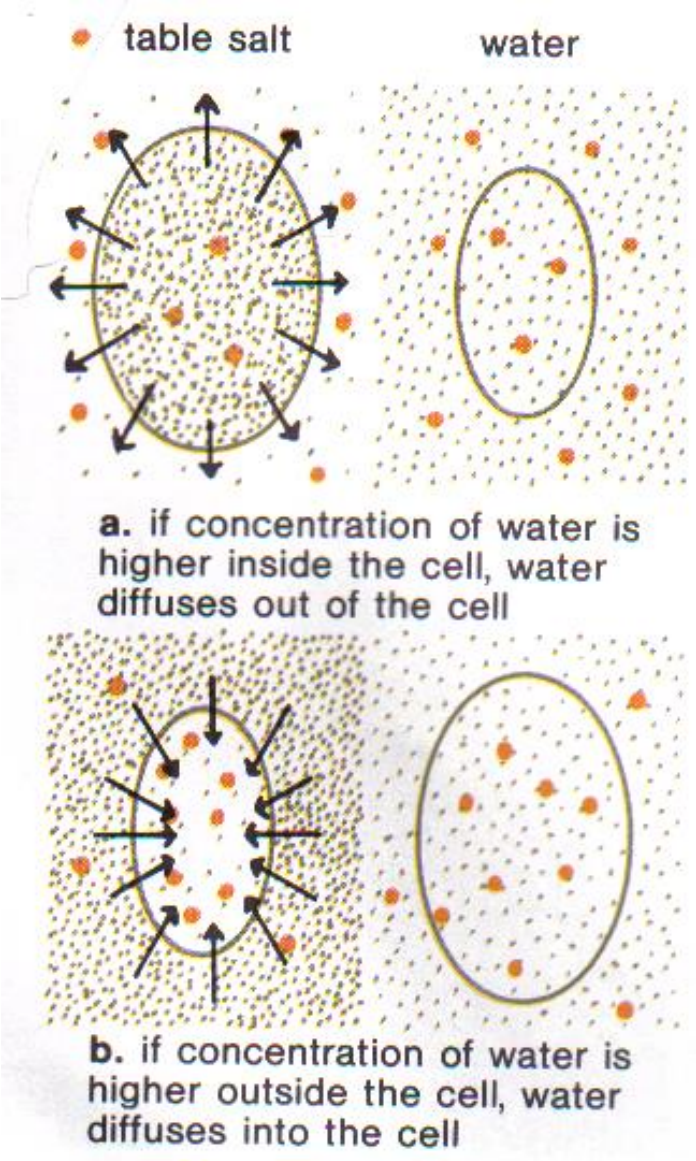
Which type of transport does it represent?



Water balance

Stranded at sea-drink ocean water? Why or why not?

What do supermarkets do to keep produce looking nice & firm?



Osmosis-Animal vs. Plant

Special terms reflect % solute OUTSIDE OF CELL, and therefore the effect on the net direction of osmosis.

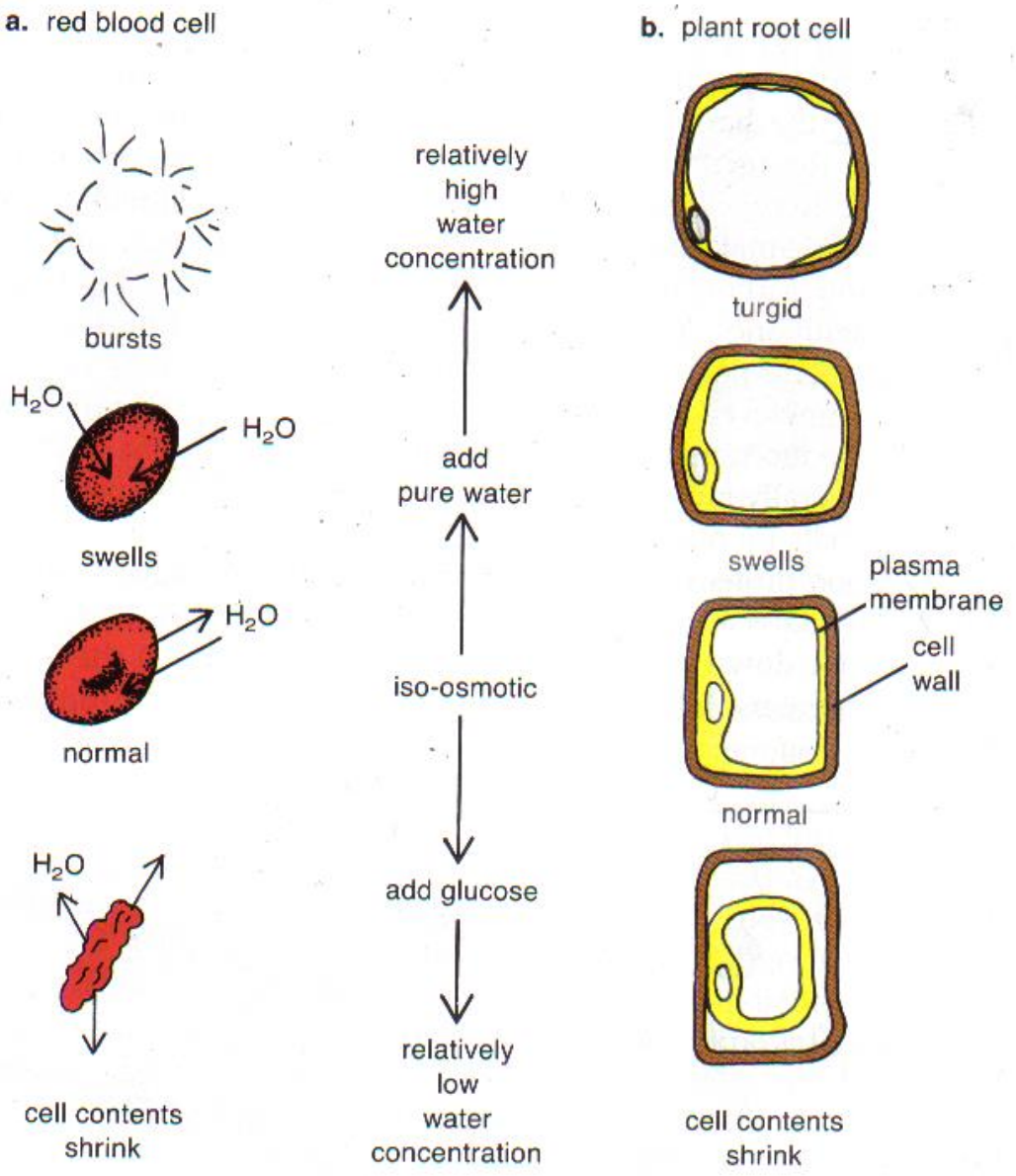
What do the following prefixes mean?

