MBS 240

Introduction to Bacteriology

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Objectives

- By the end of this lecture, you should know and understand:
    - Microbiology and Bacteriology
    - Bacteria and its classification:
        - By shape and structure
        - By cell wall
        - Metabolism
        - Food needs for growth and survival
        - Bacterial reproduction genetics
    - Bacterial Normal flora

Microbiology and Bacteriology

- Microbiology is the Study of Microorganisms, amongst them: Bacteria, Viruses, Parasites and Fungi

- Bacteriology is the studies in detail, the morphology, ecology, reproduction and genetics as well as the biochemistry of Bacteria

Bacteria

- In any direction you look, you will see bacteria at work
- Are enormously successful and diverse organisms
- Have caused some serious disease outbreaks like TB, cholera, typhoid fever and plague
- Bacteria:
  - Help some plants grow by capturing nitrogen from the air
  - Degrade waste like dead plants, oil, sewage
  - Some cause food spoilage
  - Others are useful in the food industry in making of yoghurt and cheese
  - Are found on the skin, hair, teeth, intestines….
  - Belong to a group of living organisms called PROKARYOTES: and are Eubacteria: the `True Bacteria`

- Are single-celled (unicellular)
- Reproduce asexually, by binary fission
- Lack of membrane-bound organelles
  - Mitochondria
  - Golgi bodies
  - Endoplasmic reticulum
- Have a DNA that forms long circular molecules but is not contained within a defined nucleus
- May be motile using flagella
- Surrounded by complex cell wall in two forms:
  - Gram-positive: have a thick peptidoglycan layer
  - Gram-negative: have a thinner peptidoglycan layer with an inner and outer membrane
- Sometimes surrounded by a thick capsule
- Are classified by both phenotypic and genotypic data

Bacteria composition/Structure

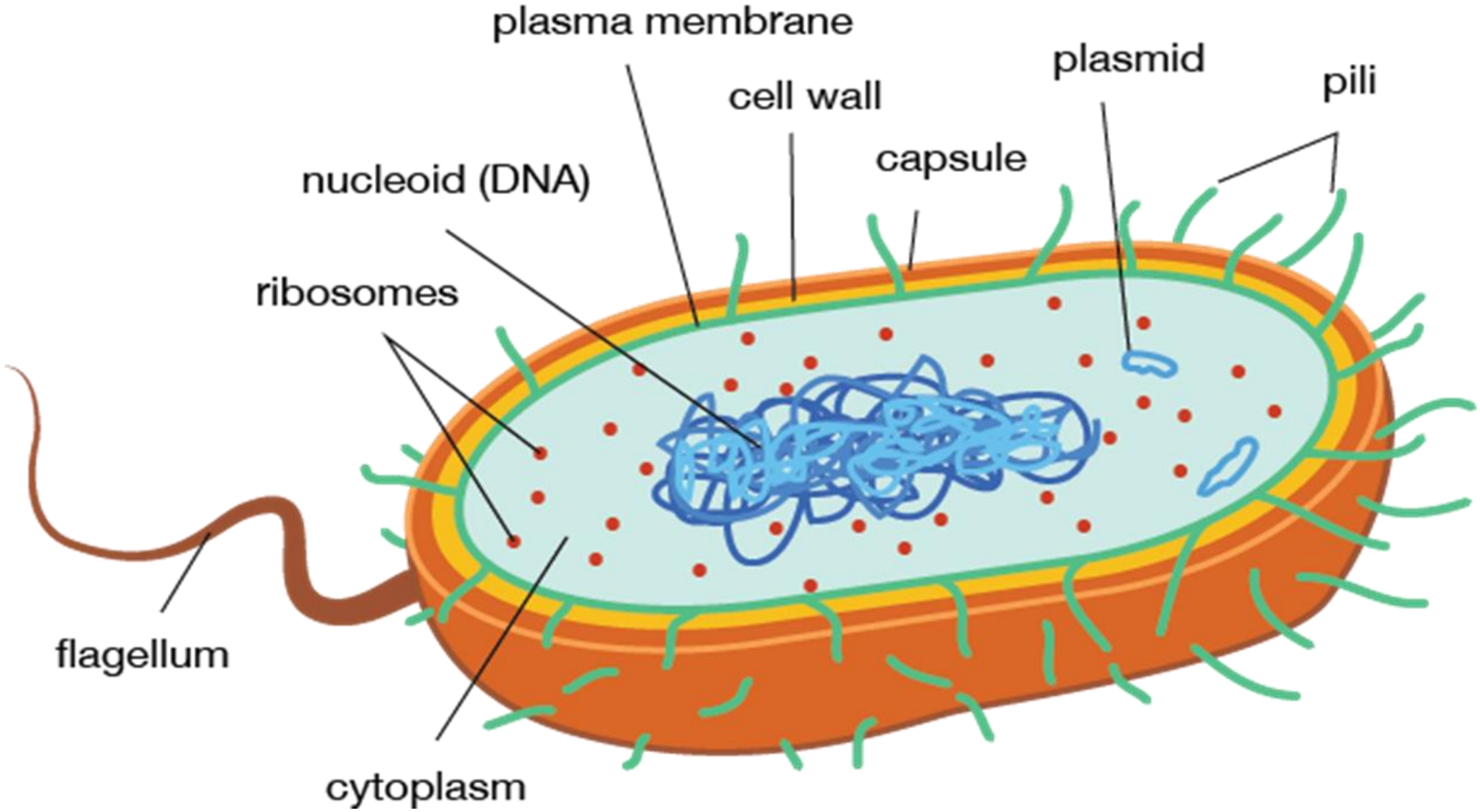
1. Flagella: Hair like structures useful for locomotion towards nutrients and away from toxic substances
   - Protein component(flagellin) which is strongly antigenic
2. Pili: Are a protein composed projections from the surface that enable bacteria to adhere to host tissue surfaces (common pilus) and special pilus(sex pilus) for conjugation
3. Capsule/Glycocalyx: Composed of polysaccharide and protein functioning as a buffer to environment, for protection and immune evasion, cell protection and surface attachment
4. Cell wall: for bacterial shape determination, cell protection and prevent cell lysis
5. Cytoplasmic membrane: composed of proteins and phospholipids. Functioning as a cell boundary, regulation of the in-flow and out-flow of materials and site for enzymatic reactions

