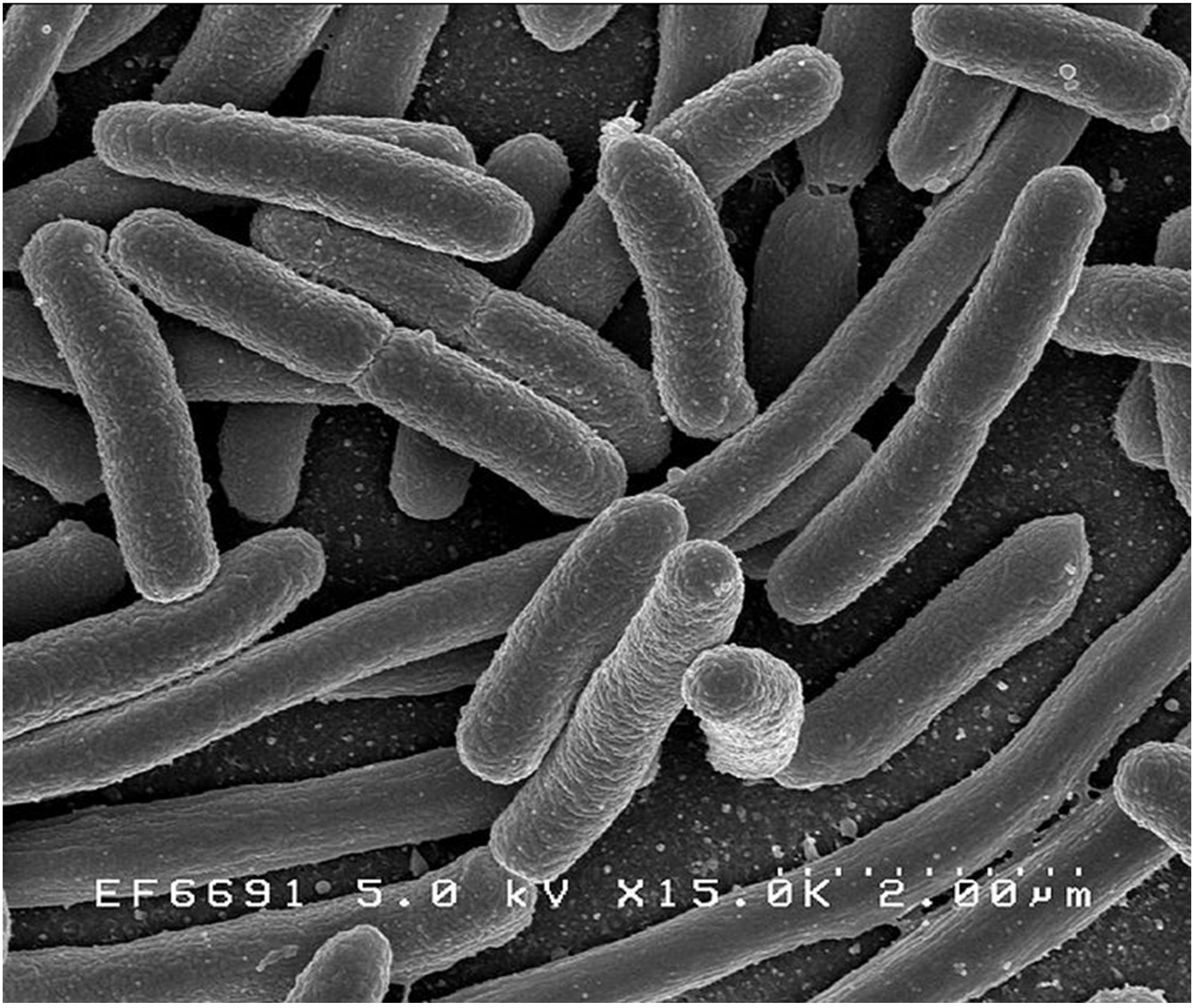
Teknisk Biologi/Mikrobiologi



Escherichia coli, förkortas ofta *E. coli*

kvalitetsledning/säkring

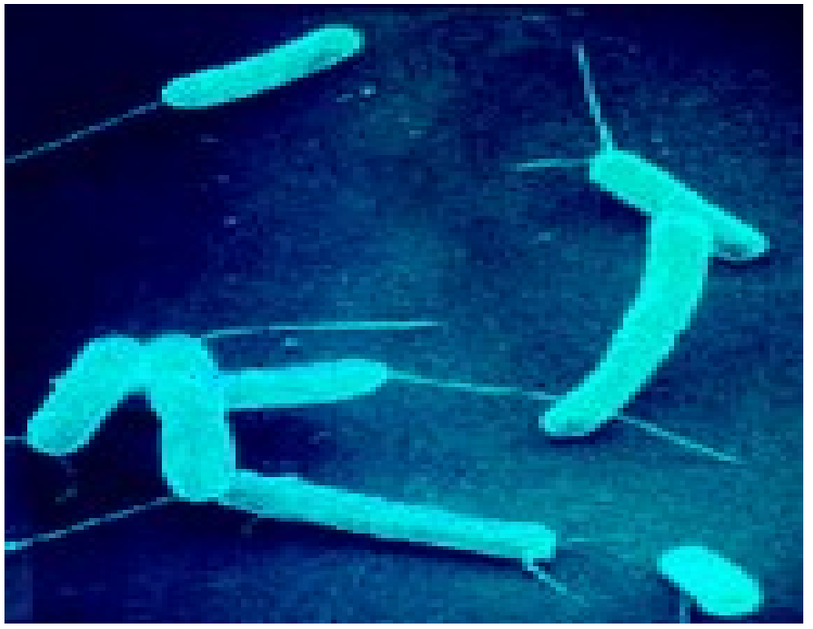
- **Quality Assurance (QA), Qualified Person (QP), GMP manager…**

- Produktkvalitet
- Produktsäkerhet
- Aseptik (sterilteknologier och avdödningskinetik kopplad till olika typer av mikroorganismer)
- Hygien (Rena Rum, resistensutveckling, desinfektion, antibiotika)

QA Engineer

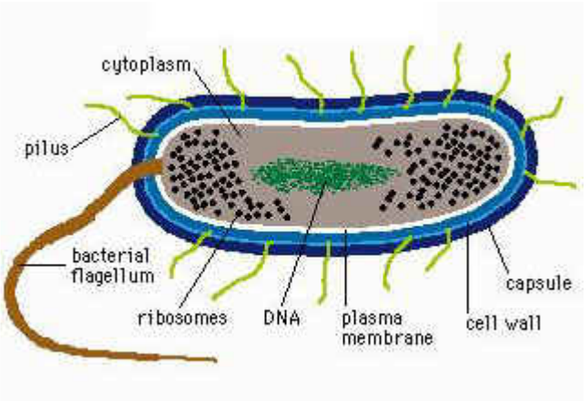
Micro-organisms

- Study of organisms too small to be clearly seen by the unaided eye (i.e., micro-organisms)
- Size < ~1 mm (microscopic organisms)
- Mainly single-celled organisms
- Lack organs and true tissue
- Some multi-celled organisms have primitive tissues
- Very heterogeneous group



Micro-organisms

Can be divided into:

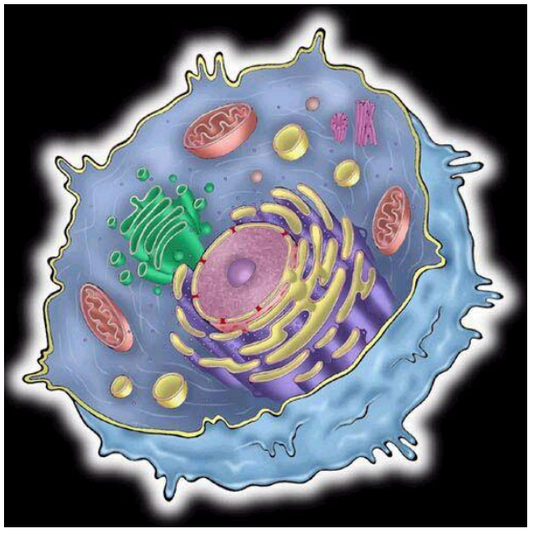


A **Procaryotes** (lack a true membrane-delimited nucleus)

A1 **Archaea or archaeobacteria**

A2 **Bacteria or eubacteria**

B **Eucaryotes** (have a membrane-enclosed nucleus, are more complex morphologically and are usually larger than procaryotic cells)

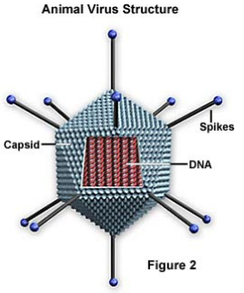


Eukaryotes have organelles!

