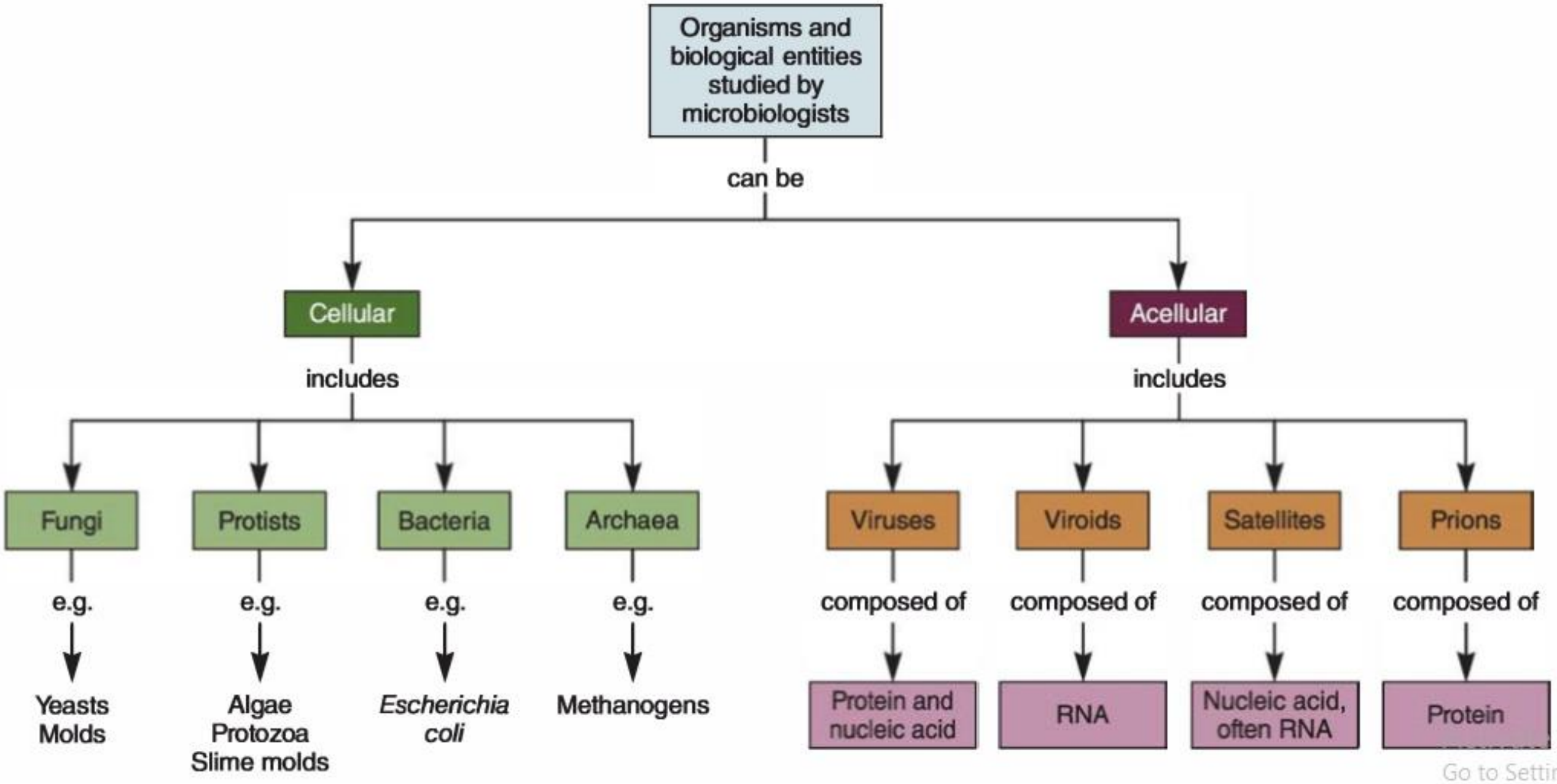
Lesson 1
Introduction to microbiology. Classification, morphology, ultrastructure and investigation methods of bacteri

Subject microbiology

***Microbiology**-(Greek **mikros**-small, **bios**-life, **logos**-science) is a science that studies the patterns of vital activity of microorganisms invisible to the naked eye.*

***General microbiology**-studies the morphology (shape and structure), physiology (nutrition, metabolism, respiration and reproduction), genetics (heredity and variability) of microorganisms.*

***Private microbiology** - studies the characteristics of individual microorganisms. In this regard, it is divided into such sections as bacteriology, virology, mycology, protozoology.*


Organisms and biological entities studied by microbiologists
can be
Cellular
includes
Fungi
Protists
Bacteria
Archaea
e.g.
e.g.
e.g.
e.g.
Yeasts
Molds
Algae
Protozoa
Slime molds
Escherichia coli
Methanogens
Acellular
includes
Viruses
Viroids
Satellites
Prions
composed of
composed of
composed of
composed of
Protein and nucleic acid
RNA
Nucleic acid, often RNA
Protein

Classification of prokaryotes

- *D.Bergey-The modern classification of prokaryotes was proposed by an American bacteriologist in 1923. It is regularly updated by the International Committee on Systematics of Bacteria.*
- *In the last 9th publication, prokaryotes are divided into four categories according to the structure of the cell wall.*

Modern classification of prokaryotes according to Bergey

- Gram-negative, cell-walled eubacteria
- Gram-positive, cell-walled eubacteria
- Eubacteria without a cell wall
- archaebacteria

In the modern classification of prokaryotes, there is24 types, 33 classes

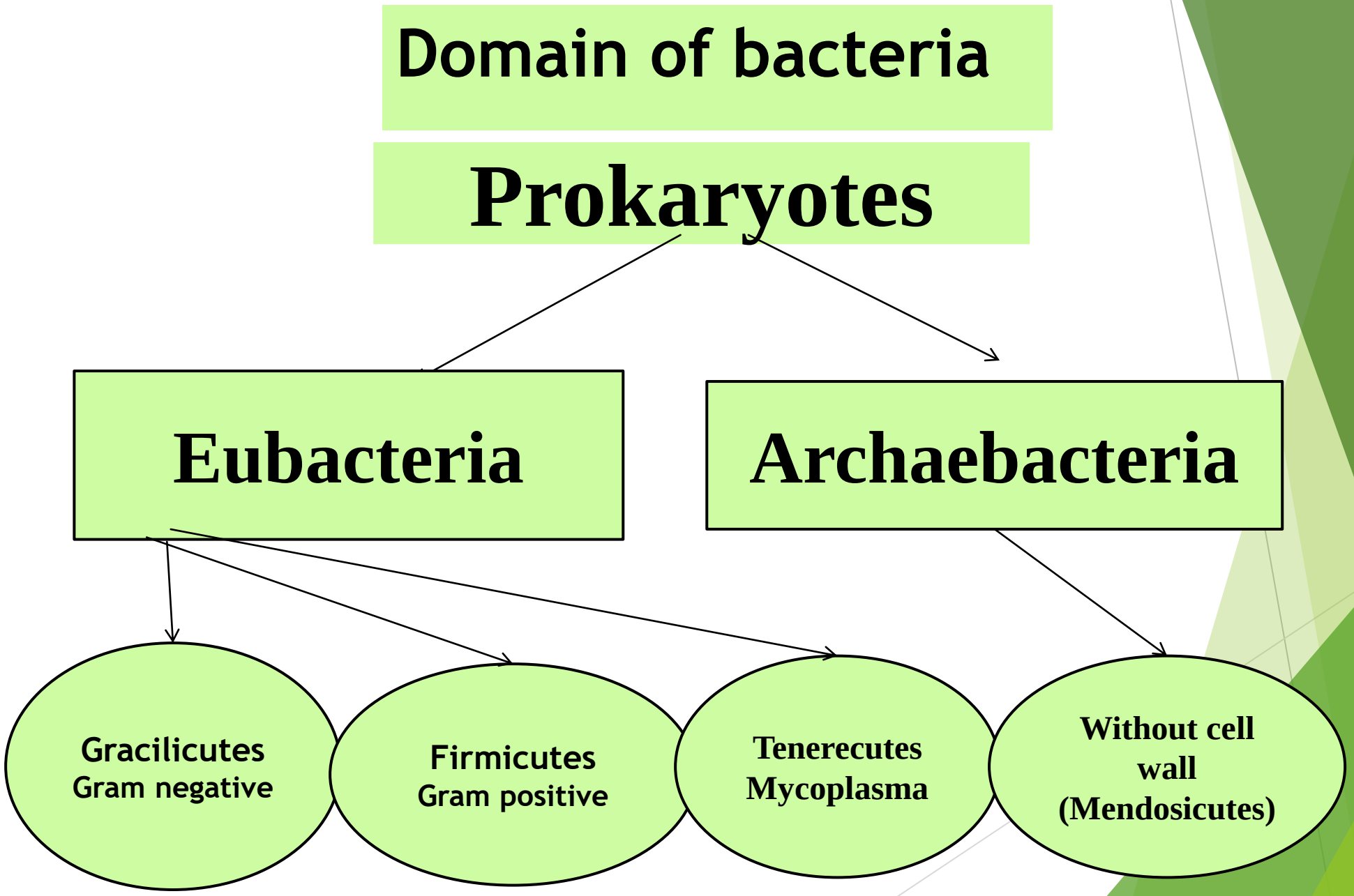
1. *Bacteria with a thin Gram-negative cell wall - Gracilicutes 16 groups*

2. *Eubacteria with a thick Gram-positive cell wall – Firmicutes 13 groups*

3. *Eubacteria that do not have a cell wall - mycoplasmas - Tenericutes class Mollicutes - 1 group*

4. *Archaebacteria lack a cell wall and peptidoglycan. They do not cause disease in humans. Includes 5 groups.*

Classification and taxonomy of bacteria



General characteristics of bacteria

- ✓***Bacteria (Greek bacteria-stick) single-celled microorganisms not visible to the naked eye***
- ✓***prokaryotes***
- ✓***Sedimentation of 70S ribosomes***
- ✓***They do not have a nuclear membrane or nucleolus***
- ✓***Have one chromosome***
- ✓***Mitochondria, lysosomes, Golgi apparatus, endoplasmic reticulum are absent***
- ✓***There are no sterols in the cytoplasmic membrane (with the exception of mycoplasmas)***

Bacteria sizes

- *The length of bacteria varies from 1.5-3 microns (for mycoplasma 0.1-0.2 microns) to 10-15 microns (for the causative agent of gas gangrene 4-8 microns).*
- *Diameter - 0.6-0.8 microns*
- *Thickness - 0.1- 2.5 microns*