Iso?

Hypo?

Hyper?

Suffix is “tonic” = tension



Osmosis & Solution Types

F. Osmotic Environments

i. Isotonic/isoosmotic solution:

- » Equal % total solute on both sides of membrane
- » Water movement: Equal in & out of cell
- » Dynamic equilibrium; Cell size constant

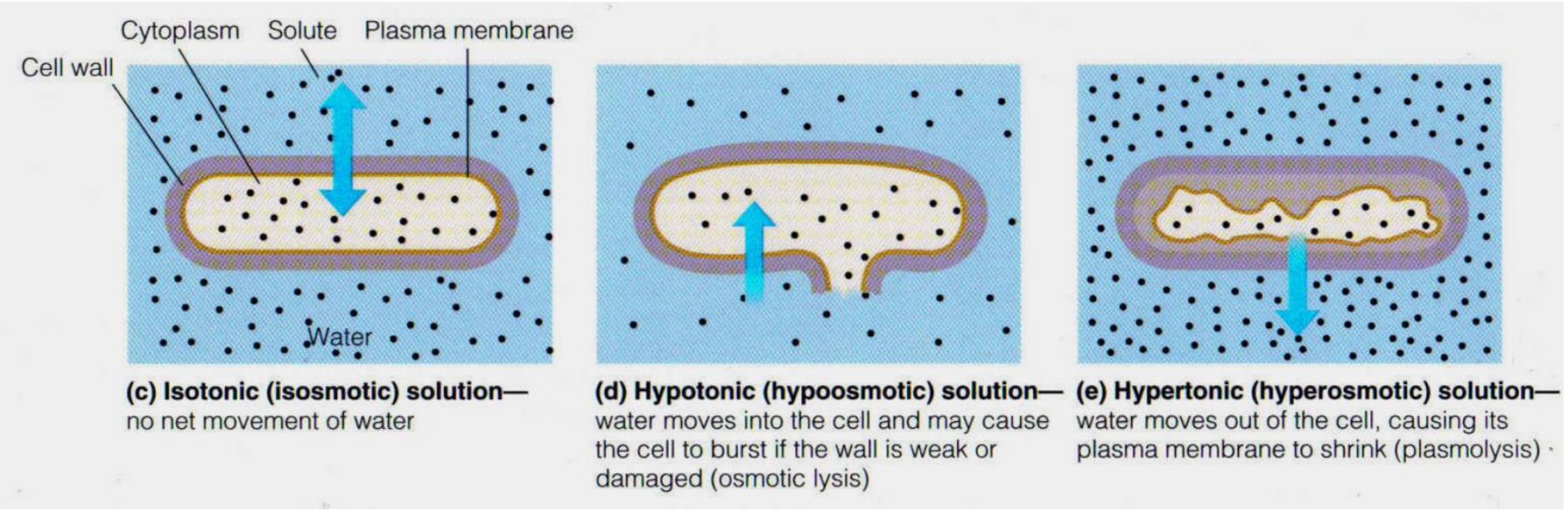
ii. Hypotonic solution: low % solute outside cell

- » Net H_2O moves into cell
- » Cell wall protects and prevents osmotic lysis
- » If there is no cell wall, lysis occurs due to osmotic lysis

iii. Hypertonic solution: high % solute outside cell

- » Net H_2O movement out of cell
- » Cell wall does NOT protect
- » Plasmolysis occurs: cell shrinks inside of wall

