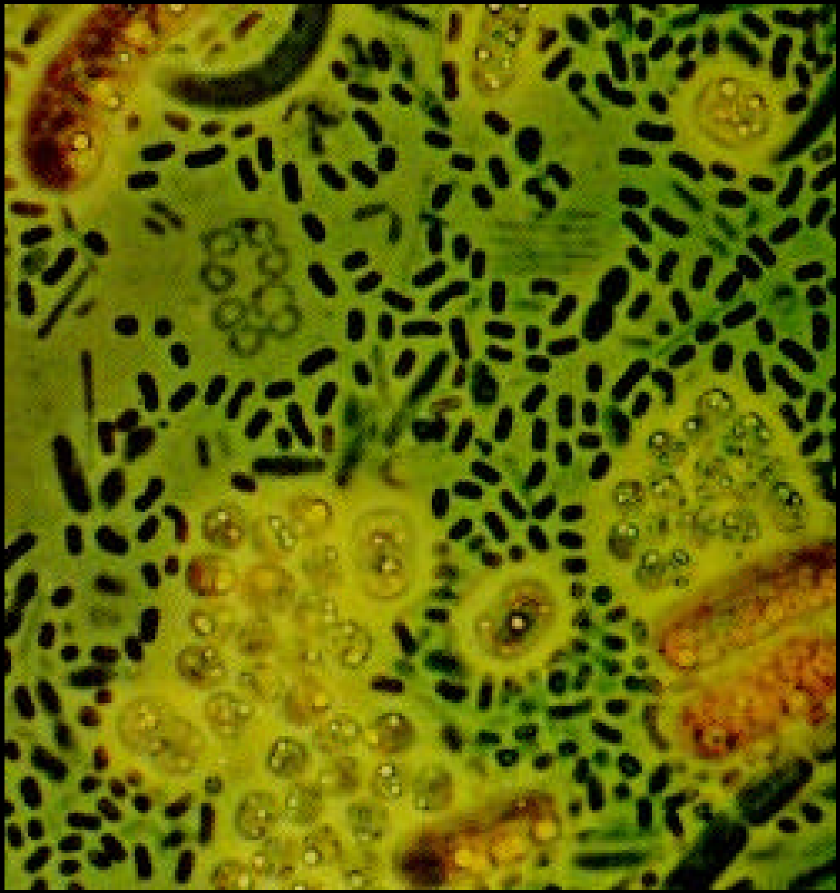
Basics of Microbiology

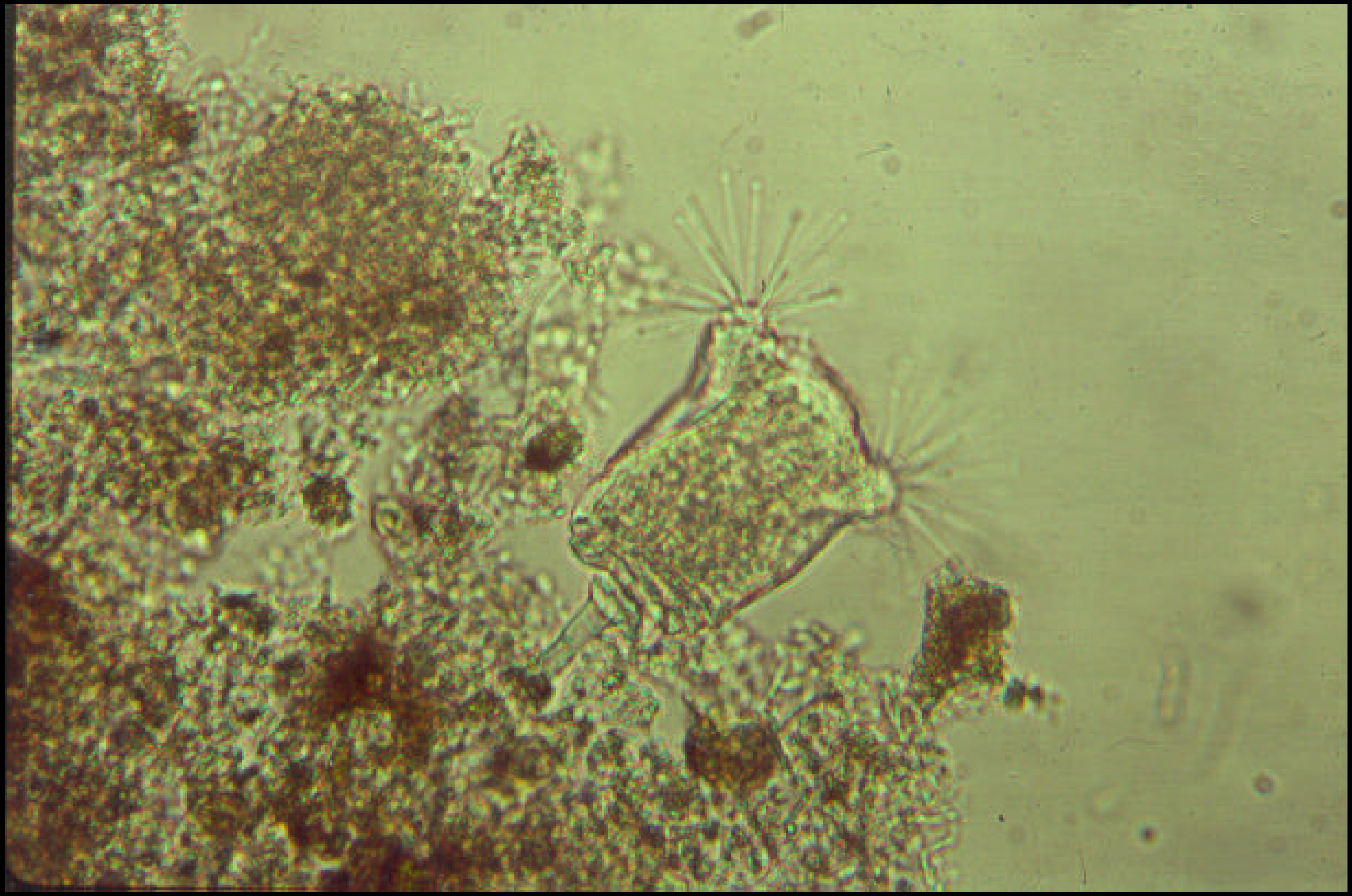
- chemical nature of life
- types of cells
- cell structure and function