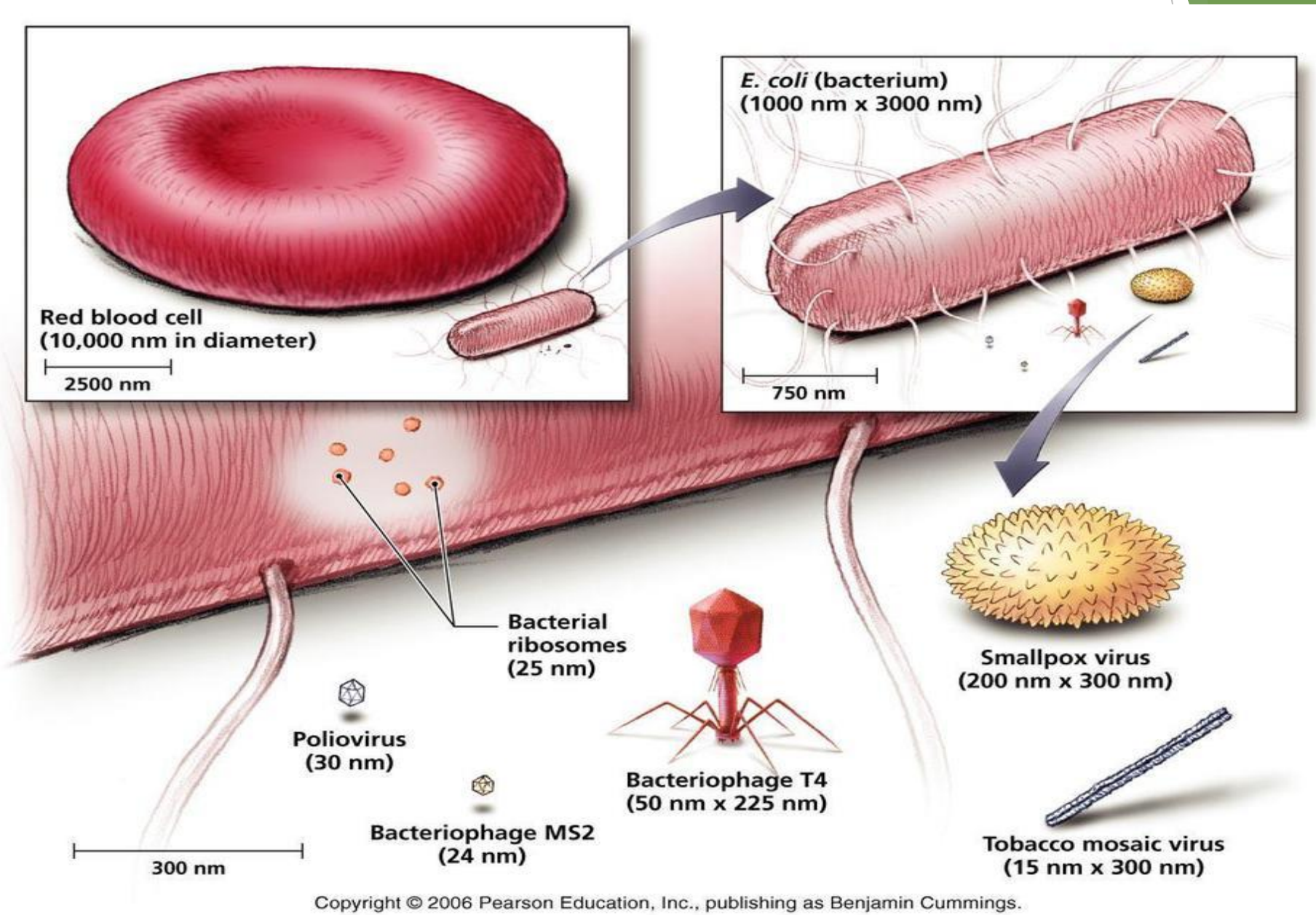
1 mkm = 10^{-6} m = 10^{-3} mm

1 nm = 10^{-9} m = 10^{-6} mm

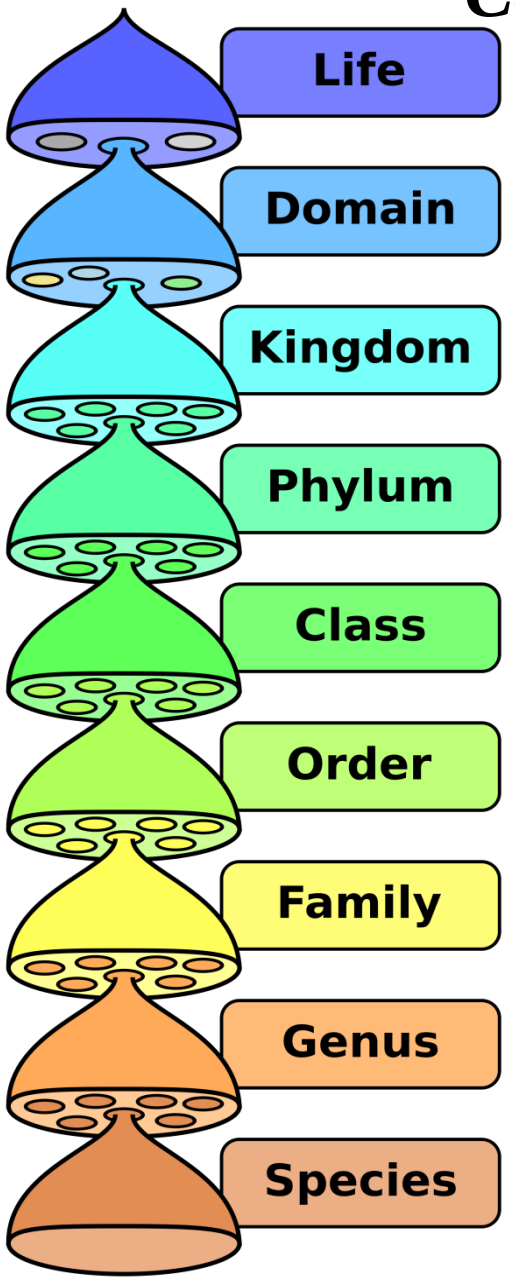
1000 nm = 1 mkm

0.001 mkm = 1 nm

Bacteria sizes

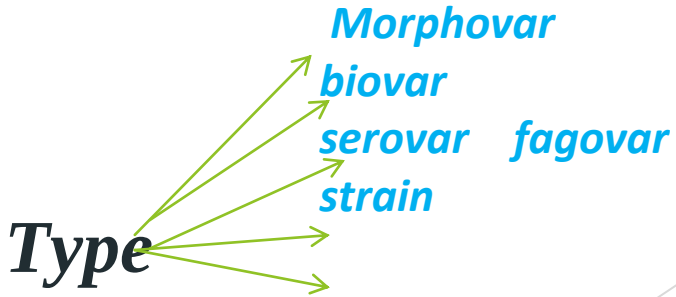


Taxonomy of microorganisms
Categories of classification



Each microorganism in systematics has a certain taxonomy (Greek taxis-place, row). The taxonomy is:

- ***Classification***
- ***Identification***
- ***Nomenclature***



ТАКСОНОМИЧЕСКИЕ КАТЕГОРИИ

Kingdom	**Prokaryotae**
Division	**Gracilicutes**
Class	**Scotobacteria**
Order	**Eubacteriales**
Family	**Enterobacteriaceae**
Genus	**Escherichia**
Species	**coli**
Subtype	**Escherichia coli O157:H7**

It is believed that for taxa of high rank, the name is not "Department", but "Type" is more suitable. (Phylum).

Nomenclature of microorganisms

- *The nomenclature of microorganisms created by K. Linnaeus is used to designate their names (except for viruses).*
- *In this case, the first word denotes the genus, it is written with a capital letter, the second word denotes the species and is written with a small letter.*

For example,

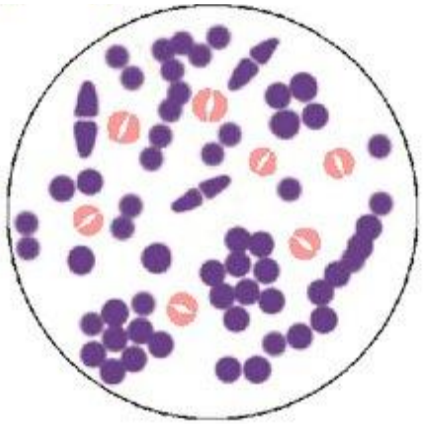
- *Mycobacterium tuberculosis*
- *Francisella tularensis*
- *Staphylococcus aureus*

- **Species**-are microorganisms that have a common origin and similar morpho-biological properties.
- **A strain-**is a pure culture of one type of microorganism obtained from different (or the same) sources at different times.
- **A clone** is a culture grown from a single microbial cell.
- **A colony** is an accumulation (population) formed by bacteria on solid nutrient media.
- **A pure culture** of a microbe means a population formed by one type of microorganism on a dense nutrient medium.

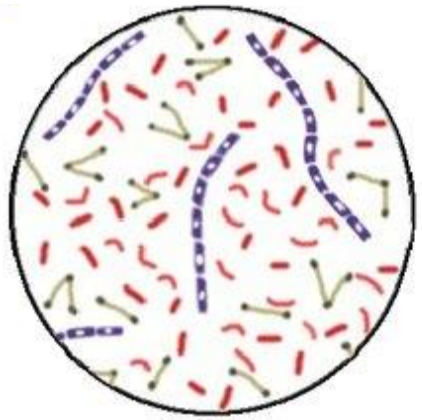
Variants of intra specises

- *morphovar - a variant that differs from the main species in morphological properties.*
- *biovar - difference in several biological properties*
- *serovar - difference in antigenic structure*
- *fagovar - by sensitivity to a particular phage*
- *chemovar - difference in biochemical properties*
- *resistovar - difference in sensitivity to antimicrobial drugs*

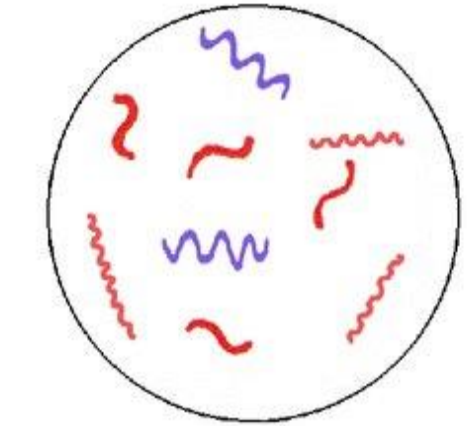
Morphology of bacteria



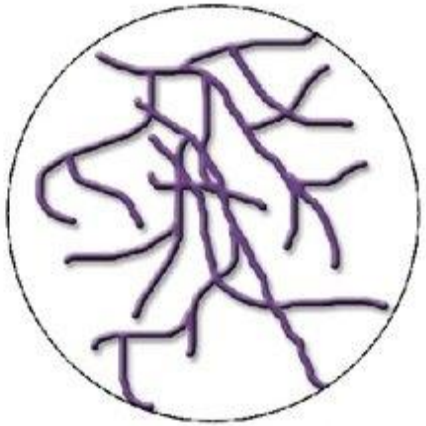
Cocci



Rod-shaped bacteria

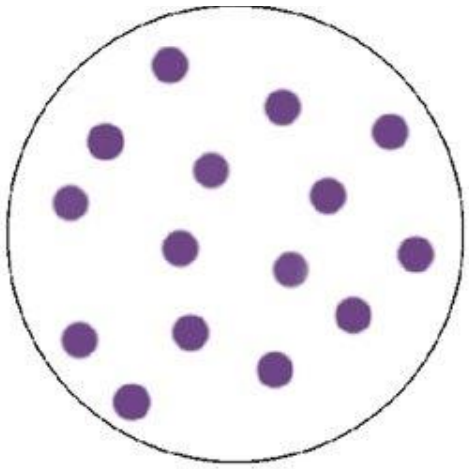


Spiral bacteria

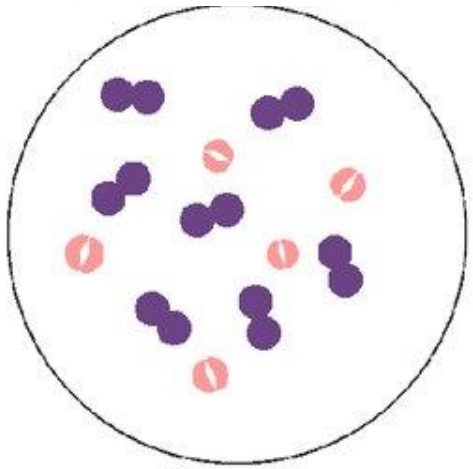


Filamentous bacteria

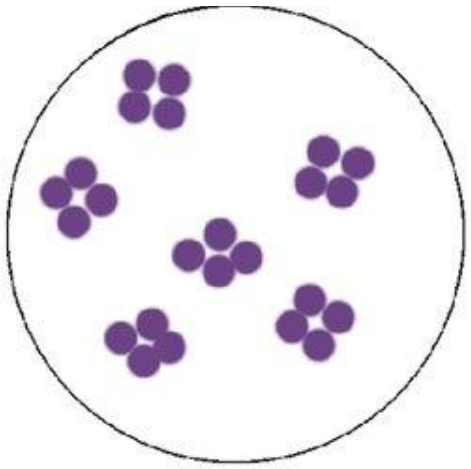
Spherical bacteria and cocci (0.5-1.5 µm)



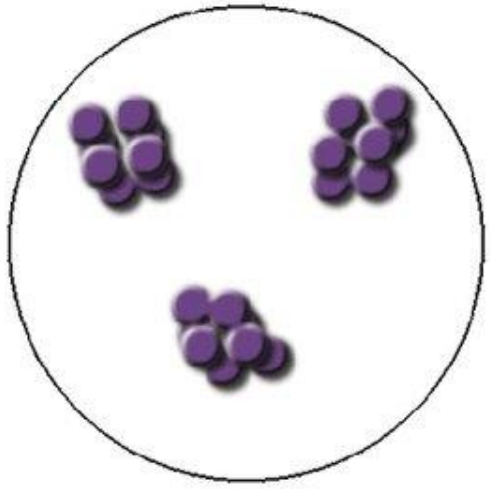
Micrococcus



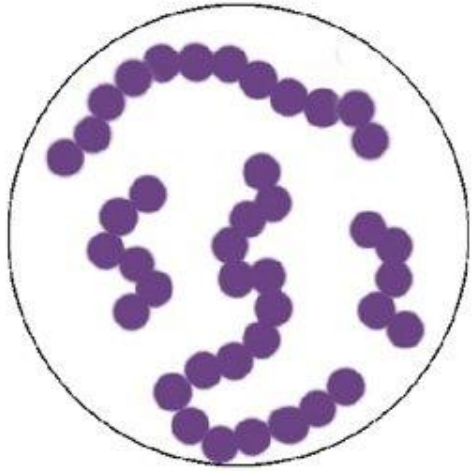
Diplococcus



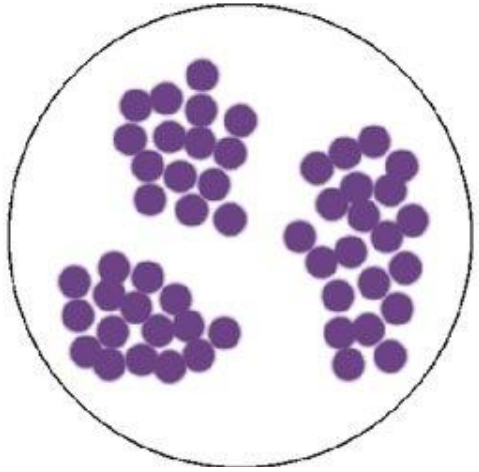
Tetracoccus



Sarcina



Streptococcus



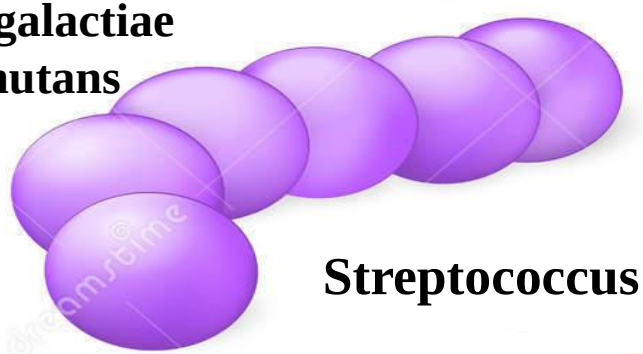
Staphylococcus

Cocci

N.meningitidis
N.gonorrhoeae
S.pneumonia

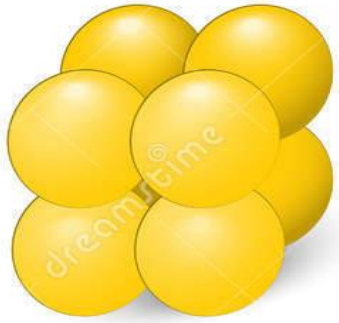
S.pyogenes
S.agalactiae
S.mutans

Diplococcus



Micrococcus

Streptococcus



Sarcina

Tetracoccus

Staphylococcus aureus
S.epidermidis,
S.saprophyticus

Cocci

***Micrococci** (Greek micros - **small**) - divided transversely, arranged separately*

***Diplococci** (Greek diplos - **together**) - divided transversely, arranged in pairs*

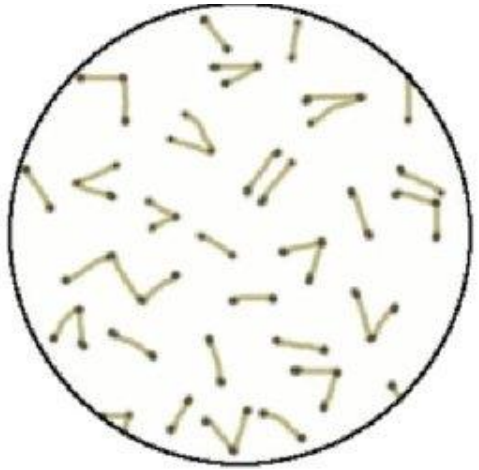
***Streptococci** (Greek Streptos - **chain**) - divide transversely, arranged in chains*