B1 **Algae**

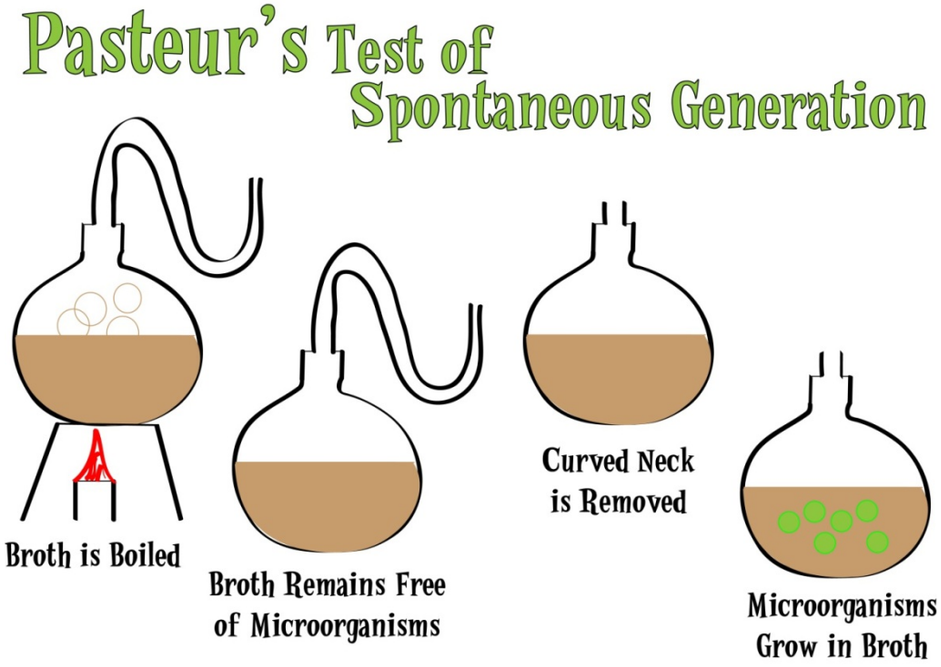
B2 **Fungi**

B3 **Protozoa**



Viruses (obligate parasites)

”Uralstring”

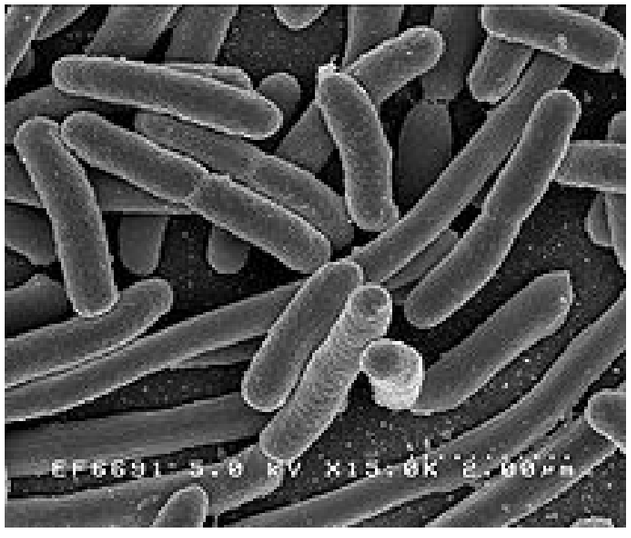


Louis Pasteur,1822-1895

Petriskål, Julius Richard Petri (1852–1921)
Robert Koch.

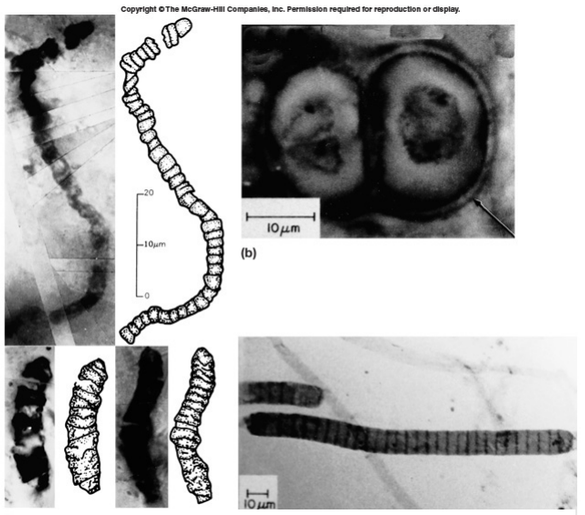
The Nobel Prize Medicine 1905
Robert Koch

Carl von Linné född 13 maj 1707 i Råshult, Stenbrohults socken, Småland, död 10 januari 1778 i Uppsala

Domän		Bakterier
Stam		Proteobacteria
Klass		Gamma Proteobacteria
Ordning		Enterobacteriales
Familj		Enterobacteriaceae
Släkte		*Escherichia*
Art		*coli*

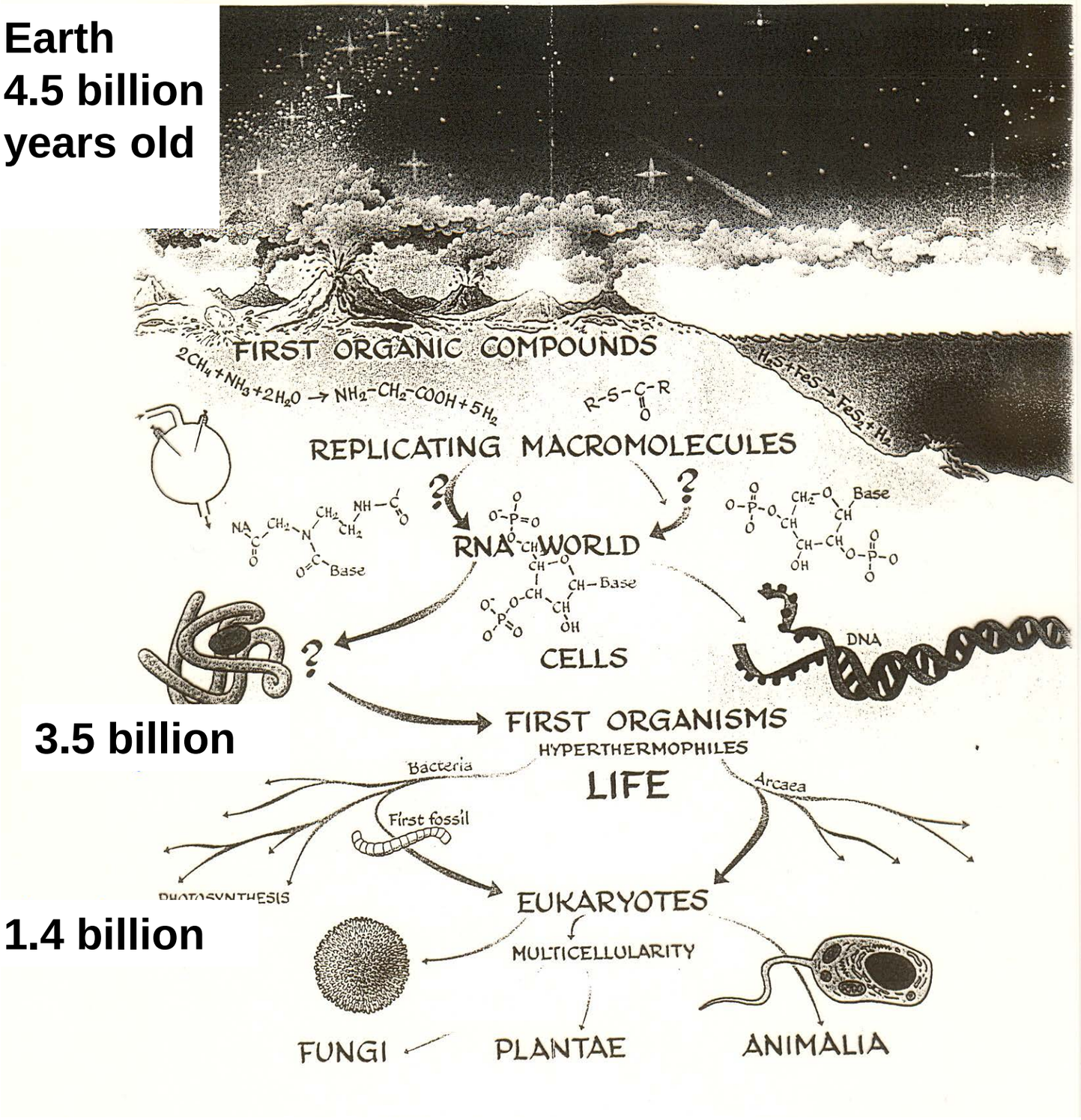
Examples of the importance of micro-organisms

- The oldest and most common form of life
- Mineralization (biogeochemical cycles, bioremediation, composting, water purification, etc.)
- Useful to macro-organisms (nitrogen fixation, nutrition, symbiosis, normal flora, photosynthesis etc.)
- Biotechnical processes (food, pharmaceuticals, chemical products, etc.)
- Pathogenic micro-organisms (infectious diseases, product spoilage, etc.)
- Model systems in biology (genetics, physiology, metabolism, etc.)

