Chapter 2: An Overview of Biological Basics

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Presentation Outline: Lectures 2 and 3

- Diversity of Microorganisms
- Naming and Taxonomy of Cells
- Procaryotes
- Eucaryotes
- Viruses
- Cell Structure - major classes of compounds
- Culture Media Components

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Diversity of Microorganism Environmental Conditions

Temperature

i. Grows best below 20˚C	*Phychrophiles*
ii. Grows best between 20 and 50˚C	*Mesophiles*
iii. Grows best above 50˚C	*Thermophiles*

pH

i. Grows best near neutral pH	
ii. Grows well at pH of 1 to 2	*Acidophiles*
iii. Grows well at pH as high as 9	

Diversity of Microorganism Environmental Conditions

Moisture

i. Most cells require a minimum moisture content

ii. Some cells grow in the near absence of moisture

Salinity

i. Most cells require a moderate level of salinity

ii. Some cells can exist in very high salt concentrations

Diversity of Microorganism
Environmental Conditions

Oxygen Availability

i. Require oxygen for growth	*Aerobic*
ii. Require lack of oxygen for growth	*Anaerobic*
iii. Aerobic or anaerobic	*Facultative*

Nutrient Availability

i. Most microorganisms require organic and inorganic nutrients to grow and survive

ii. *Cyanobacteria* grow in the absence of key nutrients: e.g. can convert CO_2 from air into organic cellular molecules.

Diversity of Microorganism
Cell Morphology

Size and Shape

i. Spherical or Elliptical	*Coccus*
ii. Cylindrical	*Bacillus*
iii. Spiral	*Spirillum*

Naming of Cells (Nomenclature)

Cell Names are in Latin

Genus Name: *Escherichia*
Species Name: *coli*

→ *E. coli*

Taxonomy of Cells

Table 2.2

Group	Cell Structure	Properties	Constituent groups
Eucaryotes	Eucaryotic	Multicellular; extensive differentiation of cells and tissues	Plants (seeds plants, ferns, mosses) Animals (vertebrates, invertibrates)
		Unicellular; coenocytic or mycelial; little or no tissue differentiation	Protists (algae, fungi, protozoa)
Procaryotes	Procaryotic	Cell chemistry similar to Eucaryotes	Most bacteria
	Procaryotic	Distinctive cell chemistry	Methanogens, halophiles, thermoacidophiles.

Archaebacteria

Taxonomy of Cells

Table 2.1

Characteristics	Procaryotes	Eucaryotes
Genome		
No. of DNA molecules	One	More than one
DNA in organelles	No	Yes
DNA observed as chromosomes	No	Yes
Nuclear membrane	No	Yes
Mitotoc and meiotic division of nucleus	No	Yes
Formation of partial diploid	Yes	No
Organelles		
Mitochondria	No	Yes
Endoplasmic reticulum	No	Yes
Golgi apparatus	No	Yes
Photosynthetic apparatus	Chlorosomes	Chloroplasts
Flagella	Single protein, simple structure	Complex structure with microtubules
Spores	Endospores	Endo- and exo-spores
Heat resistance	High	Low

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Procaryotes: Gram Negative Cells

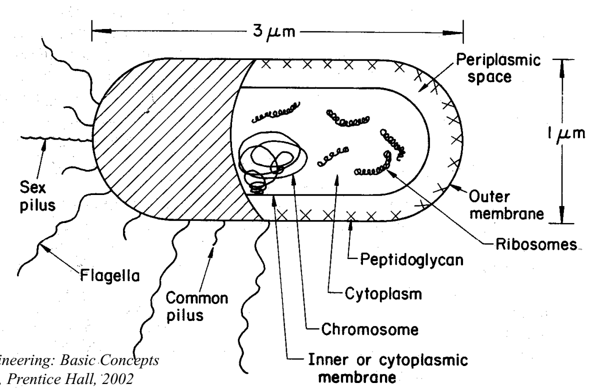

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Figure 2.2. Schematic of a typical gram-negative bacterium (*E. coli*). A gram-positive cell would be similar except that it would have no outer membrane, its peptidoglycan layer would be thicker, and the chemical composition of the cell wall would differ significantly from the outer envelope of the gram-negative cell.