Fig 4.17 cde Osmosis in Varying Osmotic Environments



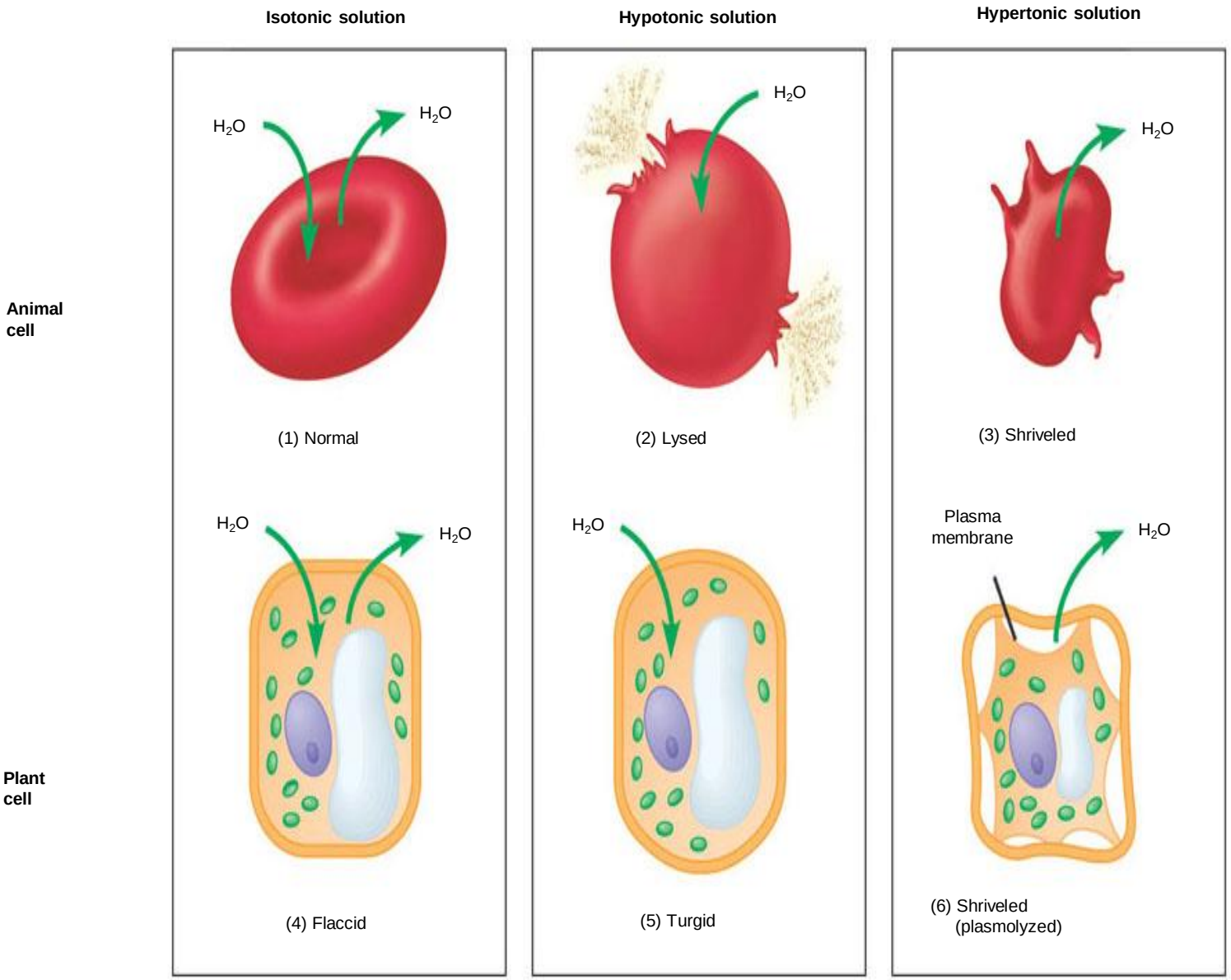
What can make a cell wall weak like the middle diagram above?

- Old age of bacteria
- Treatment with penicillin, or lysozyme in tears.

How was meat preserved in past, before refrigeration?

- Salted meat. Caused plasmolysis like diagram on right.

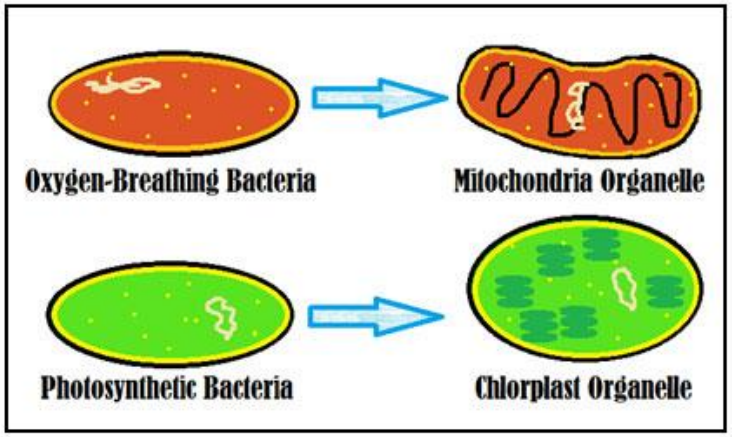
Diagram from Bio-Osmosis & Plant vs. Animal

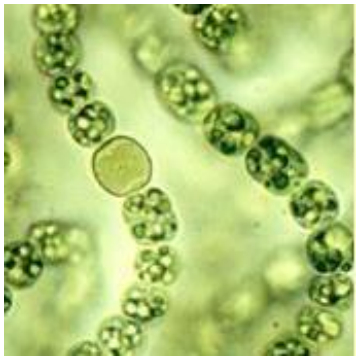


Internal Cell Structures continued

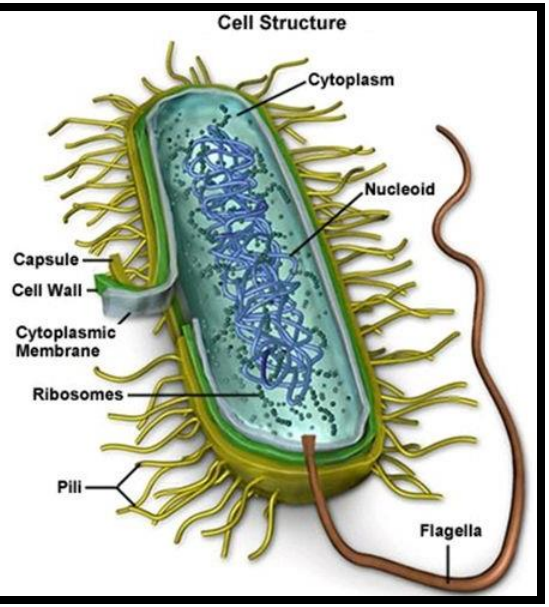
3. **Chromatophores/thylakoids: photosynthetic structures**