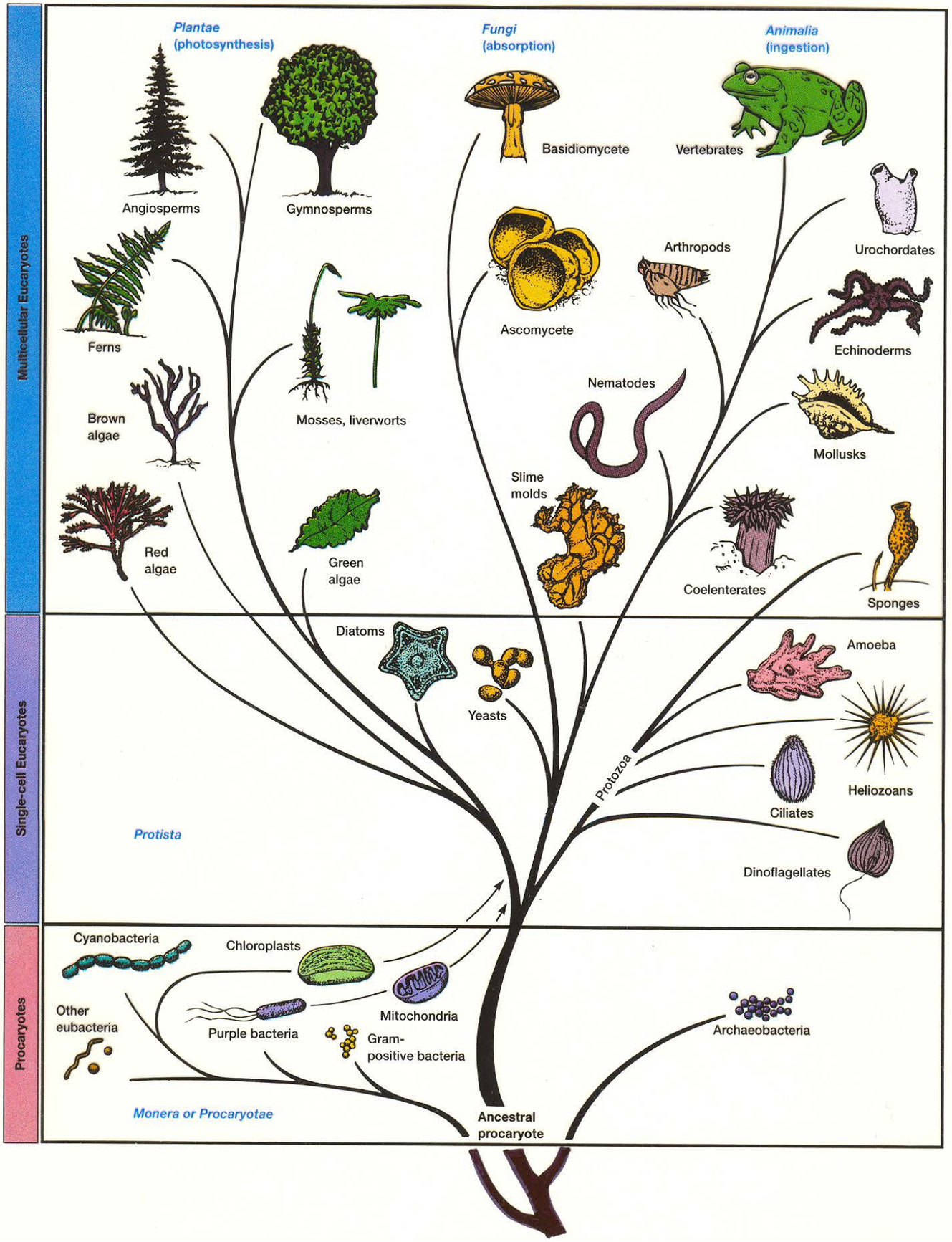


Fossilized bacteria

The oldest and most common form of life



The oldest and most common form of life



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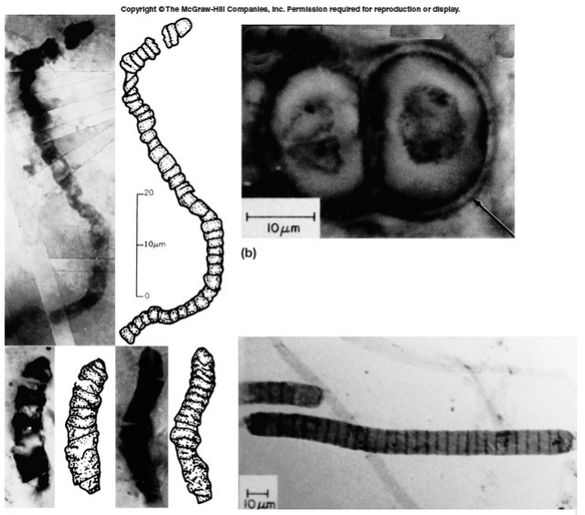
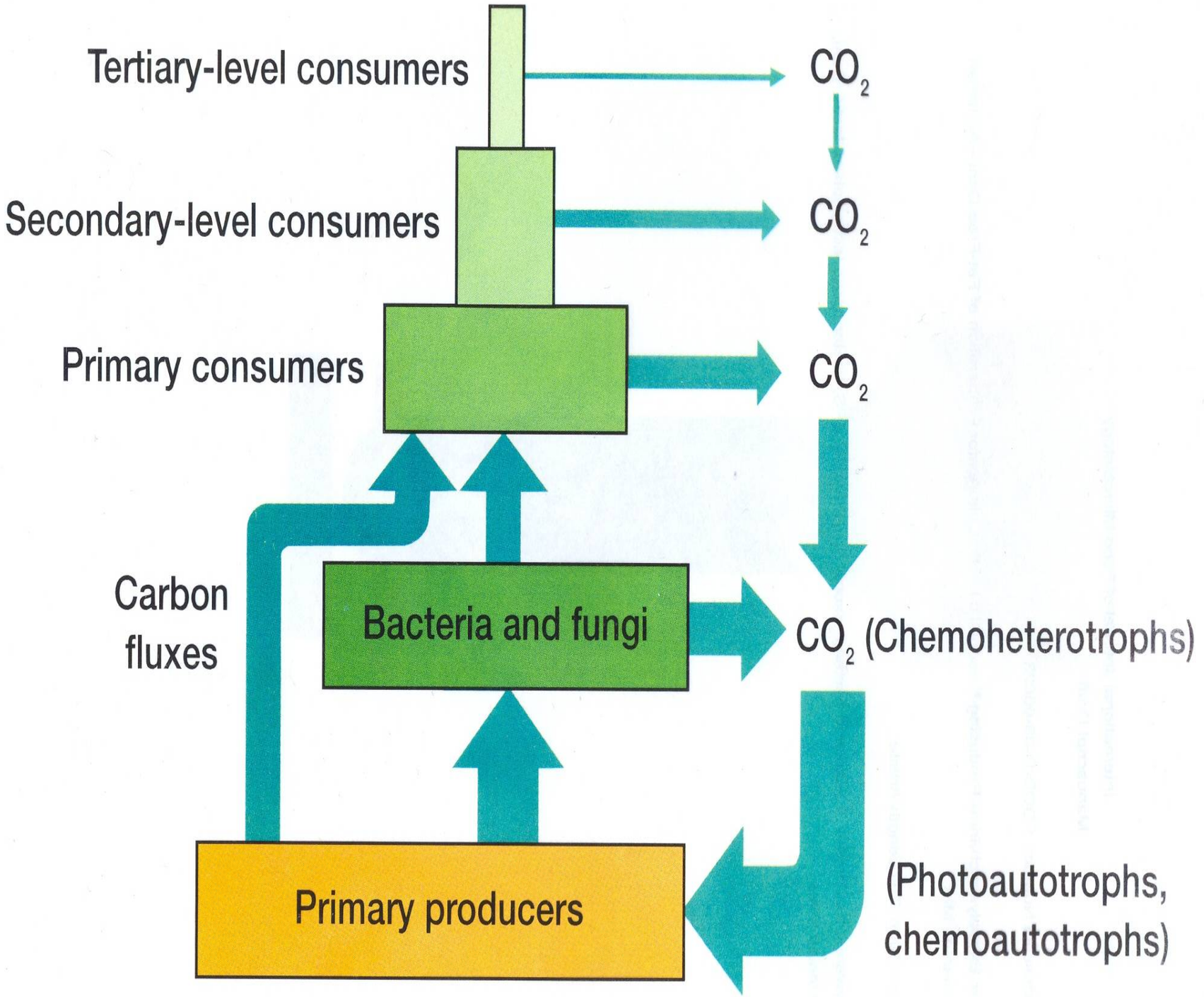


Fossilized bacteria

Mineralization (biogeochemical cycle, bioremediation, composting, water purification, etc.)



