Medical Microbiology: essence, history, taxonomy

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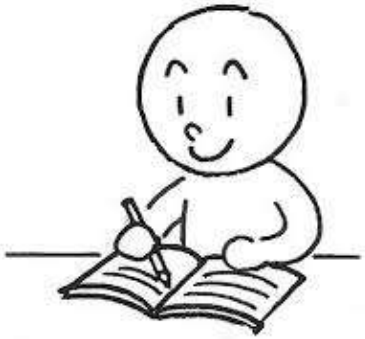
Lecture-thesis in microbiology

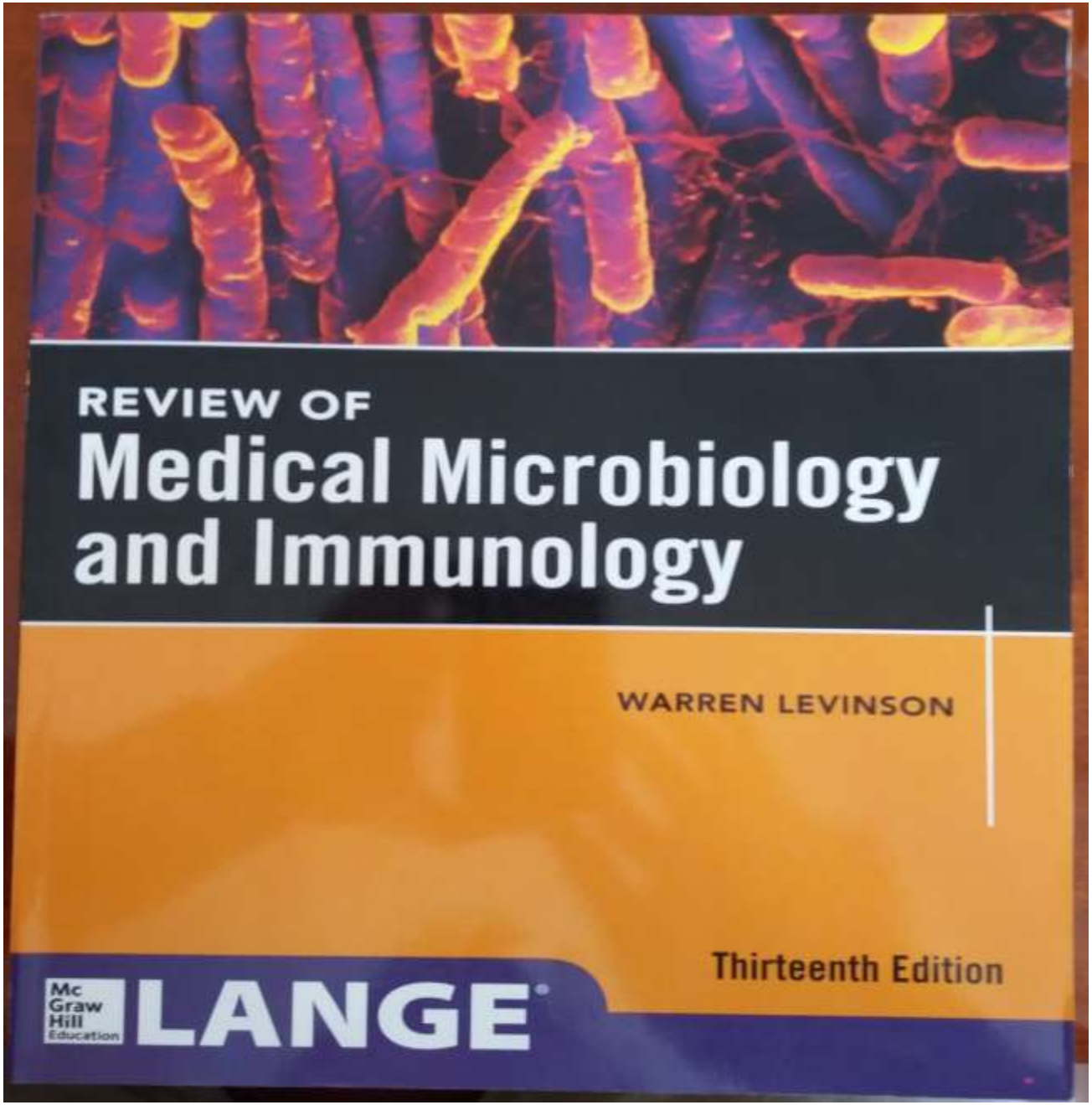
Course introduction



- **Lecture courses** – mandatory
- **Lab classes**:
  - mandatory
  - write and draw theses
  - observe and perform tests
- **Textbooks:**

  Obligatory:
  - Review of Medical Microbiology and Immunology, Warren Levinson, 13 e. McGraw Hill Education, 2016, ISBN 978-0-07-181811-7, 789 pp. or
  - Medical Microbiology. Patrick R. Murray, Ken Rosenthal, Michael Pfaller. 8 e. Elsevier, 2016, ISBN 978-0-323-29956- 5, 848 pp.

  Recommended:
  - Medical Microbiology. S. Baron, 4 edt, 2000, ISBN-10: 0-9631172-1-1, http://www.ncbi.nlm.nih.gov/books/NBK7627/
  - Todar's online textbook of bacteriology. K. Todar, 2009, http://www.textbookofbacteriology.net/kt_toc.html


REVIEW OF
Medical Microbiology and Immunology
WARREN LEVINSON
Thirteenth Edition
Mc Graw Hill Education
LANGE

What does Microbiology study?

- Microbiology studies microorganisms (too small to be seen with the naked eye) – their morphology, physiology, pathogenesis, sensitivity to antimicrobials
- A microscope is needed to view them.

- Microorganisms include:

bacteria
viruses
fungi
protozoa
helminths (worms)
algae

Sizes :

mm

µm

$\frac{1}{1.000}$ = 1 Mikrometer

nm

$\frac{1}{1.000}$ => Nanometer


length in micrometers (µm)
0
2
4
6
8
10
12
Mycoplasma genitalium 0.4 µm
Haemophilus influenzae 1.2 µm
Staphylococcus aureus 0.9 µm
Escherichia coli 1.5 µm
Bacillus megaterium 4 µm
red blood cell 8 µm

Why microorganisms are so important?

Microbes help us by:

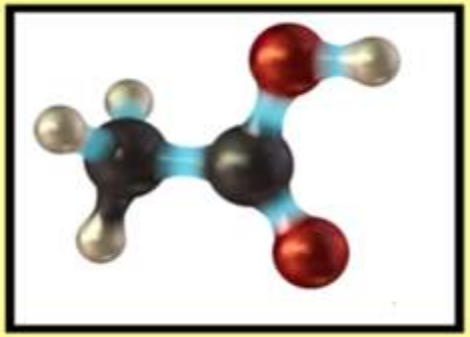
- nutrient production and energy flow
  (wine, vinegar, beer, bread, yogurt, cheese)
- decomposition of organic waste
- biotechnology and genetic engineering
  (vaccines, AB, IFNs, insulin)