6. Have cytoplasm with the following components inside:

- DNA chromosome
- Plasmids –Extrachromosomal genetic material
- mRNA
- Ribosomes
- Proteins

7. Endospores: for dormancy when nutrients and environment are no conducive e.g. Bacillus and Clostridium

Bacterial Cell



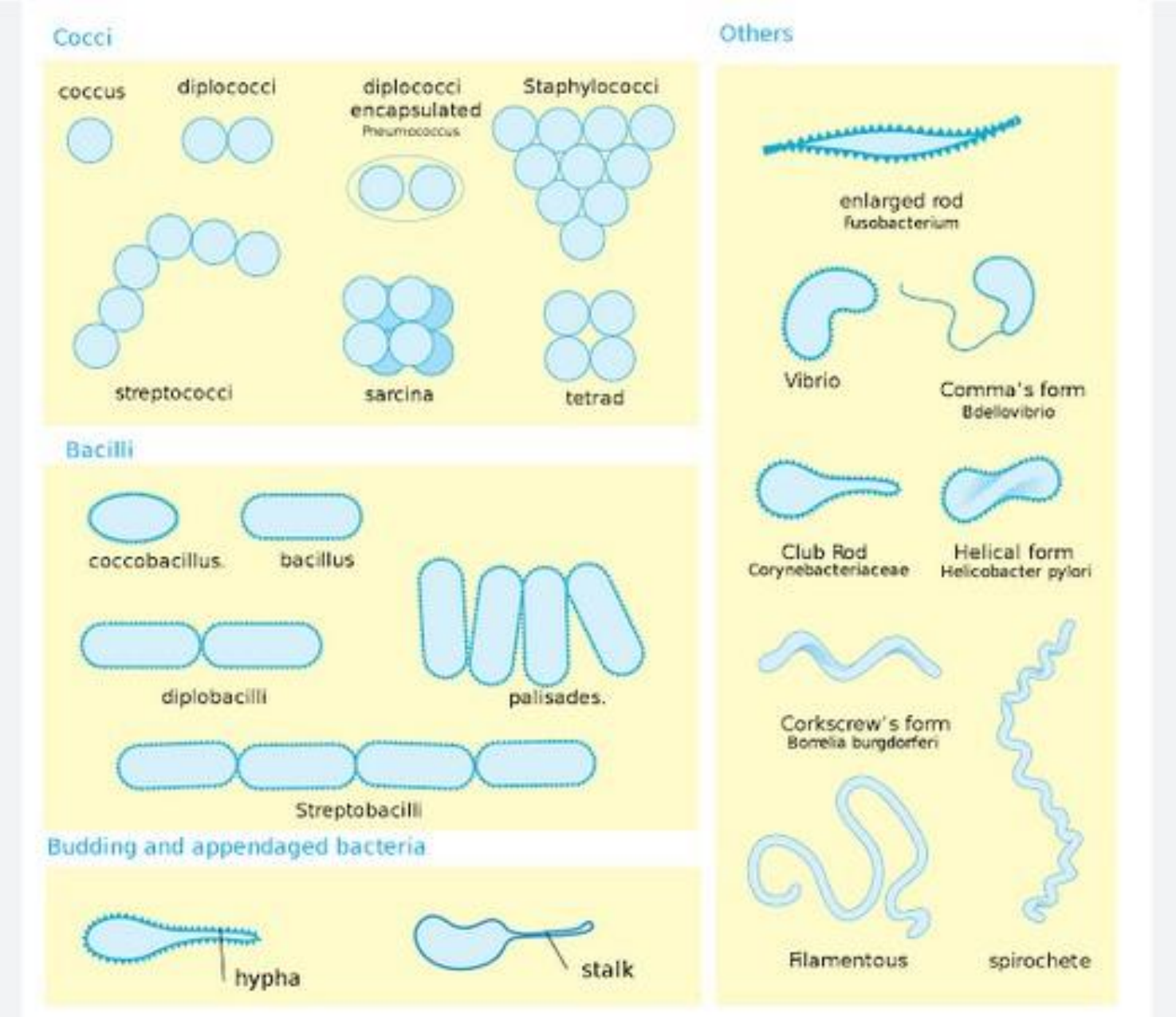
Bacteria classification

- Bacteria can be classified by:
    1. Shape
    2. Cell wall characteristics
    3. Growth requirements
    4. Oxygen/Carbon dioxide requirements
    5. Biochemical tests
    6. Flagella