Role of microorganisms in flow of organic carbon and CO_2

Examples of the importance of micro-organisms

- The oldest and most common form of life
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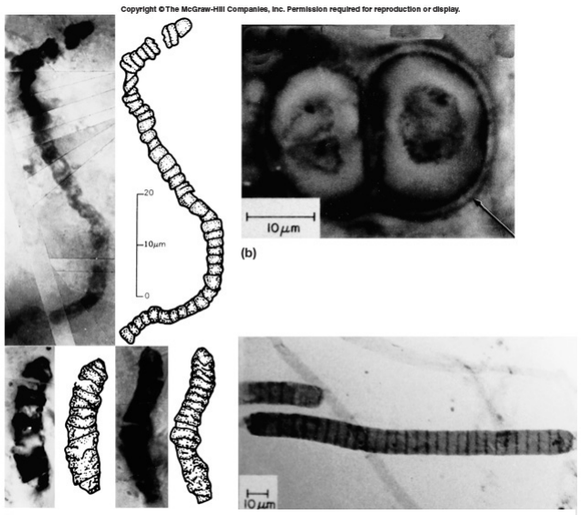

Fossilized bacteria

Useful to macro-organisms

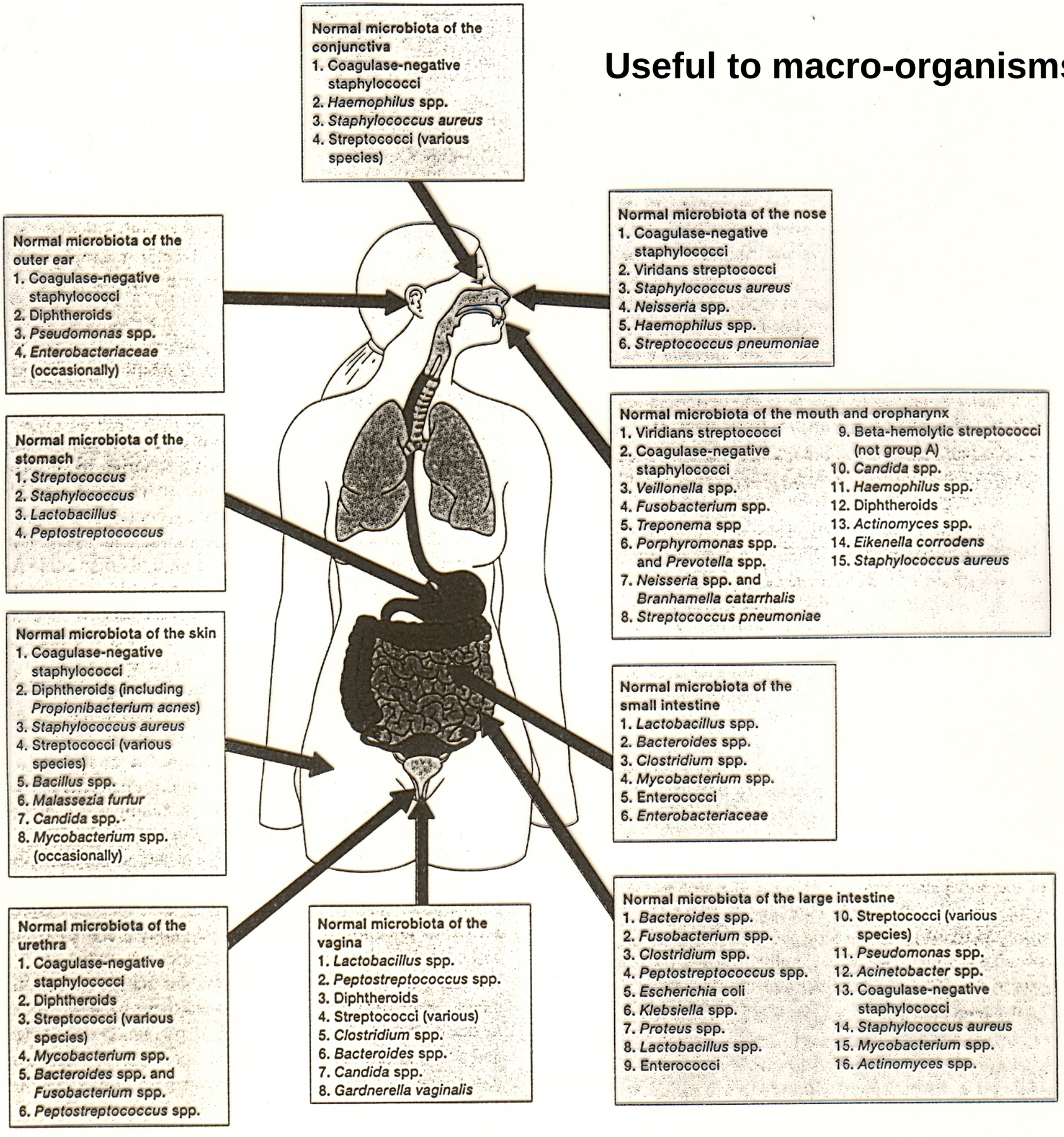

Figure 28.8 Normal Microbiota of a Human. A compilation of microorganisms which constitute normal microbiota encountered in various body sites.

Useful to macro-organisms

Examples of the importance of micro-organisms

- The oldest and most common form of life
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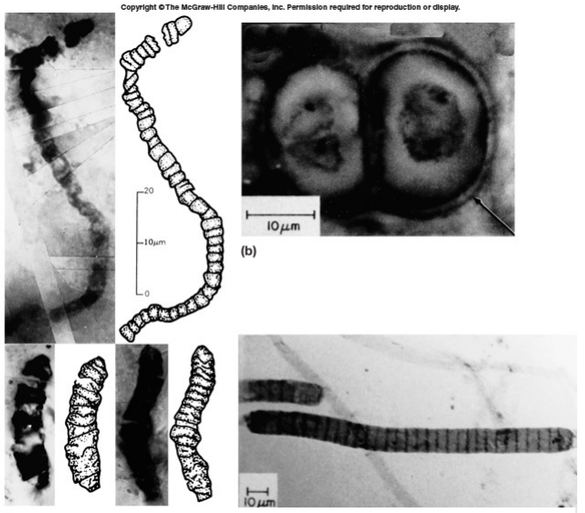

Fossilized bacteria

Novel biocatalysts for oil-industrial processes using metagenomics

Wax (long-chain alkanes) deposition in oil piplines

Starter cultures (fermented foods) “The Good Microbes”

- Lactic acid-producing bacteria (LAB) play a key role in the production of fermented foods and beverages, accounting for tens of billions of dollars in sales annually. Products constituting a fine repast, such as wine, salami, cheese, sourdough bread, pickles, yogurt, cocoa, and coffee are all enhanced by LAB.

- Probiotics and Health

Biotechnical processes

Table 13.1 Some of the compounds produced by industrial scale culture of microorganisms.