Microbes harm us by:
causing infectious disease

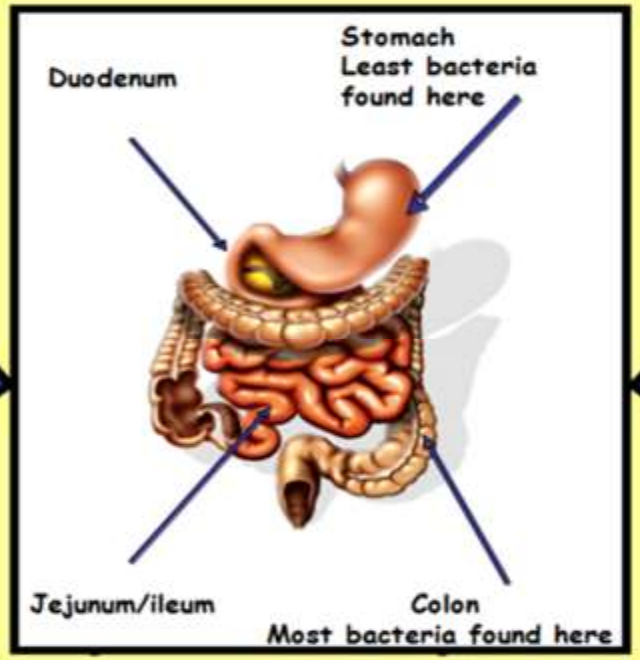
The dual role of human microbiome

- **Useful**
- **Harmful?**

The dual role of microbiome - useful bacteria



Digestion of food and the production of beneficial metabolites e.g. acetic acid

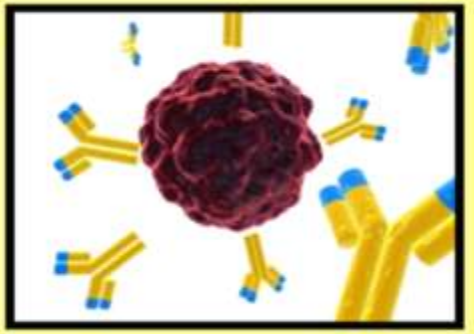


Vitamin synthesis (K and B12 vitamins)

Development of the intestinal structure

Metabolism of toxic compounds

Immune system development

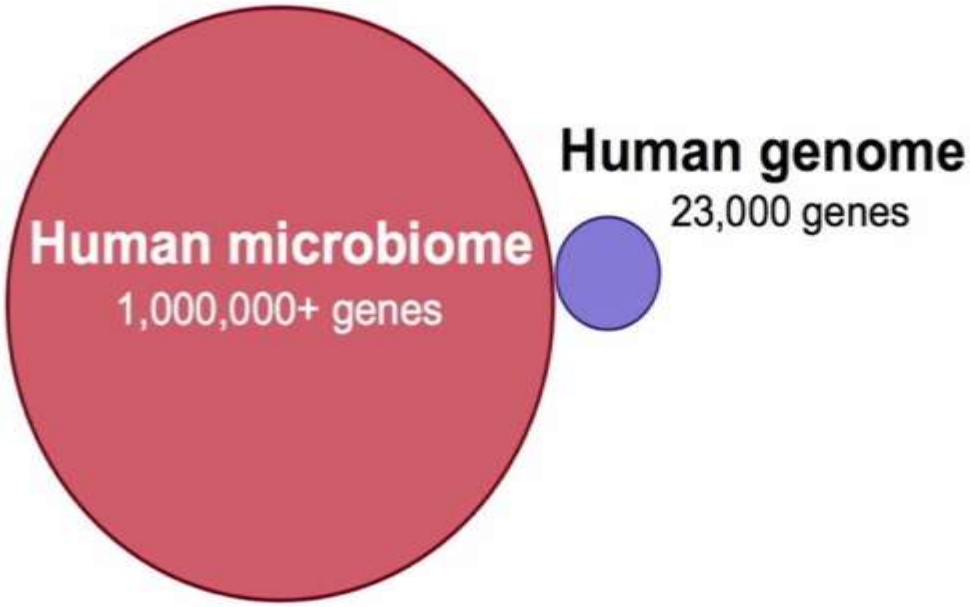


Our cells – human and microbial!

We are composed of several species:	• Eucaryotic • Bacterial • Archaea
As adults our microbial census exceeds the total number of our own human cells	• By about 10 fold
The largest collection of microbes resides within the intestine	• With 10^{13-14} cells!!!! • Several hundreds of species • «The GUT MICROBIOTA»

100 % Human ?

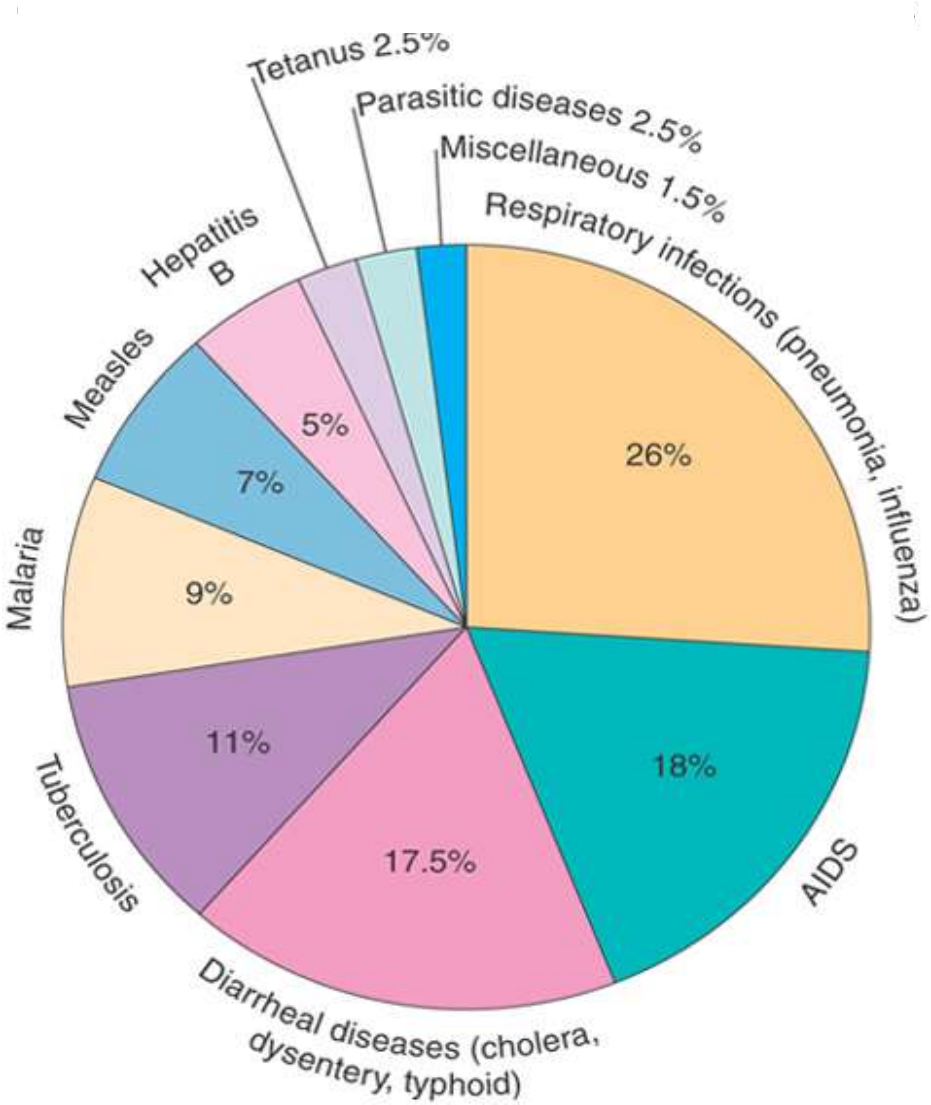
Normal microbiota of our body - objective reality and dramatic insight