4. **Nucleoid/nuclear area: No nuclear membrane**
   A. **Contains 1 circular chromosome w/o histones**



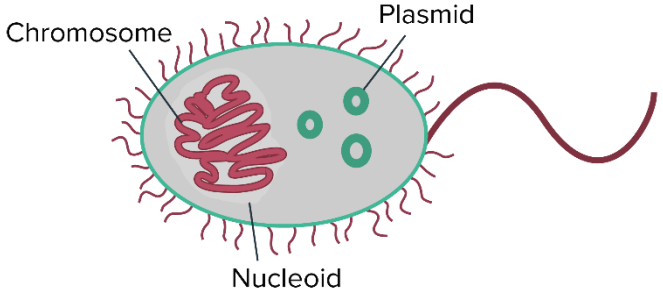


- Nucleoid region (center) contains the DNA
- Surrounded by cell membrane & cell wall (peptidoglycan)
- Contain ribosomes (no membrane) in their cytoplasm to make proteins



Internal Cell Structures continued

5. **Plasmids: circular extrachromosomal DNA** Benefits to org? To us?
   - A. **Transferred BETWEEN SPECIES of CURRENT generation (not just to offspring)**
     - **Can transfer genes for antibiotic resistance, toxin production, resistance to toxic metals, …**
     - **NOT critical for "normal" survival. Only an advantage in special circumstances.**
     - **Allows for genetic variation not found in binary fission**
   - B. **Conjugation: transfer plasmids through direct cell-to-cell contact**
     - i. **GN-sex pili We'll focus on GN, but GP do in diff way.**
   - C. **Biotech: Used as a vector for genetic manipulation**
     - **We use for genetic engineering. Example: insert human gene for insulin, gene from surface of virus to make vaccine, etc.**



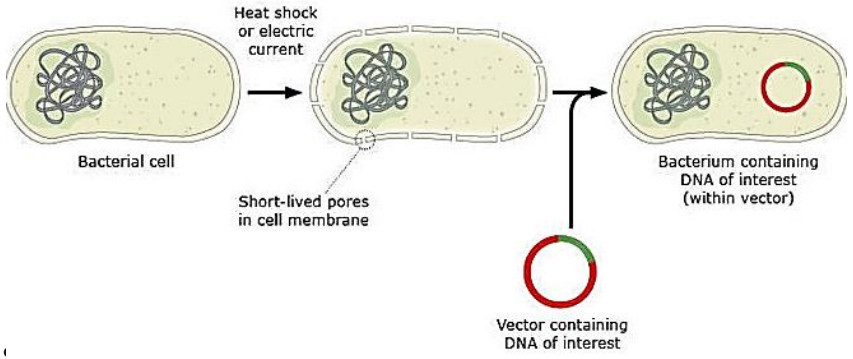
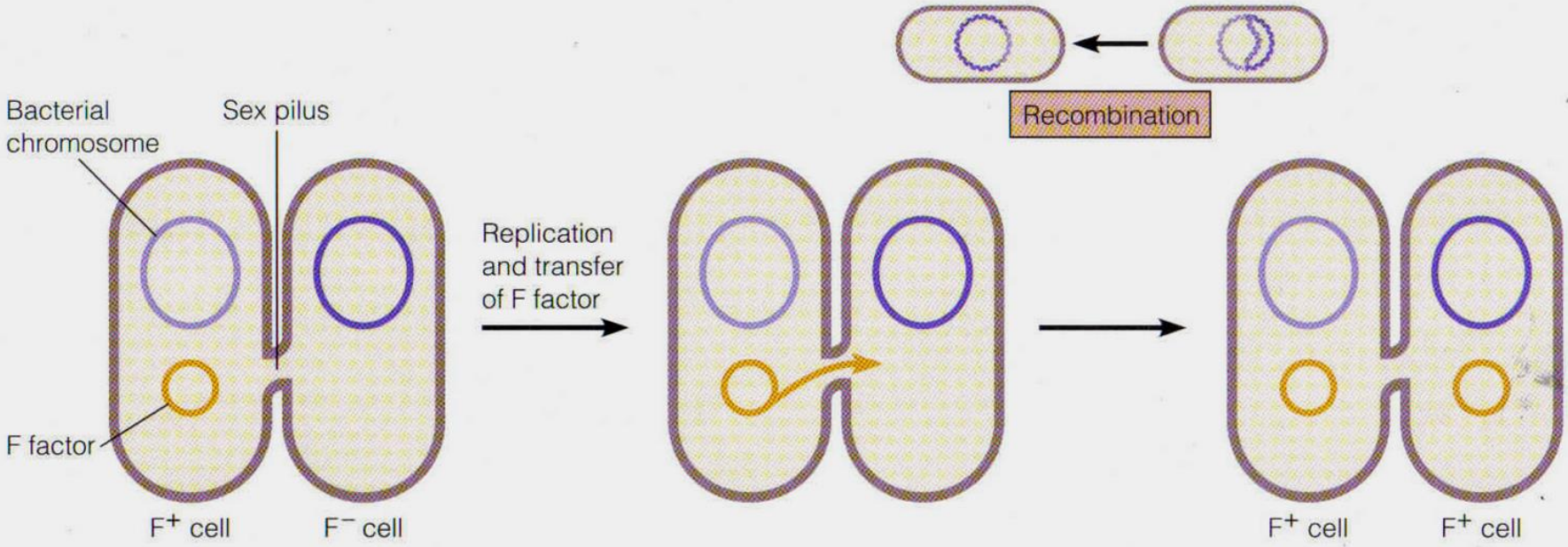