Prokaryotic

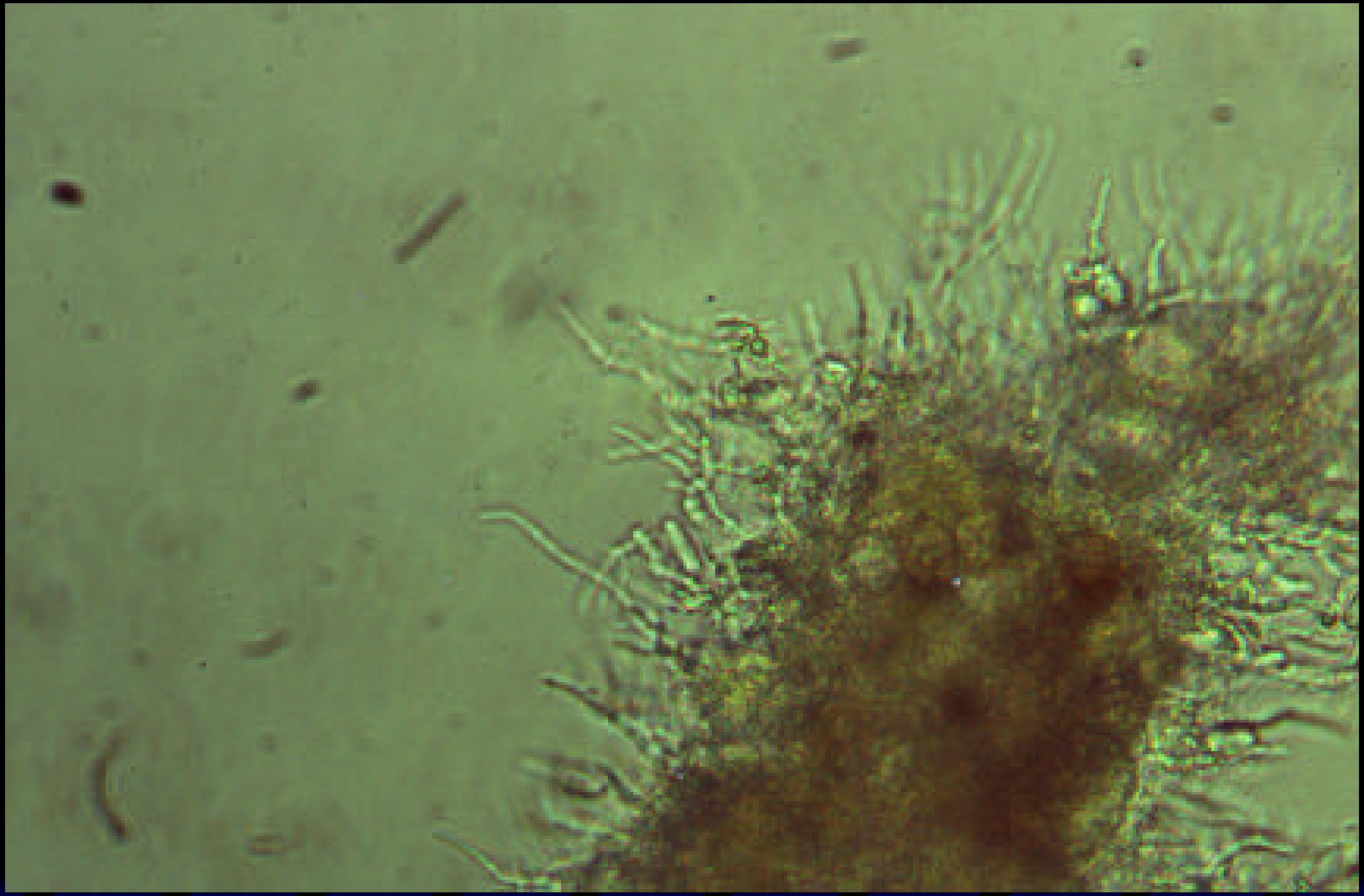


Eukaryotic

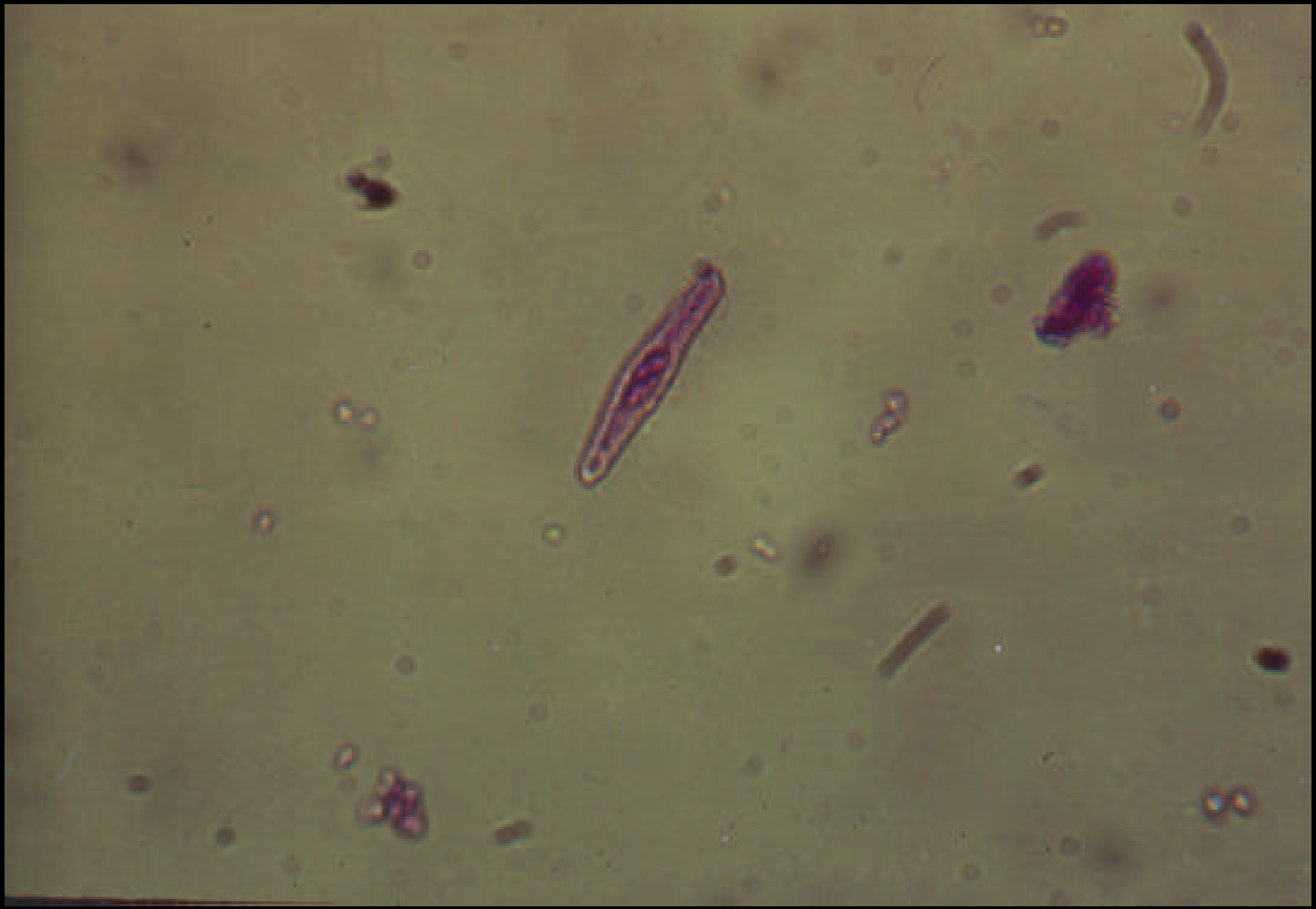
Bacteria and Protozoa



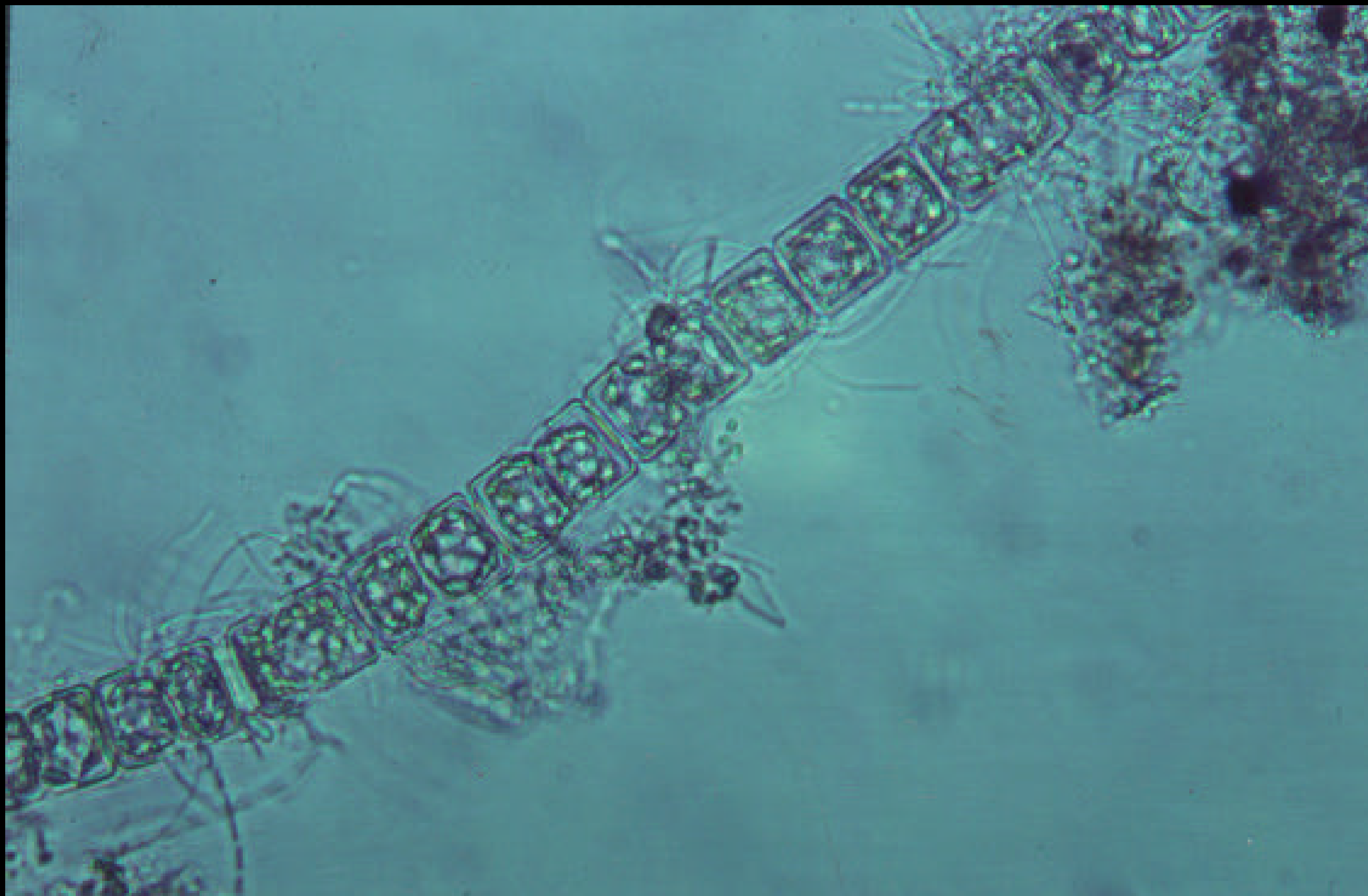
Floc and Protruding Filaments



Ciliated Protozoan

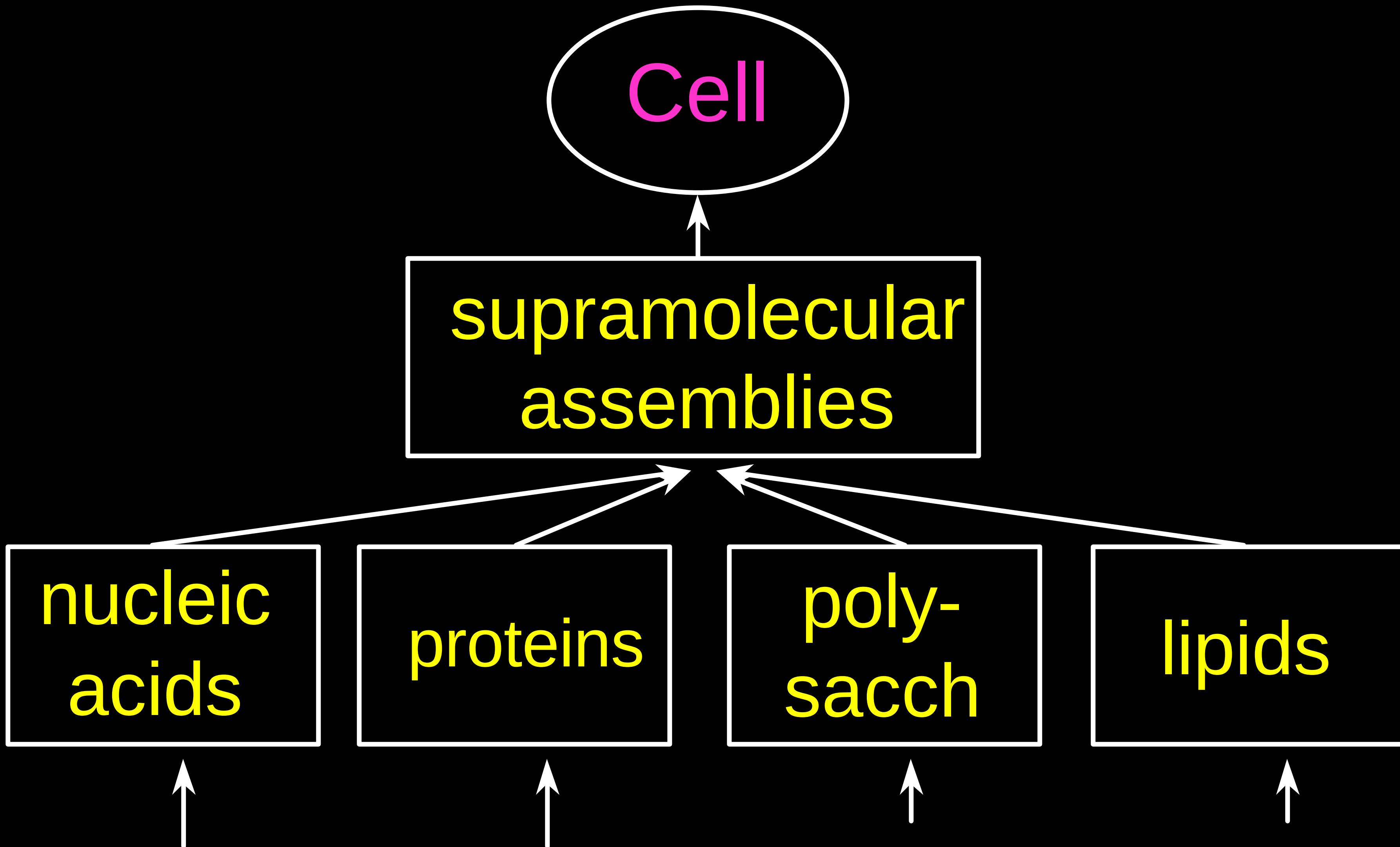


Filamentous Algae

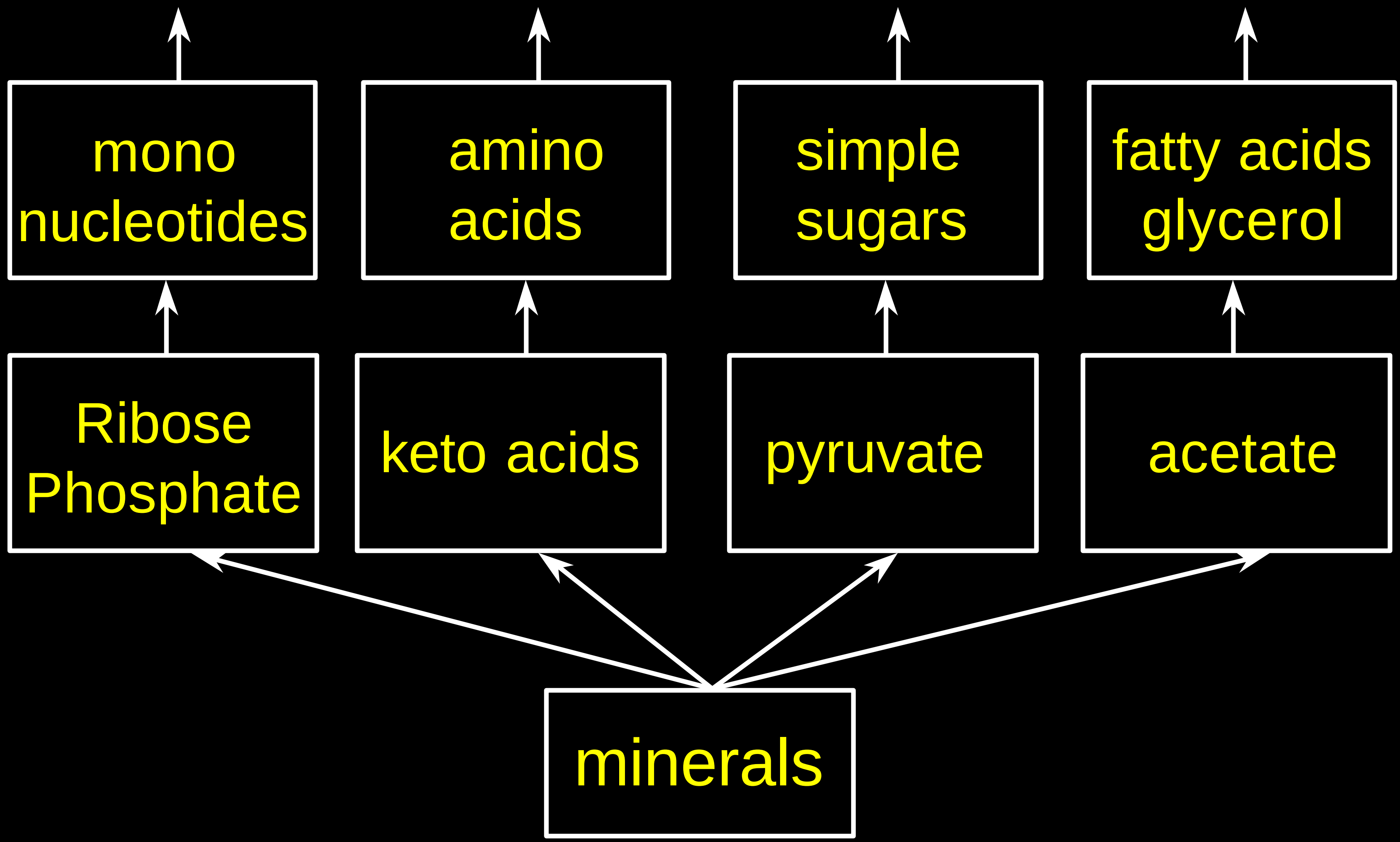


Chemical Nature of Life

Cellular Complexity



Cellular Complexity (Continued)



Chemical Composition of *E. coli*

Element	% Dry Wt
C	50
O	20
N	14
H	8
P	3
S, K, Na, Ca, Mg, Cl, Fe	4.5
Others	0.3

What do we use for Cell Composition?

- Simple formula based on typical cell composition,
  - $C_5H_7O_2N$ MW = 113
  - Should recognize this is a "typical" value that is no more than a representative number
- Alternative to include phosphorus
  - $C_{60}H_{87}O_{23}N_{12}P$ MW = 1374

Functional Groups

- Carbon is a primary element in cellular life
- Carbon may be oxidized or reduced
- These reactions give rise to different functional groups that are important in cellular chemistry. As examples