- **Curios facts:**
  - ~ 100 trillions bacteria are found in the gut of healthy adults
  - The Microbiome possesses ~3 million genes and weighs ~ 2 kg.
  - The Microbiome exists in peace with the host

Some facts about infectious diseases

- ~2 000 different microbes cause diseases.
- 10 bil new infections/year worldwide
- 13 mil deaths from infections/year worldwide

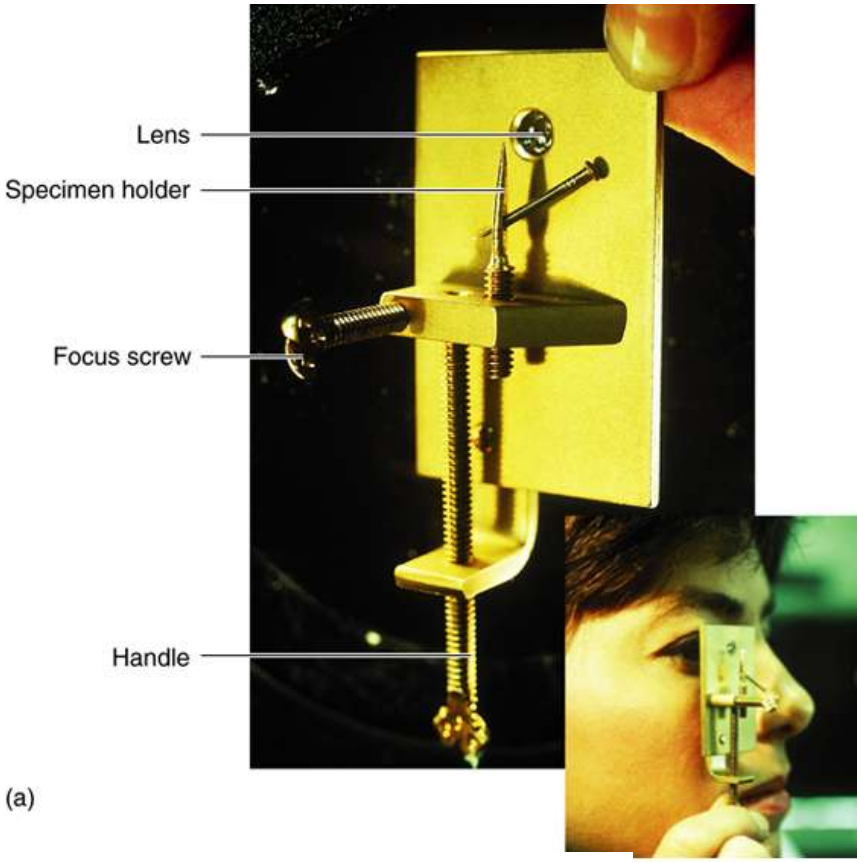


Historical development of Microbiology

- 300 years of contributions by many researchers

- Prominent discoveries include:
  - microscopy
  - scientific method
  - development of medical microbiology
  - microbiology techniques

Antonie van Leeuwenhoek (1632-1723)



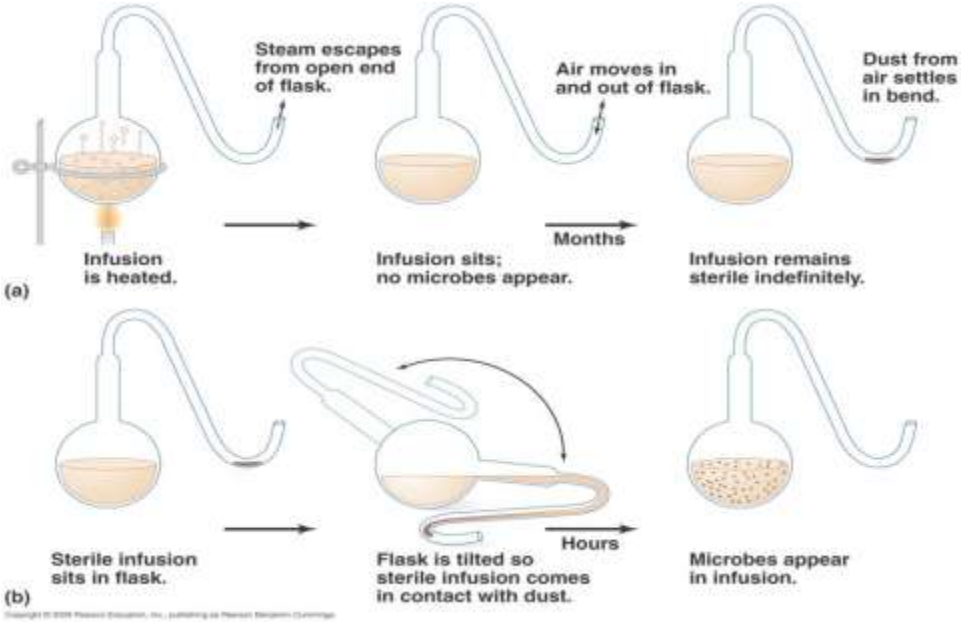
- Dutch linen merchant
- First to observe living microbes
- Single-lens magnified up to 300X

Louis Pasteur (1822-1895)

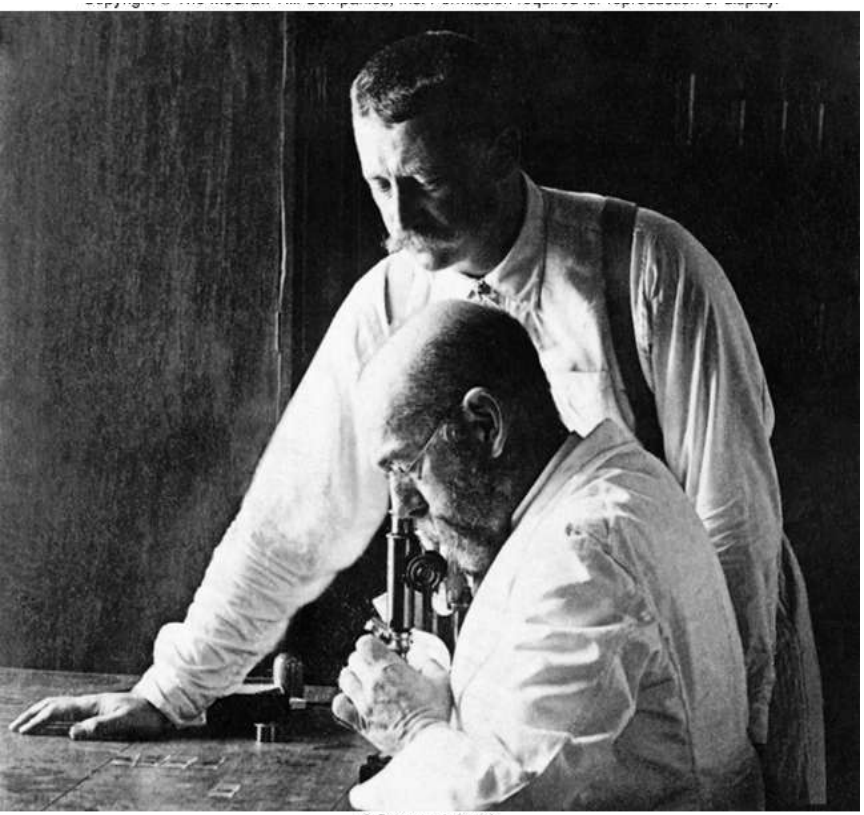
- Showed microbes caused fermentation and spoilage
- Disproved spontaneous generation of microorganisms
- Developed pasteurization
- Demonstrated what is now known as Germ Theory of Disease
- Developed a rabies vaccine