***Tetracocci** (Greek tetra - **four**) - are divided perpendicularly in two planes and are arranged in four*

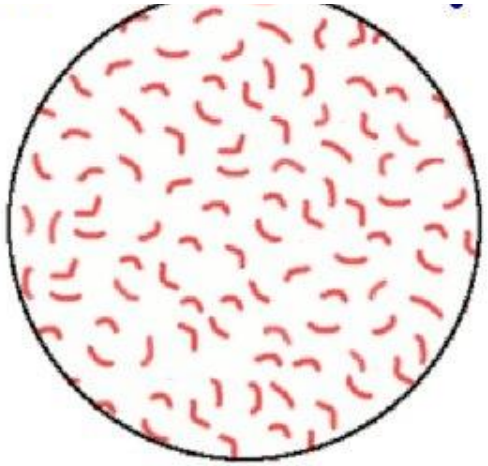
***Sarcins** (lat. sarsina - **sheaf**) - are divided perpendicularly in three planes and are arranged in the form of sheaves*

Staphylococci (Greek staphyle - a bunch of grapes) - are divided perpendicularly in two planes and are arranged in the form of bunches of grapes

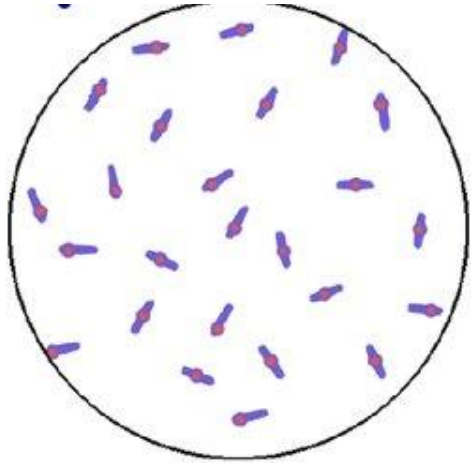
Rod-shaped bacteria (0.3-10 mkm)



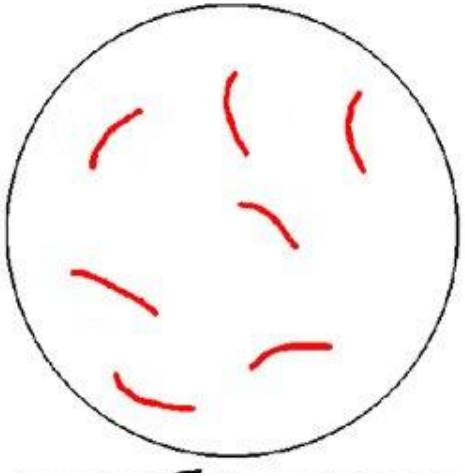
corynebacteria



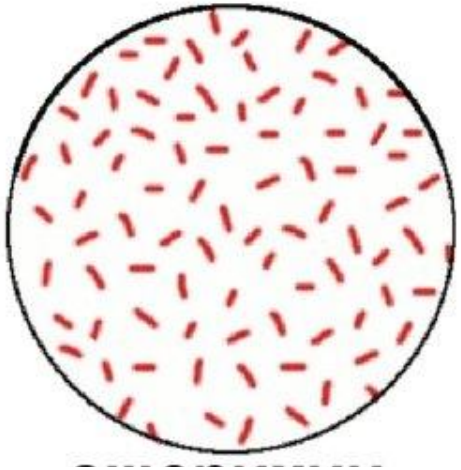
vibrios



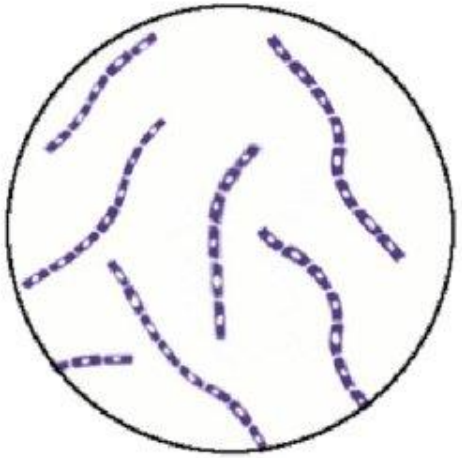
clostridia



mycobacteria



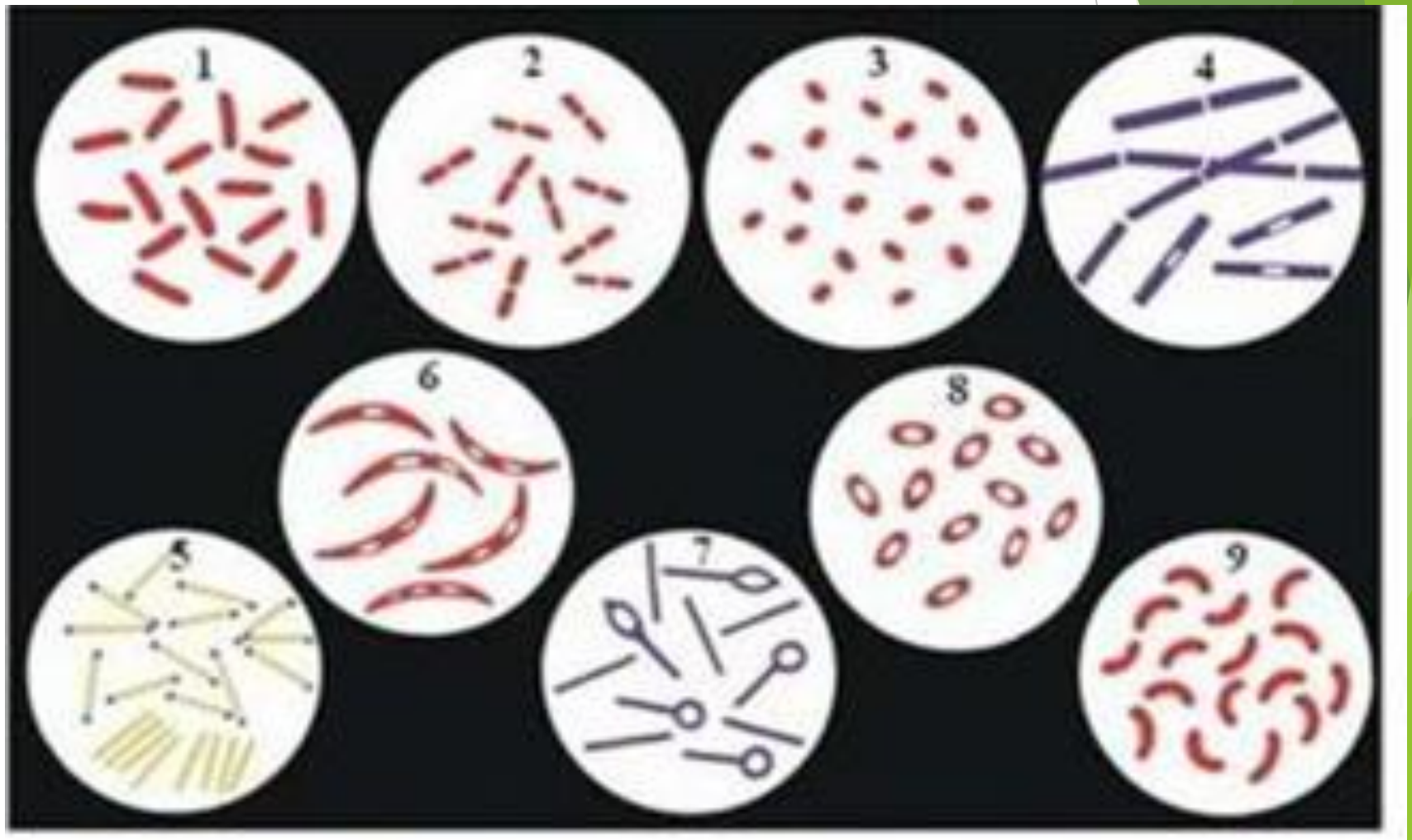
Escherichia



streptobacilli

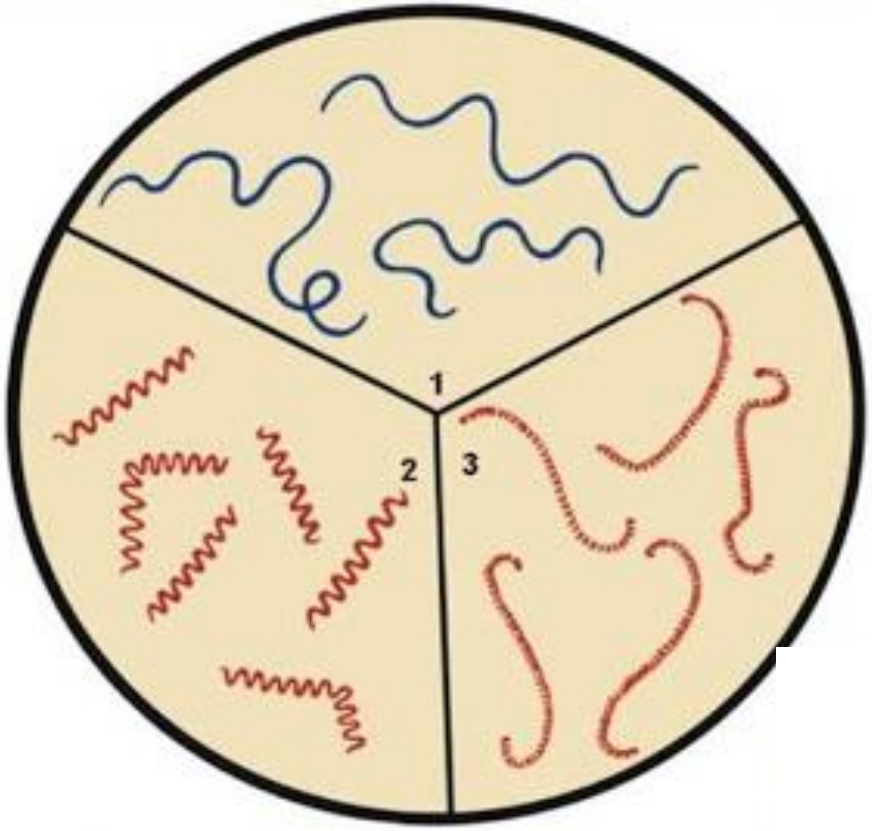
Rod-shaped bacteria

1. Escherichia
2. Klebsiella
3. Brucella
4. Bacillus
5. Corynebacterium
6. Fusobacteria
7. Clostridia
8. Yersinia
9. vibrios



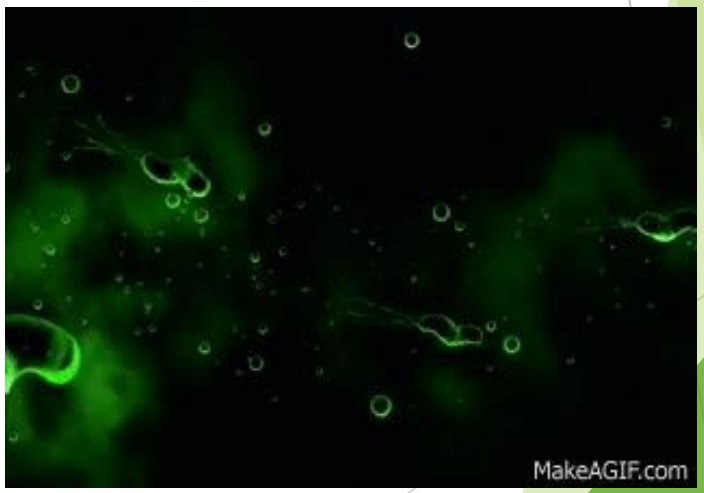
Spiral bacteria (<20 mkm)

- *Spirilla*
- *Spirochetes*



1. ***Borrelia***
2. ***Treponema***
3. ***Leptospira***

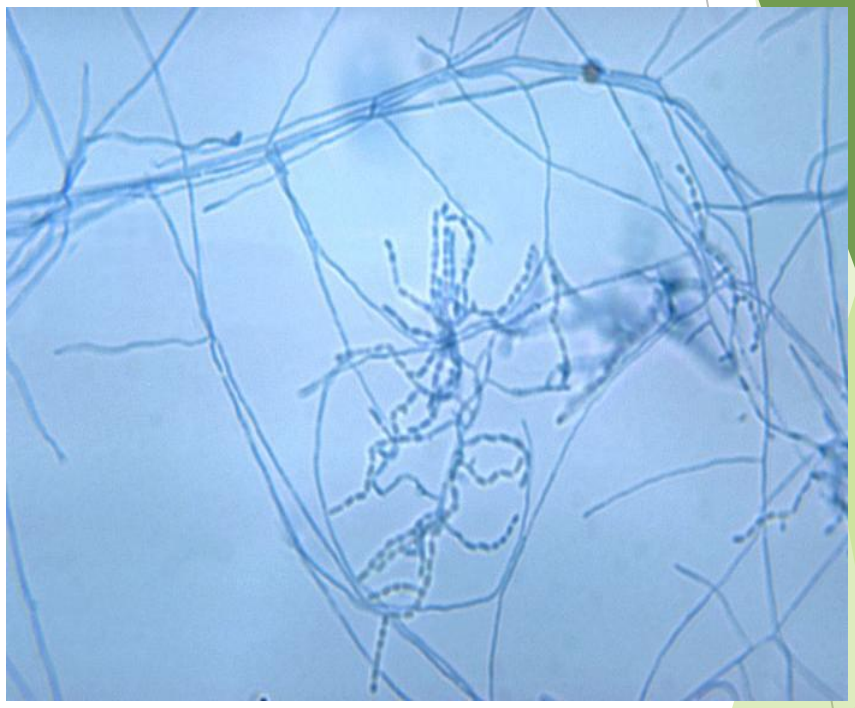
Campylobacter



Helicobacter pylori

Filamentous bacteria (10-50 microns)