Compound	Microorganism
Antibiotics	
Penicillins	*Penicillium* spp.
Cephalosporins	*Cephalosporium* spp.
Gramicidins, polymixins	*Bacillus* spp.
Chloramphenicol, streptomycin	*Streptomyces* spp.
Enzymes	
Invertase	*Saccharomyces cerevisiae*
Proteases, amylases	*Bacillus* spp., *Aspergillus* spp.
Alcohol	*S. cerevisiae*, *Saccharomyces carlsbergensis*
Glycerol	*S. cerevisiae*
Vinegar	*S. cerevisiae*, acetic acid bacteria
Dextran	*Leuconostoc* spp.
Butyric acid	Butyric acid bacteria
Acetone, butanol	*Clostridium* spp.
Citric acid	*Aspergillus niger*

Examples of the importance of micro-organisms

- The oldest and most common form of life
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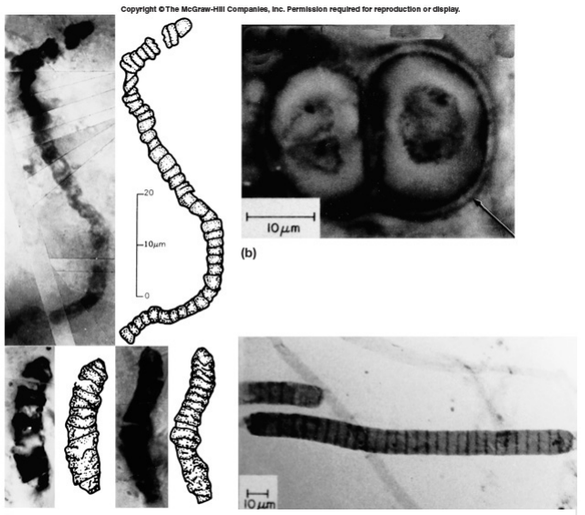


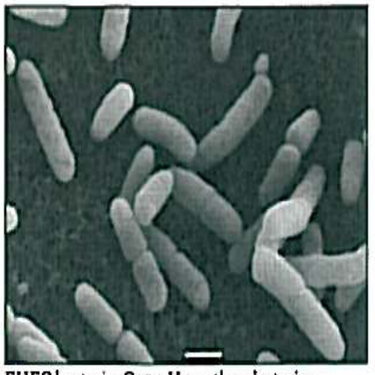
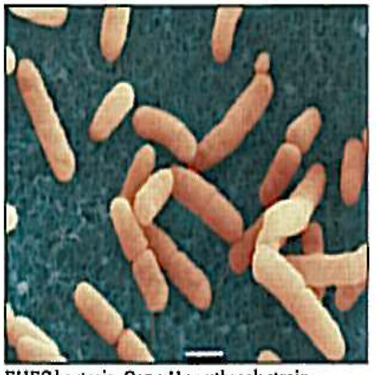
Fossilized bacteria

EHEC outbreak in Germany 2011

- On June 8, it was reckoned that the Enterohaemorrhagic *Escherichia coli* (EHEC) O104:H4 outbreak cost $2,840,000,000 in human losses (such as sick leave), regardless of material losses (such as dumped cucumbers).

Organic farming of bean sprouts identified as a source of the outbreak

Electron microscopy: EHEC bacteria, O104:H4 outbreak strain

Black and White	Color
	
EHEC bacteria, O104:H4 outbreak strain. Scanning electron microscopy. Bar: 1 µm. Source: Holland, Laue (Robert Koch Institute)	EHEC bacteria, O104:H4 outbreak strain. Scanning electron microscopy. Bar: 1 µm. Source: Holland, Laue (Robert Koch Institute)

During the outbreak period 4,321 cases were reported in Germany and in total 50 patients died. According to the European Centre for Disease Prevention and Control 76 EHEC cases were reported across other countries of the European Union.

http://en.wikipedia.org/wiki/2011_E._coli_O104:H4_outbreak#Economics

Pathogenic micro-organisms

fakta/bakterier

■ Mängder av ämnen marknadsförs som "anti-bakteriella" eller "bakteriedödare" – oftast i rengöringsmedel.

Tills för några år sedan var de vanligaste natriumhypoklorit, väteperoxid, etanol och salicylsyra. Men de nya medlen, till exempel triklosan, fungerar på ett helt annat sätt. Triklosan är en klorerad difenyleter som inte slår ut bakterien helt, utan förstör ett enzym som är viktigt för bakteriens uppbyggnad – i princip på samma sätt som antibiotika fungerar.

Läkemedelsverket har godkänt triklosan i tandkrämer, tvålar och kosmetika. För till exempel rengöringsmedel och kläder behövs inget tillstånd alls.

Examples of the importance of micro-organisms

- The oldest and most common form of life
- Mineralization (biogeochemical cycles, bioremediation, composting, water purification, etc.)
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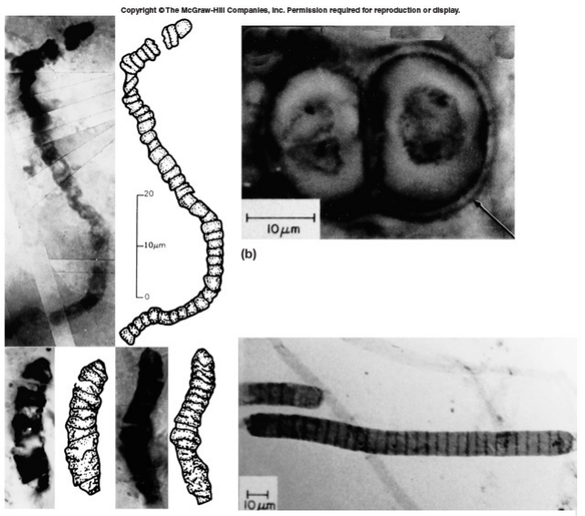
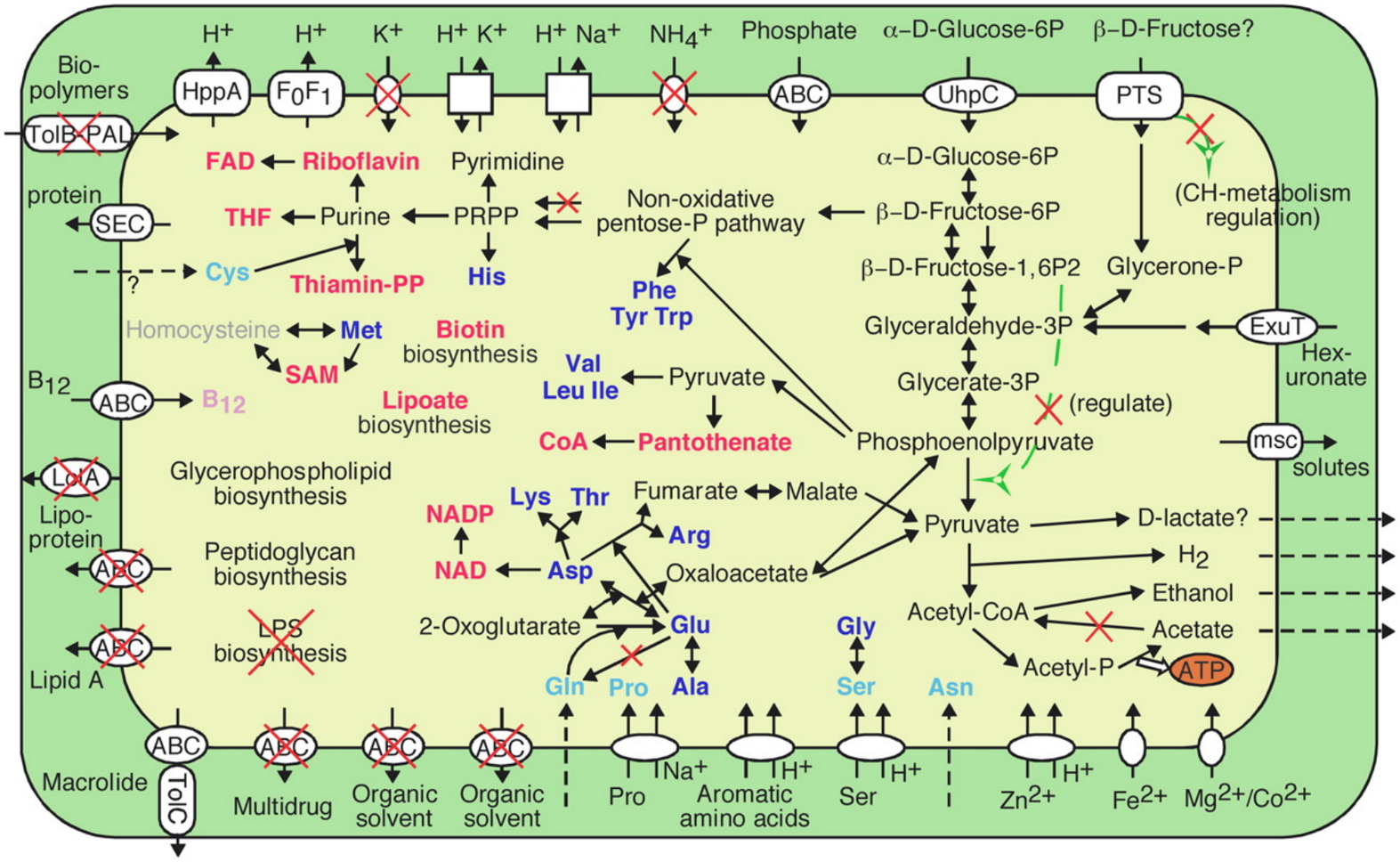


Fossilized bacteria

Model systems in biology (genetics, physiology, metabolism, biochemistry..)