The Golden Age of Microbiology

- **Pasteur's Experiments**
  - When the "swan-necked flasks" remained upright, no microbial growth appeared
  - When the flask was tilted, dust from the bend in the neck seeped back into the flask and made the infusion cloudy with microbes within a day



Robert Koch (1843-1910)


© Bettmann/Corbis

- Established **Koch's postulates** - a sequence of experimental steps that verified the germ theory
- Identified cause of anthrax, TB, and cholera
- Developed pure culture methods
- Nobel Prize 1905

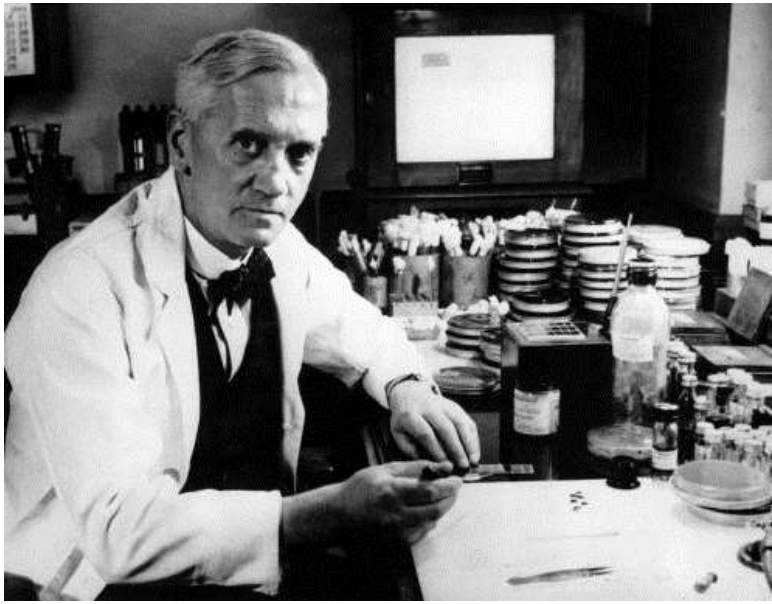
The Birth of Vaccination

- Jenner and smallpox vaccination (1796)

- ~ 100 years later: Pasteur shows how vaccinations work - creation of avirulent strains of bacteria during extended laboratory cultivation

The Birth of Modern Chemotherapy

- 1910: Paul Ehrlich developed a synthetic arsenic drug, salvarsan, to treat syphilis



- 1930s: Synthesis of sulfonamides

- 1928: Alexander Fleming discovered the **first antibiotic - Penicillin**

Selected Nobel Prizes for Microbiology Research

1901 von Behring	Diphtheria antitoxin
1902 Ross	Malaria transmission
1905 Koch	TB bacterium
1908 Metchnikoff	Phagocytes
1945 Fleming, Chain, Florey	Penicillin
1952 Waksman	Streptomycin
1969 Delbrück, Hershey, Luria	Viral replication
1987 Tonegawa	Antibody genetics
1997 Prusiner	Prions
2005 Marshall & Warren	*H. pylori* & ulcers
2008 Harald zur Hausen	HPV and Ca cervix
Françoise Barré-Sinoussi and Luc Montagnier	HIV

Contemporary challenges in medical microbiology

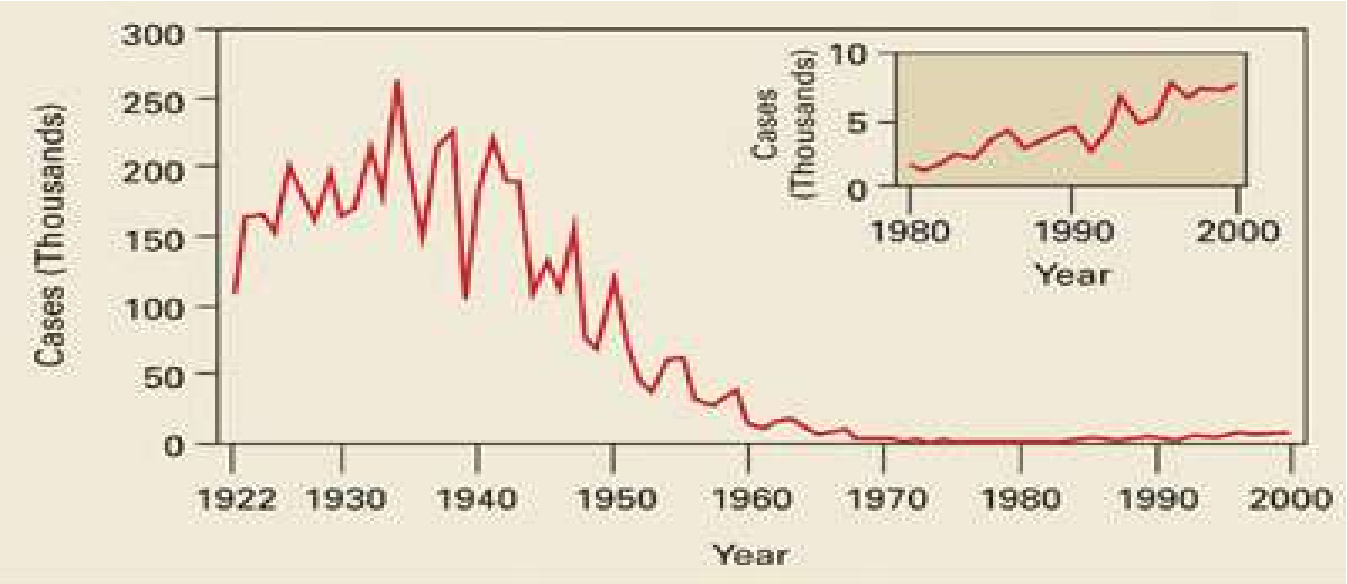
- New approaches for diagnostics and treatment of infections caused by new or hardly known agents:
  - Legionella disease
  - AIDS, Hantaviruses, West-Nile-encephalitis
  - viral hepatitis B, C, D, E, G
  - bird and swine influenza (grippe)
  - Ebola, ZIKA virus

Fig. 53.2 Canoeing in fresh water, a risk for leptospirosis.

Fig. 19.1 Cooling towers—a possible source of *Legionella* infection.

Contemporary challenges in medical microbiology

- Diagnostics of “re-emerging” infections:
  - tuberculosis
  - diphtheria
  - pertussis



Source: Pertussis—United States, 1997–2000. *MMWR Morb Mortal Wkly Rpt* 2002; 51:73–76.