  - amino acids
  - alcohols
  - DNA, RNA

Functional Groups

Name	Structure	Example
Methyl	CH_3	hydrocarbons

—OH

Name	Structure	Example
Hydroxyl	- OH	alcohols

Functional Groups

Name	Structure	Example
Carbonyl	O ‖ - C -	aldehydes ketones
Carboxyl	O ‖ - C - OH	acids

Functional Groups

Name	Structure	Example
Amine	$- NH_2$	amino acids
Sulfhydryl	$- SH$	amino acids mercaptans

Polymeric Nature of Cellular Structure

- Many of the cells components are constructed of polymeric units
- Cell walls, membranes, storage products, DNA, RNA, enzymes, etc. are all polymers made from a predetermined set of monomers
- The four primary polymers of interest are:

Polymeric Structures

- Lipids: glycerol and fatty acids
- Polysaccharides: carbohydrates
- Proteins: amino acids
- RNA and DNA: nucleotides

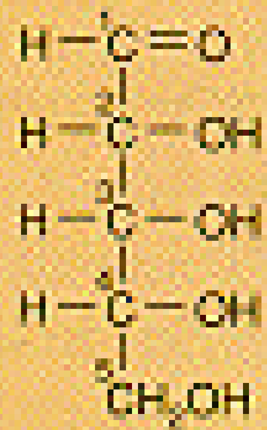
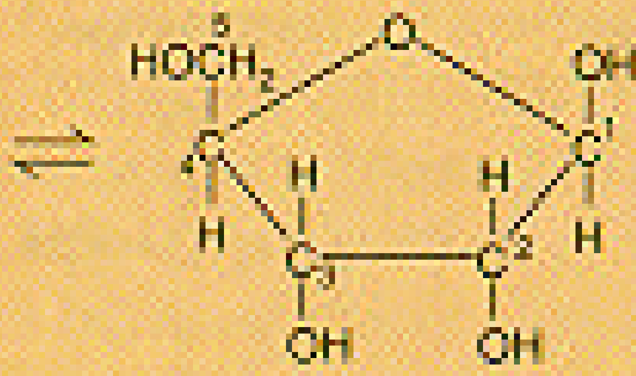
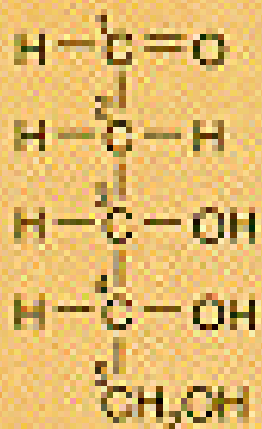
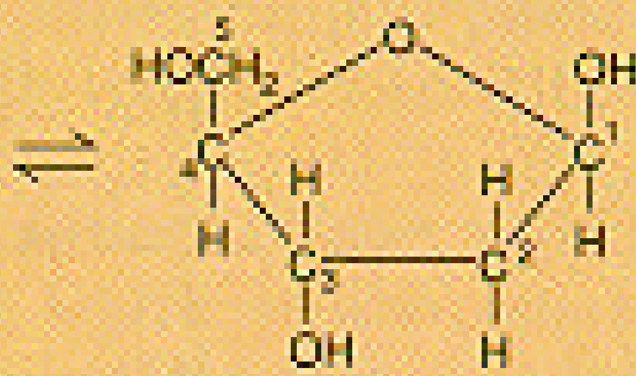
Lipids

