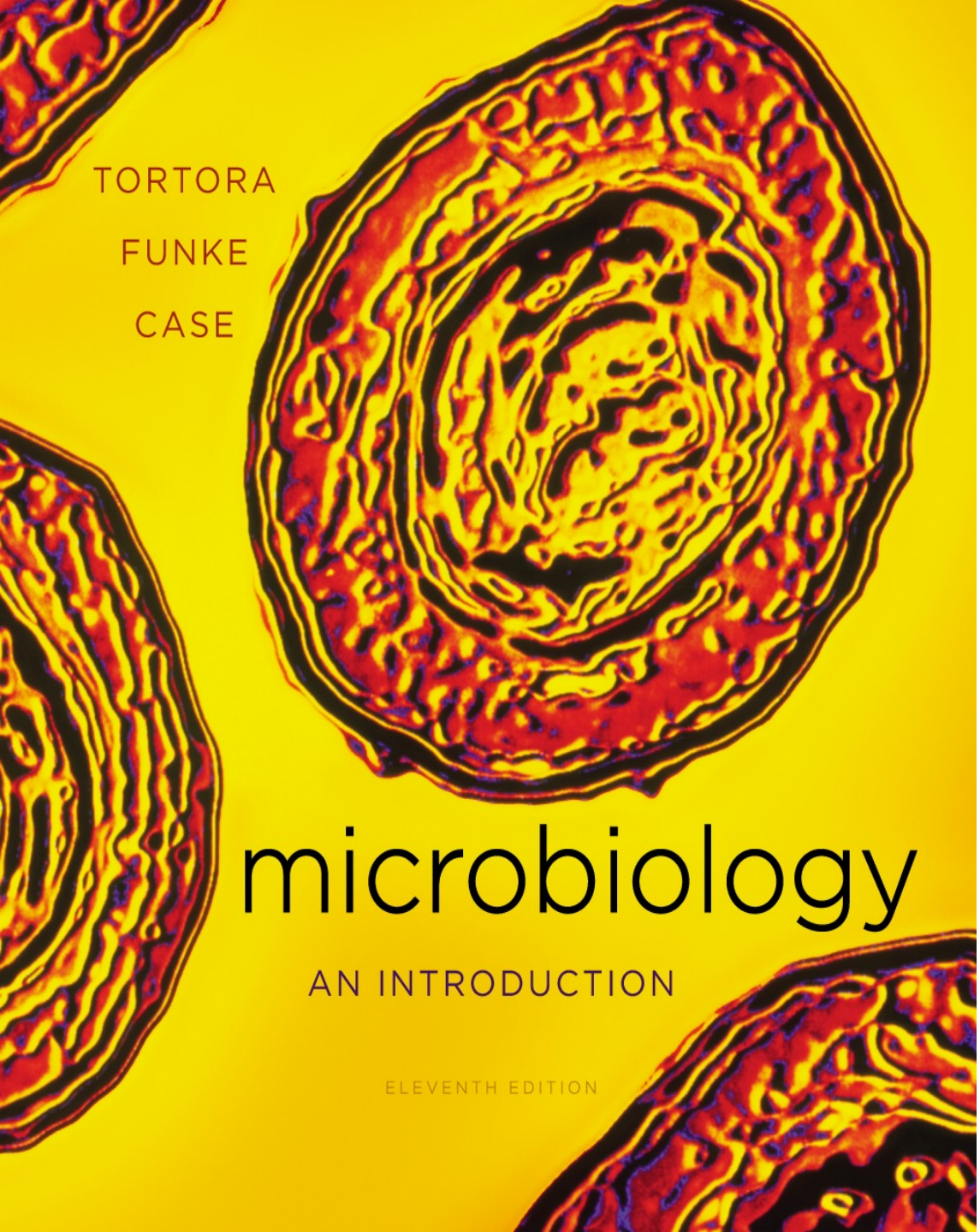


Ch 10

Classification of Microorganisms

Lecture 16
BT 206
22/02/23

Taxonomy and Phylogeny

Taxonomy: Science of classifying organisms.

Provides universal names for organisms.