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Gram Negative Cells

A. Cell Envelope

Outer membrane: 10 - 20 nm thick, a protein-polysaccharide-lipid complex

Inner membrane: 5-10 nm thick, 50% protein - 30% lipid - 20% carbohydrate

Pariplasmic space: space between membranes

Flagellum: 10-20 nm thick hair-like structures, provides mobility

Gram Negative Cells

B. Cytoplasm

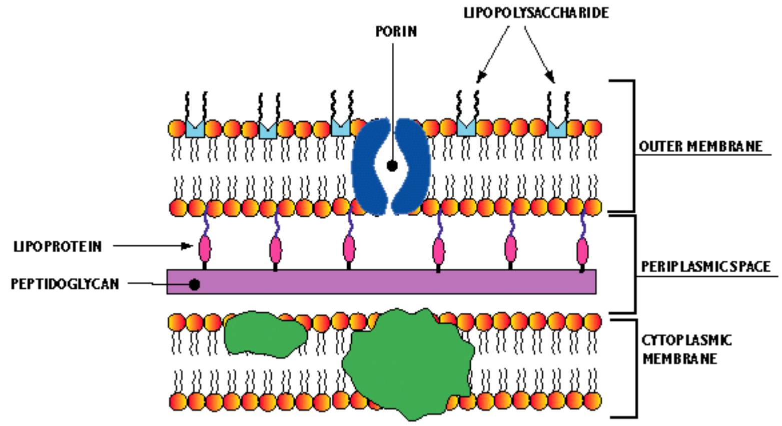
Nuclear material: a single chromosome of DNA with no nuclear membrane.

Ribosomes: sites of protein synthesis. Cells contain about 10,000 of them. Size is about 10 - 20 nm. 63% RNA and 37% protein.

Storage granules: storage of key metabolites. 0.5-1 μm each.

Spores: used by cell to survive harsh conditions of high heat, dryness, and antibiotic agents. One spore formed per cell.

Gram Negative versus Gram Positive Cells



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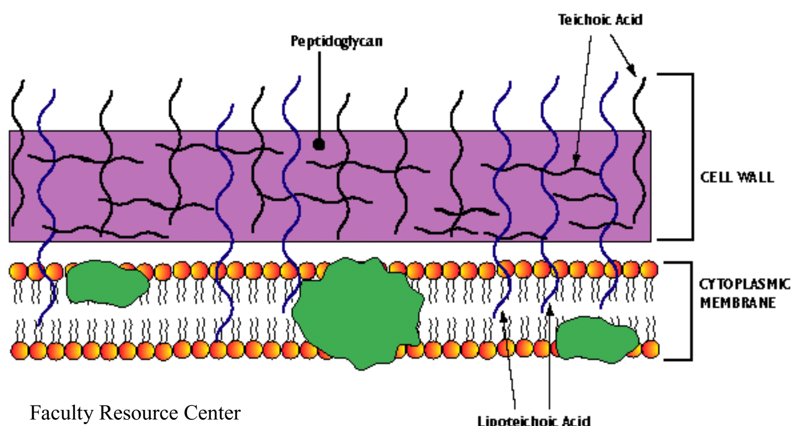
Gram Negative versus Gram Positive Cells



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Eucaryotes

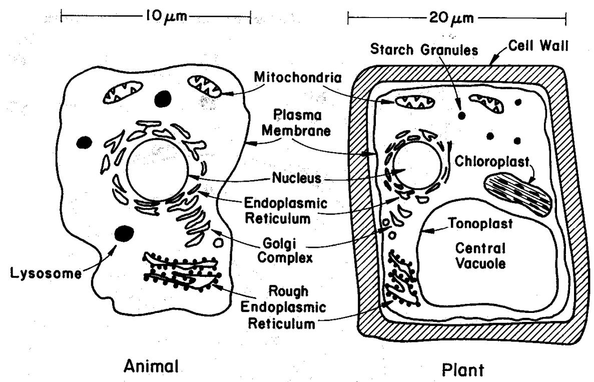

Figure 2.3. Sketches of the two primary types of higher eucaryotic cells. Neither sketch is complete, but each summarizes the principal differences and similarities of such cells.