- Soluble in non-polar solvents
- Found primarily in cell membranes
- Found in many industrial wastestreams
- Lipids can be classified as Simple or Complex
- An important component of simple lipids are short chain fatty acids which are important intermediates in anaerobic metabolism leading to methane formation

Carbohydrates

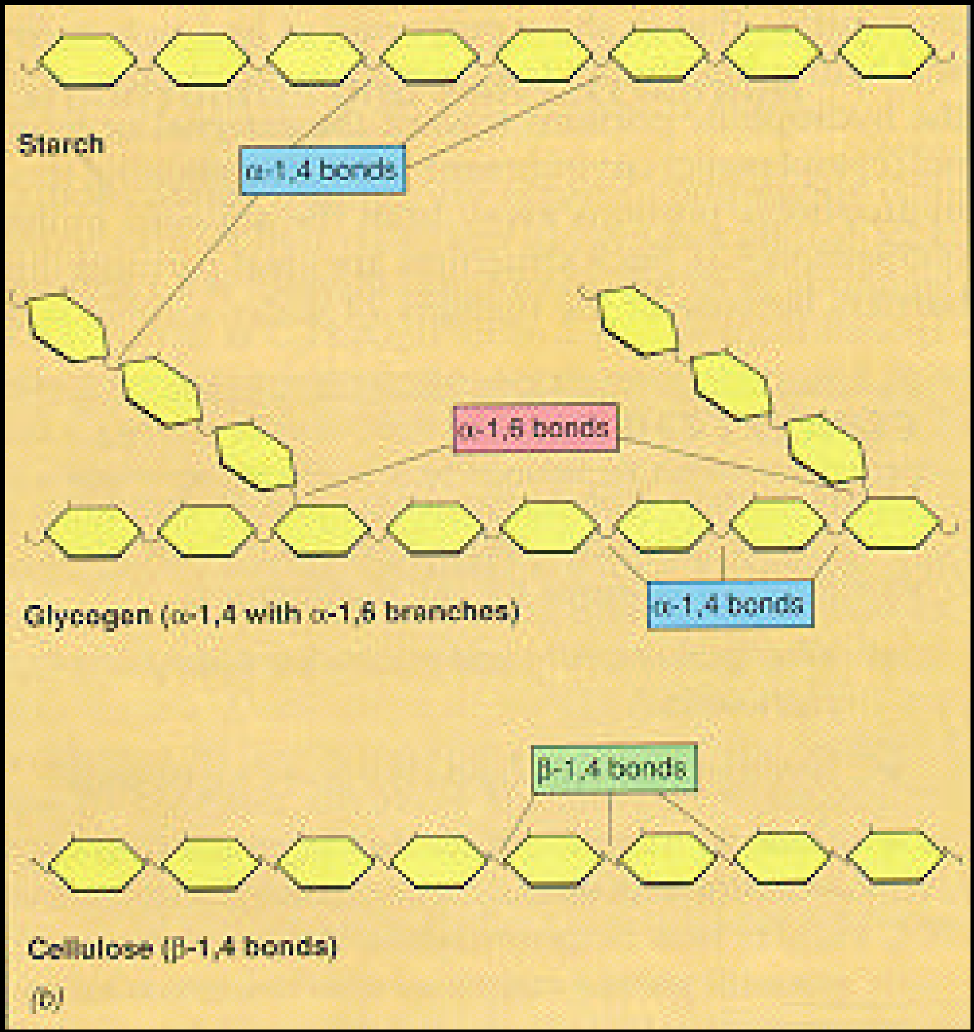
- general group of compounds ($C_nH_{2n}O_n$)
- found in all cells, structural or storage
- most prevalent form of organic matter in biosphere
- basic component of diet (>50%)
- present in large amounts in domestic and industrial wastes

Pentose Sugars

Sugar	Open chain	Ring	Significance
Pent-oses Ribose			Sugar-phosphate backbone of RNA
Deoxy-ribose			Sugar-phosphate backbone of DNA

Complex Sugars: Polysaccharides

Carbohydrate Polymers



Amino Acids

- Sequences of amino acids make proteins (peptide bond)
- All amino acids have an amine group and a carboxyl group
- There are approximately 20 different amino acids found in natural proteins
- Amino acids classified based on their hydrophobicity

Proteins

- Proteins most abudent matter in cell
- Typically 30 - 70% as dry wt.
- All contain C H N O
- Some contain S which contributes to structure
- Proteins contribute to nitrogen loadings in treatment plants
- Found in a variety of wastewaters

Protein Function

- Biological Catalyst (Enzyme)
  - oxidoreductases
  - transferases
  - hydrolases
  - lyases
  - isomerases
- Contractile proteins
- Transport proteins
- Glycoproteins

Nucleotides

- Informational polymer for cell heredity (RNA, DNA)
- Energy carriers (adenosine disphosphate and adenosine triphosphate)

$$ATP + H_20 \rightarrow ADP + PO_4 + \text{energy}$$

- Electron carriers (nicotinamide adenine dinucleotide)

$$NAD^+ + 2e^- + 2H^+ \longrightarrow NADH + H^+ \; (NADH_2)$$

Basic Structure of Nucleotides

- Phosphoric Acid
- Ribose or Deoxyribose sugar
- Nitrogenous base

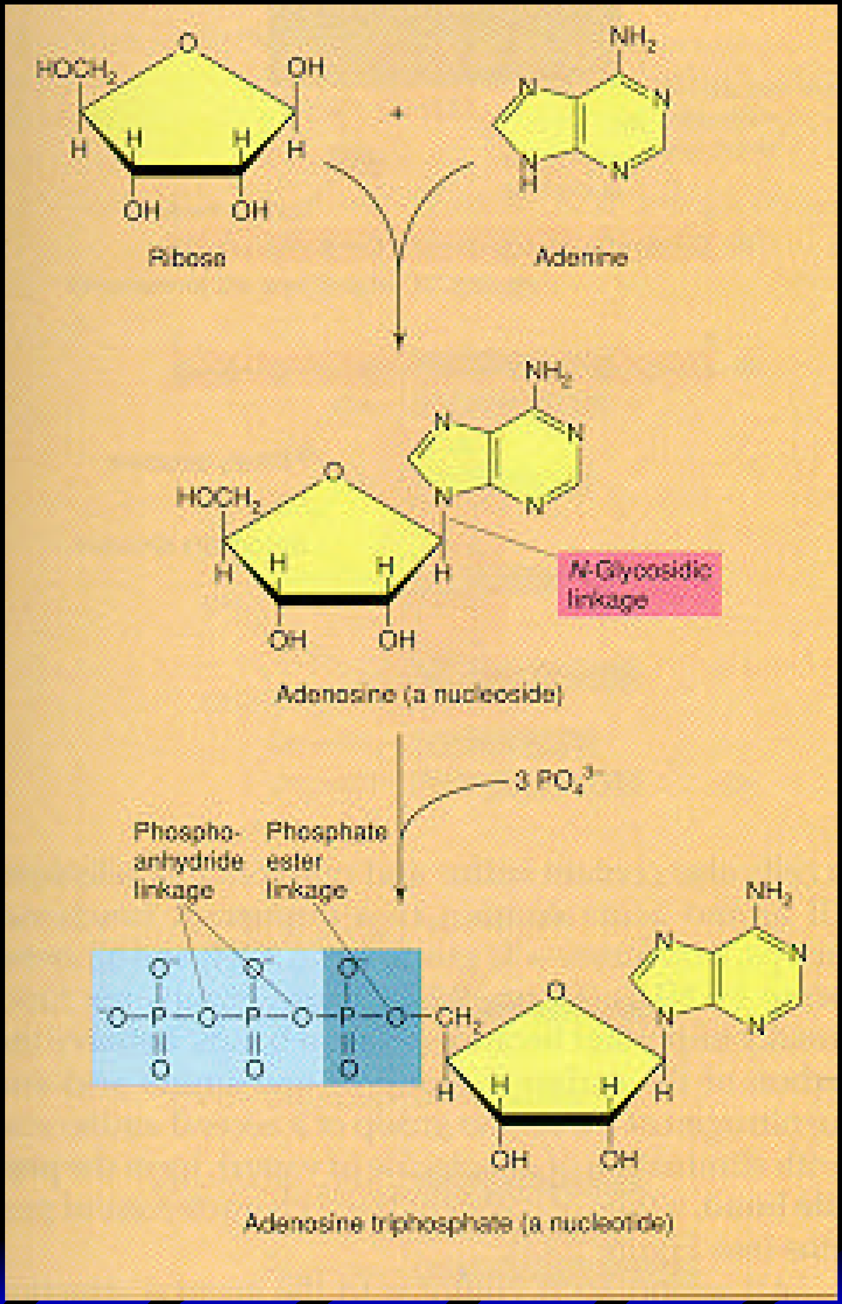
Nitrogenous Bases

DNA (double strand)

- Purine bases
  - Adenine
  - Guanine
- Pyrimidine bases
  - Thymine
  - Cytosine
- A/T
- G/C

RNA (single strand)

- Purine bases
  - Adenine
  - Guanine
- Pyrimidine bases
  - Uracil
  - Cytosine