Taxonomic categories: Taxon / Taxa

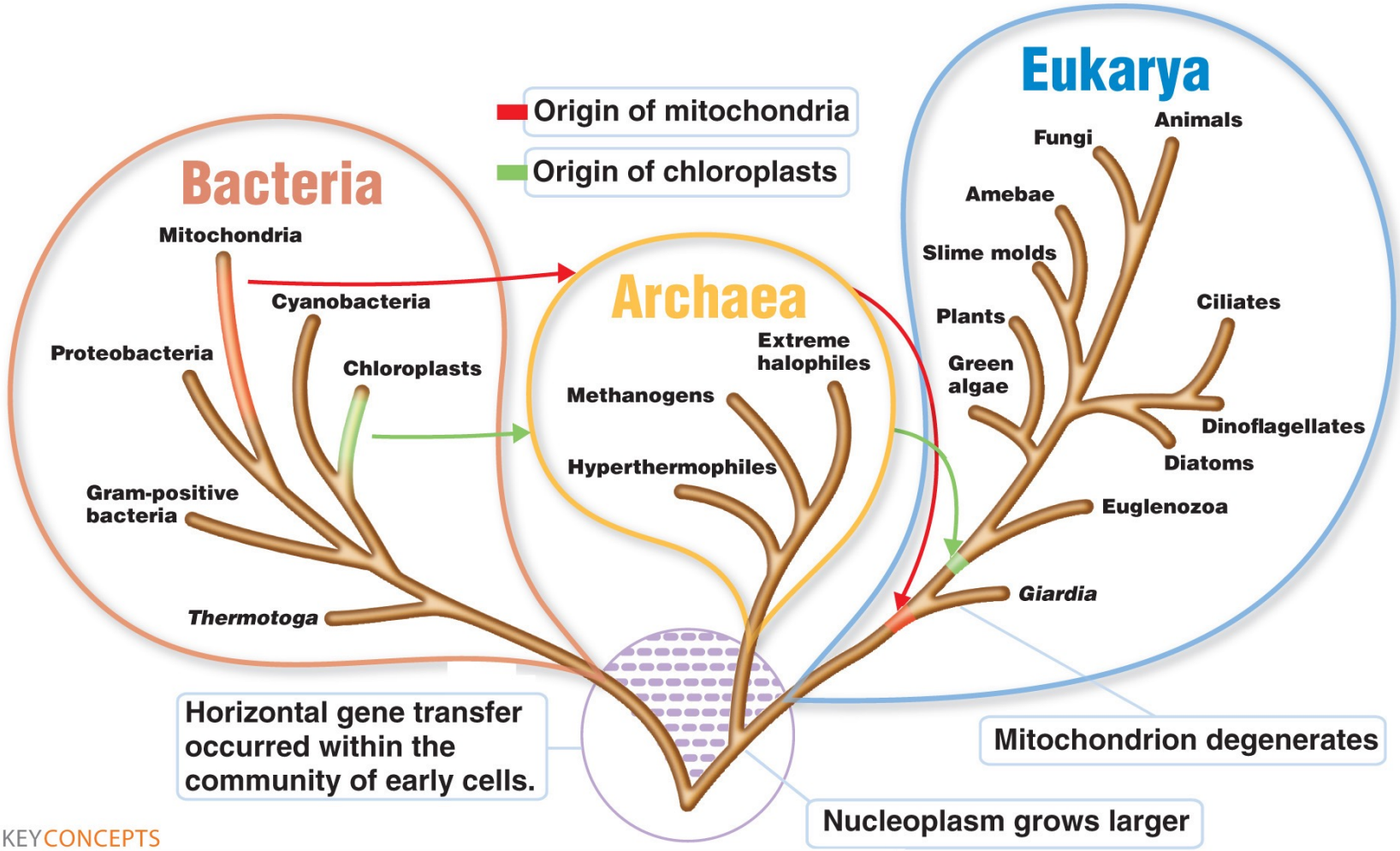
(Domain, Kingdom, Phylum, Class, Order, Family, Genus, Species)

Phylogeny or **Systematics**: Evolutionary history of group of organisms.

- Taxonomic hierarchy shows phylogenetic (evolutionary), relationships among organisms.
- **1969:** Living organisms divided into **five kingdoms**.
- **1978**: Two types of prokaryotic cells found. Prokaryotic relationships determined by rRNA sequencing. **Domains**!
- All Species Inventory (2001–2025)

The Three-Domain System

Foundation Fig 10.1



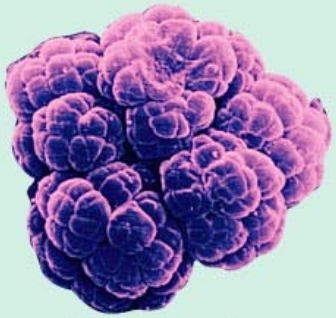
KEY CONCEPTS

- All organisms evolved from cells that formed over 3 billion years ago.
- The DNA passed on from ancestors is described as *conserved*.
- The Domain Eukarya includes the Kingdoms Fungi, Plantae, and Animalia, as well as protists. The Domains Bacteria and Archaea are prokaryotes.