Fig 8.27a Conjugation-Plasmid Transfer

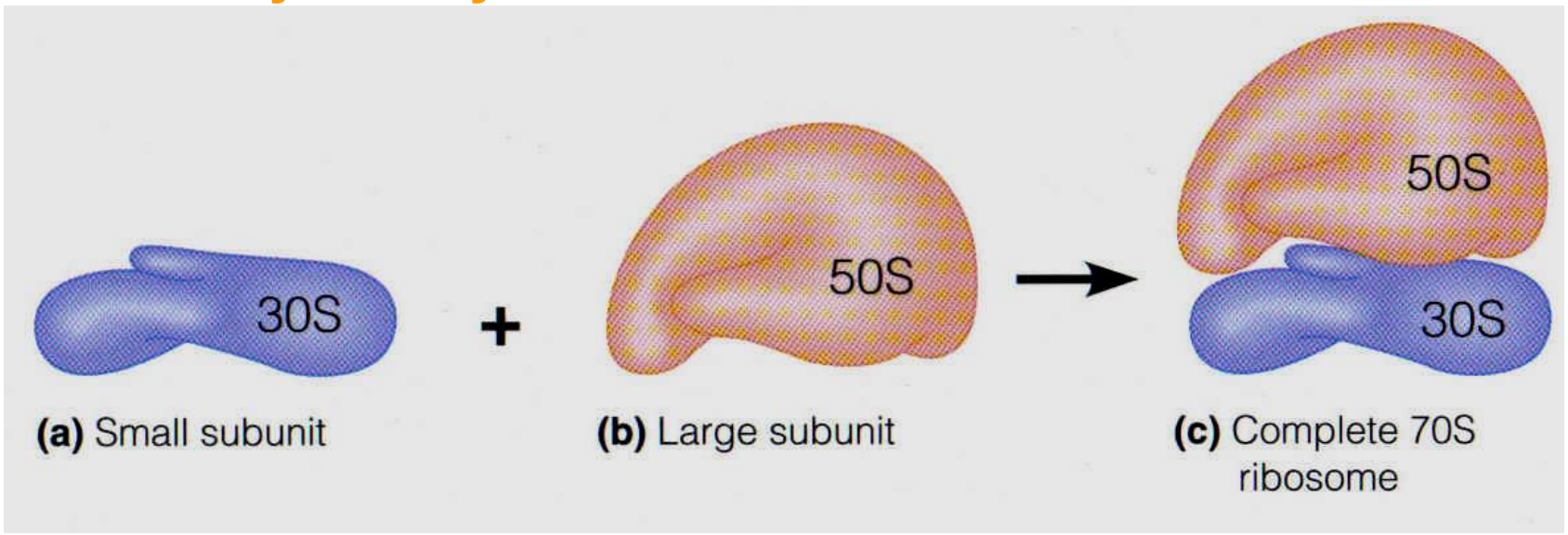
Figure 8.27a: Conjugation in *E. coli*.



(a) When an F factor (a plasmid) is transferred from a donor (F^+) to a recipient (F^-), the F^- cell is converted into an F^+ cell.

Internal Structures cont'd-Ribosomes

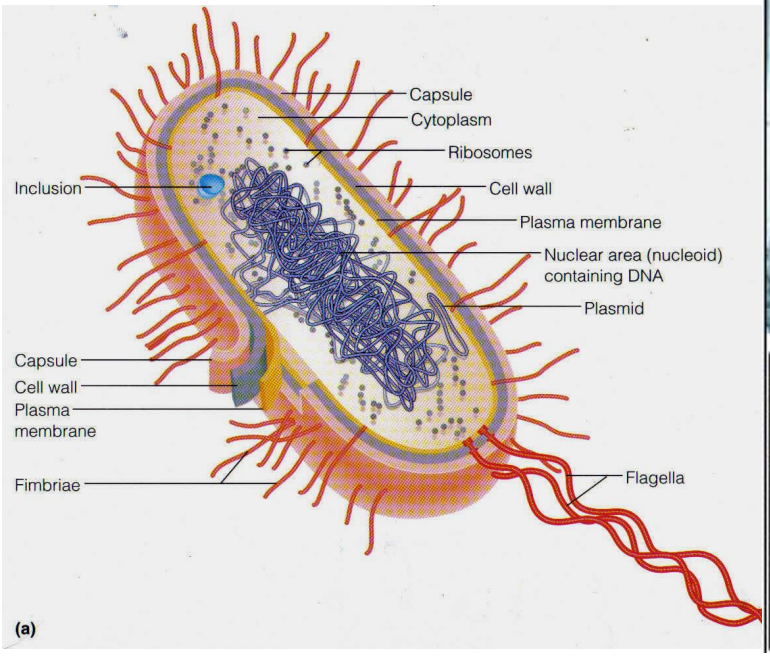
6. Ribosomes: protein synthesis
   A. 2 subunits: protein & RNA
      i. Prokaryotic size: 70S (30S & 50S subunits)
      ii. Euk: 80S (60S & 40S subunits)
      iii. S related to rate of "sedimentation" in a tube
   B. Antibiotics attach to subunit of prokaryotic size and not eukaryotic size. Examples: Gentamycin & Erythromycin.