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Eucaryotes

A. Cell Envelope - provides rigidity

Cell wall: animal cells have no cell wall (fragile). Plant cells have a wall containing peptidoglycan, polysaccharides and cellulose.

Plasma membrane: phospholipid bilayer structure with imbedded proteins similar to procaryotes. Major difference is the presence of sterols, which impart rigidity.

Eucaryotes

B. Cytoplasm

Nucleus: chromosomes surrounded by a membrane.

Mitochondria: 1-3 μm cylindrical bodies. The powerhouses of the cell where *respiration* and *oxidative phosphorylation* occur.

Endoplasmic reticulum: Membrane complex extending from cell membrane, sites of protein synthesis and modification.

Lysosomes: Small membrane-bound particles that contain digestive enzymes.

Eucaryotes

B. Cytoplasm (continued)

Glogi bodies: small particles composed of membrane aggregates responsible for excretion of proteins and other products.

Vacuoles: membrane bound organelles of plant cells responsible for nutrient digestion, osmotic regulation, and waste storage.

Chloroplasts: chlorophyl-containing structures that are responsible for photosynthesis in plants and algae.

Fungi

Yeasts

5-10 µm in size; spherical, cylindrical or oval in shape. Most common yeast used in industry is *Saccharomyces cerevisiae*, which is used for baker's yeast production under aerobic conditions and for alcohol production under anaerobic conditions.

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Fungi

Molds

Filamentous fungi with a mycellial structure. Mycelium is a highly branched system of tubes that contain mobile cytoplasm with many nuclei. Molds are used for production of citric acid and many antibiotics.

See Figure 2.7 of the text.

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Algae

Single or multi-celled organisms that contain chloroplasts and perform photosynthesis. Size is 10 - 30 μm. Diatoms contain silica in their cell walls and are used as filter aids in industry. Some algae are used in the wastewater treatment industry with simultaneous production of single-cell protein. Certain gelling agents such as agar and alginic acid are obtained from marine algae and seaweed.

Protozoa

Are unicellular, motile, relatively large (1-50 μm) eucaryotes that lack a cell wall. They cause a number of human diseases (malaria, dysentery) but may have beneficial roles in removing bacteria from wastewater.

Viruses

Obligate Parasites - not considered life

Structural Components of Viruses

i. Genetic material: DNA (DNA viruses) or RNA (RNA viruses)

ii. Capsid: a protein coat over the genetic material

iii. Outer envelope: some contain a lipoprotein outer envelope

Types of Viruses

i. Bacteriophage: virus that infects a bacteria

ii. Plants: tobacco mosaic virus

iii. Humans: polio virus, SARS virus

Viruses

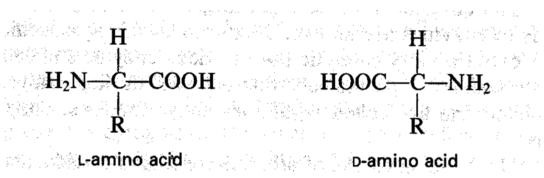
Beneficial Uses of Viruses

Viruses are used as vectors for the insertion of desired DNA into a host cell in genetic engineering.

Lecture 3: Cell Construction

Amino Acids

Amino acids are the building blocks (monomers) of proteins and enzymes. Amino acids have acidic (COOH) and basic (NH_2) groups. Both groups can exchange protons (H+) and alter the charge as a function of pH. This pH - charge behavior allows for their separation using a column apparatus.



The L-isomer is the most common form. The D-isomer (switch COOH and NH_2 groups) is rare.