Level Above Kingdom: The Three-Domain System

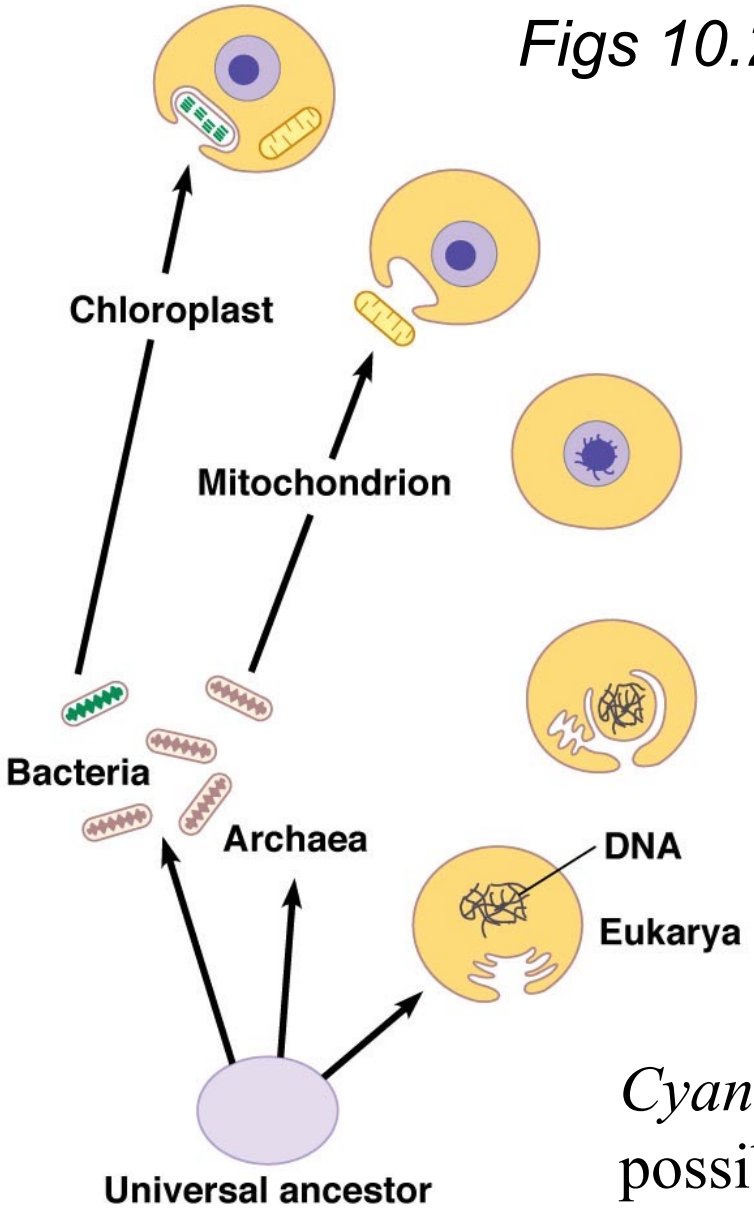
Carl Woese 1978

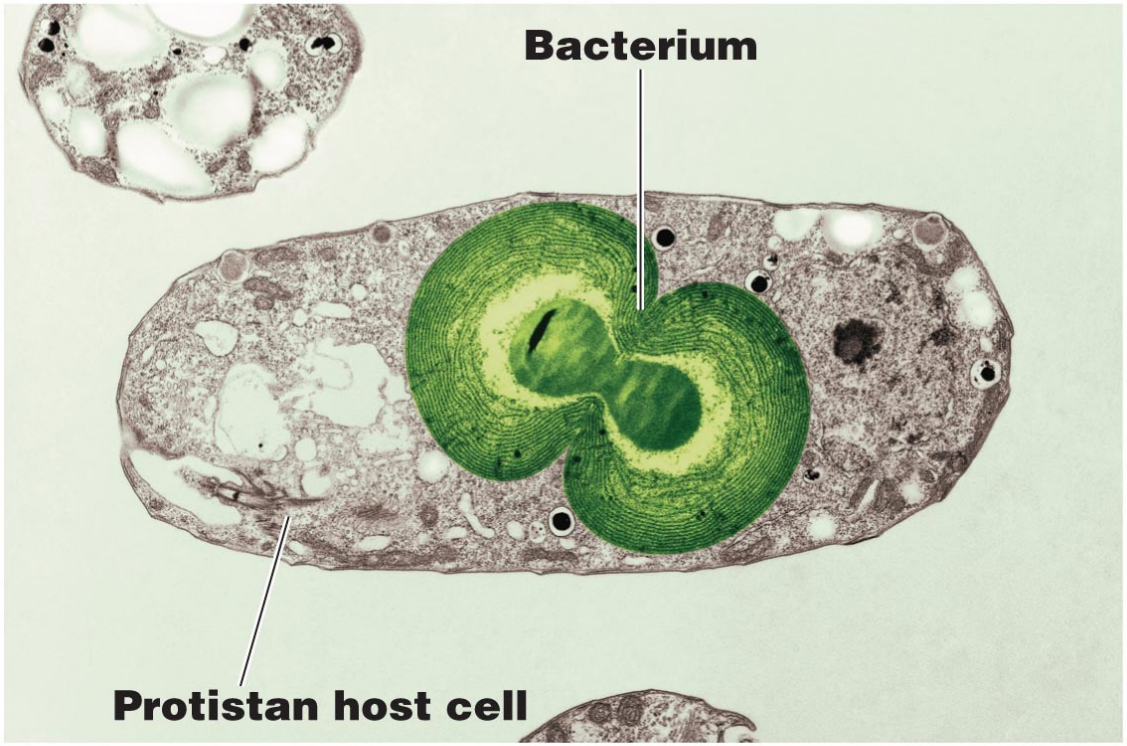
TABLE 10.1 Some Characteristics of Archaea, Bacteria, and Eukarya

	Archaea	Bacteria	Eukarya
	 *Methanosarcina* SEM 10 μm	 *E. coli* SEM 1 μm	 *Amoeba* SEM 10 μm
Cell Type	Prokaryotic	Prokaryotic	Eukaryotic
Cell Wall	Varies in composition; contains no peptidoglycan	Contains peptidoglycan	Varies in composition; contains carbohydrates