Actinomycetes



Ultrastructure of a bacterial cell

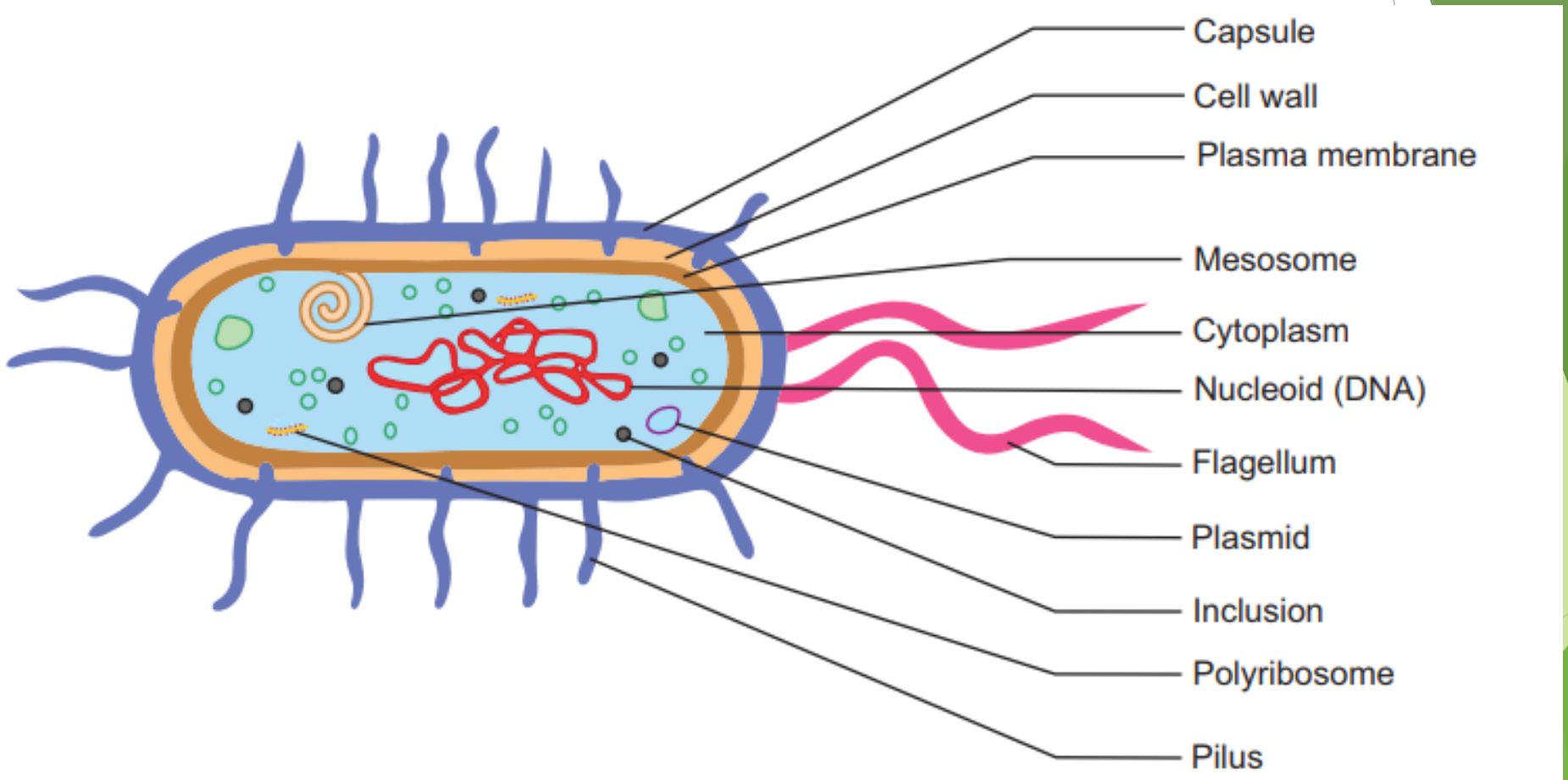
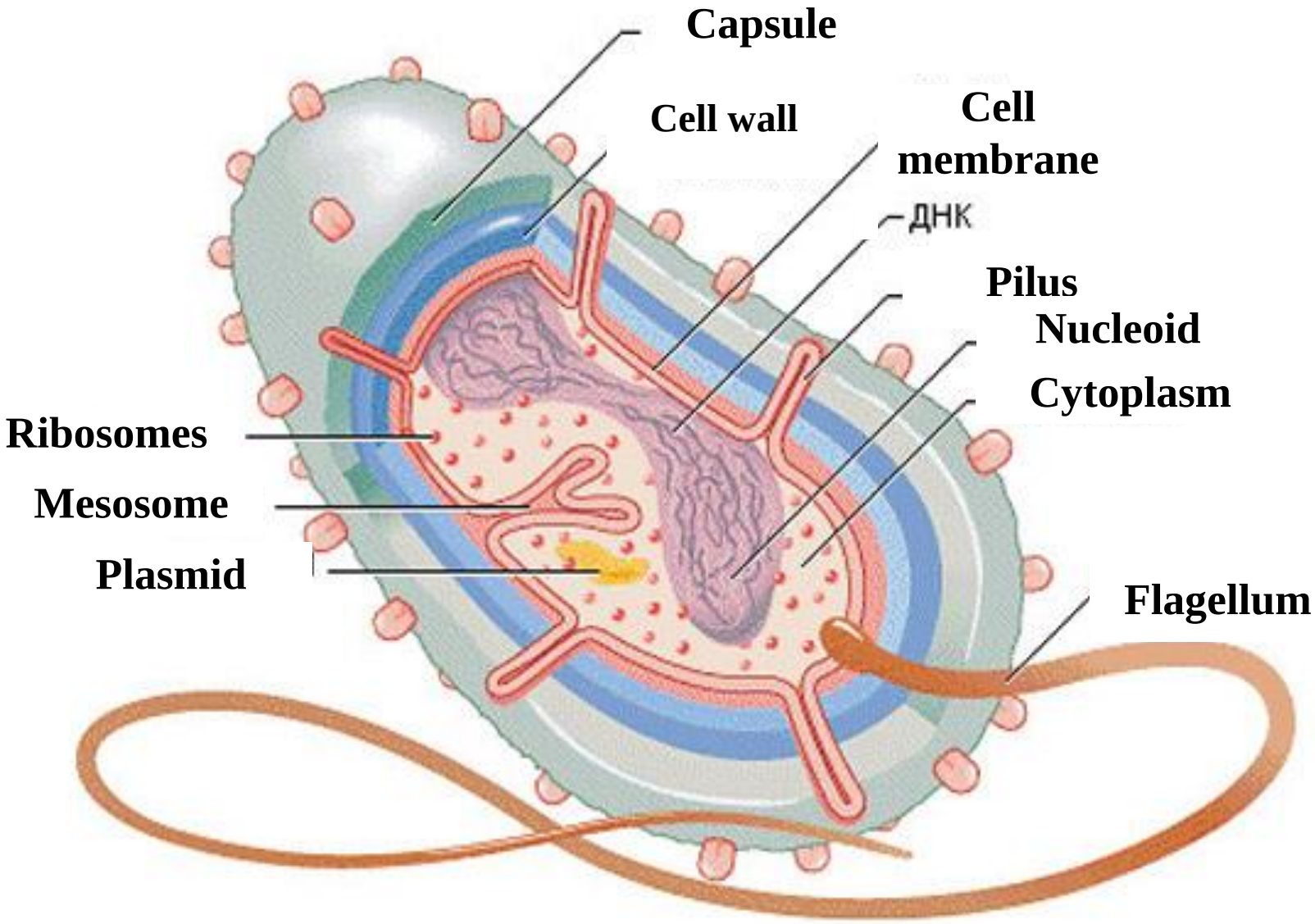
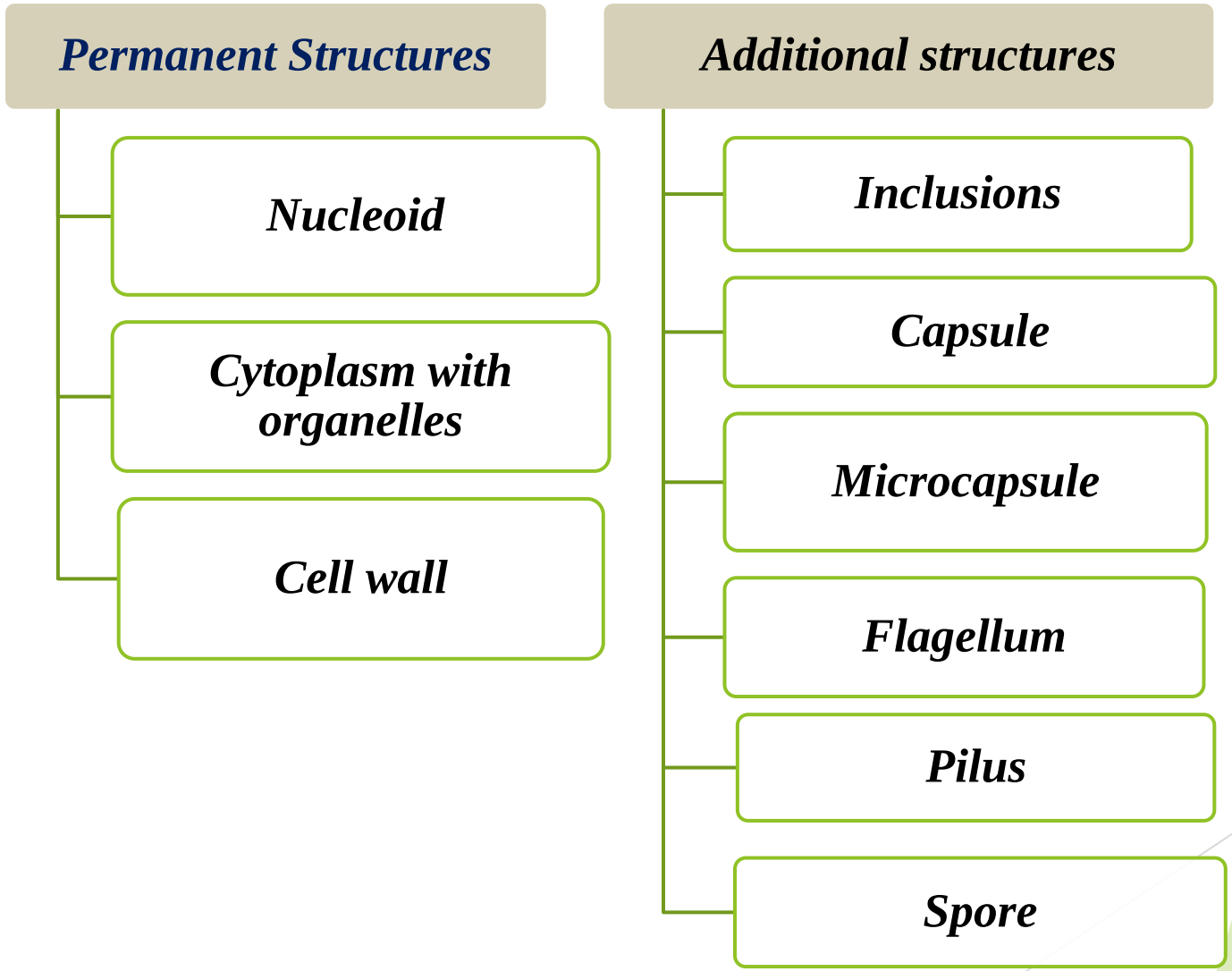


Figure 1.9: Ultrastructure of a bacterial cell

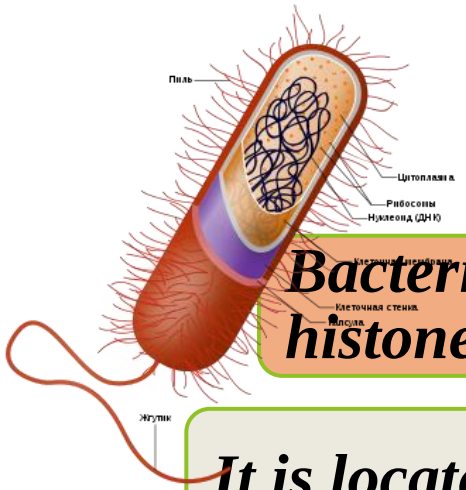
The structure of a bacterial cell



The structure of a bacterial cell



Nucleoid



Bacteria lack a nucleus, nuclear membrane, nucleolus, and histones

It is located in the cytoplasm, consists of 10 million bp.

Usually, a cell contains one haploid chromosome, represented by double-stranded DNA closed in a ring.

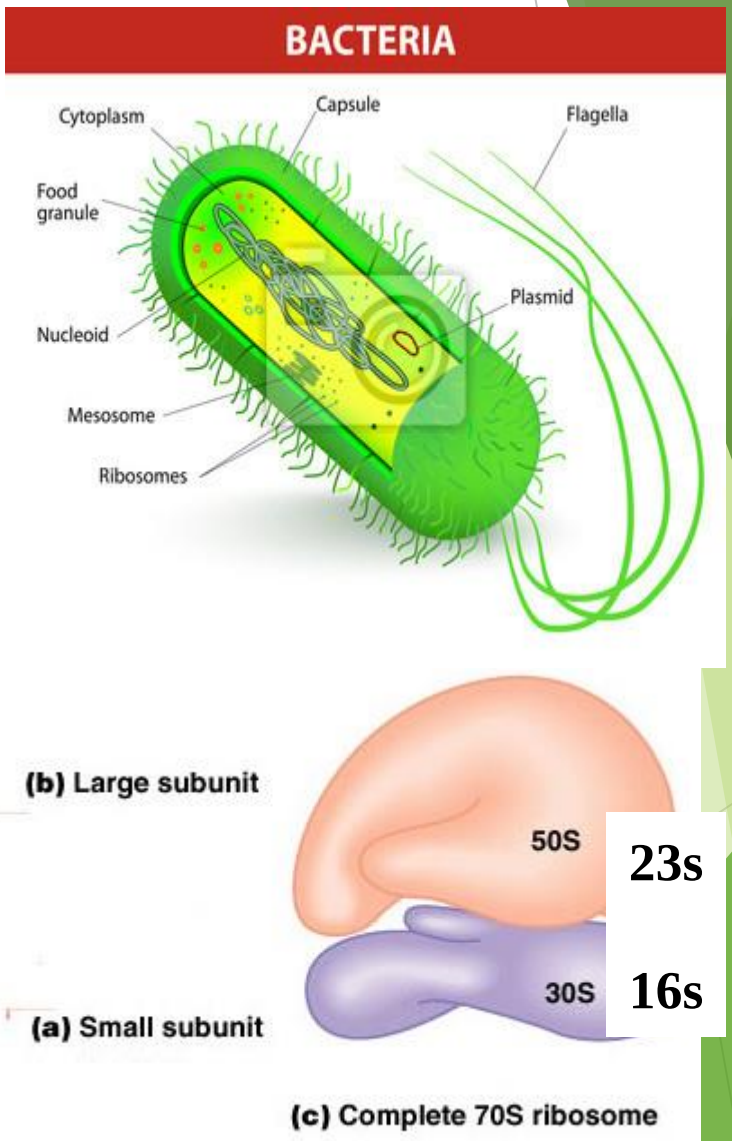
Borrelia burgdorferi has linear DNA

In addition to the chromosome, there are extrachromosomal factors of heredity - plasmids