Contemporary challenges in medical microbiology

- Emerging antimicrobial resistance
- Bioterrorism (anthrax, cholera, plague, etc…)
- Infections in immunocompromised
- New technology for microbiological diagnosis (PCR, *MALDI TOF, microarray)*

Taxonomy

- Formal system originated by **Carl von Linné** (1701-1778)

- Concerned with:
  - *classification* – orderly arrangement of organisms into groups
  - *nomenclature* – assigning names
  - *identification* – discovering and recording traits of organisms for placement into taxonomic schemes

Classification

Domain - Archaea, Bacteria & Eukarya

Kingdom

Phylum

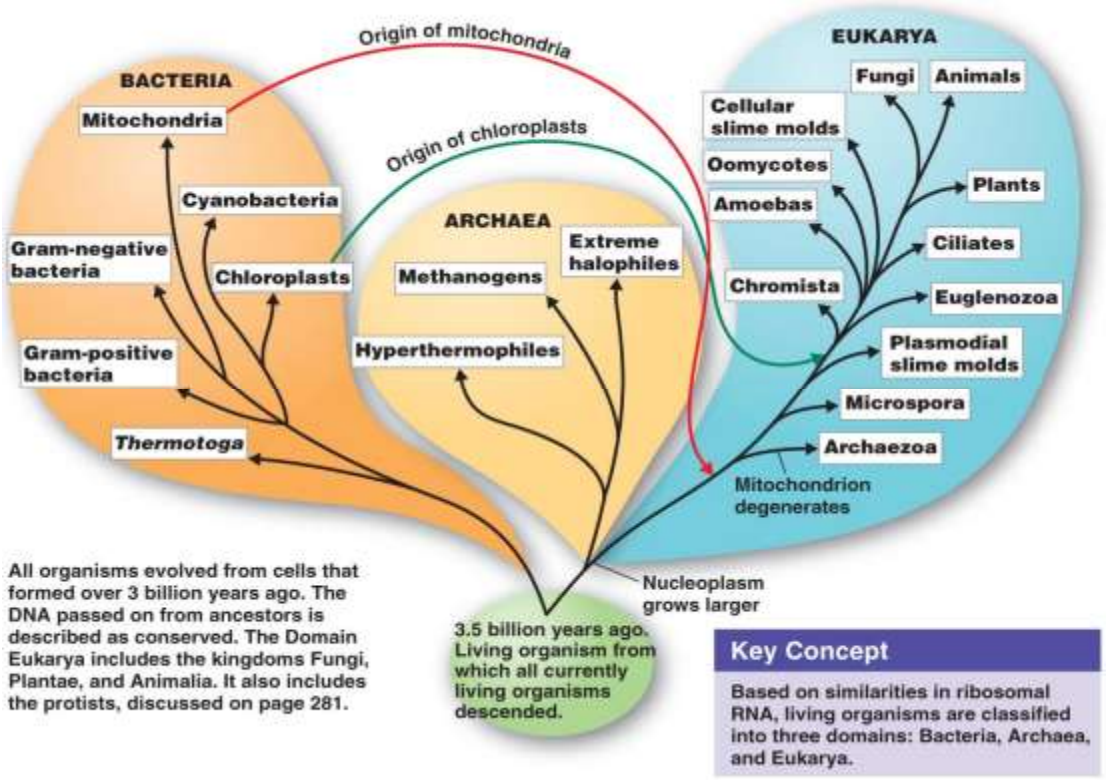
Class

Order

Family

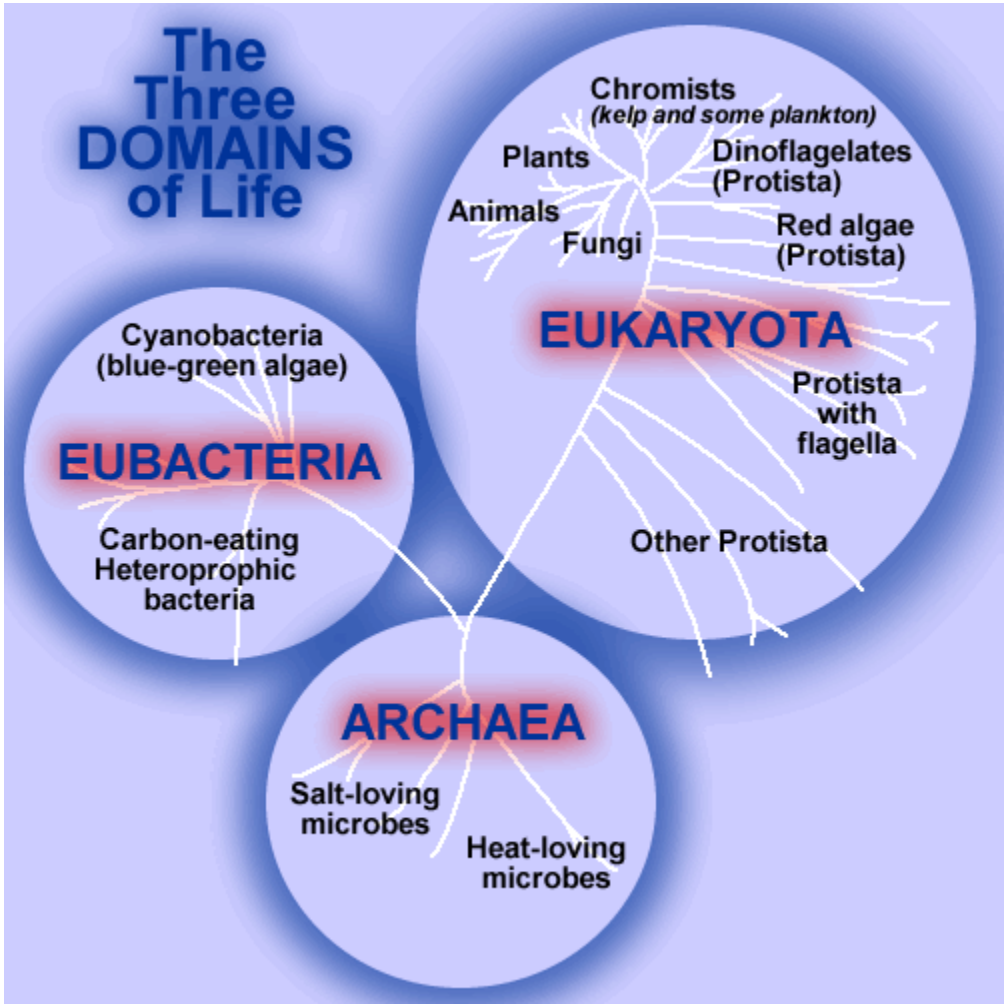
Genus

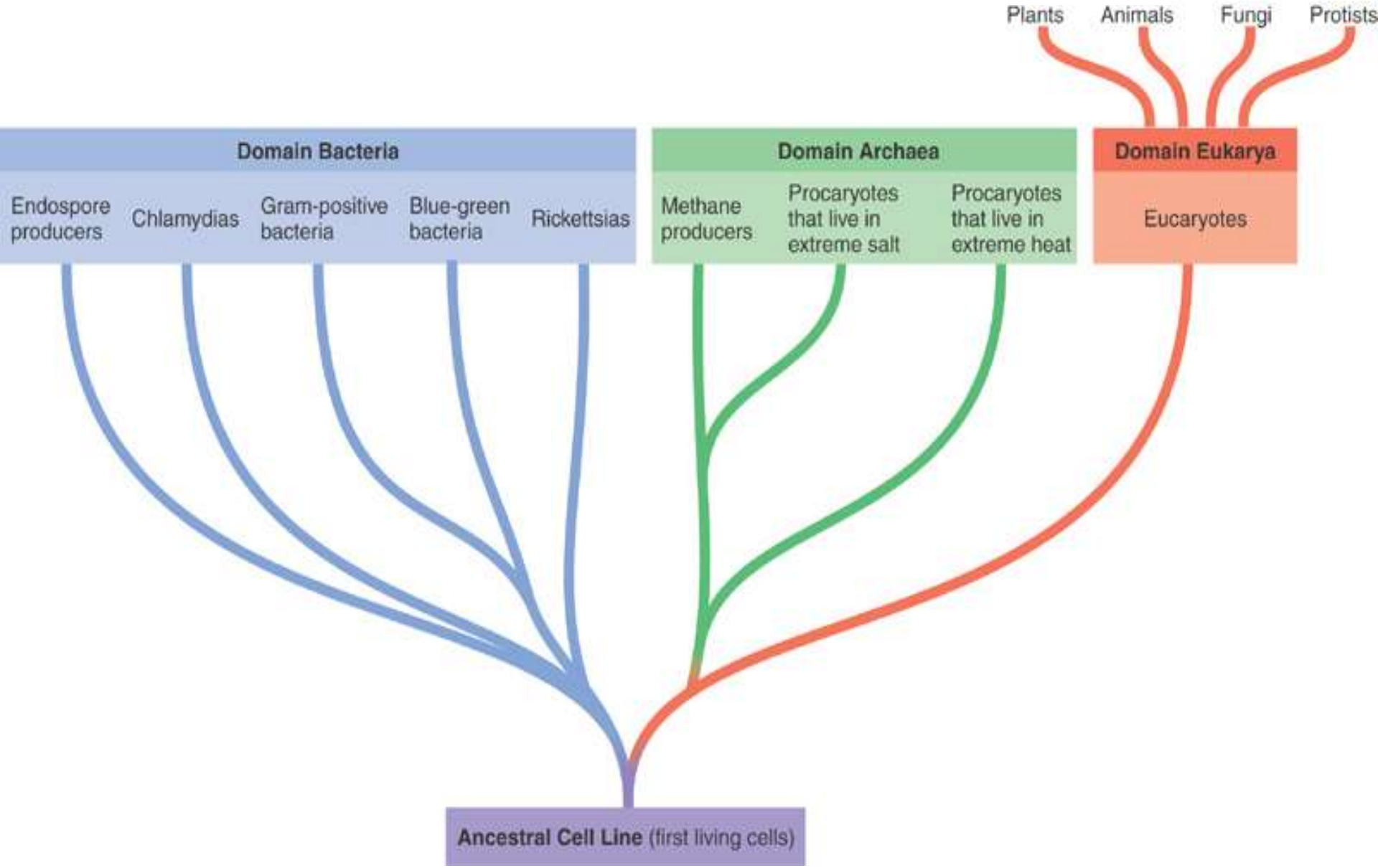
species



3 domain classification

- **Bacteria** - true bacteria, peptidoglycan
- **Archaea** - odd bacteria that live in extreme environments, high salt, heat, etc.
- **Eukarya**- have a nucleus and organelles




Plants
Animals
Fungi
Protists
Domain Bacteria
Endospore producers
Chlamydias
Gram-positive bacteria
Blue-green bacteria
Rickettsias
Domain Archaea
Methane producers
Procaryotes that live in extreme salt
Procaryotes that live in extreme heat
Domain Eukarya
Eucaryotes
Ancestral Cell Line (first living cells)