Endosymbiotic Theory: Origin of Eukaryotes

Figs 10.2, 10.3





Cyanophora paradoxa: modern example of possible evolutionary process

Scientific Nomenclature

Common names

- Vary with languages
- Vary with geography

Binomial Nomenclature: _____+

_____

- Used worldwide
- *Examples: ?*

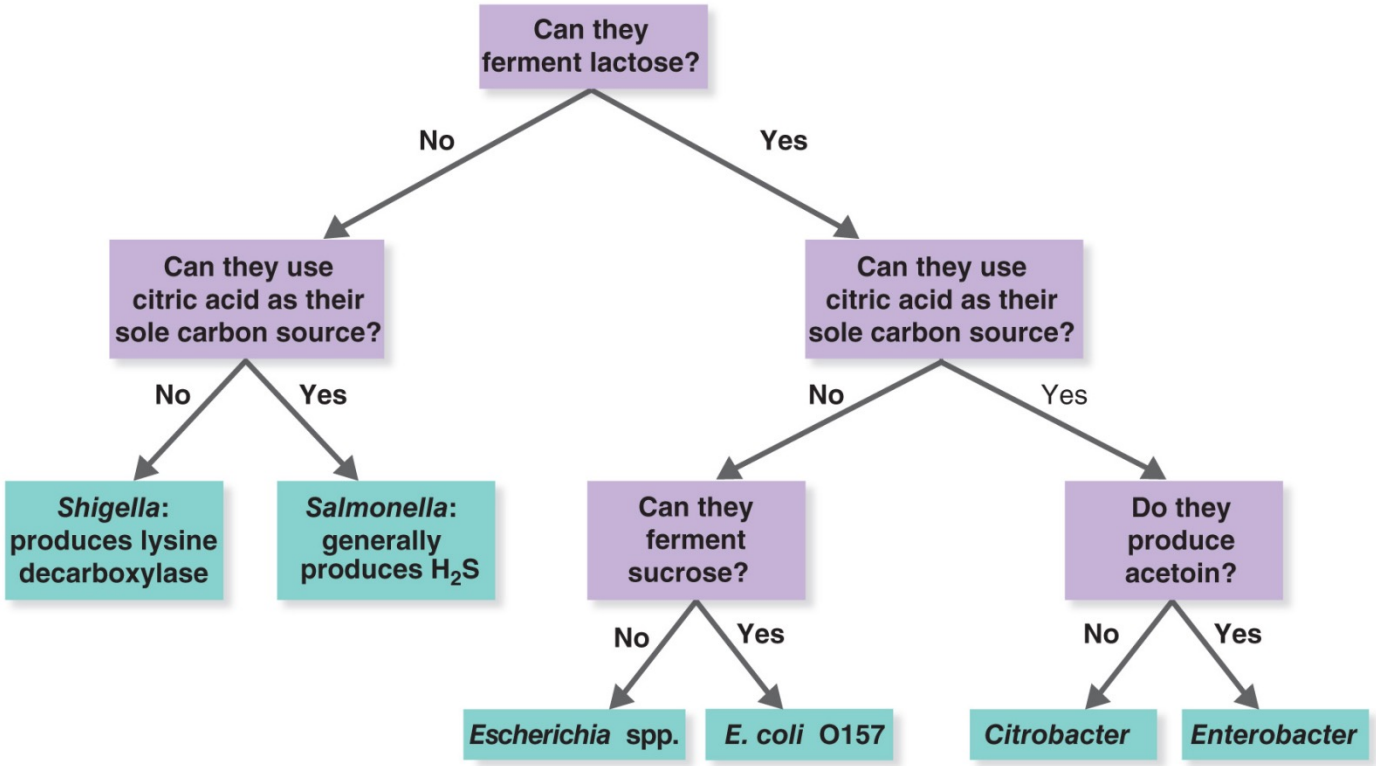
Classification: Species Definition

1. **Eukaryotic species**: Interbreeding organisms
2. **Prokaryotic species**: A population of cells with similar characteristics (*Bergey's Manual of Systematic Bacteriology* is standard reference on bacterial classification).
   - **Culture**: Bacteria grown in laboratory media
   - **Clone**: Population of cells derived from a single cell
   - **Strain**: Genetically different cells within a clone or species
3. **Viral species**: Population of viruses with similar characteristics occupying a particular ecological niche. Viruses not placed in kingdom or domain – *why not?*

Domain Eukarya

- **Animalia**: Multicellular; no cell walls; chemoheterotrophic
- **Plantae**: Multicellular; cellulose cell walls; usually photoautotrophic
- **Fungi**: Chemoheterotrophic; unicellular or multicellular; cell walls of chitin; develop from spores or hyphal fragments
- **Protista**: A catchall kingdom for eukaryotic organisms that do not fit other kingdoms
  - Grouped into **clades** based on rRNA