Nucleoid is detected by the method of Feulgen and Giemsa

Cytoplasm and intracytoplasmic inclusions

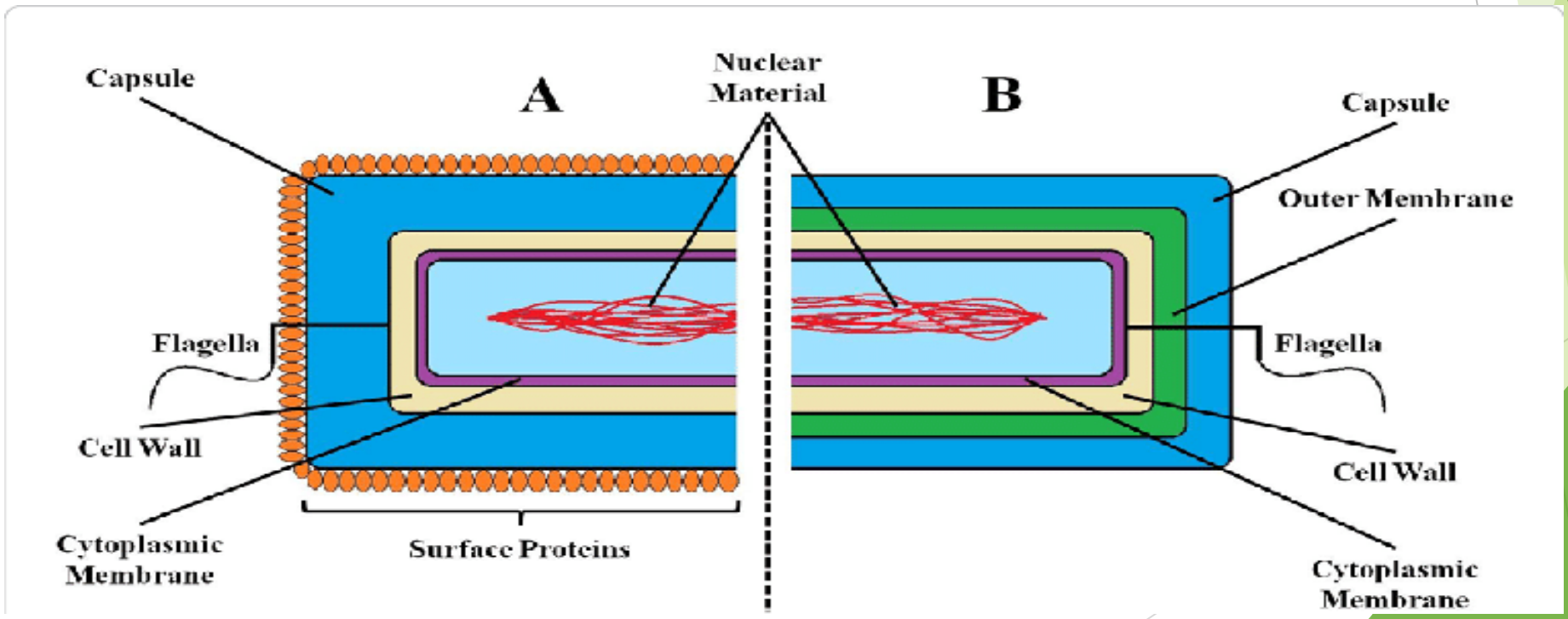
- ***Cytoplasm** is a colloidal matrix containing soluble proteins, inclusions and ribosomes (RNA)*
- *Bacterial ribosomes are about 20nm in size and have a sedimentation coefficient of 70S (50S and 30S are Svedberg units)*
- *23S-rRNA is part of 50S*
- *16S rRNA is part of 30S*
- *Various inclusions (glycogen granules, polysaccharides, lipids and polyphosphates) accumulate in the cytoplasm as reserve nutrients and an energy source.)*



Shell of a bacterial cell

The bacterial cell wall includes:

- ***cytoplasmic membrane***
- ***cell wall***
- ***Mucous layer - capsule, microcapsule, glycocalyx***



Functions of the cytoplasmic membrane

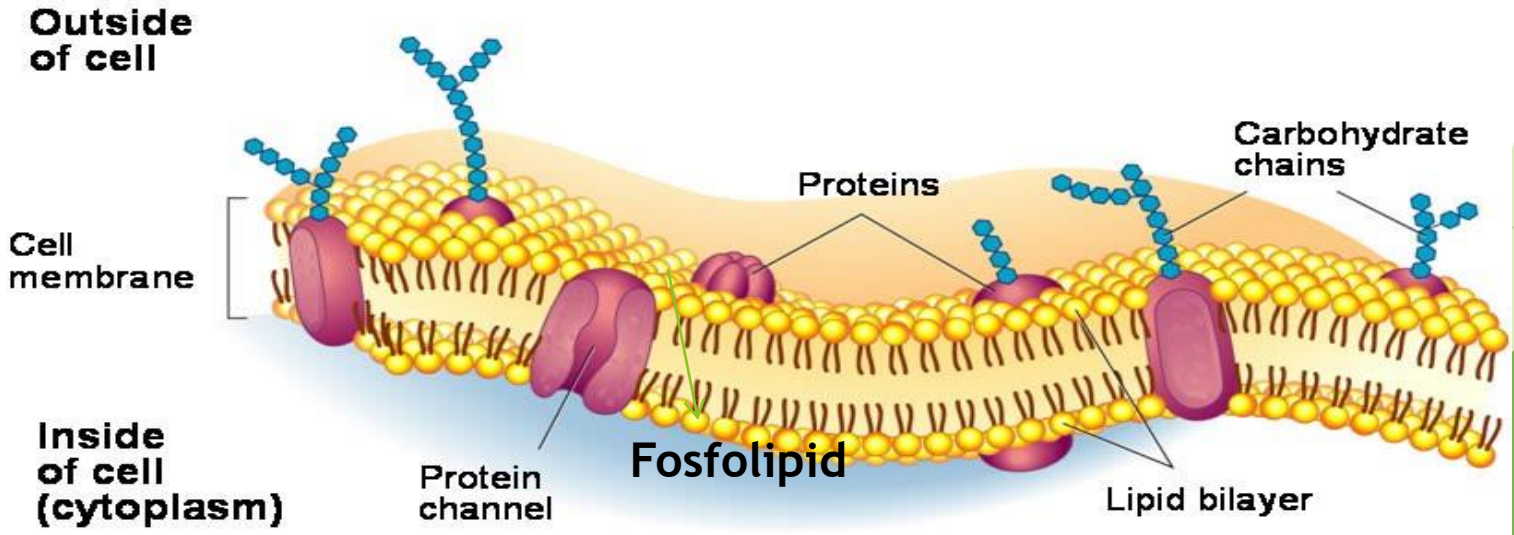
- ✓*Osmotic pressure regulation*
- ✓*Transmembrane proteins are involved in signal transmission, lipid layers determine the biological properties.*
- ✓*Possesses selective permeability.*
- ✓*Causes the transfer of substances through active transport*
- ✓*Uses an electron transport system for breathing.*

Functions of the cytoplasmic membrane

- ✓ *Participates in the transfer of biosynthetic and hydrolytic enzymes, transport and signal proteins.*
- ✓ *It has specific sites for binding to the chromosome and plasmids.*
- ✓ *The inner layers of the CPM contain actin-like protein fibers that determine the morphology of bacteria. These fibers provide the helical shape of the treponema.*

Cytoplasmic membrane

- *Free of sterols (except for mycoplasmas)*
- *Consists of a bilipid layer (phospholipids) and embedded membrane proteins*
- *The main function is energy synthesis and electron transport*
- *Contains transpeptidase (penicillin-binding protein)*
- *mesosome invagination of the membrane into the cytoplasm*
- *(in gram-positive bacteria, they function as mitochondria)*
- *central mesosome DNA replication*
- *lateral mesosome protein-enzyme synthesis*



Cell wall

Protective layer surrounding the cytoplasmic membrane

- *Gives shape to the bacterial cell*
- *Performs a barrier function*
- *Protects the cell from osmotic lysis*
- *Provides interaction with the host cell*
- *Detected by the Gram method*
- *Plays a role in the pathogenesis of bacterial infections*

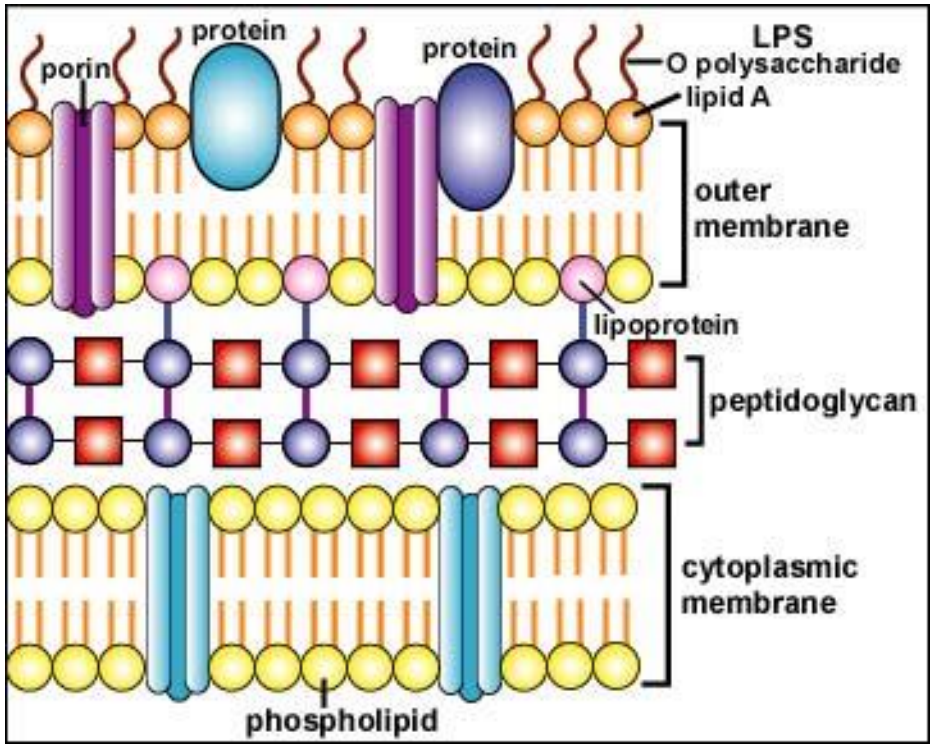
Cell wall

- *The cell wall of bacteria has a thickness of 15–20 nm and makes up 20–30% of the dry residue.*
- *The cell wall is a strong structure that gives bacteria a certain shape, has a complex structure and consists of several layers.*
- *Different attitudes towards Gram staining, and the division of bacteria into two groups - Gram-positive and Gram-negative, is based on the difference in the structure of their cell wall.*

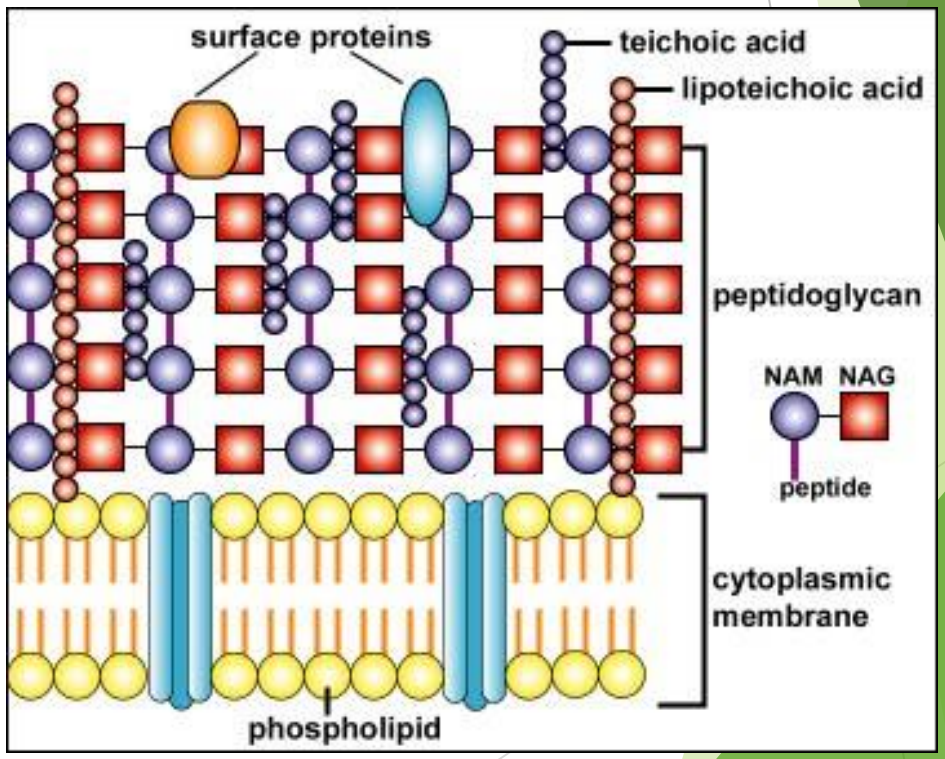
Differences between the cell wall of Gram-positive and Gram-negative bacteria

- ***Gram-positive** bacteria have a thicker cell wall with a thickness of 50 nm or more, 40-80% of it is peptidoglycan.*
- ***Gram-negative** bacteria have a thinner cell wall, 15-20 nm thick, peptidoglycan makes up 5-10% of the cell wall mass*