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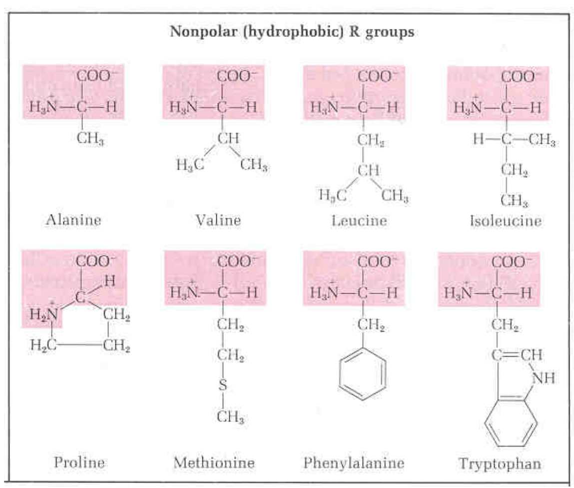
There are 21 Amino Acids: Nonpolar R Groups



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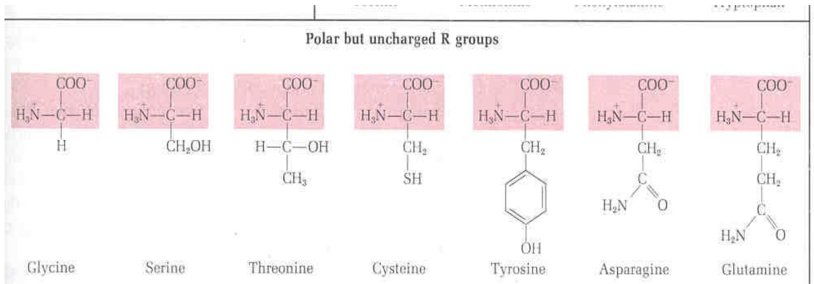
Amino Acids:
Polar R Groups



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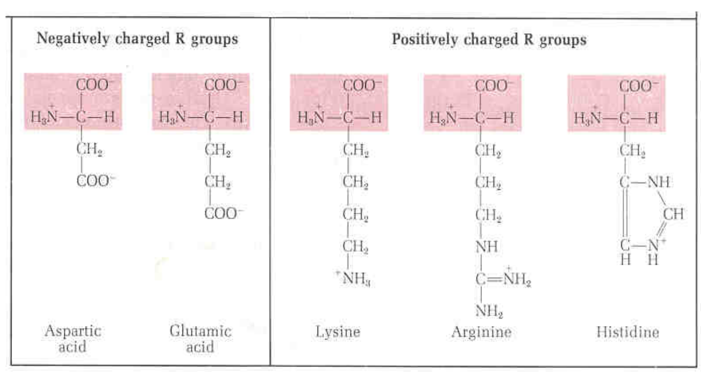
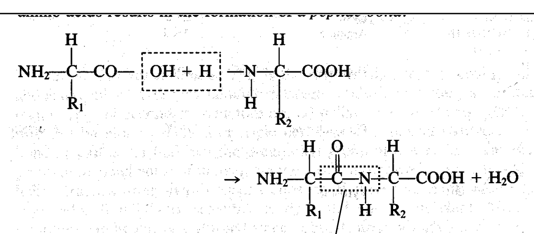
Amino Acids:
Charged R Groups



The pH at which there is no net charge on the molecule is termed the *Isoelectric Point*. *"Principles of Biochemistry", Lehninger, Worth*

Proteins

Proteins are biopolymers composed of numerous amino acid units, created through enzyme-mediated <u>condensation reactions</u> forming a *peptide bond*.



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Peptide Bond

Proteins: Prosthetic Groups

In addition to amino acids, proteins can also contain organic or inorganic components termed *prosthetic groups*. For example, <u>hemoglobin</u> contains four heme groups (iron-containing organometallic complex). Prosthetic groups can impart catalytic activity to proteins. Proteins containing prosthetic groups are termed *congugated proteins*.

Structure of Proteins

Primary Structure:

The linear sequence of amino acids.

Each protein has a unique sequence of amino acids.