Bergey's Manual:

Classifying and Identifying Prokaryotes

Bergey's Manual of Determinative Bacteriology Provides *identification* schemes for identifying bacteria and archaea	Morphology, differential staining, biochemical tests
Bergey's Manual of Systematic Bacteriology Provides *phylogenetic* information on bacteria and archaea	Based on rRNA sequencing

Clinical Lab Identification

Morphological characteristics

Useful for identifying eukaryotes

Differential staining

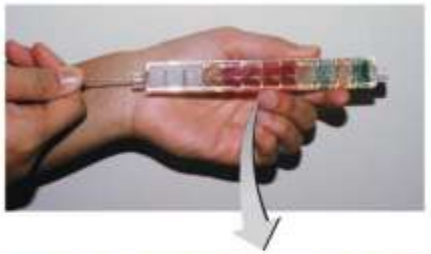
Gram staining, acid-fast staining

Biochemical tests

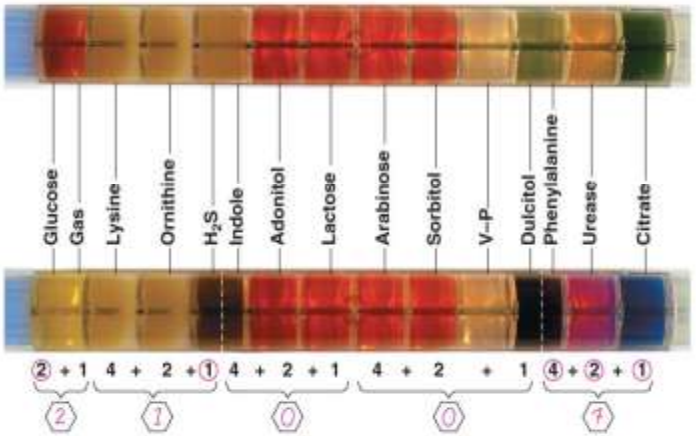
Determines presence of bacterial enzymes

Numerical Rapid Identification

1. One tube containing media for 15 biochemical tests is inoculated with an unknown enteric bacterium.



2. After incubation, the tube is observed for results.



3. The value for each positive test is circled, and the numbers from each group of tests are added to give the ID value.

4. Comparing the resultant ID value with a computerized listing shows that the organism in the tube is *Proteus mirabilis*.

ID Value	Organism	Atypical Test Results	Confirmatory Test
21006	*Proteus mirabilis*	Ornithine⁻	Sucrose
21007	*Proteus mirabilis*	Ornithine⁻	
21020	*Salmonella choleraesuis*	Lysine⁻	