Scheme of the structure of the cell wall of gram-negative bacteria



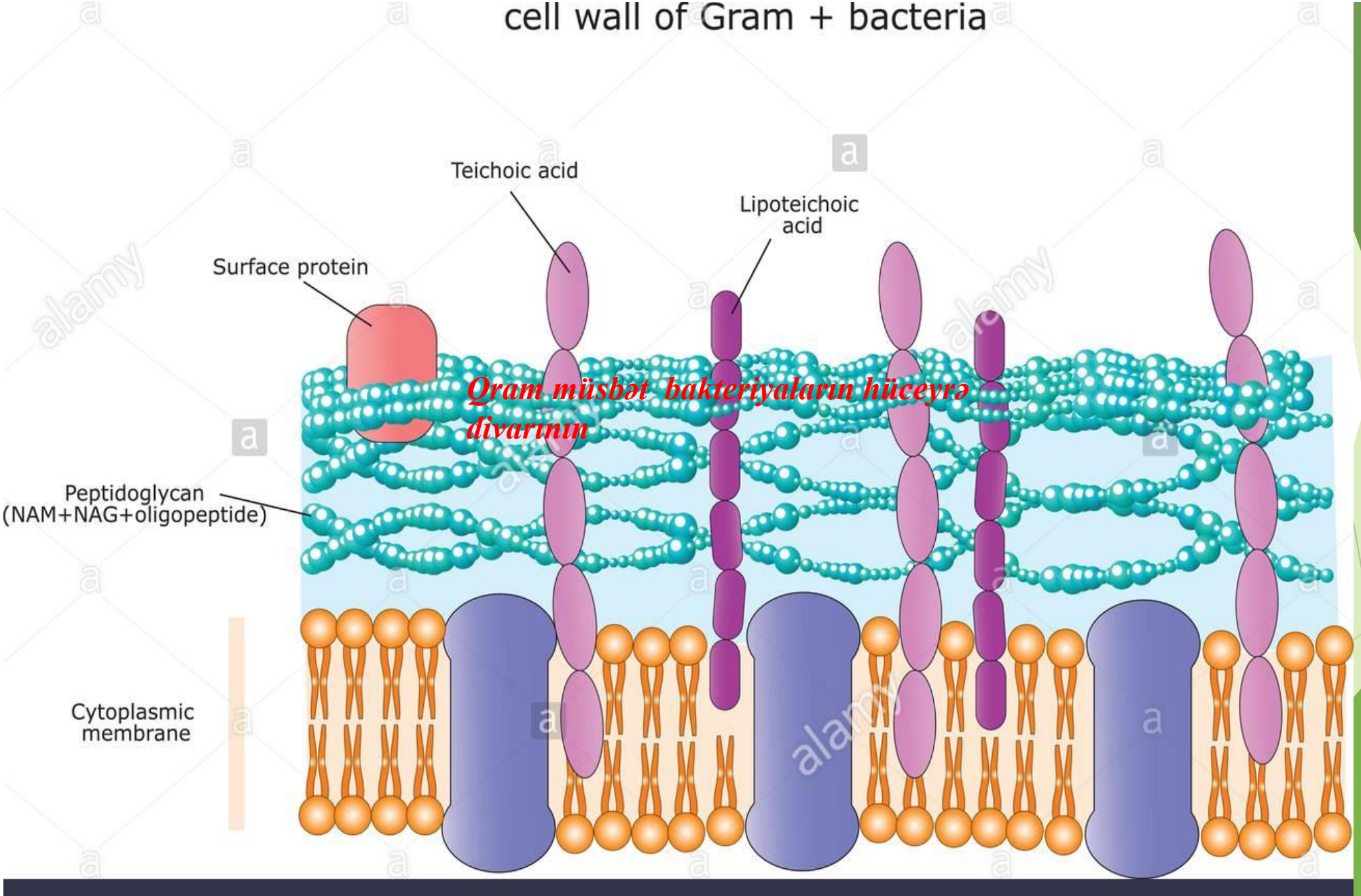
Scheme of the structure of the cell wall of positivebacteria



Differences between the cell wall of Gram-positive and Gram-negative bacteria

Features	Gram+	Gram-
Thickness	20-80 nm	10 nm
Peptidoglycan content	>50%	10 -20 %
Teichoic acids	+	-
Lipids and lipoproteins	0-3%	58%
Proteins	0%	9%
Lipopolysaccharide	0%	13%
Sensitivity to Penicillin	+	-
Sensitivity to Lysozyme	+	-

Cell wall of Gram-positive bacteria



Qram müsbət bakteriyaların hüceyrə divarının

The structure of the cell wall of gram-positive bacteria

✓*Teichoic acids (from the Greek teichos-wall) are covalently bound to the peptidoglycan of the cell wall.*

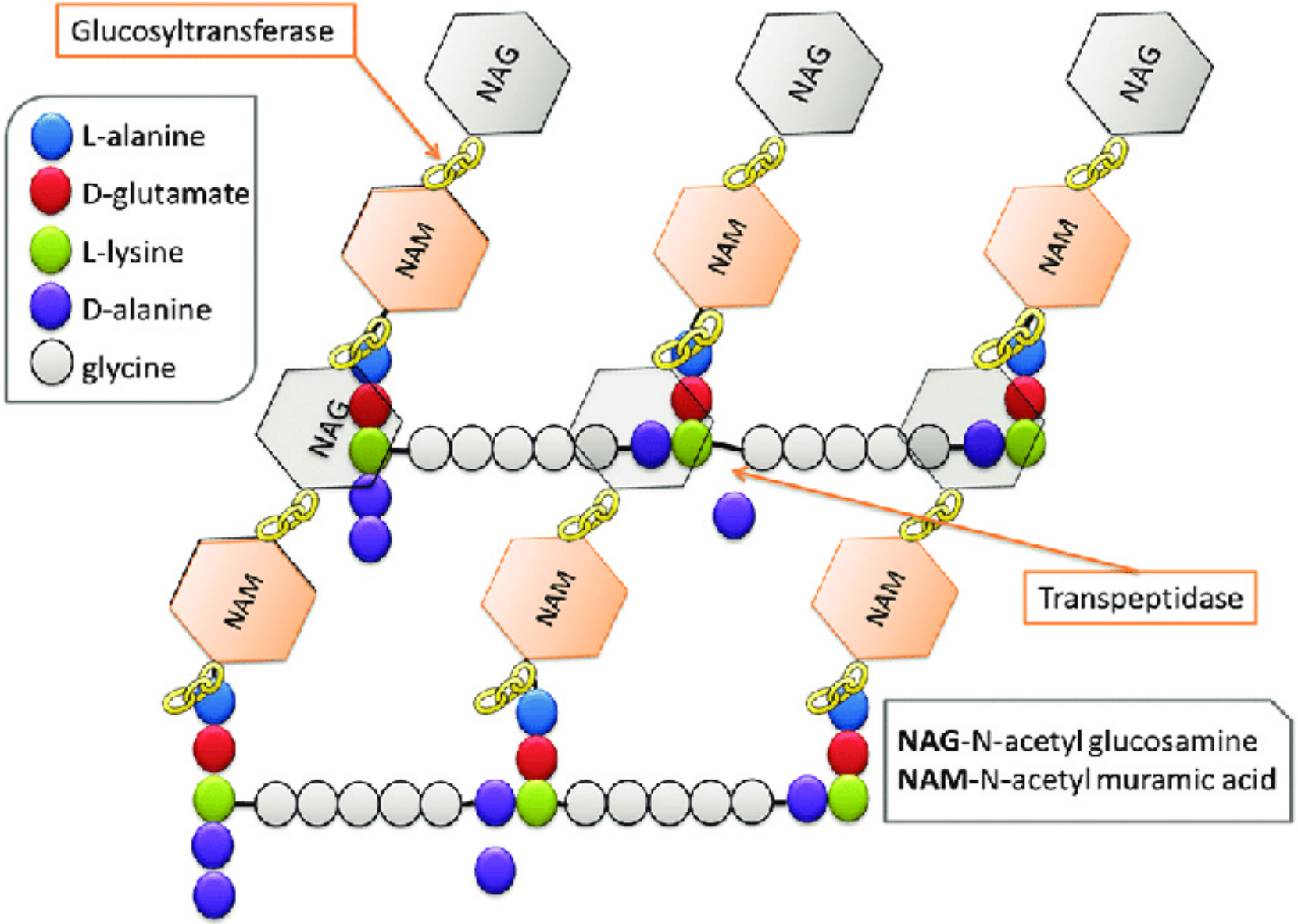
✓*Teichoic acid molecules are polymers of glycerol phosphate and ribitol phosphate.*

✓*Teichoic acids are soluble in water.*

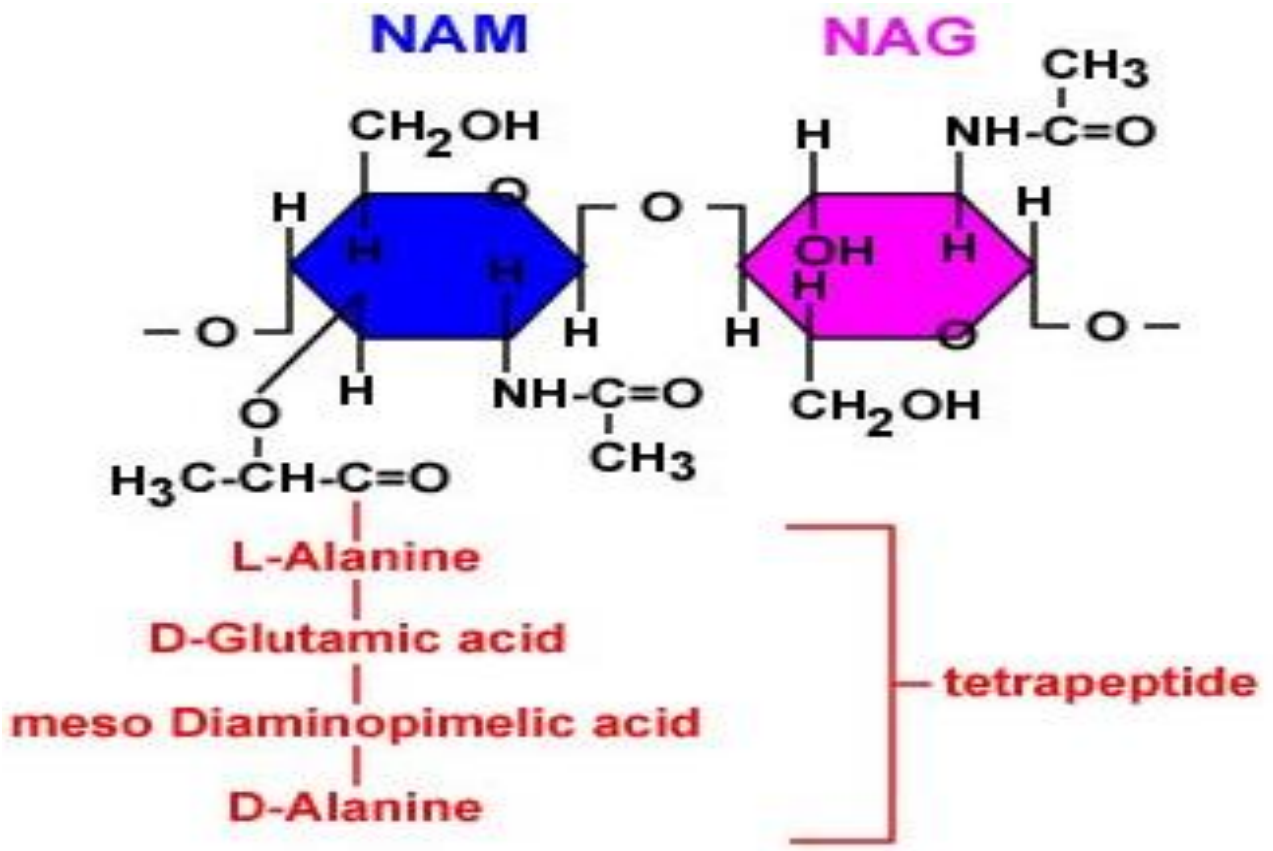
✓ *Teichoic acids are involved in cell division, in the regulation of the synthesis and disintegration of the cell wall, in adhesion to the cells of the body.*