Structure of ATP

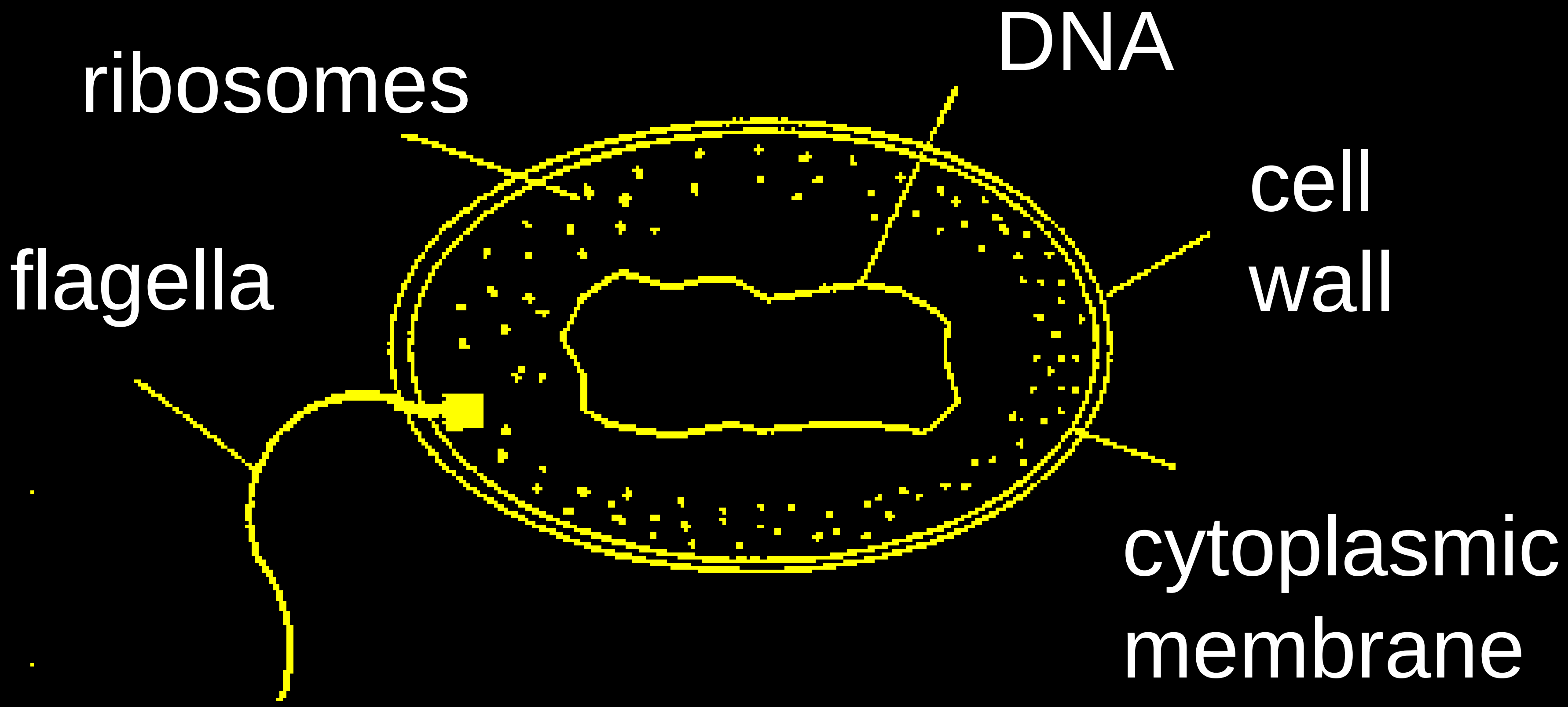
Microorganisms Types

- Basic microorganisms of interest are
  - Bacteria
  - Algae
  - Protozoa
  - Fungi
- Bacteria are Prokaryotic organisms
- Algae, Protozoa, and Fungi are Eukaryotic organisms

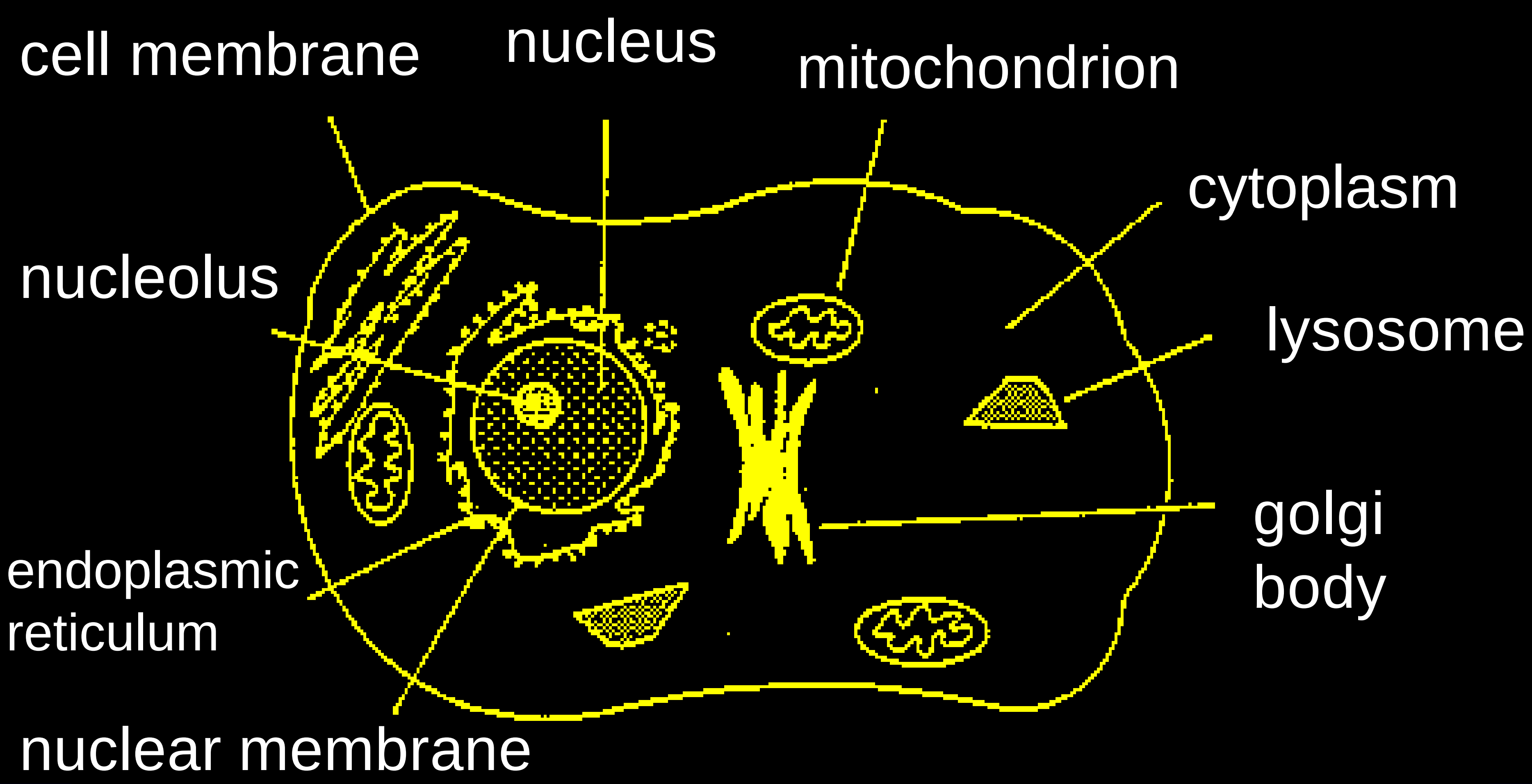
Microorganism Classification

	Prokaryotic	Eukaryotic
Macroorganisms	None known	Eukarya: Animals Plants
Microorganisms	Archaea	Eukarya: Algae Fungi Protozoa
	Bacteria	

Prokaryotic Organism Structure



Eukaryotic Organism Structure



Microorganism Classification

	Prokaryotic	**Eukaryotic**
Size	very small, 1 - 5 μm long	bigger, 2 - 100 μm long
Nuclear Structure	None Single DNA molecule	well define nucleus Several chromosomes
Internal Structures	none, other than storage	many, membrane bound

Prokaryotic Bacteria

- Two Prokaryotic kingdoms
- Archaea: includes those bacteria that have traits typically associated with harsh environments

  examples included halophyles, thermophyles, methanogens,
- Bacteria: includes a variety of bacteria including most "typical" groups

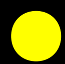
Bacteria and Archaea

- prokaryotic bacteria (Bacteria and Archaea) are nutritionally diverse
- assimilate soluble substrates which are either soluble initially or have been solubilized by exocellular enzymes
- live in anaerobic and aerobic environments
- key component of the decomposers
- disease

Prokaryotic Organisms Classified by Metabolic requirements

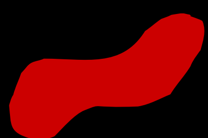
- autotrophic (CO_2) and heterotrophic (organics) for cell carbon
- chemotrophic (chemical)and phototrophic (light) for energy
  - oxidize inorganics for energy (chemolithotrophs)
  - oxidize organics for energy (chemoorganotrophs)

Prokaryotic Organisms Also Classified by Shape

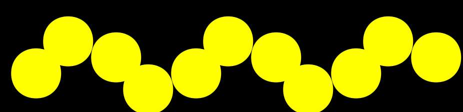


Coccus

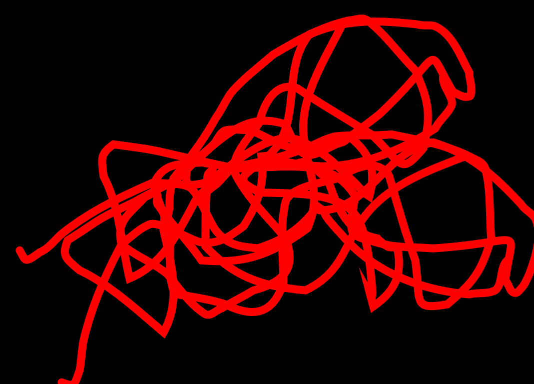
Rod



Spirilum



Spirochete



Filamentous

Algae

- microscopic and macroscopic
- microscopic (single cell/filamentous)
- most are obligate photoautotrophs
- characterized by:
  - nature of chlorophylls
  - carbon reserves or storage
  - motility
  - cell wall structure

Protozoa

- unicellular Eukaryotic organism which lack cell wall
- chemoorganoheterotrophs
- typically fulfill nutritional needs by grazing
- Grazing on bacteria is an important process in producing clear effluents in biological treatment plants

Protozoa Continued

- often parasitic
  - *Giardia*
  - *Cryptosporidium*
- often motile, means of motility is used to classify
  - flagella
  - cilia