Bacterial shapes

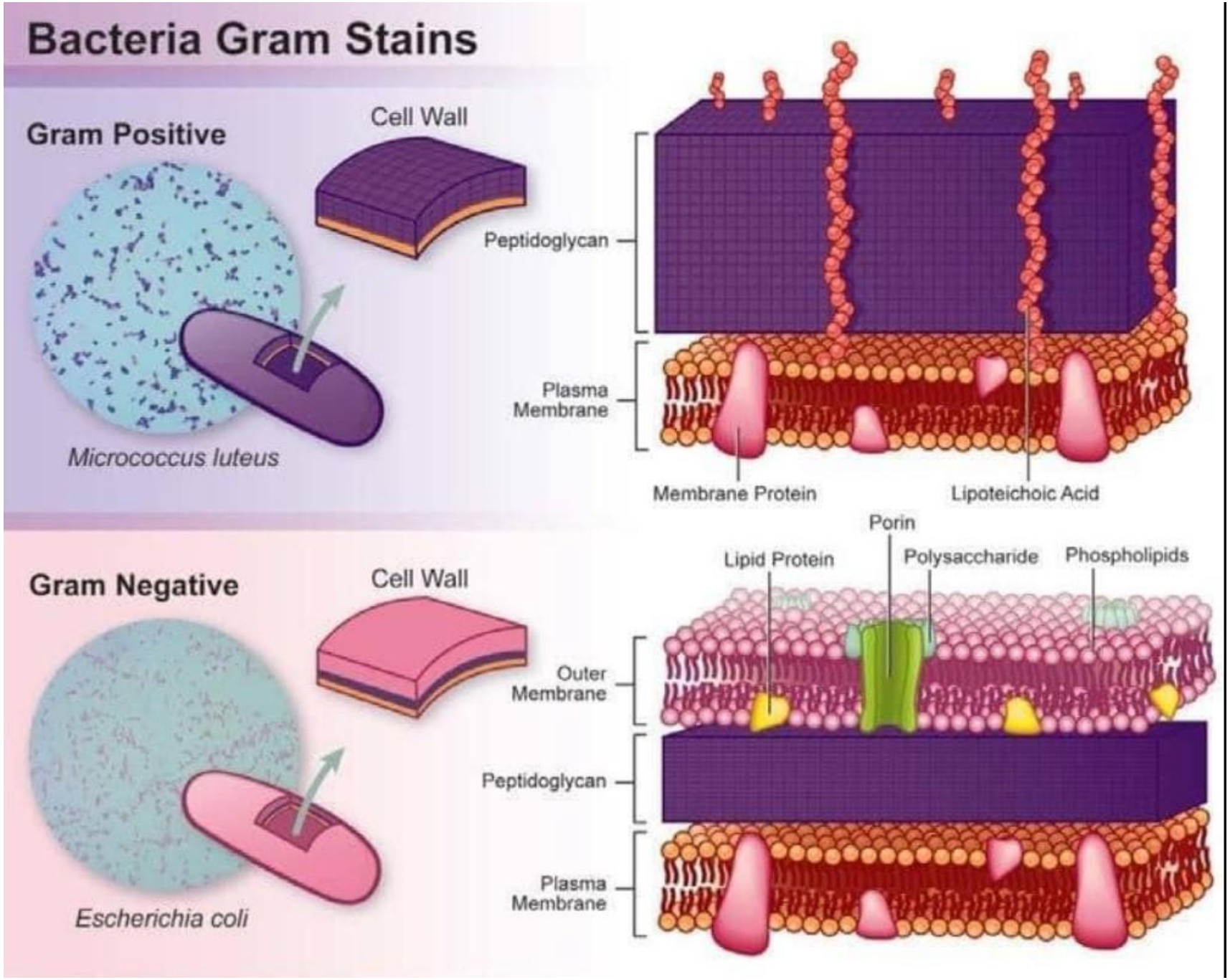
Bacterial shapes:

1. Coccus
2. Coccobacillus
3. Bacillus
4. Vibrio
5. Spirillum
6. Spirochete



Cell wall structures

- There are 2 main types of bacteria according to the cell wall structure. They can be:
  1. Gram positive
  2. Gram negative
- Mycoplasma are none of the above because they lack a cell wall
- Mycobacteria are also an exception due to a peculiar cell wall structure


Bacteria Gram Stains
Gram Positive
Cell Wall
Micrococcus luteus
Peptidoglycan
Plasma Membrane
Membrane Protein
Lipoteichoic Acid
Gram Negative
Cell Wall
Escherichia coli
Porin
Lipid Protein
Polysaccharide
Phospholipids
Outer Membrane
Peptidoglycan
Plasma Membrane