Example - Ribonuclease containing 124 amino acids

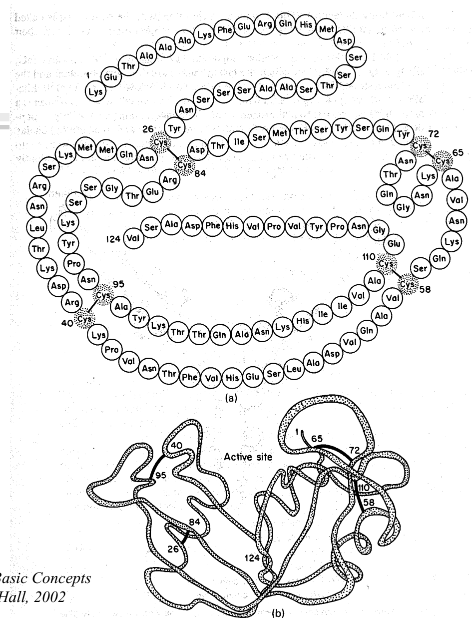

Figure 2.12. Structure of the enzyme ribonuclease. (a) Primary amino acid se-

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Secondary Structure of Proteins

The way the molecule is extended. *Hydrogen bonding* between adjacent R groups that are not widely separated

α-helix

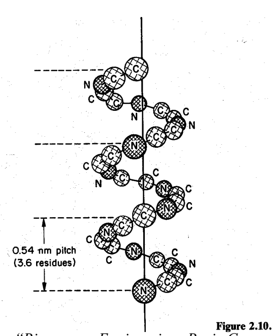
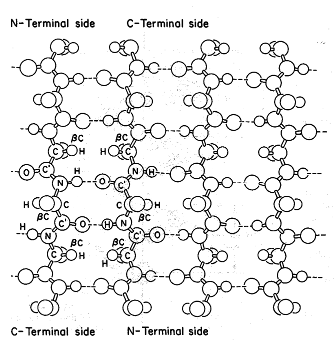


Figure 2.10.

β-sheet



...re 2.11. Representation of an antiparallel β-pleated sheet. Dashed lines indi-

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Tertiary and Quaternary Structure of Proteins

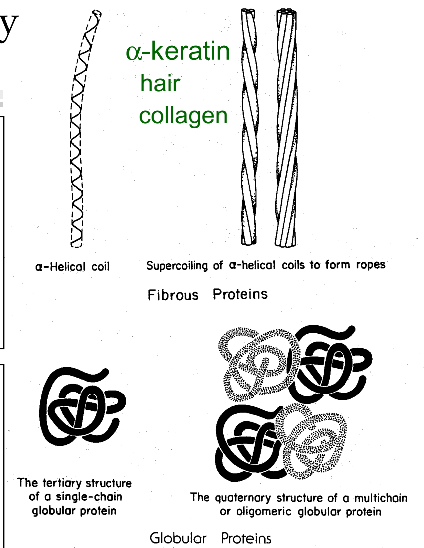

Figure 2.9. Fibrous and globular proteins. (With permission from A. Lehninger, *Biochemistry*, 2nd ed., Worth Publishing, New York, 1975, p. 61.)

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Tertiary Structure

3-dimensional form of the protein. Interactions of R groups far apart on the chain (hydrogen, covalent, and disulfide bonding, hydrophobic/hydrophillic).

Quaternary Structure

Assembly of multiple polypeptide chains. Some enzymes (catalytic proteins) have this structure.

Antibodies (Immunoglobulins)

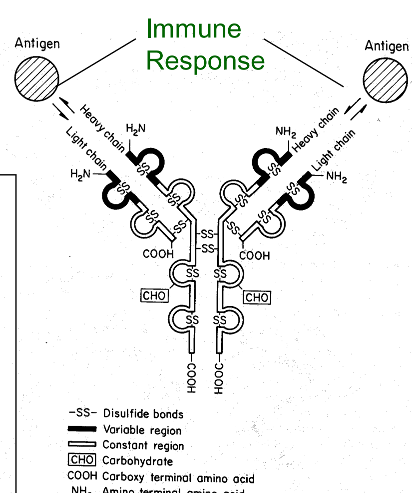

Figure 2.13. Structure of immunoglobulin G (IgG). Structure showing disulfide

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Antibodies

Are proteins that bind to foreign macromolecules (antigens) with high specificity (immune response).