Fungi

- lack chlorophyll
- are chemoorganoheterotrophs
- most are obligates aerobes
- structure often characterized by long filaments called hyphae
- grow well under low nutrient and acidic conditions

Fungi, Continued

- because they grow well under adverse conditions and form filaments, they are often problematic in wastewater treatment plants where settling is important
- play major role in nutrient cycling in soil and aquatic environments

Biological Structure and Function

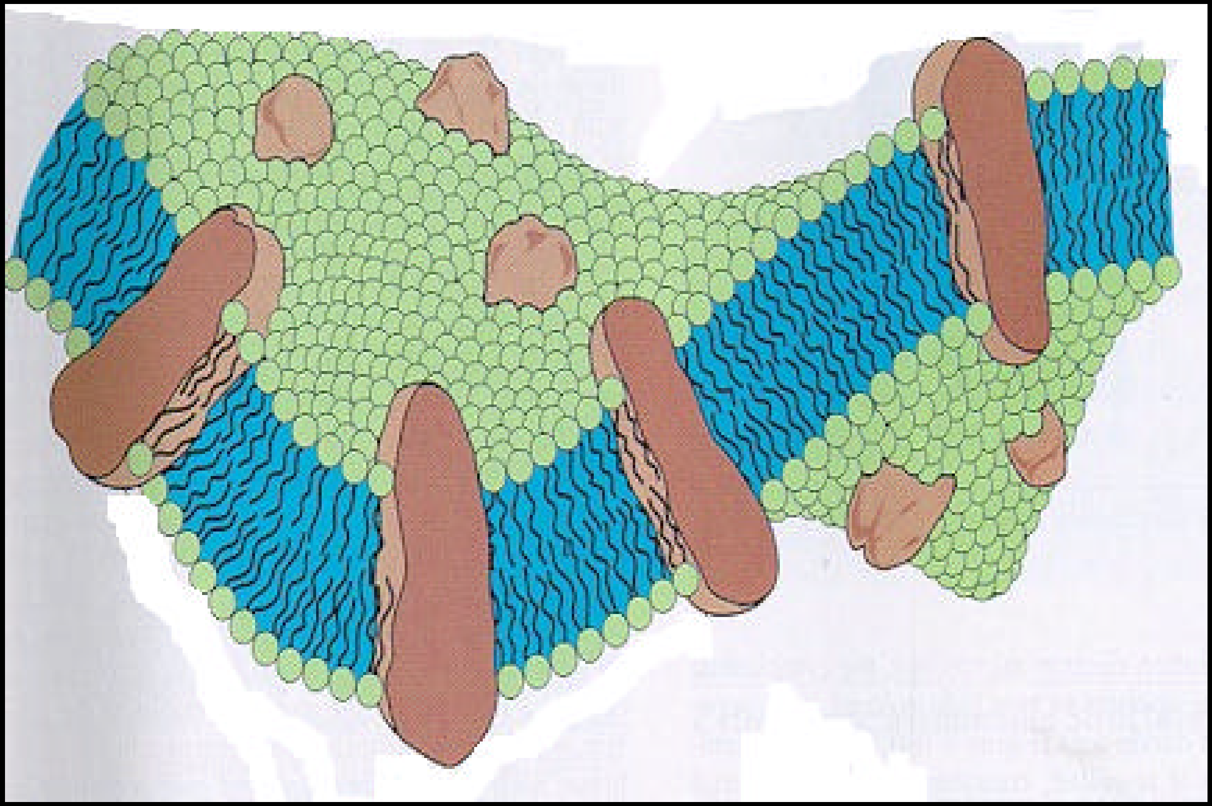
All cells need:

- capture and excretion of nutrients and wasteproducts
- protection from environment
- metabolic conversion of nutrients
- preservation and replication of genetic information

Capture of Nutrients: Cytoplasmic Membrane

- Thin structure that completely encloses cell
- Selective to regulate nutrient and waste flow
- Phospholipid bilayer structure
  - hydrophilic phosphoric head
  - hydrophobic lipid tail
  - hydrophobic interactions give stability to membrane

Phospholipid Bilayer of Membrane



Protection from the Environment: Cell Wall

- Structural Protection
- In Eukarya, cell wall constructed of
  - cellulose (fungi, algae, plants)
  - chitin (fungi)
  - silica (diatoms)
  - polysaccharides (yeasts)
- Prokaryotic and Archaea (different amounts of peptidoglycan
  - Gram + versus Gram -

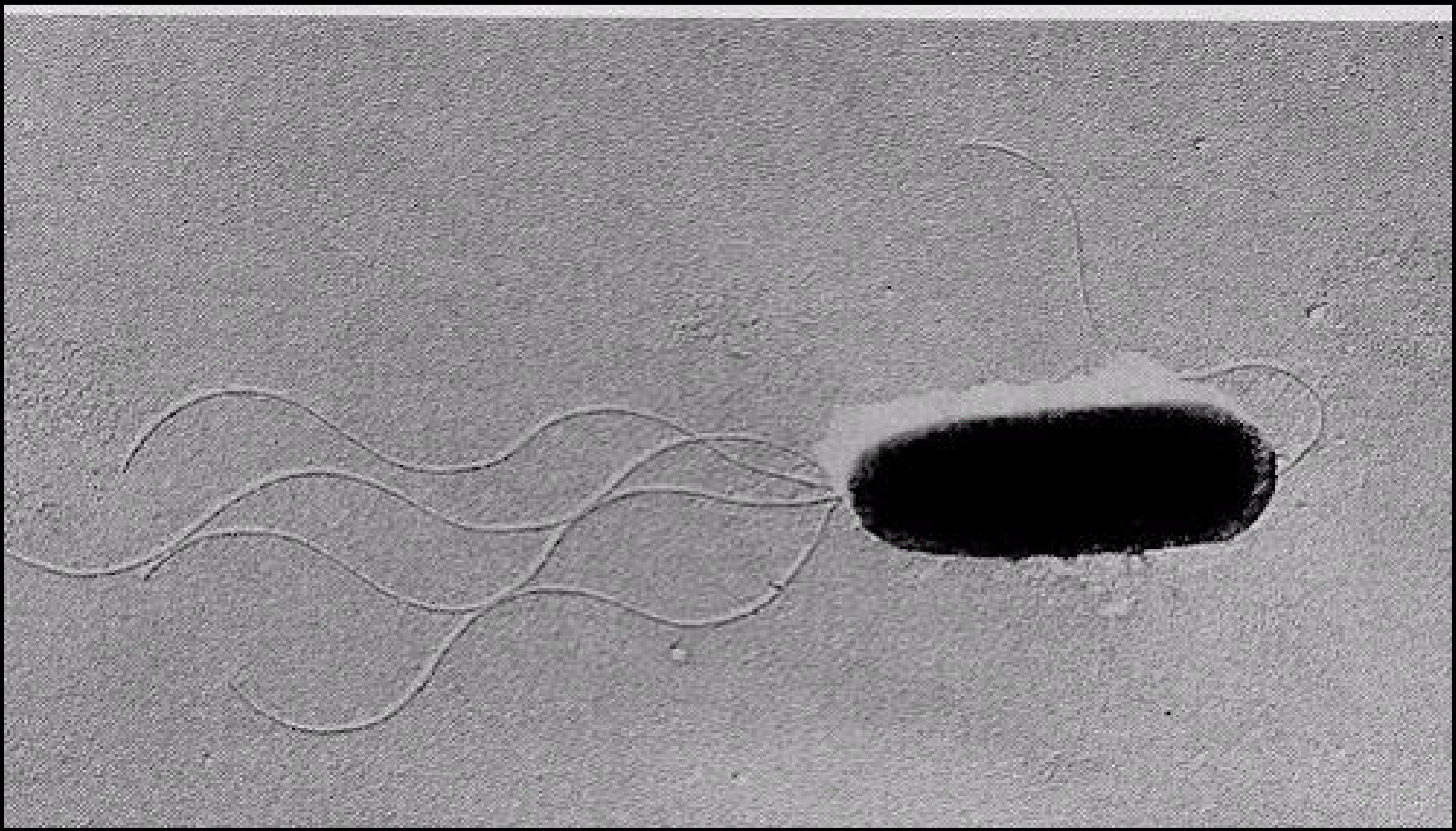
Additional Cell Structures Related to Protection

- Flagella
- Cilia
- Gylcocalyx (capsule or slime layer)
- Fimbriae

Flagella

- Flagella provide means to move towards or away from chemicals (chemotaxis), light (phototaxis), or oxygen (aerotaxis)
- From an ecological view, chemotaxis provides a competitive advantage in environments
- Organisms can have a single polar flagella (montrichous), a tuft of flagella (lophotrichous), or many (peritrichous)