Comparison of G-ve and G+ve cell walls

Characteristic	G+ve	G-ve
Gram stain	Purple as they keep the crystal violet stain	Pink/Red as they keep the safranin stain
Periplasmic region	No	Yes
Outer membrane	No	Yes
Teichoic acid	Yes	No
Peptidoglycan	Yes, thick layer ~60-90 %	Yes, thin layer ~10 %
Lipopolysaccharides	No	Yes
Porin proteins	No	Yes

Mycobacteria

- A supposed G+ve bacteria, but is not, is Mycobacteria- acid-fast bacteria
- This is so because:
  - The peptidoglycan layer has a different chemical basis for cross linking to the lipoprotein layer
  - Acid-fast bacteria have a very thick lipid cell wall (60% lipid and little peptidoglycan). The outer envelope has a variety of complex lipid: mycolic acids
  - Cell wall is impermeable to gram stain

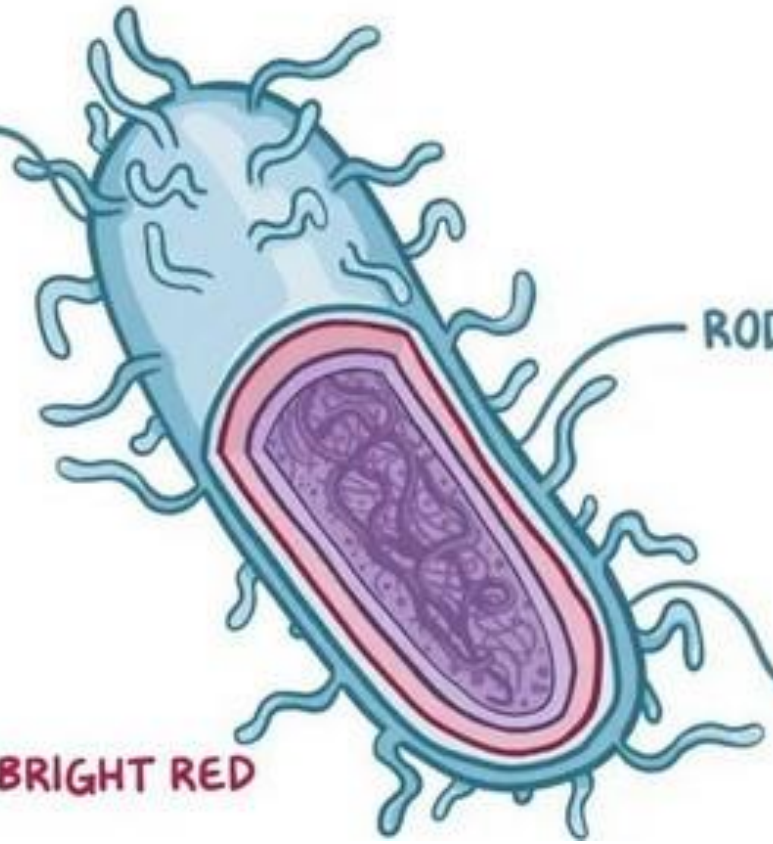
- Mycolic acids create a waxy layer which alters both the staining properties of Mycobacteria giving it the ability to resist drying and other environmental factors

- Cell wall also impermeable to nutrients

  - Hence slow growth

MYCOBACTERIUM TUBERCULOSIS (TB)

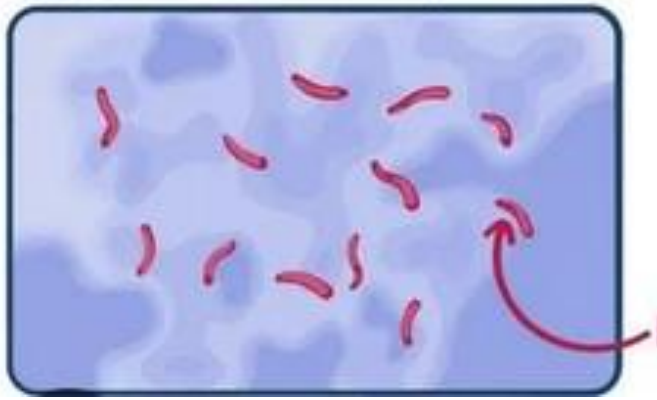
WAXY CELL WALL
- from MYCOLIC ACID
- "ACID-FAST"



ROD-SHAPED

NEEDS OXYGEN
STRICT AEROBES

ZIEHL-NEELSEN STAIN



BRIGHT RED

Growth requirements

- Minimum requirements for bacterial growth: C, N, an energy source, water & various ions
- Fastidious bacteria:
  - Need special nutrition supplements and conditions for their growth
  - They are difficult to grow in laboratories as they require specific nutrients and conditions.
  - E.g - *Neisseria gonorrhoeae* need haemoglobin or blood to grow

    -*Campylobacter* spp. and *Helicobacter* spp need elevated carbon dioxide levels to grow

- Non-fastidious bacteria:
  - do not require any special nutrition supplements and conditions for their growth
  - No specialised substances or conditions are essential for its growth.
  - *E.g Pseudomonas aeruginosa, Staphylococcus aureus*