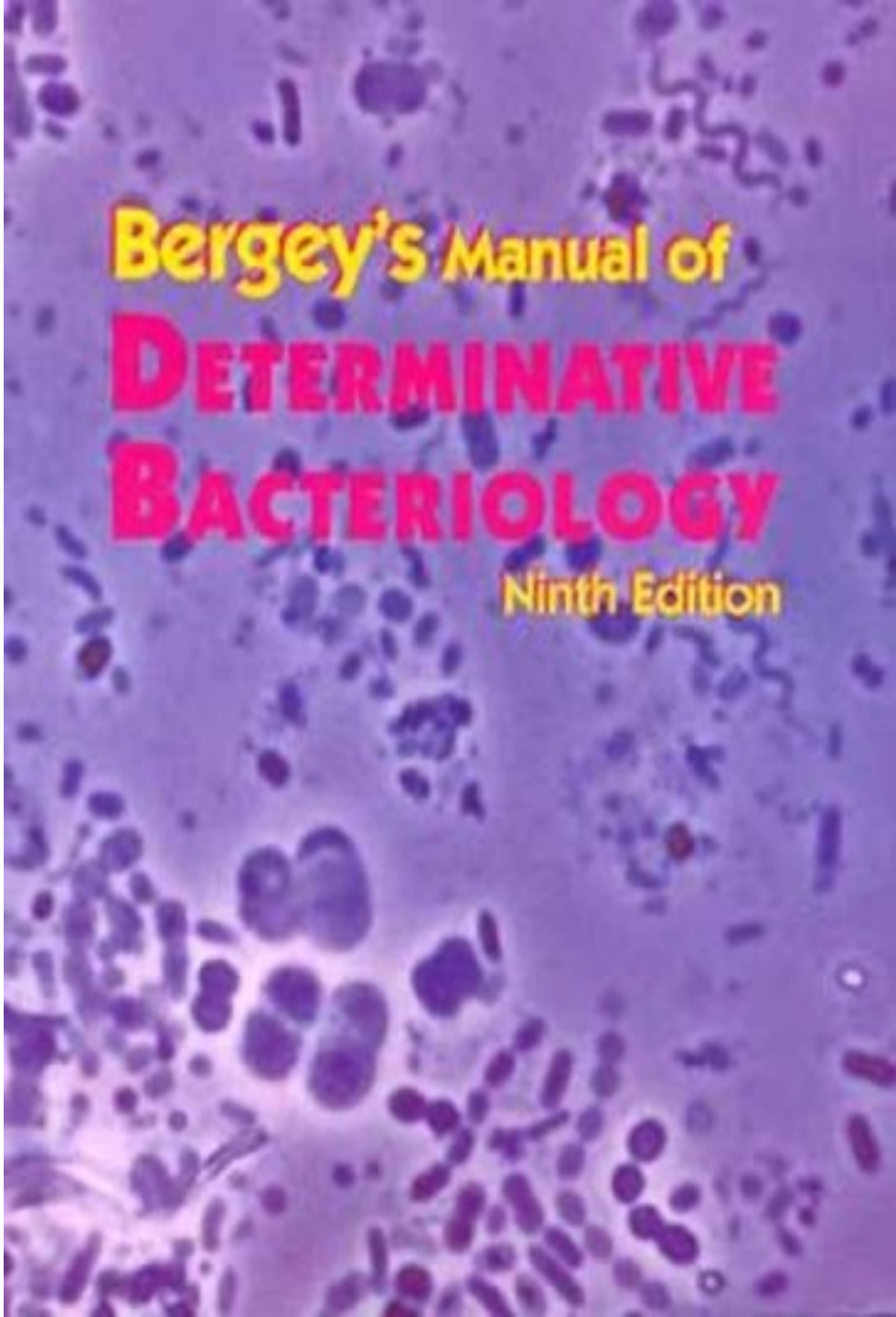
Classification and Identification



Classification: Placing organisms in groups of related species. Lists of characteristics of known organisms.

Identification: Matching characteristics of an "unknown" to lists of known organisms.

David Hendricks Bergey, 1870 - 1937



Bergey's Manual: Classifying and Identifying Prokaryotes

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

Lecture 17
BT 206
23/02/23

Methods for Classifying and Identifying Bacteria

Criterion or Method	Used for	
	Classification	Identification
Morphological characteristics	No (yes for cyanobacteria)	Yes
Differential Staining	Yes (for cell wall type)	Yes
Biochemical Testing	No	Yes
Serology	No	Yes
Phage Typing	No	Yes
Fatty Acid Profiles	No	Yes
Flow Cytometry	No	Yes
DNA Base Composition	Yes	No
DNA Fingerprinting	Yes	Yes
PCR	Yes	Yes
Nucleic Acid Hybridization Techniques	Yes	Yes
rRNA Sequencing	Yes	No

Clinical Lab Identification

Morphological characteristics only useful for identifying eukaryotes.