Applications: Biosensors, Biogas, Bioremediation Processes, Industrial Fermenters, Biopolymers and Bioplastics, Pharmaceutical Biologics, Enzymes for Pulp & Paper, Antibiotic Production, Novel Biocatalysts for Oil-Industrial Processes, Bio-Diesel, Biorefinery etc.

Main morphological groups – Bacteria

- size, appearance and cell arrangement
- two basic forms: cocci and bacilli

Cocci: spherical cells, ~1-2 µm in diam. Cell arrangement upon cell division:

- Diplococci
- Streptococci
- Staphylococci
- Tetrads
- Sarcina

Exceptions

- Flat, quadratic bacteria: 0.25 x 2 x 2-4 µm
- Pleomorphic: no particular shape, lack cell wall

Bacilli or rods: Cylindrical shape, diam 1 µm, length 2-5 µm

- Coccobacilli: very short bacilli, reminiscent of cocci

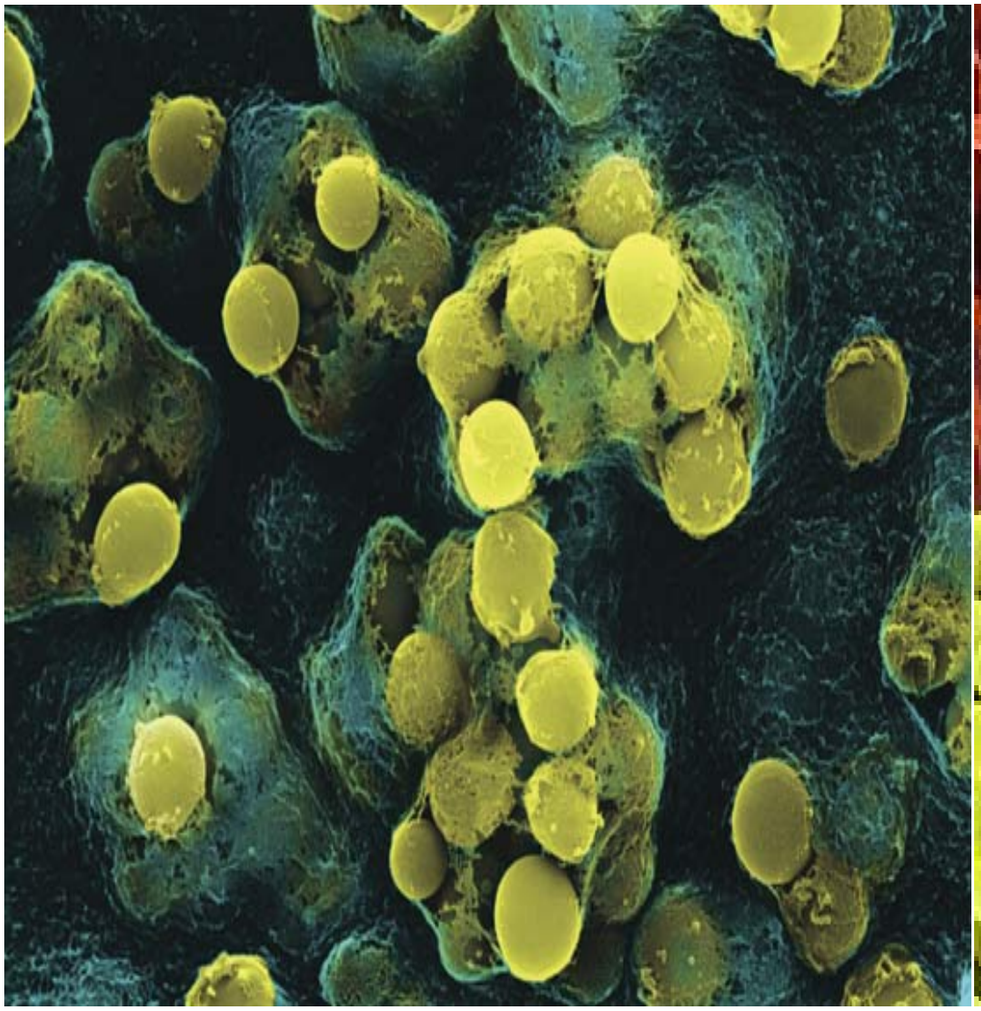
- Vibrions: a twisted bacillus, not a complete spiral, 2-3 µm long

- Spirilla bacilli: long, rigid, spiral bacilli with flagellae, 6-15 µm long

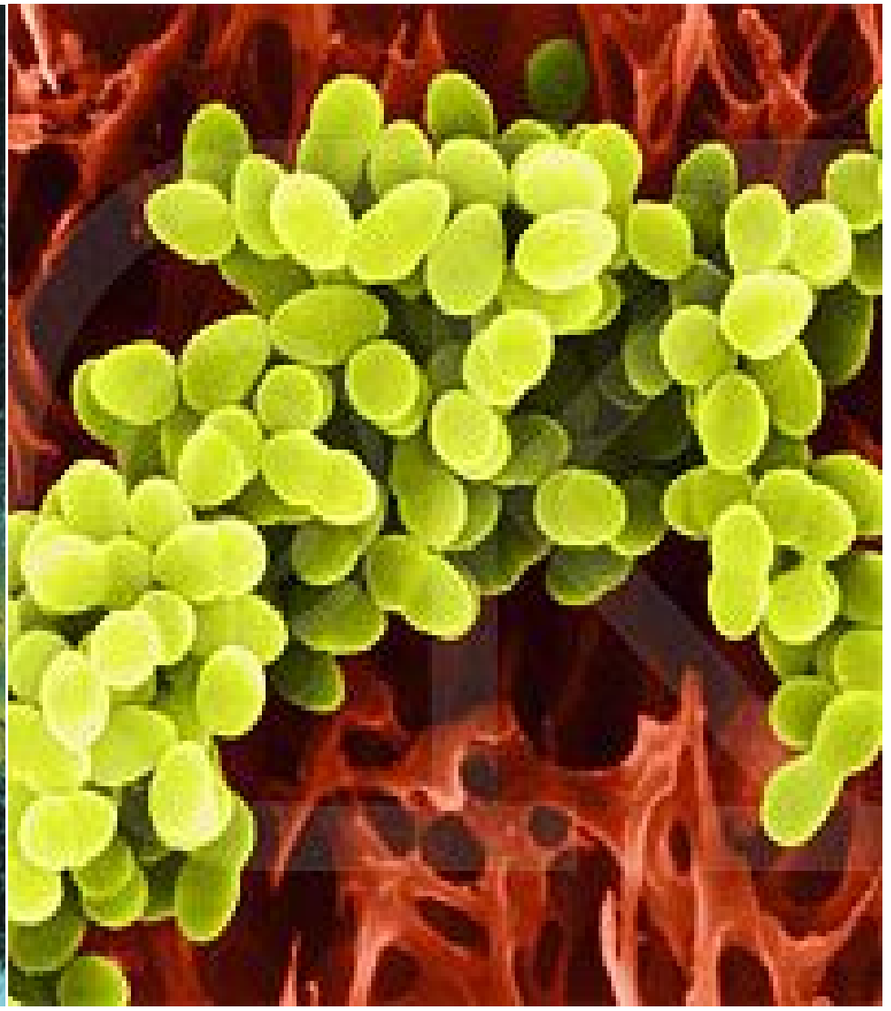
- Spirochet bacilli: long, flexible, spiral bacilli with an axial filament

- Hyphae that form a micelle

Cocci: spherical cells, ~1-2 μm in diam.