✓*They are a pathogenic factor.*

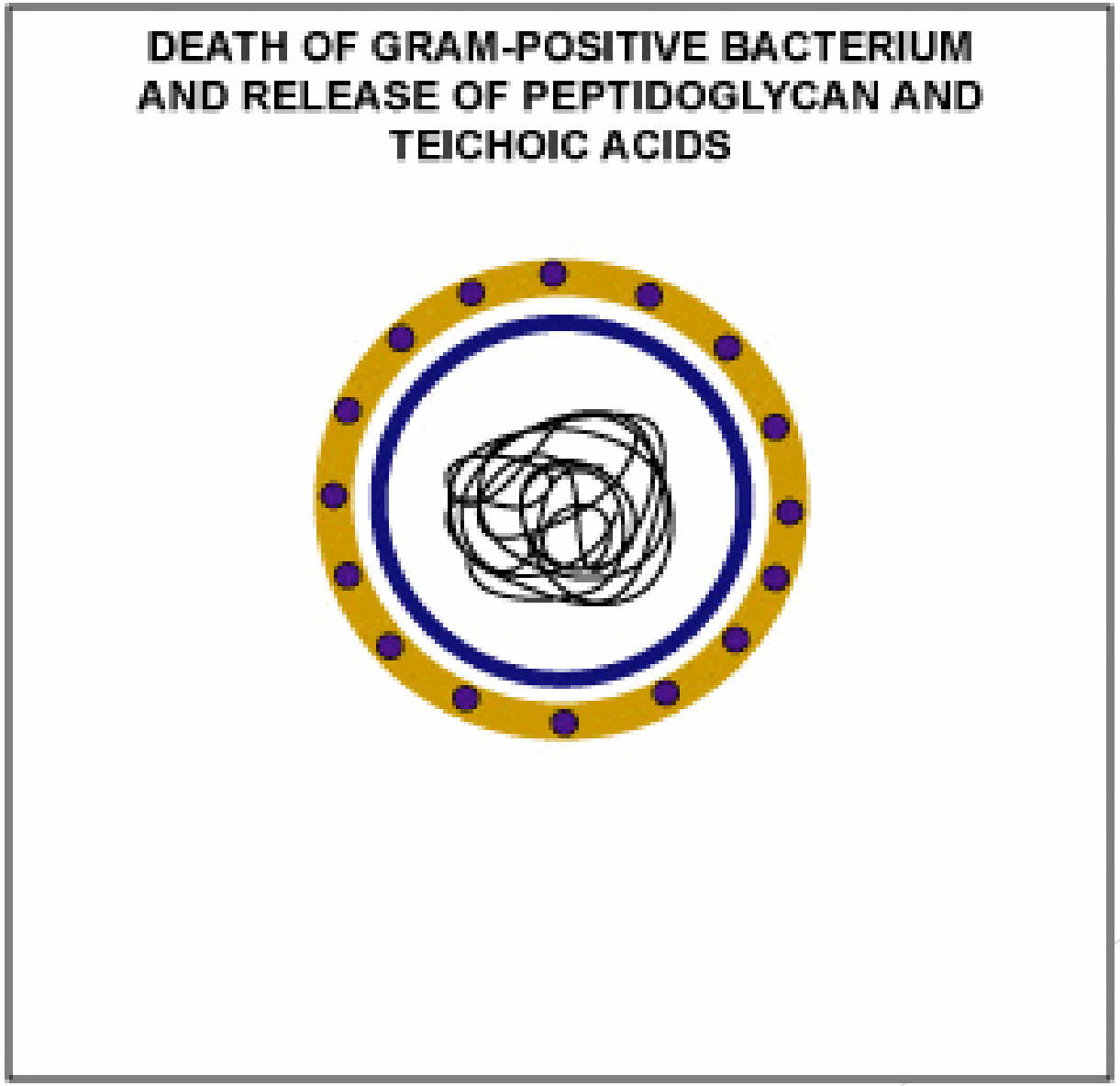
Structure of peptidoglycan



Structure of peptidoglycan



Biological activity of peptidoglycan



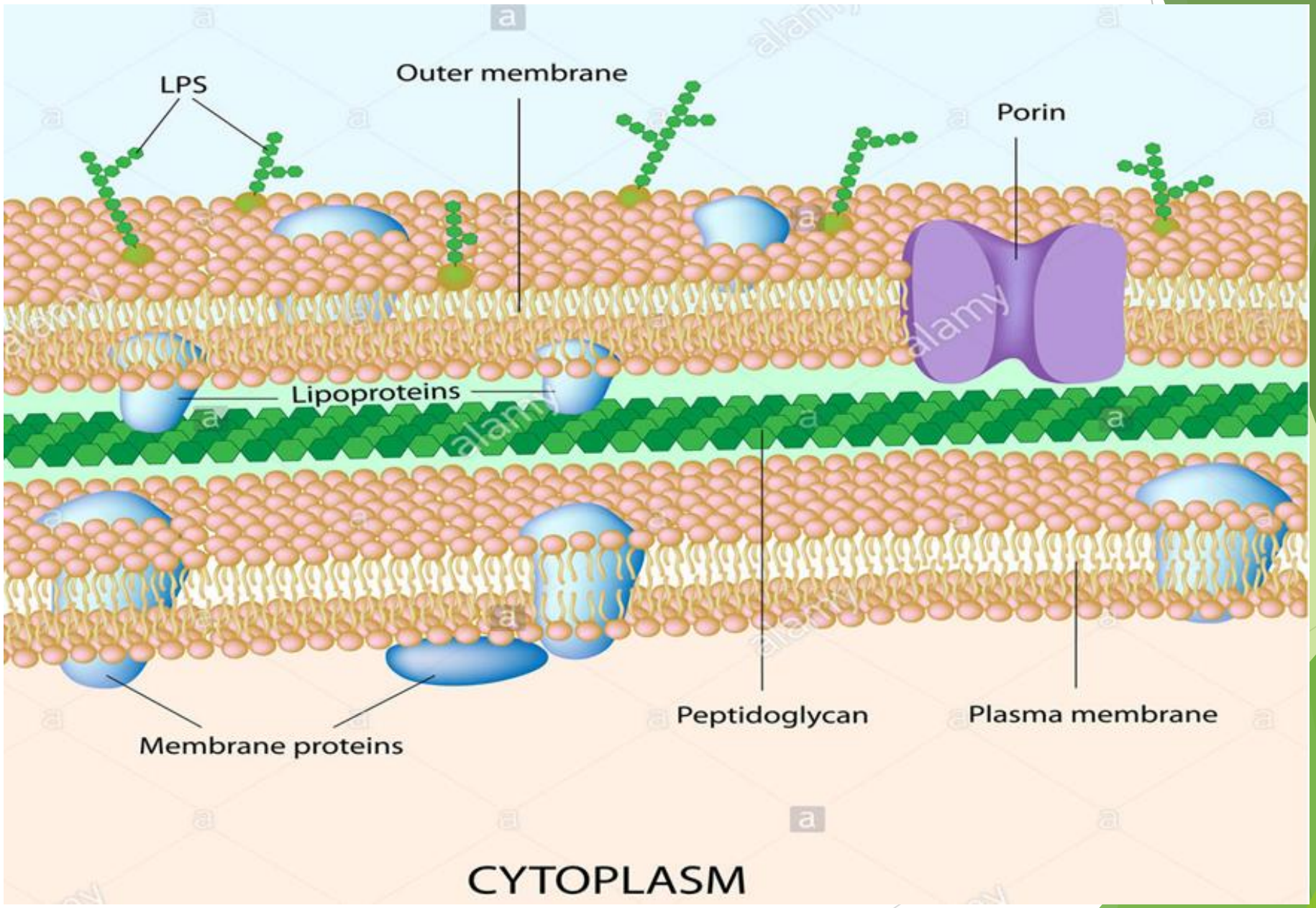
Cell wall of Gram-negative bacteria

- *The outer layer of the cell wall of Gram(-) bacteria is the outer membrane.*
- *The outer membrane contains phospholipids and LPS.*
- *Proteins (porins) of the outer membrane reduce the permeability of the cell wall of gram-negative bacteria.*
- *Between the outer and cytoplasmic membranes is the periplasmic space.*
- *The periplasm (makes up 20-40% of the cell wall) contains peptidoglycan and proteins.*
- *The periplasmic space contains adaptive enzymes and enzymes involved in metabolic processes (eg, beta-lactamase, etc.).*

Cell wall of Gram-negative bacteria

- The cell wall of gram-negative bacteria includes an outer membrane linked by a lipoprotein to the underlying layer of peptidoglycan. It consists of:
- Phospholipids,
- Lipoproteins,
- Lipopolysaccharides (LPS)

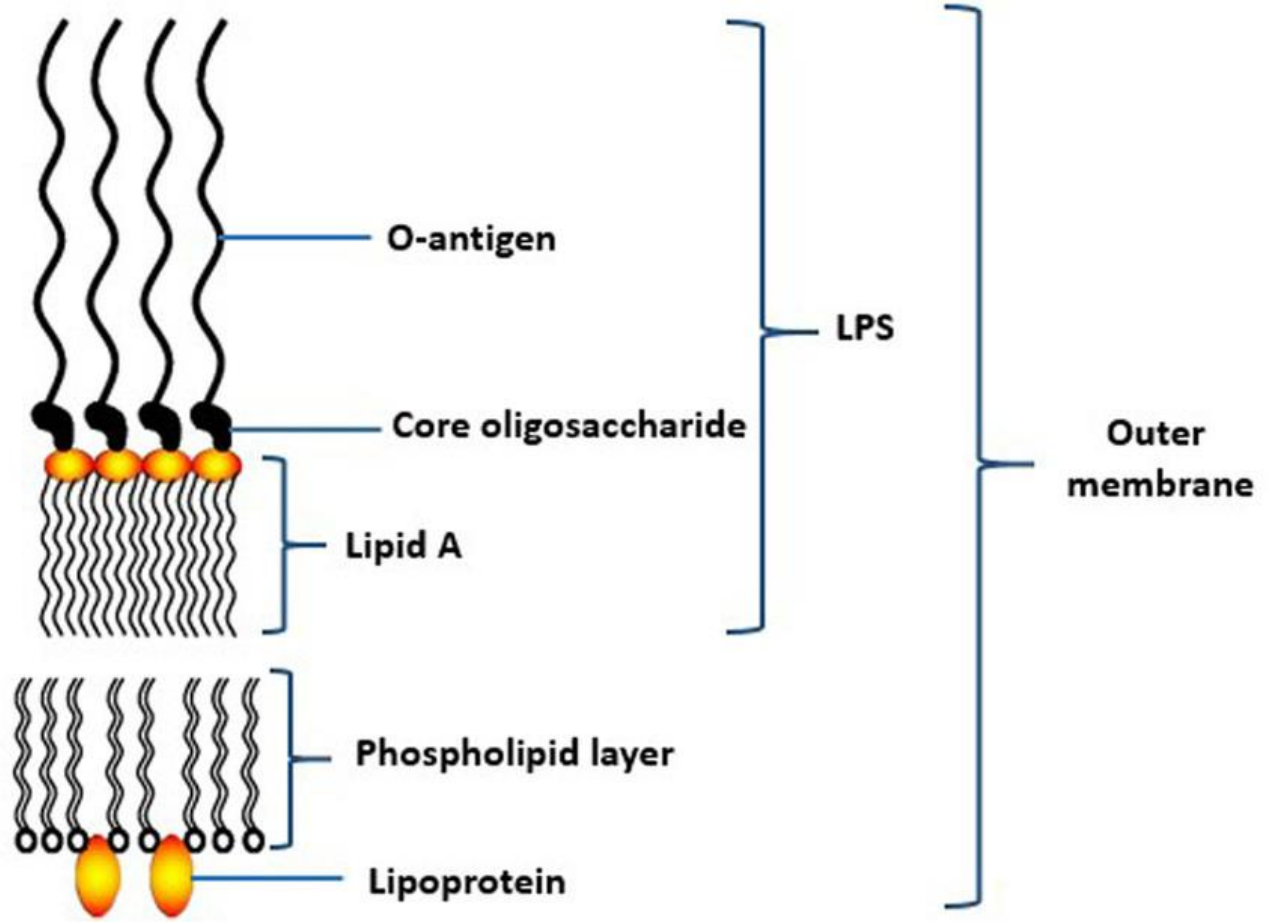
The structure of the cell wall Gram-negative bacteria



Outer membrane of the cell wall of Gram-negative bacteria

- The inner layer of the outer membrane is represented by phospholipids, and lipopolysaccharide is located in the outer layer.
- The outer membrane of Gram-negative bacteria differs in permeability from other biological membranes.
- Due to the content of lipids, it is characterized by hydrophobicity.
- Protein molecules called porins line hydrophilic pores in the outer membrane through which water and small hydrophilic molecules (sugars, amino acids, etc.) pass by passive diffusion.

Cell wall of Gram-negative bacteria (outer membrane)

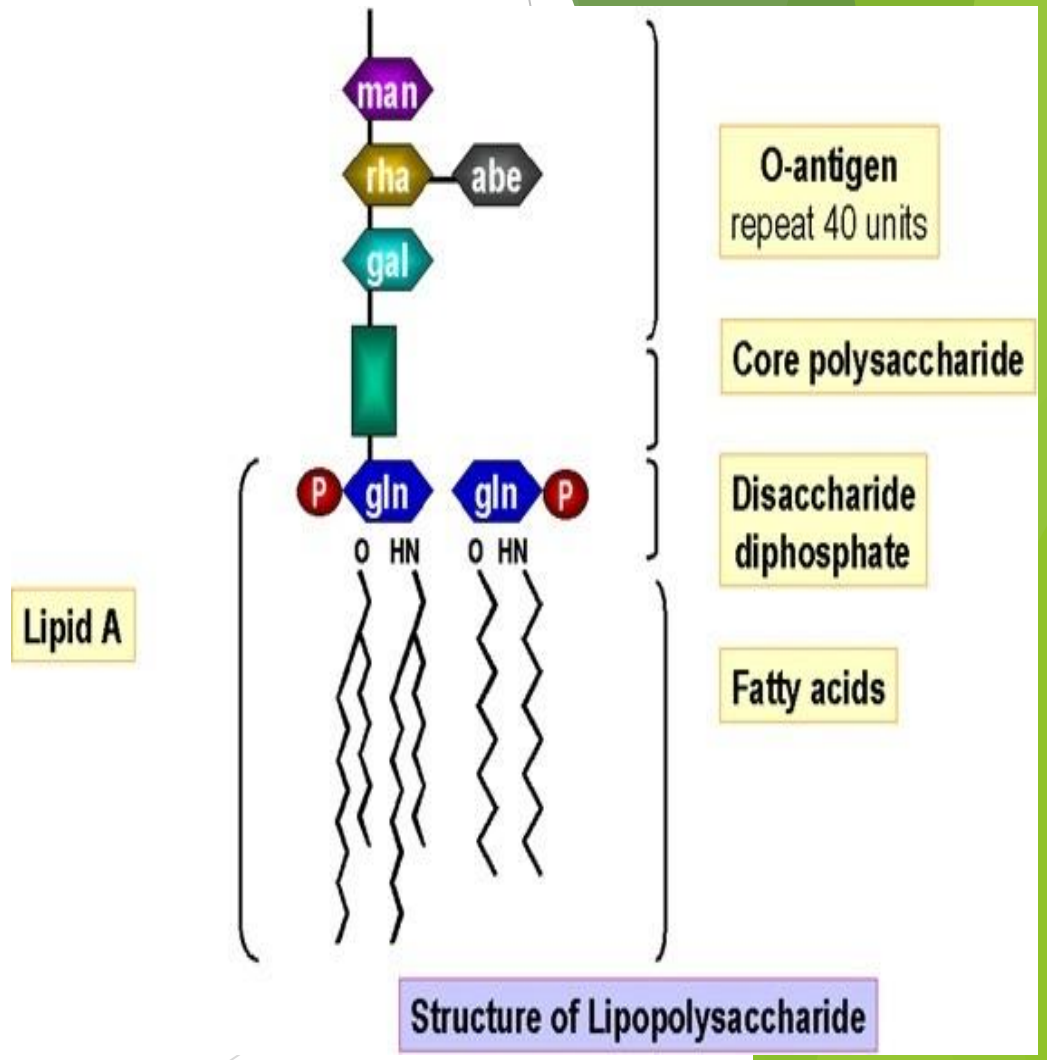


Cell wall of gram-negative bacteria (lipopolysaccharide-LPS)