Differential stains:

_____

_____

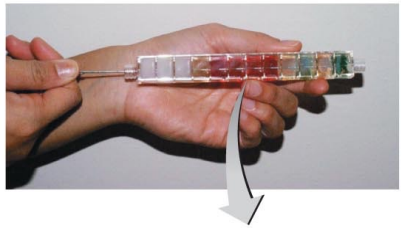
Biochemical tests:

Determine presence of

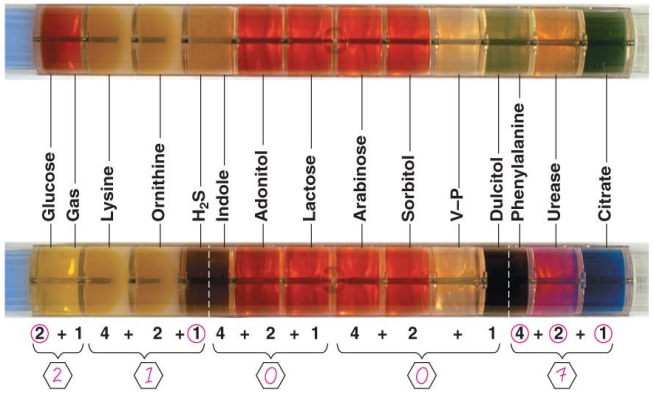
_____

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.

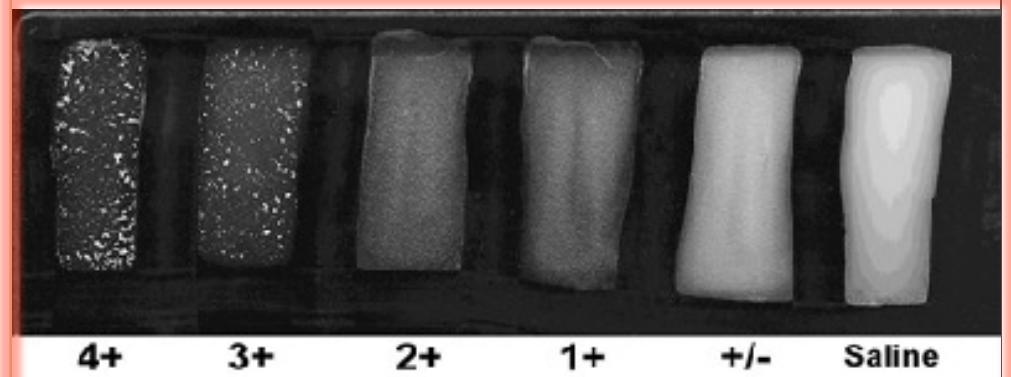
4. Comparing the resultant ID value with a computerized listing shows that the organism in the tube is *Proteus mirabilis*.

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

Serological Testing - Serology

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

Compare to Fig 10.10: Slide Agglutination

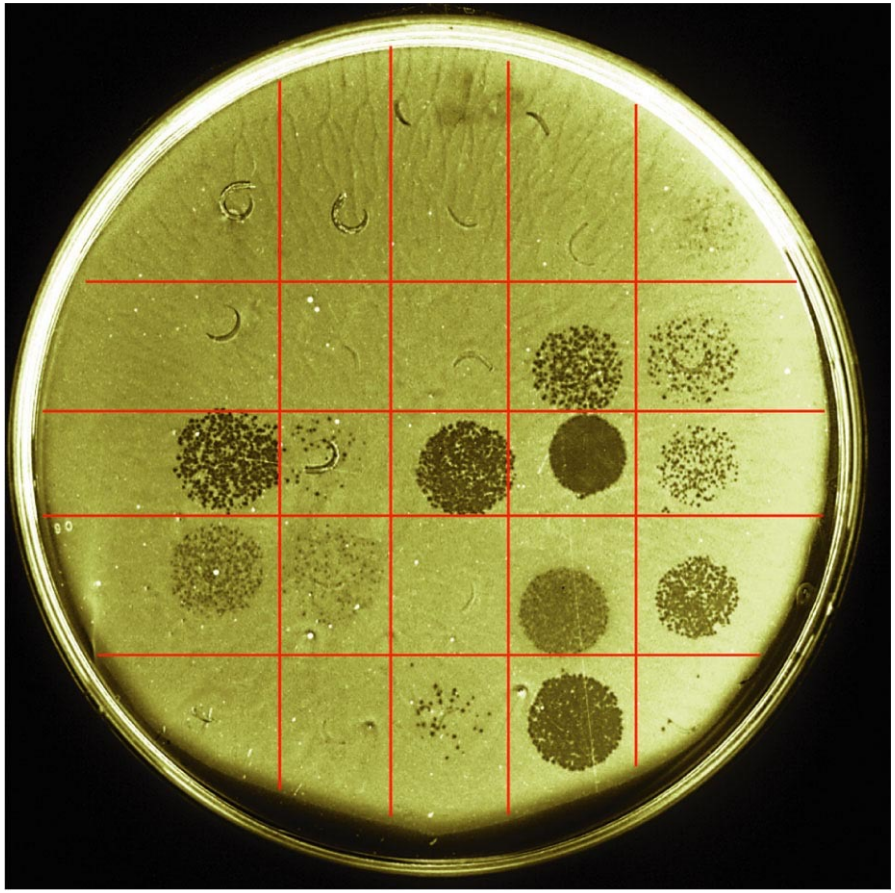


Rating the intensity of the agglutination reaction *of N. meningitidis*

- Examples:
  - Slide agglutination
  - ELISA
  - Western blot

Phage Typing

Fig 10.13



- Phage typing of a strain of *Salmonella enterica*.
- Identification of bacterial species and strains by determining their susceptibility to various phages.