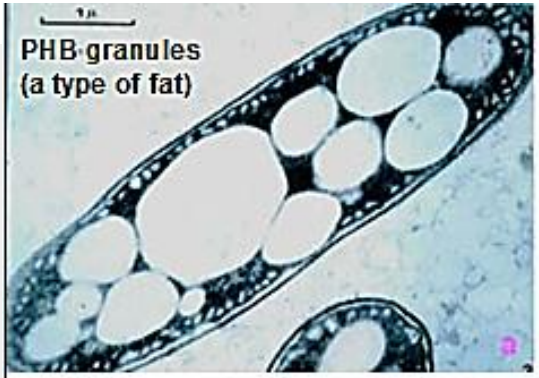


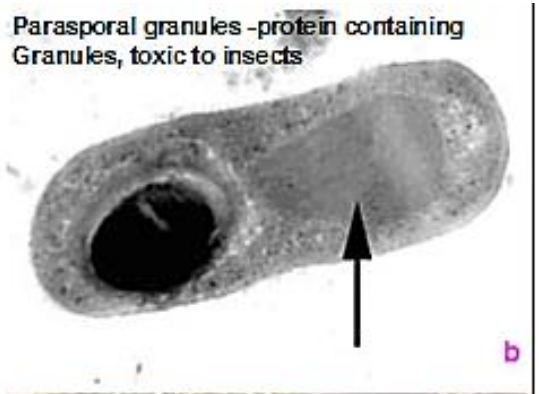
Internal Structures cont'd-Inclusions

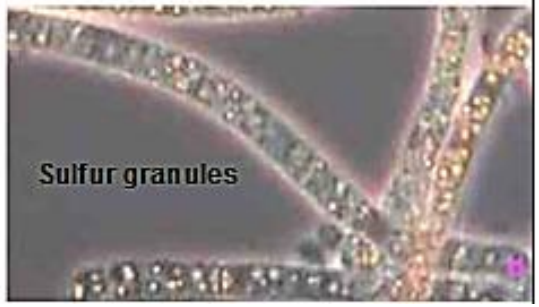
7. **Inclusions: Reserve deposits.**
   - A. **Examples: Storage of sulfur or lipids for energy, gas for buoyancy**
   - B. **Prevent internal ⇑ in osmotic pressure as solute is "packaged" and not counted as %solute in the cytoplasm**
   - C. **Can be mistaken for endospores & is why an endospore stain is needed for clear areas in the cell**

Review Structures:









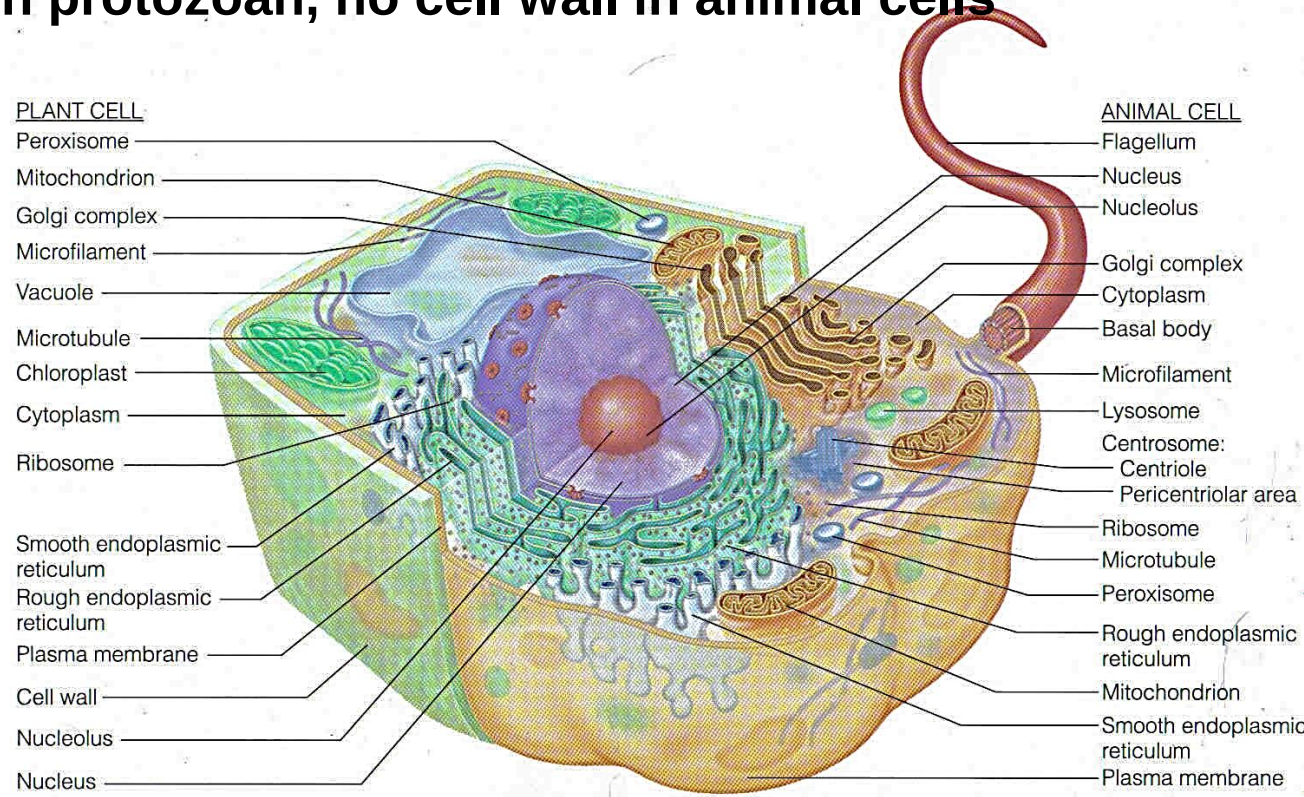
Eukaryotes

Eukaryotes

1. **Larger overall cell size**
2. **Have a nucleus & membrane bound organelles**
3. **If there is a cell wall, it is made of a substance other than peptidoglycan:**
   - A. **Fungi: Chitin**
   - B. **No cell wall in protozoan, no cell wall in animal cells**