Industrial Applications

- diagnostic kits
- protein separation
- drug delivery

Proteins (Continued)

The mass of proteins is often given in terms of *Daltons*, where one dalton is equal in mass to one hydrogen atom. For example, a protein having a molecular mass of 150 kilodaltons has a molecular weight of (150,000)(1.00797)=151,195.5 grams / mole of protein.

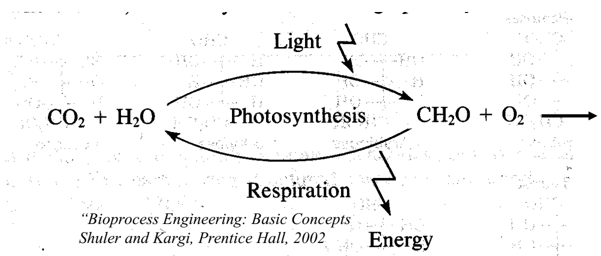
Proteins (even more!)

Protein Function

1. Structural proteins (collagen, keratin, glycoproteins)
2. Catalytic proteins (enzymes)
3. Transport proteins (hemoglobin, serum albumin)
4. Regulatory proteins hormones (insulin, growth hormone)
5. Protective proteins (antibodies, thrombin)

Carbohydrates

Carbohydrates are compounds containing *carbon*, *oxygen*, and *hydrogen*. They are created by photosynthesis primarily from plants by fixation of CO_2 using energy from sunlight. Their general formula is $(CH_2O)_n$ where n 3.



Monosaccharides, polysaccharides

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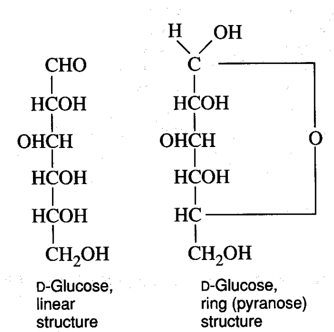
Monosaccharides

The smallest carbohydrates with carbon numbers between 3 and 9. They are organized into two groups; Aldoses and Ketoses. The difference is the location of the carbonyl group (C=O); on the terminal carbon or next to the terminal carbon.

Molecular structure



D-refers to optical properties

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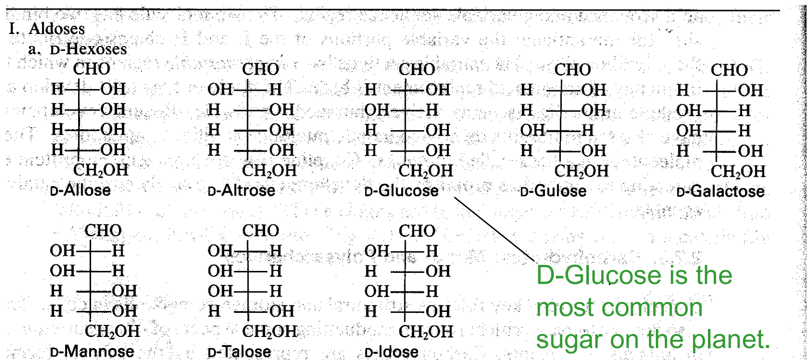
Aldoses - D-Hexoses



D-Glucose is the most common sugar on the planet. A key component of woody biomass and starch.

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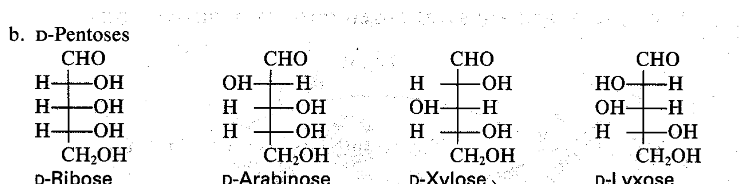
Aldoses - D-Pentoses