Genetics

- **DNA fingerprinting:** genetic similarities determined by # and sizes of DNA fragments (fingerprints) produced by RE digests
- **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.

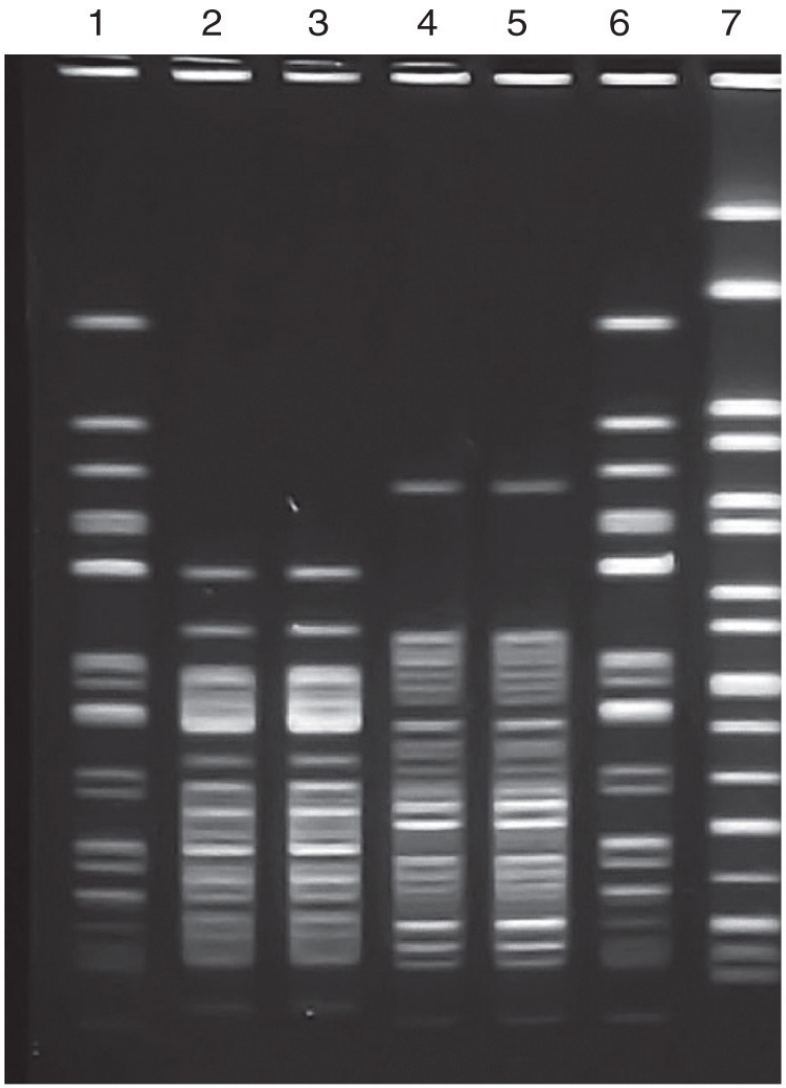
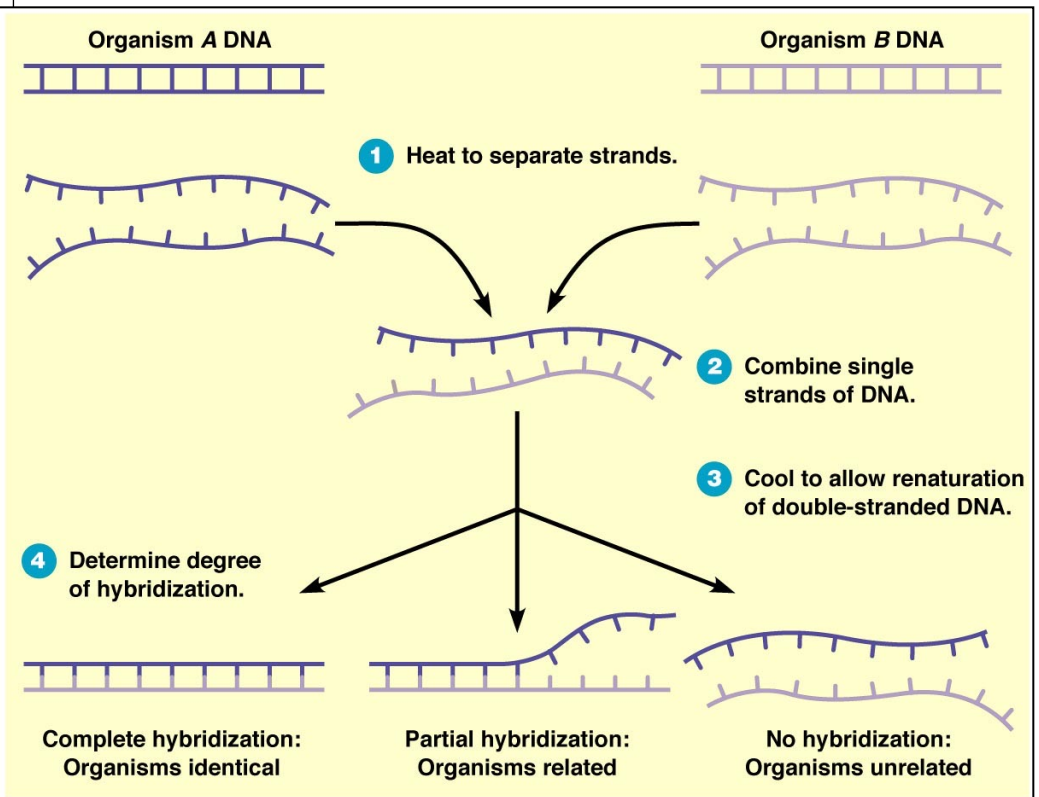


Fig 10.14: Electrophoresis of RE digest of plasmid DNA

Nucleic Acid Hybridization

Single strands of DNA or RNA, from related organisms will hydrogen-bond to form a double-stranded molecule; this bonding is called nucleic acid hybridization.