Don't need to know eukaryotic organelles except for:

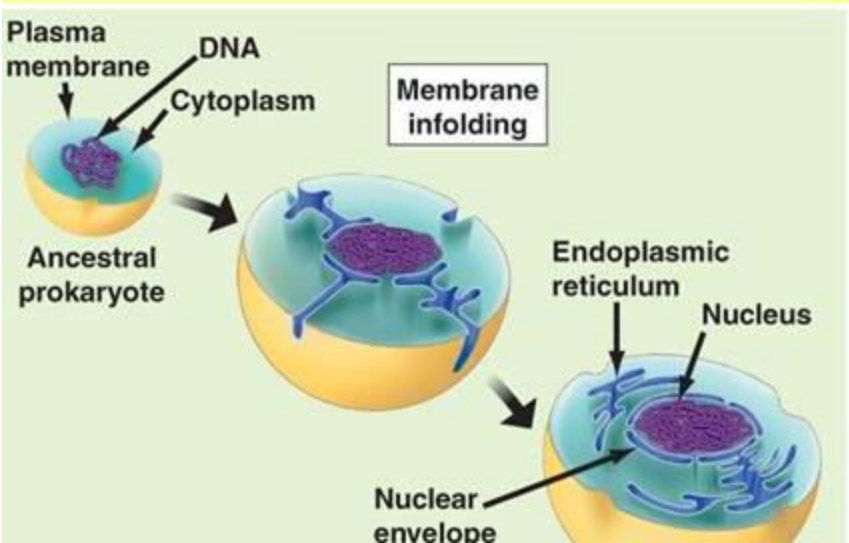
- **General info above**
- **Evidence how eukaryotes came from prokaryotes – as discussed on next slides**



Endosymbiotic Theory

Endosymbiotic Theory

1. Eukaryotes evolved from prokaryotes living inside other prokaryotes
2. Evidence
   - A. Organelle membranes of eukaryotes have phospholipids similar to the plasma membranes in bacterial
   - B. Chloroplasts in eukaryotes are similar to photosynthetic prokaryotes – have same photosynthetic enzymes
   - C. Mitochondria & chloroplasts are similar to bacterial cells in that these eukaryotic organelles:
     - i. Reproduce by binary fission
     - ii. Contain circular DNA
     - iii. Contain 70S ribosomes



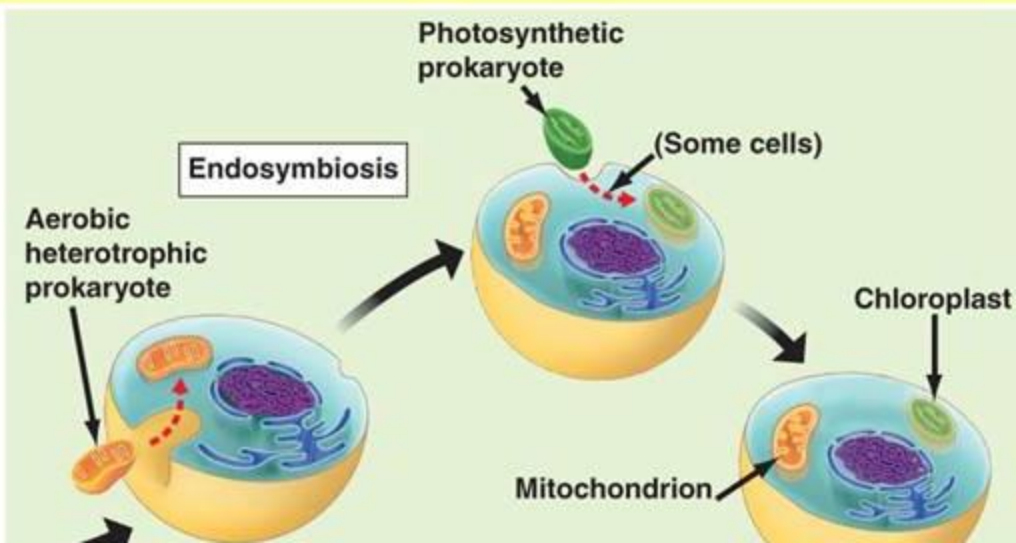
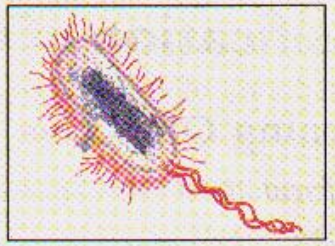


Table 10.2 Prokaryotic Cells vs. Eukaryotic Organelles

Table 10.2 *Prokaryotic Cells and Eukaryotic Organelles Compared*

	Prokaryotic Cell	Eukaryotic Cell	Eukaryotic Organelles (Mitochondria and Chloroplasts)
DNA	Circular	Linear	Circular
Histones	No	Yes	No
Ribosomes	70S	80S	70S
Growth	Binary fission	Mitosis	Binary fission