Oxygen requirements: Anaerobic Vs aerobic environments

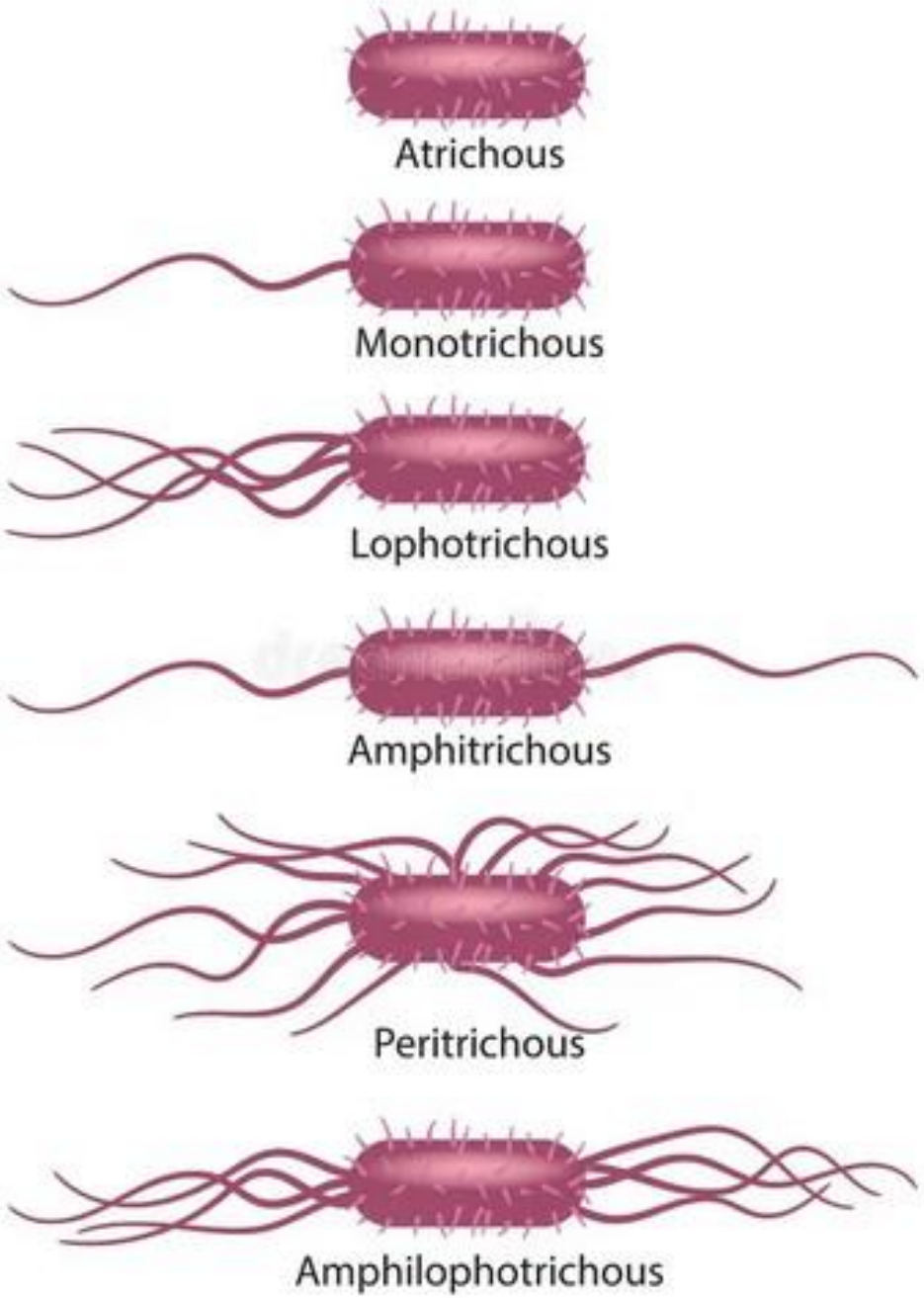
- Obligate anaerobes
  - Bacteria that cannot grow in the presence of O_2
  - E.g. *Clostridium perfringens* (causes gas gangrene)
- Obligate aerobes
  - Require O_2 for growth & metabolism
  - E.g. *Mycobacterium tuberculosis* (Mtb)
- Facultative anaerobes
  - Grow in either the presence or absence of O_2
  - Most bacterial species are facultative
- Microaerophilic
  - reduced Oxygen conc
- Capnophilic
  - high conc of carbon dioxide

Other ways of classifying bacteria

- Biochemical test reactions: these help distinguish between pathogenic and non pathogenic bacteria:
  - Bile solubility
  - Catalase
  - Citrate utilization
  - Beta-glucuronidase
  - DNA-ase
  - Oxidase
  - Urease
  - Litmus milk decolourization
  - Lysine decarboxylase
  - Coagulase
  - Indole
  - Motility

  *Read more about these tests

Flagella


Atrichous
Monotrichous
Lophotrichous
Amphitrichous
Peritrichous
Amphilophotrichous