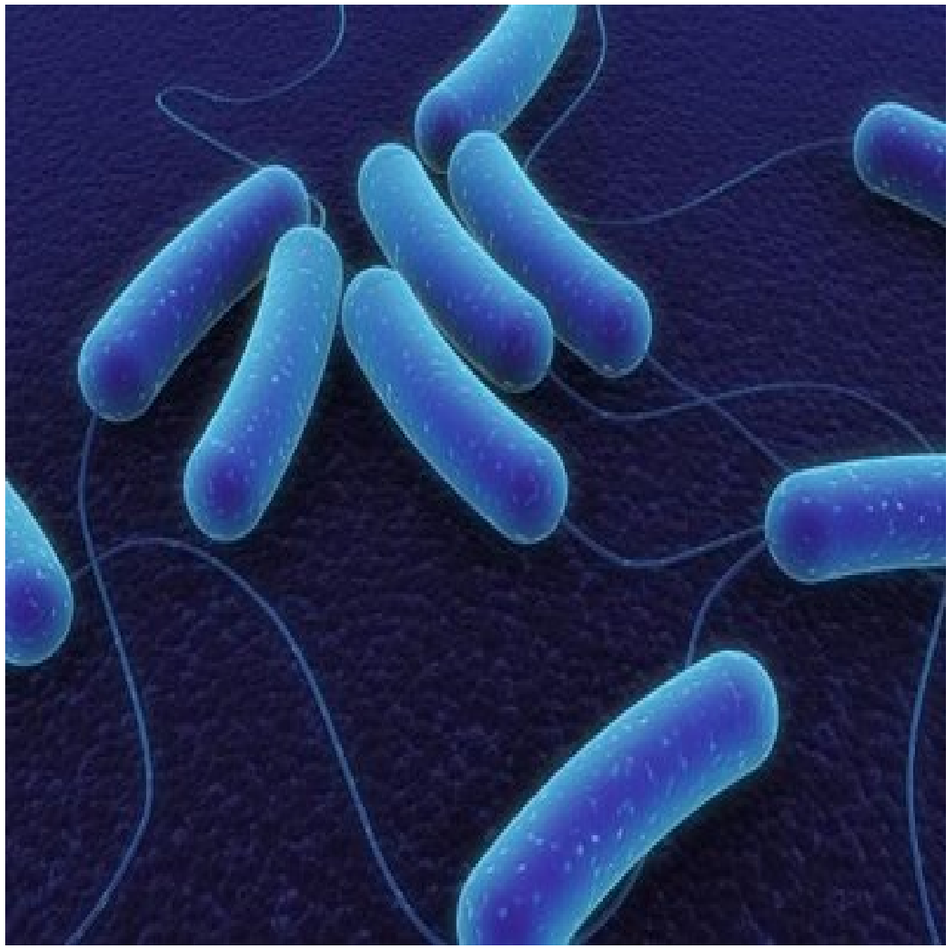


Streptococcus pyogenes



Staphylococcus aureus

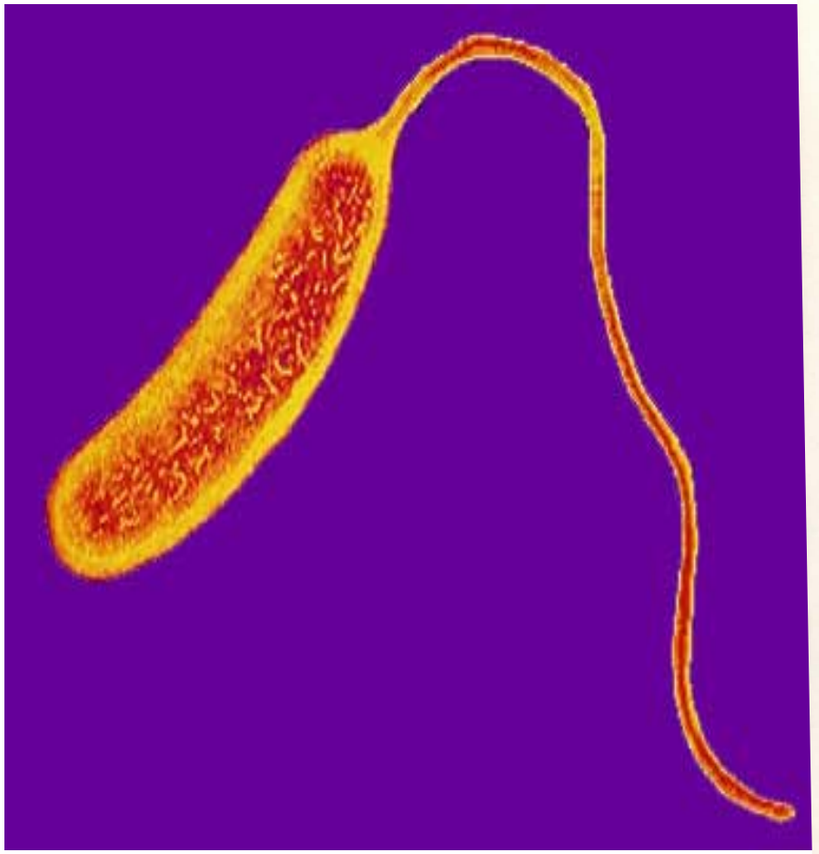
Bacilli or rods: Cylindrical shape, diam 1 μm, length 2-5 μm



Bacillus subtilis

Escherichia coli

Vibrios: a twisted bacillus



Vibrio cholerae

Figure 37.21 Cholera. *Vibrio cholerae* adhering to intestinal epithelium; scanning electron micrograph (× 12,000). Notice that the bacteria is slightly curved with a single polar flagellum.

Gram-negative, facultatively anaerobic, catalase and oxidase-positive, polar flagella

Temp; opt. ~37°C, range 5-43°C

marine environment, waterborne infection
- washed fruits and vegetables, plus seafoods can serve as a vehicle.