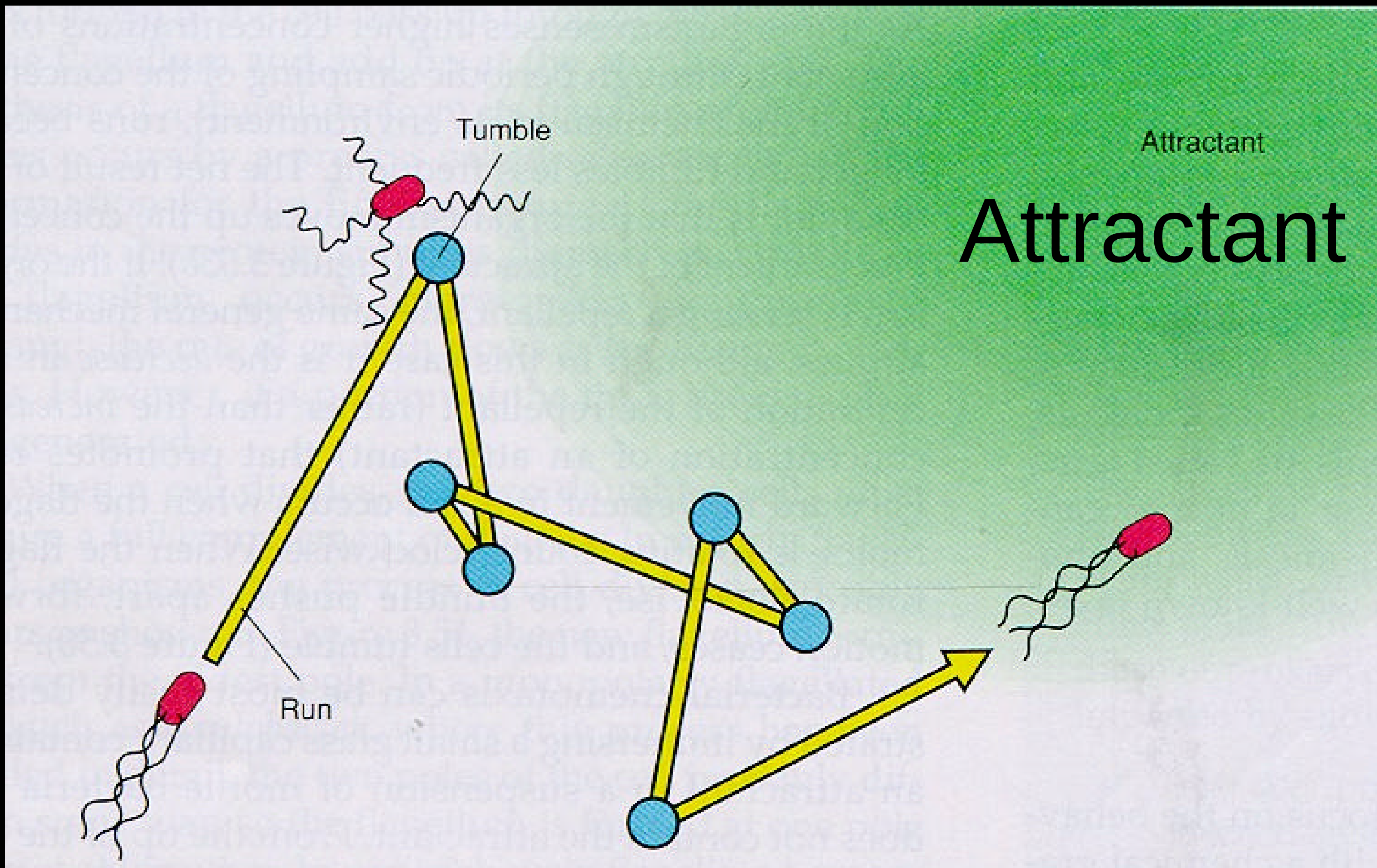
Flagella



Chemotaxis

- Chemotaxis consists of runs (nearly straight) and tumbles (random redirection)
- Runs are longer when bacteria move in favorable direction
- Response based on relative change, not absolute concentration

Chemotaxis



Other Movement Strategies

- Eukaryotic cells also move by cilia
- Cilia are shorter and more numerous than flagella
- Paramecia move by cilia
- Amoebae move by cytoplasmic streaming (amoeboid movement)

Survival in Low Nutrient Conditions

- Organisms growth in low nutrient waters is limited by supply of nutrients
- Rather than move around to capture nutrients, organisms in these environments fix themselves in place and let nutrients come to them
- Thus attachment mechanisms are important

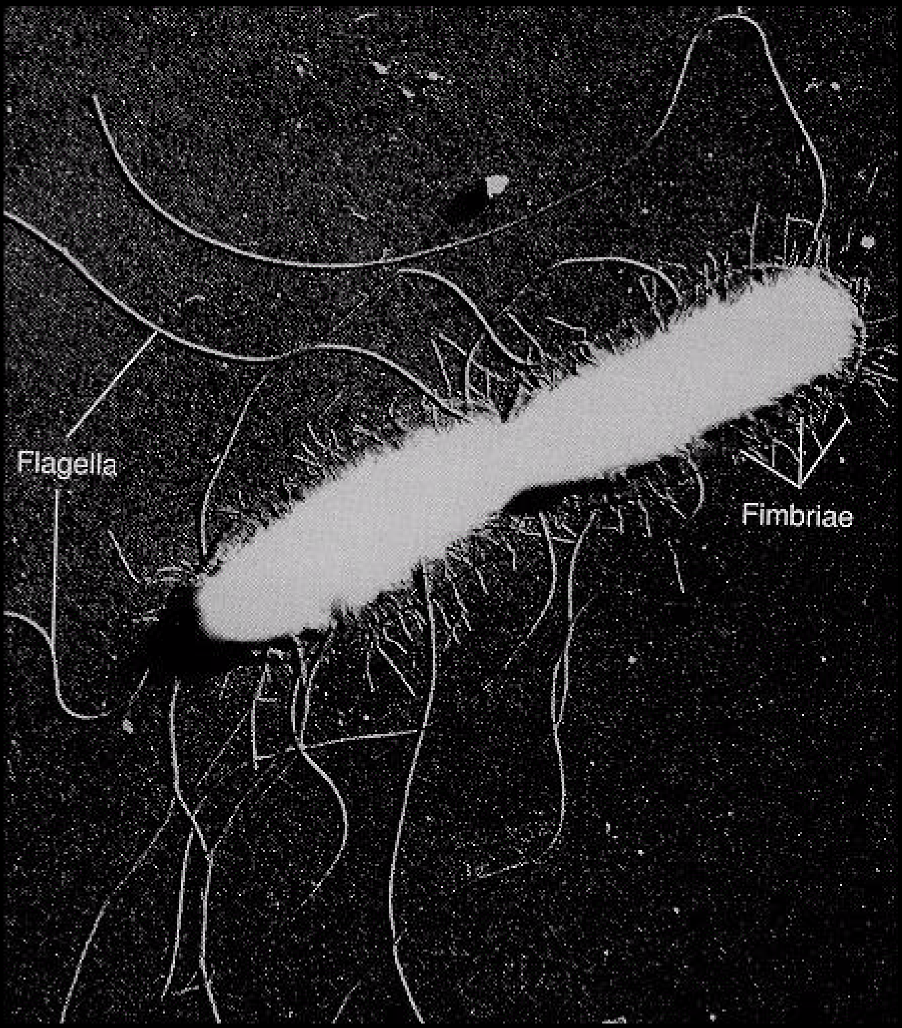
Gylcocalyx

- capsule or slime layer is comprised of a polysaccharide and protein matrix

Gylcocalyx Function

- Attachment to surfaces
- Protection from desiccation
- Microbial flocculation
- Metal complexation
- Protection from phagocytosis
- Pathogen virulence

Other External Appendages



- Fimbriae: attachment mechanisms

Eukaryotic Organelles for Metabolism

- Mitochondria: site of cellular respiration, contains enzymes for aerobic energy production
- Chloroplasts: large organelles for energy production in photosynthetic organisms

Microbial Replication

- For growth to occur, DNA must be replicated before cell division
- As reported, DNA is a double stranded macromolecule consisting of a sugar-phosphate backbone and purine or pyrimidine bases
- The double strands are linked by hydrogen bonding between base (T-A) and (G-C)

DNA Structure (Simple)

T T T G T T A A T G A G C A T C T T

A A A C A A T T A C T C G T A G A A

- Base pair hydrogen bonding between adenine and thymine
- Base pair hydrogen bonding between guanine and cytosine

DNA Replication (Simple)