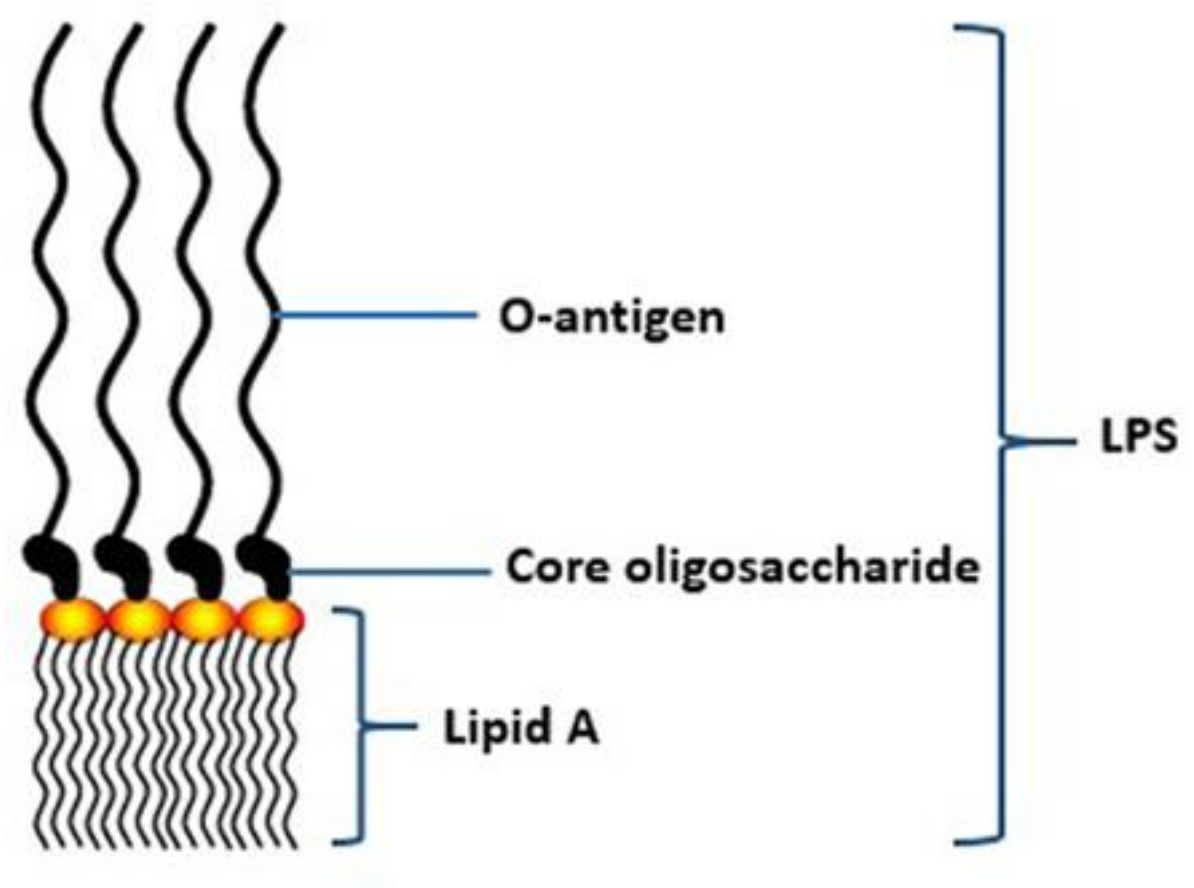
- The LPS of the outer membrane consists of three fragments:
- lipid A - a conservative structure, almost the same in gram-negative bacteria; LPS is anchored in the outer membrane by lipid A
- core, or core, core part (from lat. core - core), relatively conservative oligosaccharide;
- a highly variable O-specific polysaccharide chain formed by repeating identical oligosaccharide sequences. . O-specific chain determines the serogroup, bacterial serovar (O-antigen)
- Thus, the polysaccharide part determines the antigenicity, the lipid part determines the toxicity of LPS

Lipopolysaccharide (LPS)

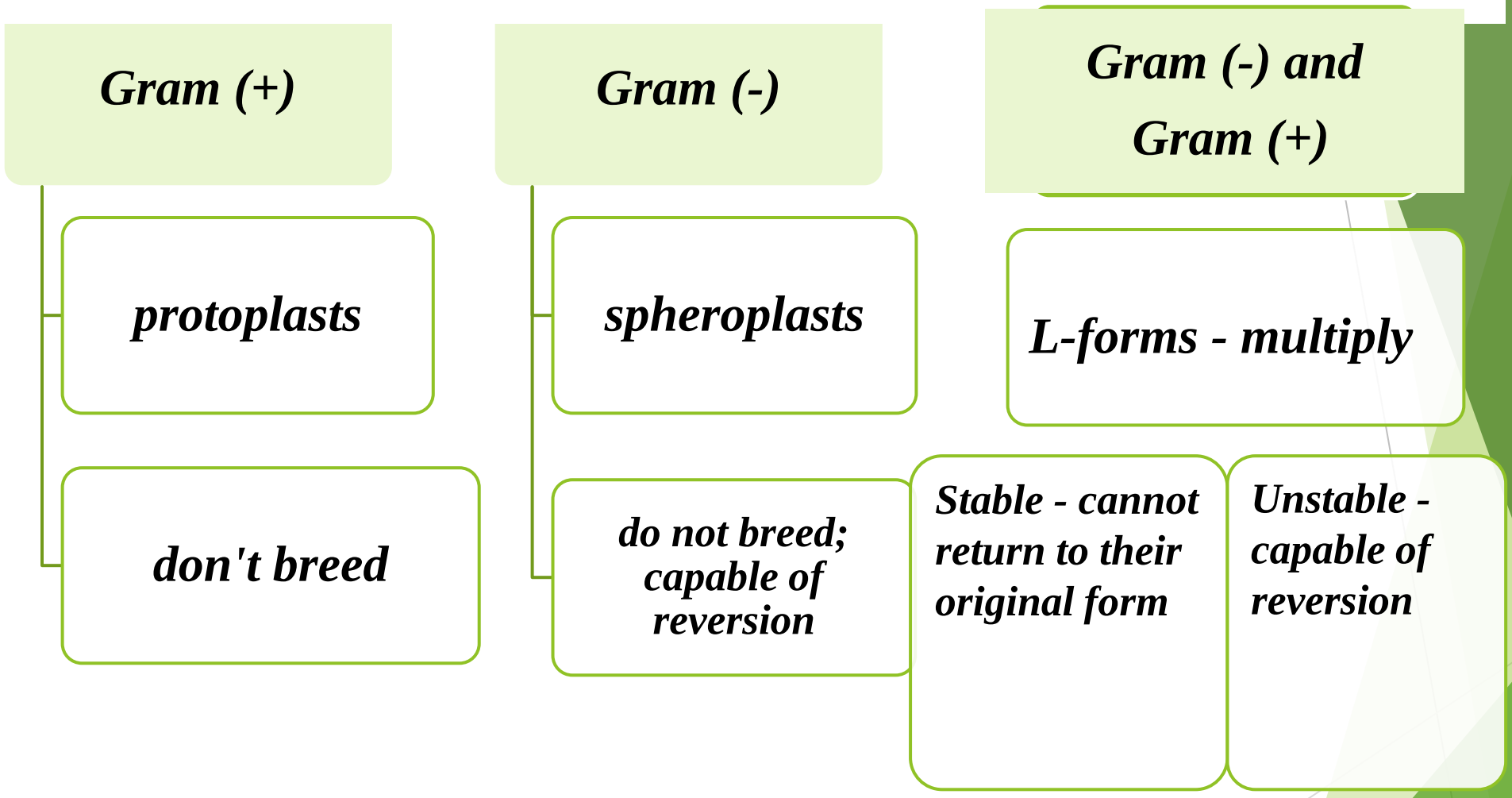
Lipid A-glycolipid
Long chain fatty acids + disaccharide glucosamine
Causes toxicity (endotoxin)
The core is ketodeoxyoctonic acid, heptose.
polysaccharide common to all Gram(-) bacteria
O-specific polysaccharide moiety
Repeating polysaccharide sequences (tri-, tetra-, penta-)
Determines serogroup



The structure of lipopolysaccharide



Loss of the cell wall



- *Latent infection*
- *Development of antibiotic resistance*

Methods of microbiological research

1.Microscopy method

2. *Cultural (bacteriological, virological, mycological, parasitological) method*

3. Biological method

4. Immunological method

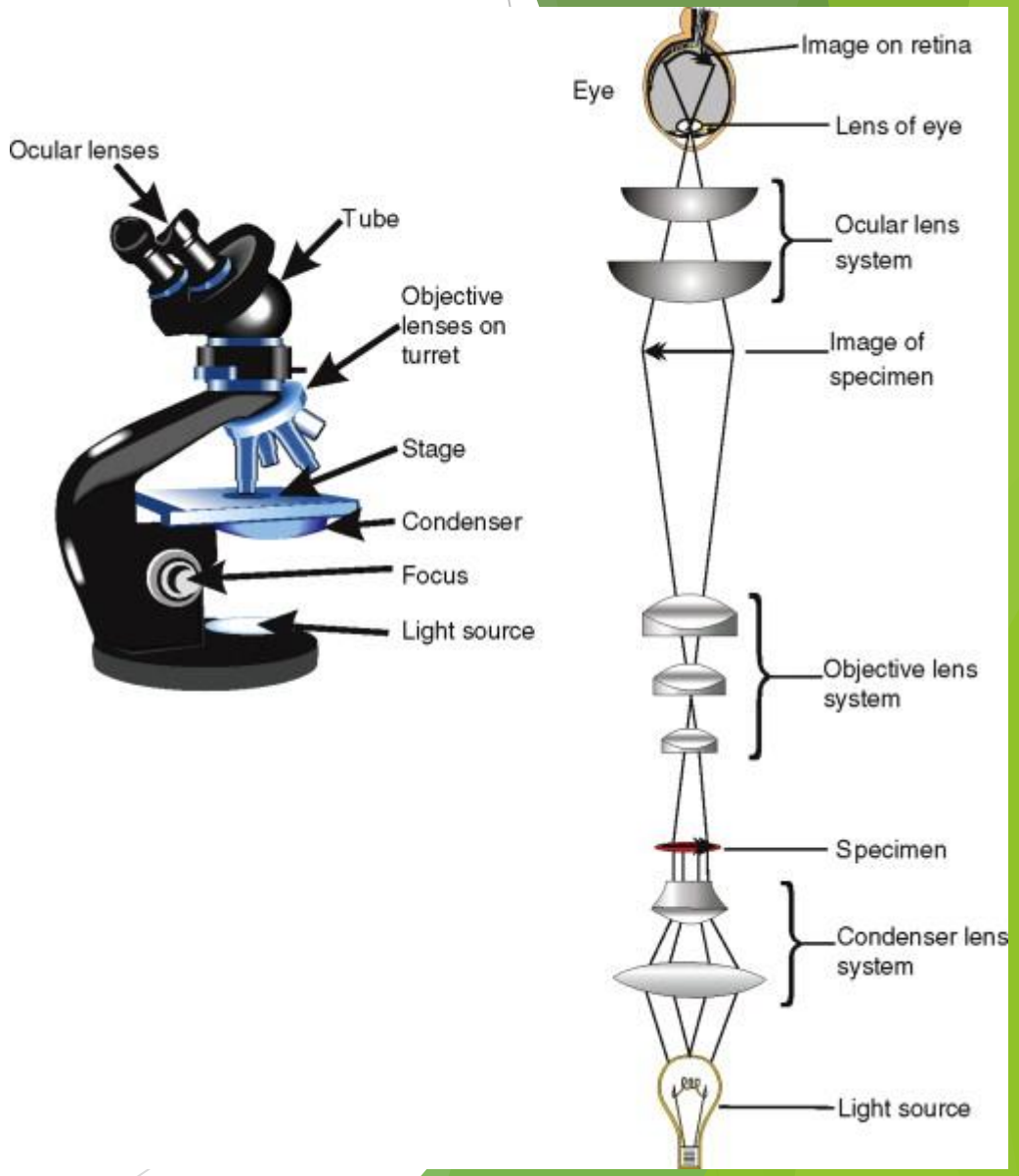
- serological

- skin allergy tests

5. Molecular genetic method

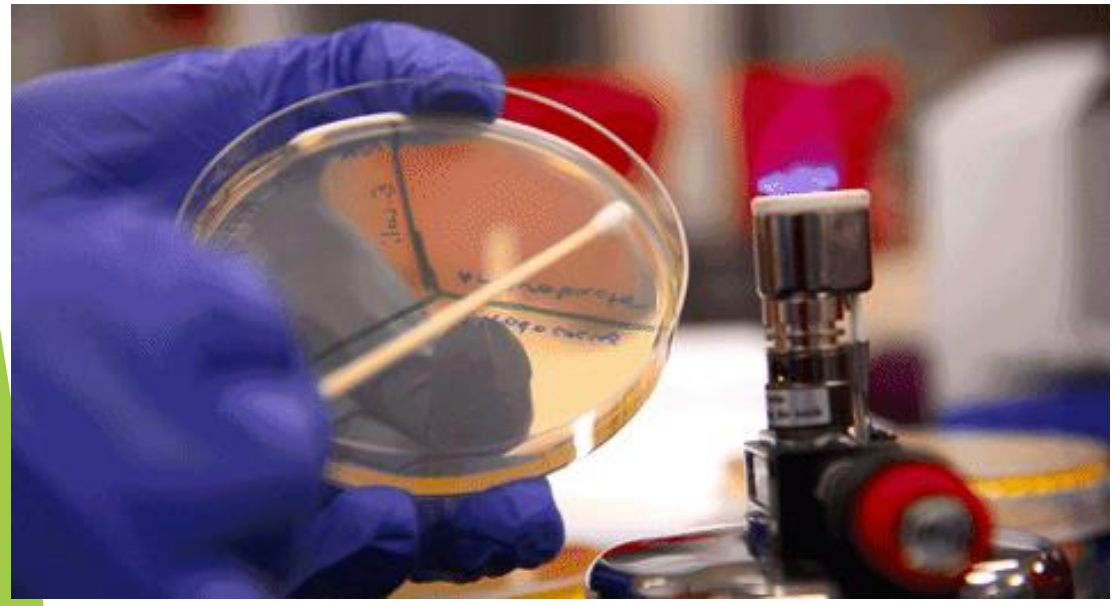
Microscopy method

- *Using a microscopic method, the presence of microorganisms and their morphology are determined in the test material.*
- *Additional elements - determine the presence of a capsule, spores, flagella, other elements (volutin grains).*
- *Since many microorganisms cannot be identified on the basis of morphology and tinctorial properties, the microscopic method is therefore considered an approximate diagnostic method.*



Cultural (bacteriological) method

- *When conducting a study by this method, pathological material is sown on appropriate nutrient media, incubated, a "pure culture" is obtained, and identification is carried out.*
- *Being the "gold standard" of microbiological diagnostics, the method allows you to correctly identify the pathogen.*



Biological and experimental method

- *Laboratory animals are infected with pathological material*
- *The biological method is used if it is impossible to obtain a pure culture by the bacteriological method.*
- *The pathogenicity, virulence and toxigenicity of the microbe is being studied.*
- *Experimental studies of new drugs are underway.*

Immunological method

***Serological method** - in the blood serum, the antigens of the pathogen, or antibodies against the pathogen, are determined, as well as using a known immune serum, the type and serovar of an unknown microbe are determined (**serological identification**).*

Skin allergy test

- *Due to the fact that the antigens of pathogens cause sensitization, allergic reactions are used to diagnose infectious diseases.*
- *tuberculosis - Mantoux test,*
- *brucellosis - Burne test,*
- *in tularemia - a reaction to tularin, etc.*

Molecular genetic method

- ***Polymerase chain reaction.*** *It is based on the principle of multiplication (amplification) and determination of the nucleic acid of the pathogen in pathological material or in pure culture.*
- *DNA and molecular hybridization of RNA. Based on the determination of genomic fragments characteristic of the pathogen.*
- *The main advantage of the molecular genetic method is its high sensitivity and specificity.*