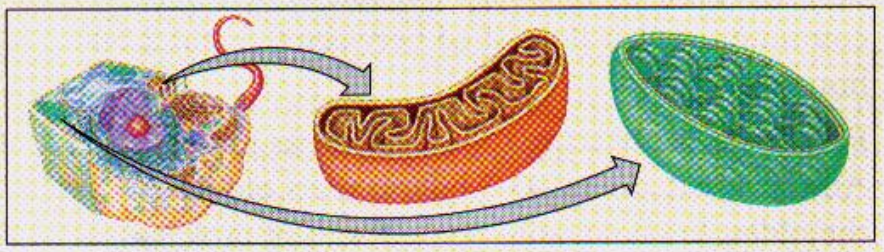


Fig 10.2 Endosymbiotic Theory

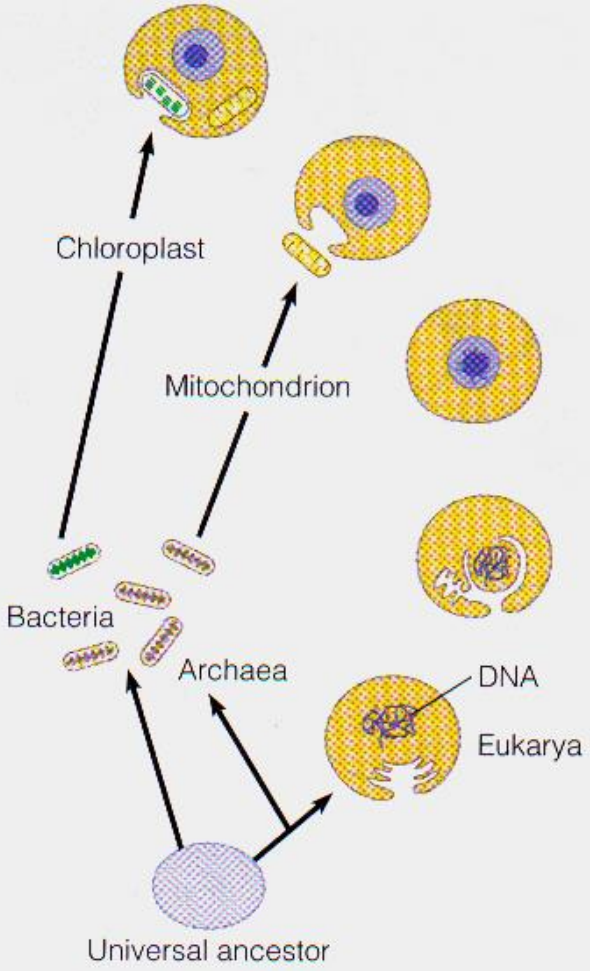
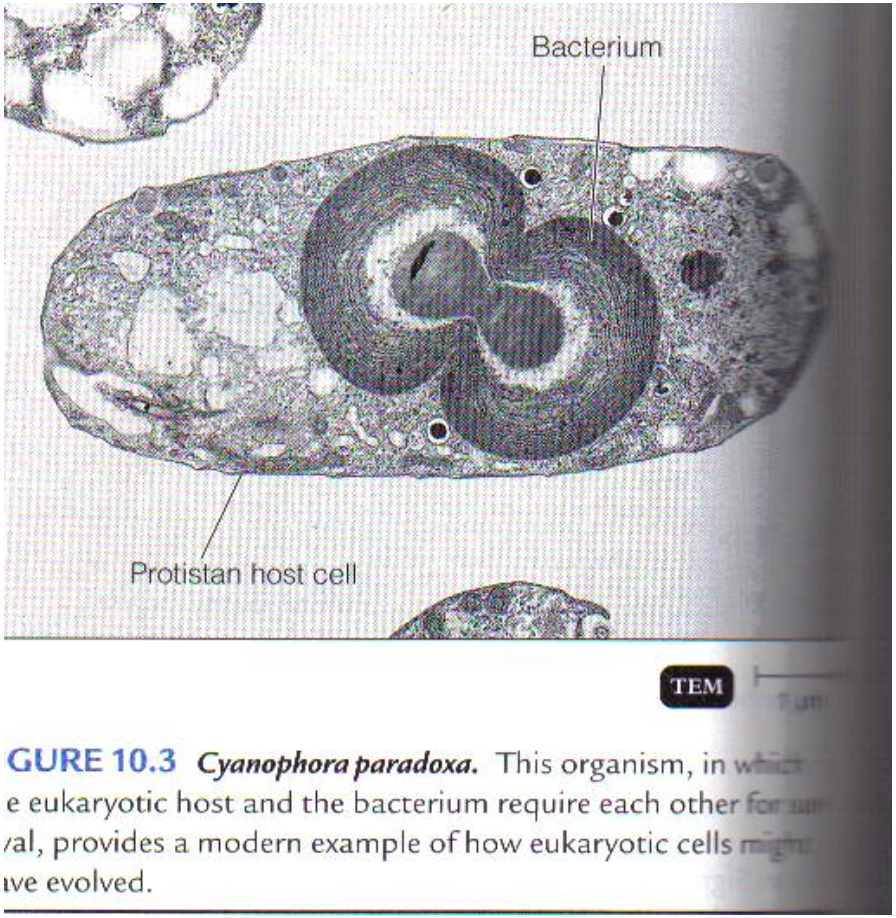


FIGURE 10.2 A model of the origin of eukaryotes. Invagination of the plasma membrane may have formed the nuclear envelope and endoplasmic reticulum. Similarities, including rRNA sequences, indicate that endosymbiotic prokaryotes gave rise to mitochondria and chloroplasts.



GURE 10.3 ***Cyanophora paradoxa.*** This organism, in which e eukaryotic host and the bacterium require each other for sur val, provides a modern example of how eukaryotic cells might ave evolved.