TTTGTTAATGAGCATCTT
AAACAATTACTCGTAGAA

TTTGTTAATCAGCATCTT
AAACAATTACTCGTAGAA

TTTGTTAATCAGCATCTT
AAACAATTACTCGTAGAA

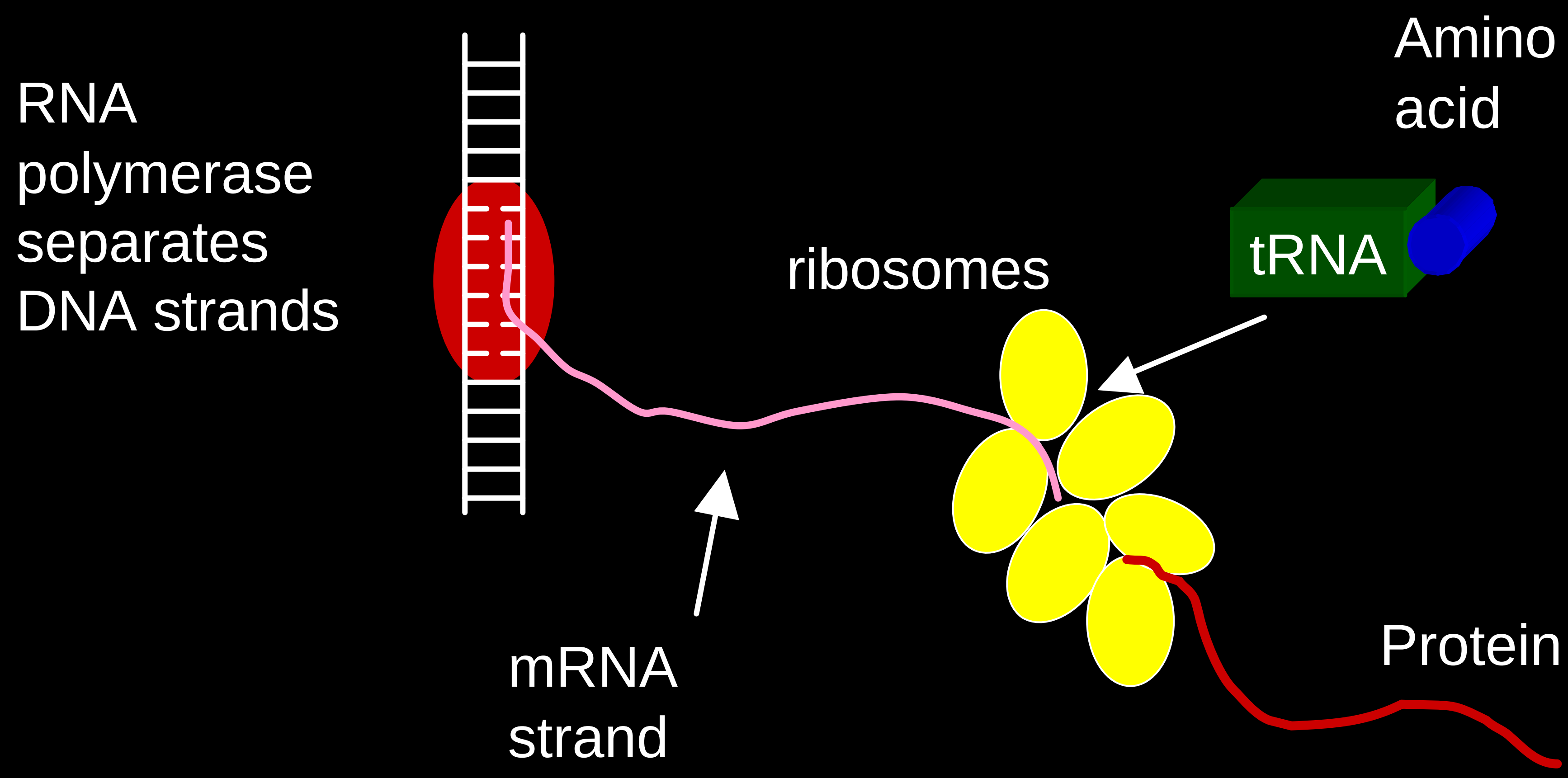
Protein Synthesis

- All information needed for protein synthesis is located on DNA
- However, this information can not be used directly
- Ribonucleic acid (RNA) is used as an intermediate to take information from DNA to make proteins
- The RNA used for this transcription is called messenger RNA (mRNA)

Translation in Protein Synthesis

- The specific sequence of amino acid in each protein is directed by the specific sequences of purine or pyrimidine bases in mRNA
- Proteins are synthesized by translating the mRNA base sequence in a system consisting of ribosomes, transfer RNA (tRNA), and a number of enzymes.
- The translation of each amino acid requires three bases (codon) in mRNA

Diagram of Translation

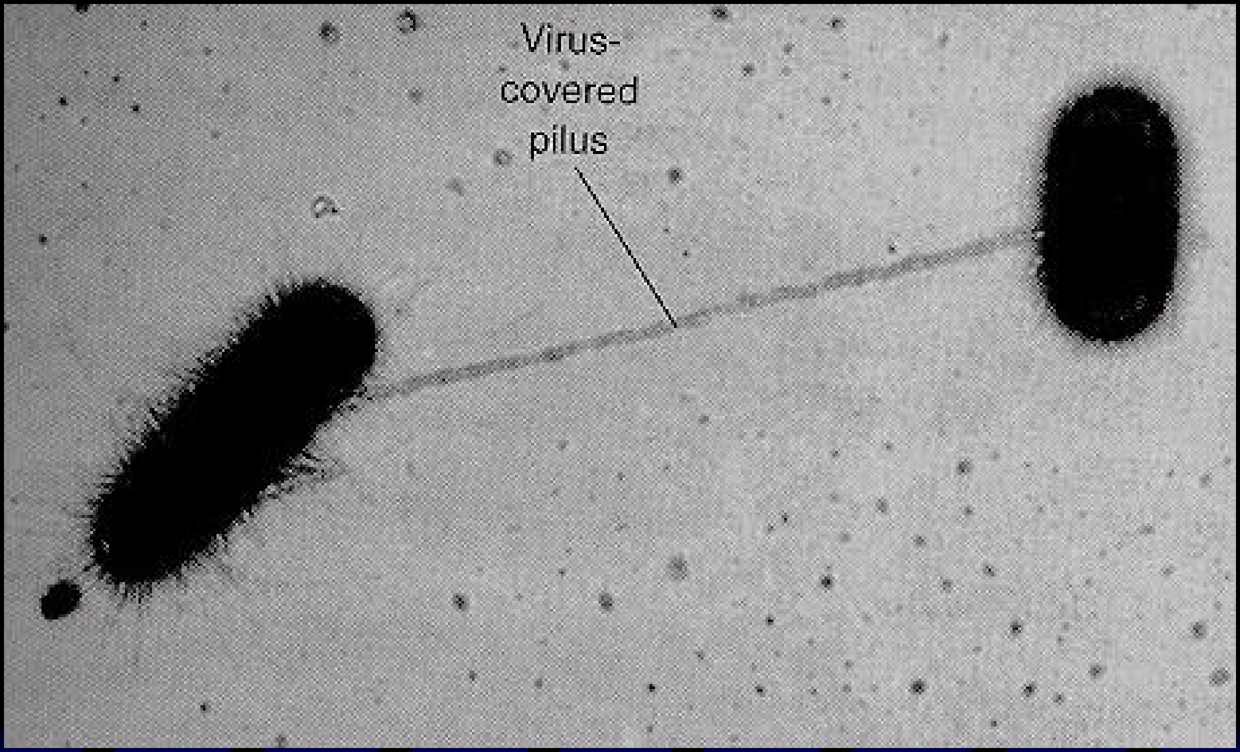


Prokaryotic Genetic Material

- Single circular strand of DNA supercoiled to fit in cell
- Plasmid**:** extrachromosomal DNA, smaller units of non-essential DNA
  - Conjugative plasmids (DNA exchange)
  - Resistance plasmids (antibiotics, metals)
  - Catabolic plasmids (degradation of unusual, non-essential substrates, PAHs, PCBs, chlorophenols, etc.

Plasmid Transfer from Cell to Cell

- important in virus reception and DNA transfer (conjugation: transfer through cell to cell contact)



Eukaryotic Genetic Material

- Eukaryotic cells have a distinct nucleus surrounded by a nuclear membrane which has very small pores to allow the exchange of material between the nucleus and cytoplasm.
- DNA is present as multiple chromosomes
- Nucleolus: an area rich in RNA, site of ribosomal RNA synthesis

Other Eukaryotic Organelles

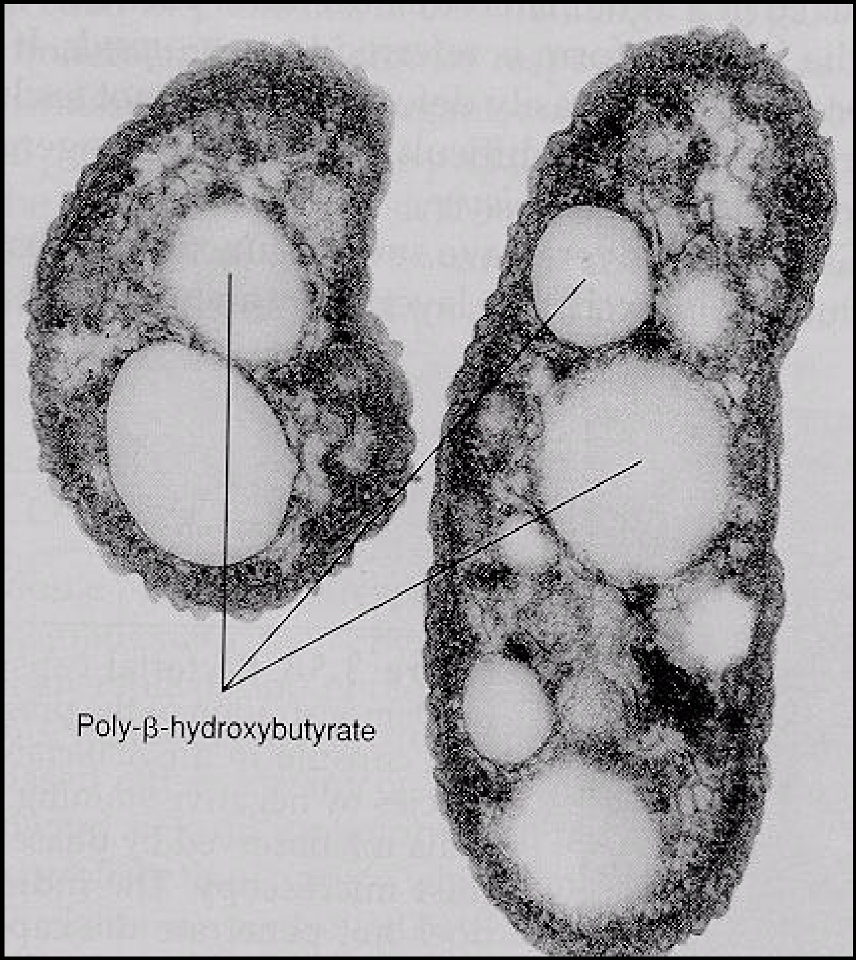
- In Eukaryotic organisms the locations of mRNA and protein synthesis are separated by the nuclear membrane characteristic of Eukaryotic organisms
- Endoplasmic Reticulum: folded membrane system which forms channels through cytoplasm. Attached to both cytoplasmic membrane and nuclear membrane. Houses ribosomes for protein synthesis.

Storage Products in Cells

- Carbon storage polymers
- Phosphate storage
- Sulfur storage

Carbon Storage

- Carbon storage as glycogen, starch, polyhydroxybutyric acid (PHB)
- PHB is very important in the biological removal of phosphorus



Phosphorus and Sulfur Storage

- Polyphosphate granules, storage of energy increased phosphorus uptake over stoichiometric needs
- Sulfur granules, elemental sulfur used as an energy source in sulfur filamentous bacteria

Gas Vacuoles

- Gas vacuoles found in prokaryotic organisms, both Bacteria and Archaea
- Cyanobacteria and other photosynthetic bacteria float because of gas vacuoles and form massive blooms at water surface.
- Allows photosynthetic organisms to "float" to optimal light intensity

Endospores

- Form inside bacteria cells
- Physical and chemical agents trigger spore formation
- Spores are very resistant to heat, chemicals, desiccation, very difficult to kill