Bacterial reproduction (Asexual reproduction)

- Reproduce by binary fission-producing 2 identical daughter cells with no genetic recombination
- This is where the cell elongates and the DNA chromosome replicates
- Binary fission can be fast for some bacteria, esp. the ones that cause food poisoning like E.coli O157:H7
- Growth rate dependent on nutritional status of surrounding environment
    - requires sufficient metabolites to support synthesis of bacterial components
    - E.g. *E. coli* replicates in 20-30 min in nutrient rich environment.
        - 1-2hrs in nutrient depleted environment
    - MTB take up to 24hrs to replicate
- Replication follows cascade of regulatory events
    - Key proteins and RNA
- Chromosomal replication initiates cell division

BACTERIA REPRODUCE BY BINARY FISSION

Most bacteria reproduce by an asexual process called **binary fission**. In this process, the cell elongates and the chromosome (DNA) replicates (FIGURE 4.14).

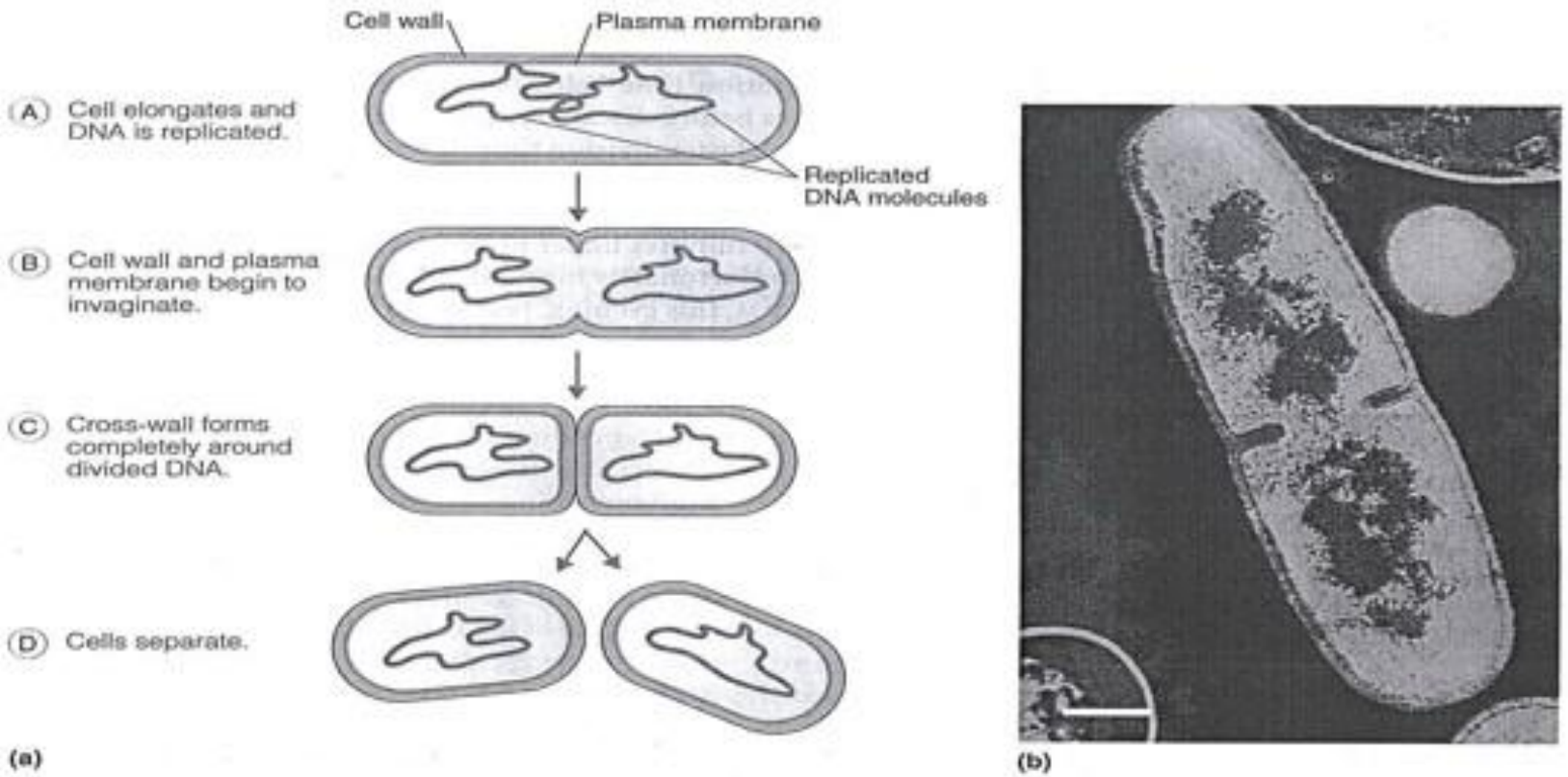


FIGURE 4.14

Binary Fission of Bacteria

(a) As a result of DNA replication and binary fission, two cells are formed, each genetically identical to the parent cell. (b) A false color transmission electron micrograph of a cell of *Bacillus licheniformis* undergoing binary fission. The invagination of the cell membrane is evident. (Bar = 0.25 μm.)