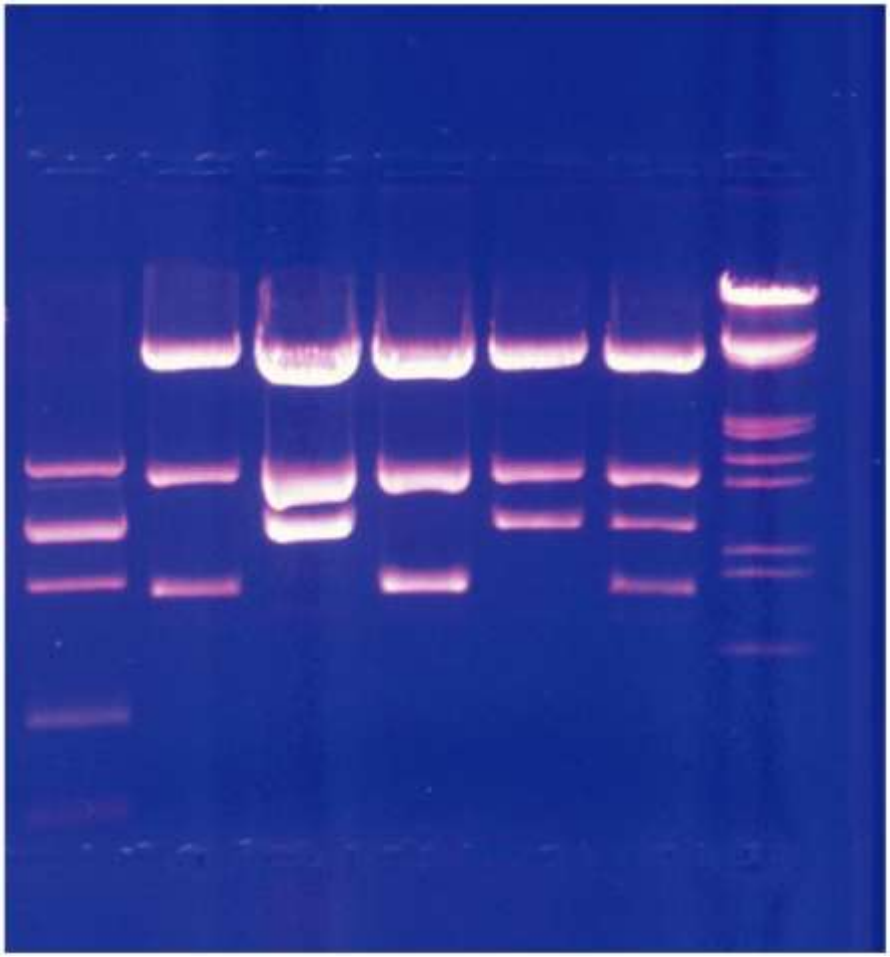
Serology

- Involves reactions of microorganisms with specific antibodies: *Combine known anti-serum with unknown bacterium*
- Useful in determining the identity of strains and species, as well as relationships among organisms.

- Examples:
  - Slide agglutination
  - ELISA
  - Western blot

Genetics

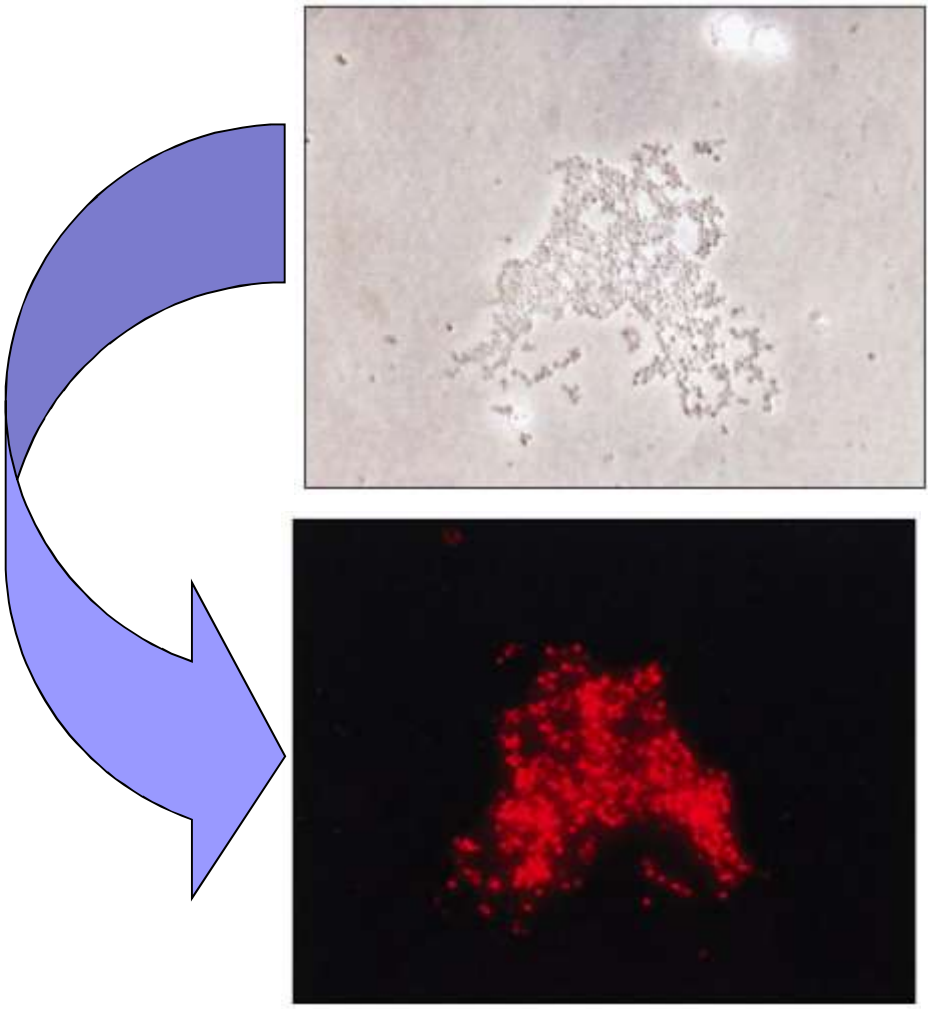
- **DNA fingerprinting:** Number and sizes of DNA fragments (fingerprints) produced by RE digests are used to determine genetic similarities.
- Ribotyping: rRNA sequencing
- Polymerase chain reaction (PCR) can be used to amplify a small amount of microbial DNA in a sample. The presence or identification of an organism is indicated by amplified DNA.



Electrophoresis of amplified by PCR plasmid DNA

Fluorescent In Situ Hybridization (FISH)

Add DNA or RNA probe attached to fluorescent dye
for *S. aureus*



Nomenclature

- Binomial (scientific) nomenclature

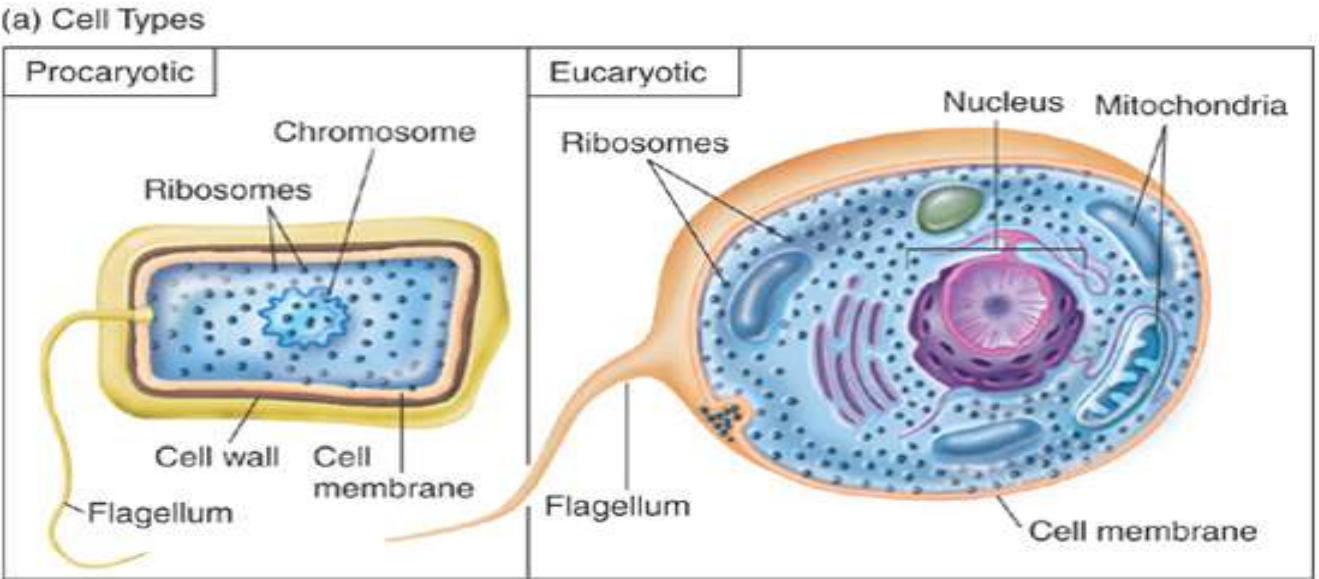
- Each microbe has 2 names:
  - **Genus** - noun, always capitalized
  - **species** - adjective, lowercase