Examples of Applications:

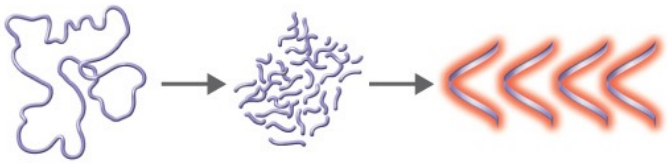
- Southern blotting,
- **DNA chips,** and
- **FISH**

Fig 10.15

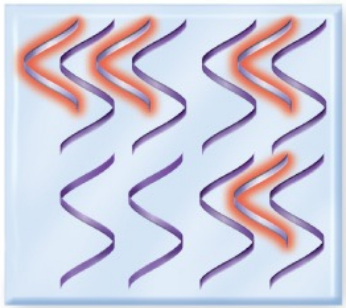
Nucleic Acid Hybridization: DNA Chip

(a) A DNA chip can be manufactured to contain hundreds of thousands of synthetic single-stranded DNA sequences. Assume that each DNA sequence was unique to a different gene.

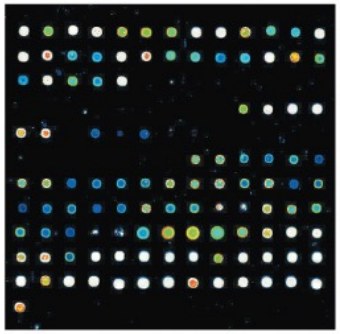
Fig 10.17



(b) Unknown DNA from a sample is separated into single strands, enzymatically cut, and labeled with a fluorescent dye.



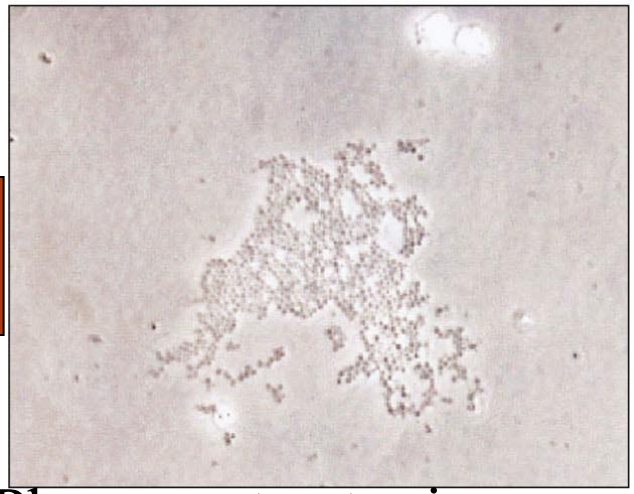
(c) The unknown DNA is inserted into the chip and allowed to hybridize with the DNA on the chip.



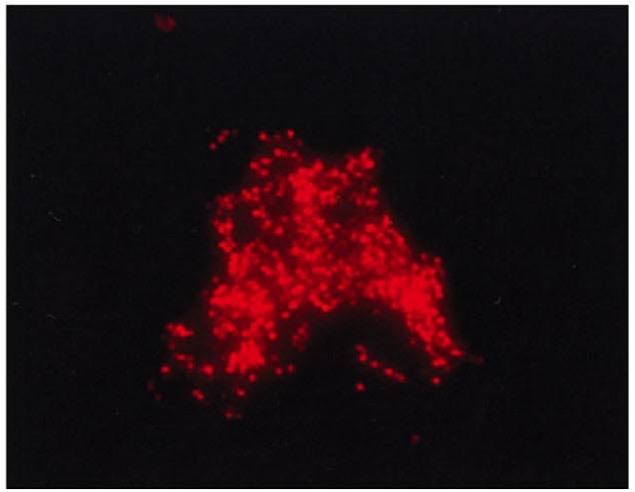
(d) The tagged DNA will bind only to the complementary DNA on the chip. The bound DNA will be detected by its fluorescent dye and analyzed by a computer. In this *Salmonella* antimicrobial resistance gene microarray, *S. typhimurium*-specific antibiotic resistance gene probes are green, *S. typhi*-specific resistance gene probes are red, and antibiotic-resistance genes found in both serovars appear yellow/orange.

Fluorescent In Situ Hybridization (FISH)

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



Phase contrast microscopy



Fluorescent microscopy

- *Detect bacteria in drinking water*
- *Detect bacteria in patient without Waiting for 24 h long culturing*

Fig 10.18a–b