Spirilla
spiral bacilli with flagellae, 6-15 µm long

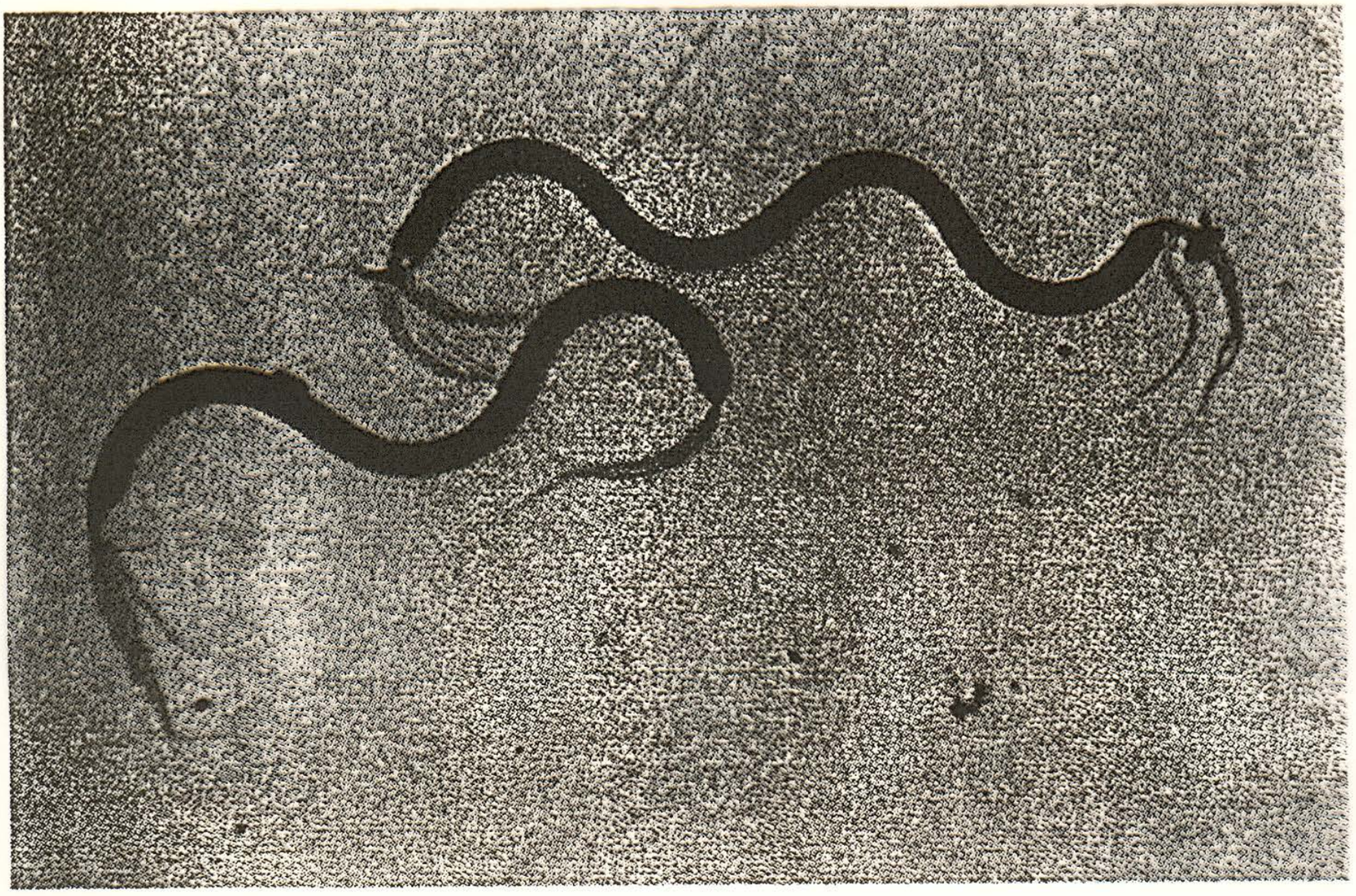
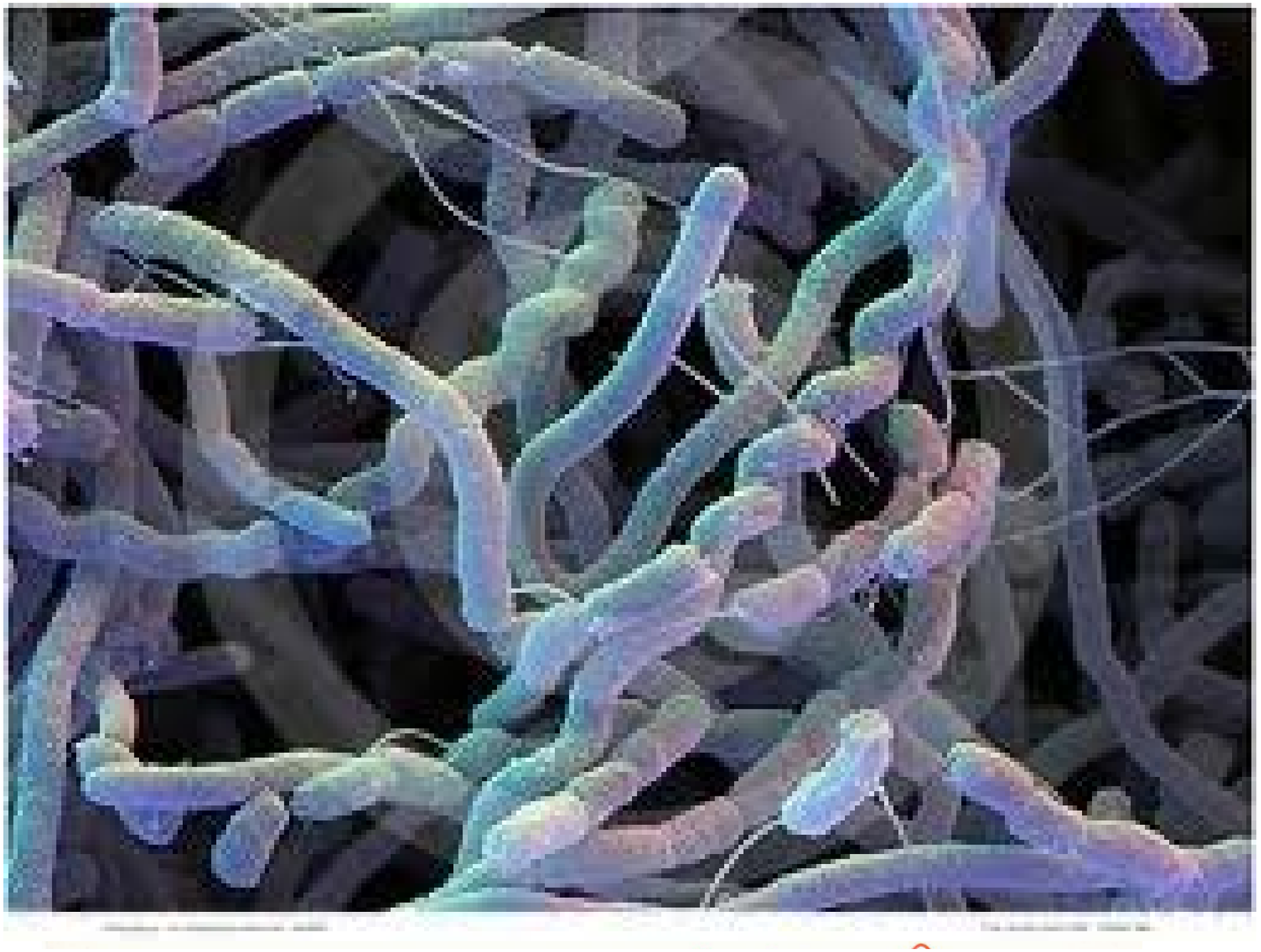


Figure 2.18 **Example of Flagella Staining.** *Spirillum volutans* with bipolar tufts of flagella (×400). (*See also figure 3.33.*)

Spirochet
spiral bacilli with an axial filament

Micelle-forming bacteria



The genus *Streptomyces* includes about 500 species. Constitutes 1-20% of the cultivable earth flora. Forms *geosmin*, or earth smell, and a large number of antibiotics.

Bacterial staining methods

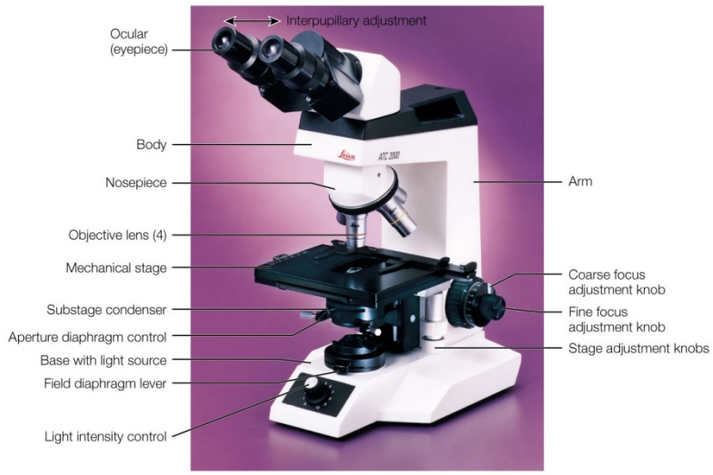
- **Aim:**
  To increase contrast in microscopy slides for the systematisation of bacteria

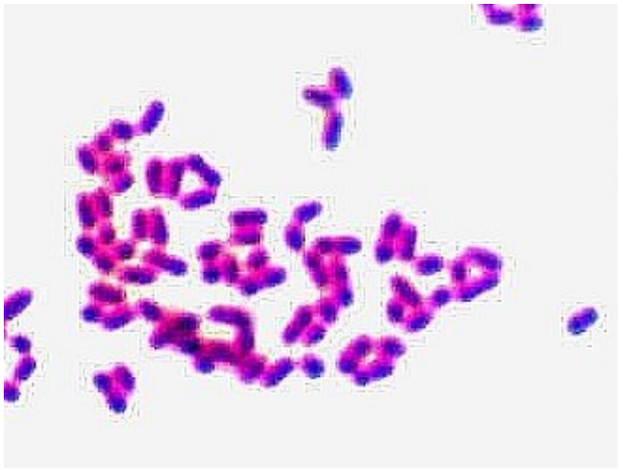
- **Principle:**
  - Basic (cationic) dyes, e.g. violet, methylene blue and malachite green

  - Acidic (anionic) dyes, e.g. eosin and Congo red

  - Fat-soluble dyes such as Sudan black

- **Fixing:**
  Heat or chemicals (formaldehyde, acetic acid or alcohol) are used to fix bacteria on slides in order to increase the penetration of the dye.





Bacterial staining methods

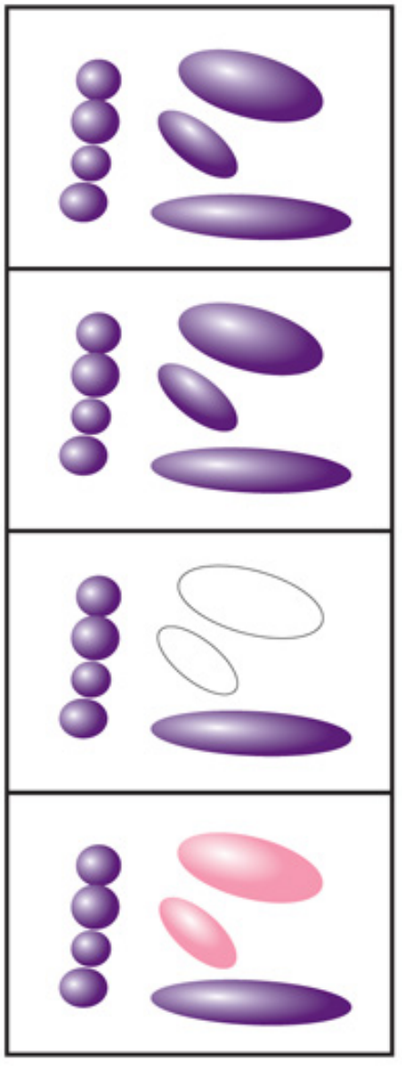
Gram staining:

(Christian Gram, 1884)

- Heat fixing
- Crystal violet
- Lugol's solution (KI + I_2)
- EtOH or acetone
- Safranin (counterstain)

Gram-positive bacteria = blue

Gram-negative bacteria = red



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

(a)