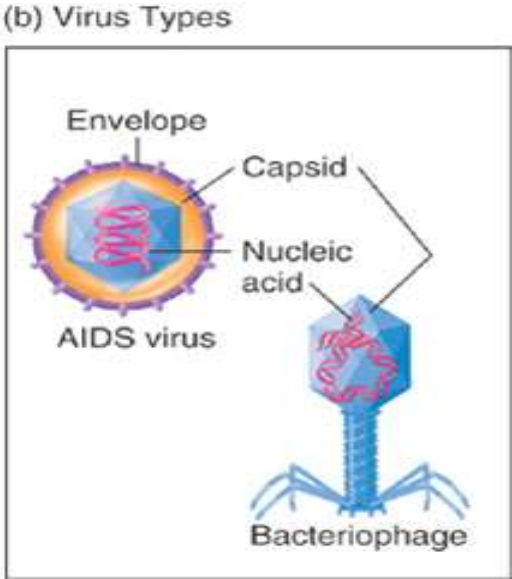
- Both *italicized* or <u>underlined</u>
  - *Staphylococcus aureus (S. aureus)*
  - *Bacillus subtilis (B. subtilis)*
  - *Escherichia coli (E. coli)*

Microorganisms are:

- Procaryotes and eukaryotes
  - **Procaryotes** – Microscopic, unicellular organisms. Lack nuclei and membrane-bound organelles
  - **Eucaryotes** – Unicellular and multicellular organisms. Have nuclei and membrane-bound organelles
- Viruses

  Acellular, parasitic particles composed of a nucleic acid and protein

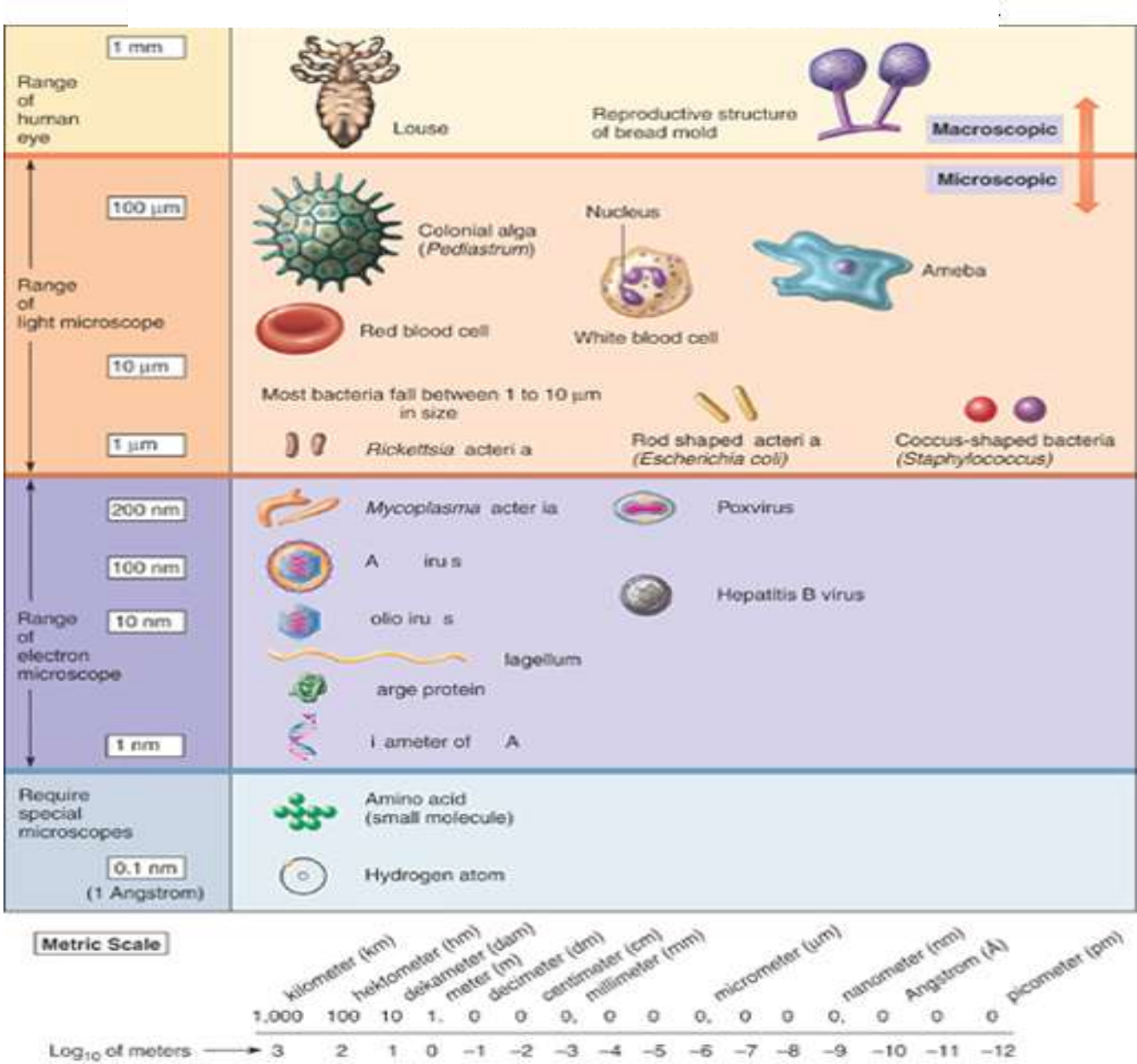




CLASSIFICATION OF MAJOR PATHOGENS					
	viruses	**bacteria**	**fungi**	**protozoa**	**worms**
nucleic acids	DNA or RNA	DNA and RNA	DNA and RNA	DNA and RNA	DNA and RNA
nuclear membrane	no	no	yes	yes	yes
external cell wall	no	yes (usually) rigid peptidoglycan	yes rigid chitin	no	no
antibiotic sensitivity	no	yes	no	some	no
replication/ reproduction	within host cells	within and outside host cells by binary fission	within and outside host cells by binary fission and sexually	within and outside host cells by binary fission and sexually	outside host cells, sexually

Microbial dimensions

- Procaryotes are measured in micrometers
- Viruses in nanometers
- Helminths are measured in millimeters



Take home points

- Microorganisms are causative agents of infections diseases although many of them live happily in our body as “microbiome”;
- In nearly 300 years of history many researchers contributed to the development of medical microbiology – the discovery of antibiotics, germ theory of infectious diseases, vaccines and sera, development of aseptic techniques, different microbiological tests for isolation and identification of bacteria;
- Taxonomy of microbes includes classification, identification and nomenclature;
- Microorganisms are procaryotes (bacteria), eukaryotes (fungi) and viruses. They have different classification and characteristics.

Next time…

- Morphology and structure of bacteria