Bacterial reproduction (Sexual reproduction)

- In sexual reproduction, 2 parents are involved and the offspring will not be genetically identical

- The 3 ways in which bacteria exchange genes in nature are:

1. Transformation- is when DNA is acquired directly from the environment having been released from another species

2. Transduction-a Bacteriophage which is a virus that attacks the bacteria, transfers the genes between bacteria

3. Conjugation-which is a cell to cell contact as DNA crosses a sex pilus from donor to recipient

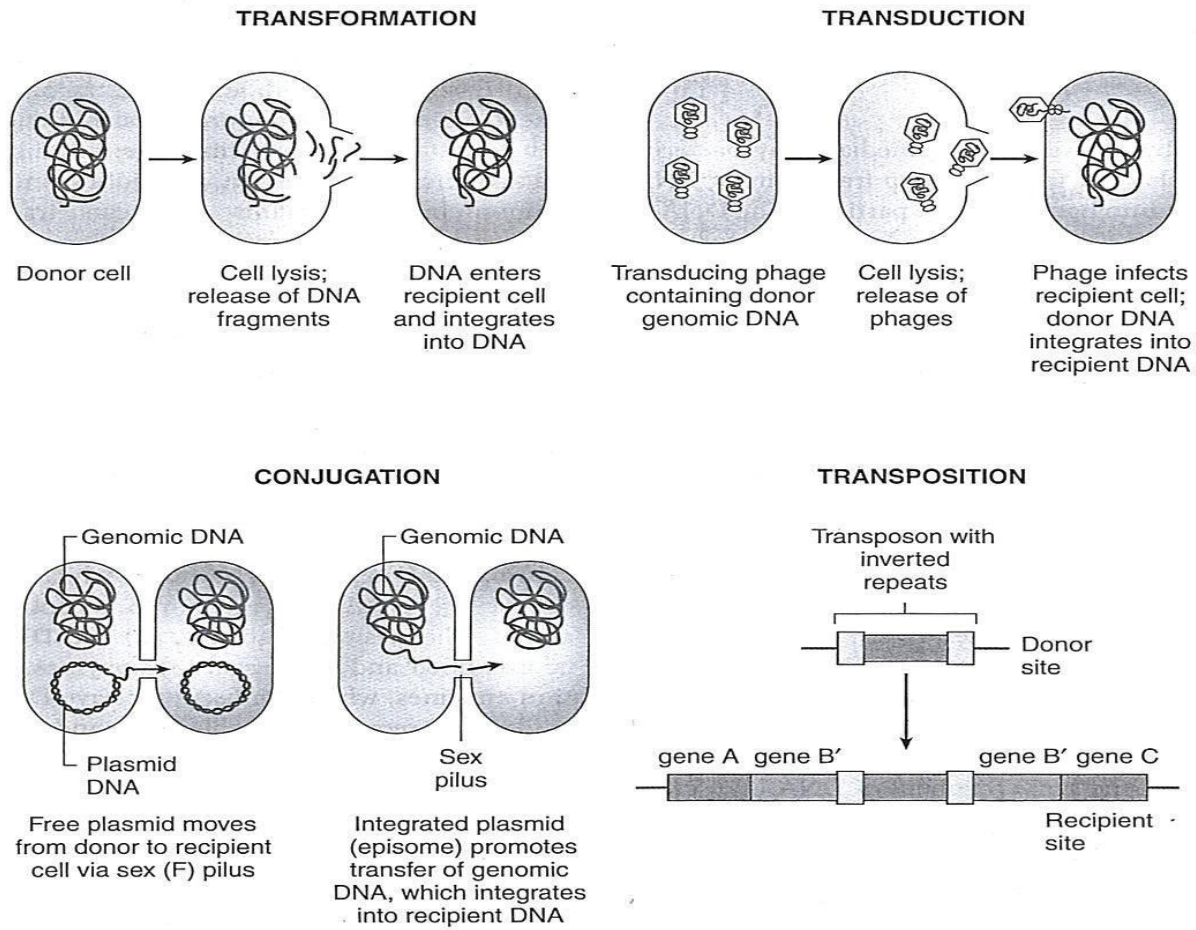
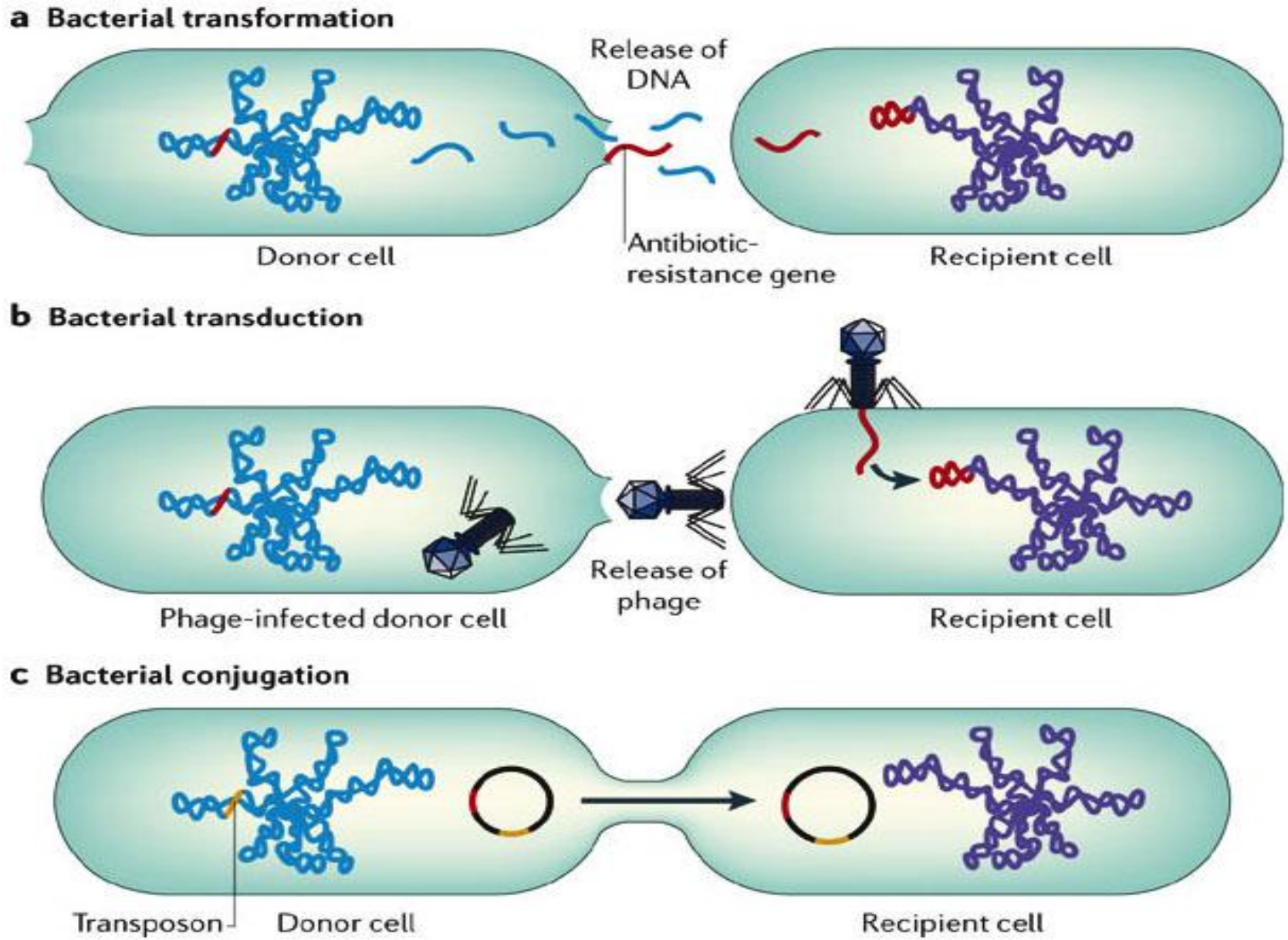


Figure 3–14. Mechanisms of bacterial gene transfer. *(From Rosenthal KS, Tan J: Rapid Reviews Microbiology and Immunology. St. Louis, Mosby, 2002.)*

An Transposon is a genetic element cell that can copy itself and insert the copy into another DNA molecule. They are called `jumping genes` and can transfer an antibiotic resistance gene from a plasmid to a chromosome and vice versa



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Normal flora

- Human body harbors >10 million microbes-the Normal flora
- They live outside and inside the human body without causing disease
- Some establish a lifelong relationship with the body eg E.coli in the large intestines
- Others reside for a short period of time
- There is a Symbiotic relationship between the body and the microorganism

- Symbiotic relationship:
  - Mutualism: both the body and microbe benefit-Lactobacillus in the vagina gets nutrients producing acid that prevents overgrowth of other microbes
  - Commensalism: the relationship only favours the microbe

- But some parts of a normal healthy human hosts are free of these normal flora like the CSF, Blood, Urinary bladder, fallopian tubes, uterus, middle ear, kidney

- Normal flora derived from the host:
  - Supply of nutrients
  - Stable environment
  - Constant temperature
  - Protection
  - Transport

- Host benefits from normal flora;
  - Produce vitamins and nutrients
  - Competition with pathogens thus inhibit pathogens
  - Production of substances that inhibit pathogens
  - Stimulate development and activity of immune system

- Bacteria at one site may be a commensal but might be pathogenic at another e.g.
  - *E. coli* is commensal in GIT but pathogenic in the urinary tract
- Antibiotic overuse causes antibiotic associated diarrhoea
  - *C. difficile* antibiotic associated diarrhoea (CDAD) and Pseudomembranous colitis (inflammation of the large intestines)

References and further readings

- Patrick .R. Murray, (5th Edition), *Medical Microbiology* (currently in library)
- Mims et al, (4th Edition), *Medical Microbiology* (currently in library)
- Monica Cheesbrough, Part 2 (2nd Edition), *District Laboratory Practice in Tropical Countries* (currently in library)
- Jeffrey Pommerville (7th Edition) *Alcamo`s Fundamentals of Microbiology*