D-Ribose is a key component of DNA and RNA

D-Xylose is a key component of woody biomass

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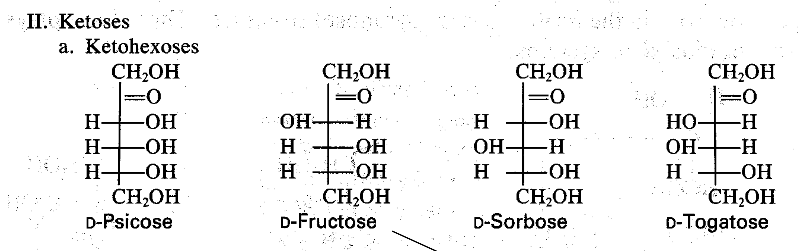
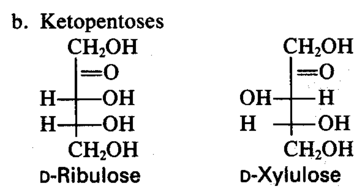
Ketoses - D-Hexoses and D-Pentoses





D-Fructose is an important sweetener

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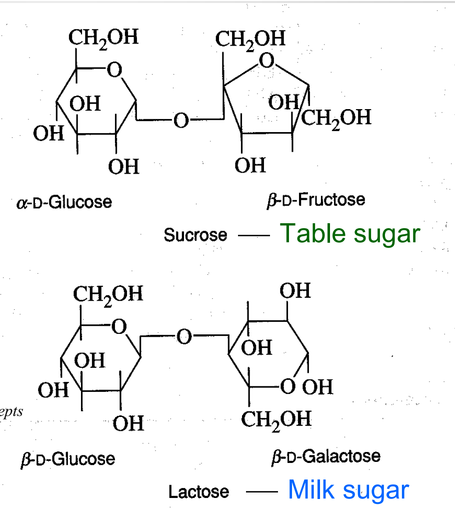
Disaccharides

Disaccharides are formed by condensation reactions between two monosaccharides.



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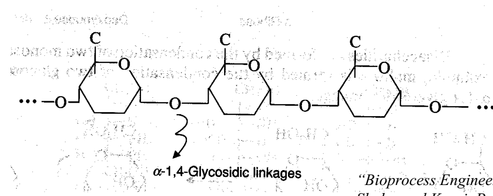
Polysaccharides

Polysaccharides are formed by condensation reactions between two or more mono-saccharides by *glycosidic bonds*.



Amylose - a polymer of glucose molecules

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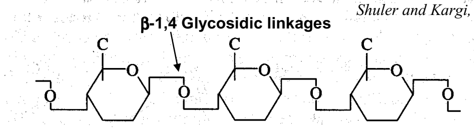
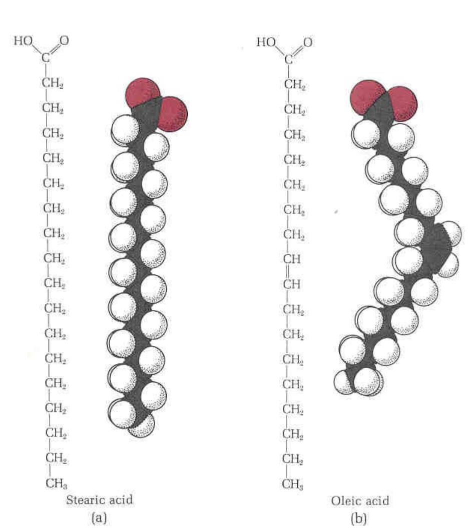


Cellulose - a polymer of glucose molecules

Fatty Acids

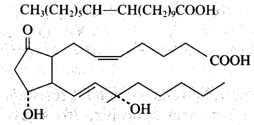
Fatty Acids are composed of a hydrocarbon *tail* and hydrophobic *head*. They are major compo-nents of cell membranes.



"Principles of Biochemistry", Lehninger, Worth

Fatty Acids

TABLE 2.6 Examples of Common Fatty Acids

Acid	Structure
Saturated fatty acids	
Acetic acid	CH_3COOH
Propionic acid	CH_3CH_2COOH
Butyric acid	$CH_3(CH_2)_2COOH$
Caproic acid	$CH_3(CH_2)_4COOH$
Decanoic acid	$CH_3(CH_2)_8COOH$
Lauric acid	$CH_3(CH_2)_{10}COOH$
Myristic acid	$CH_3(CH_2)_{12}COOH$
Palmitic acid	$CH_3(CH_2)_{14}COOH$
Stearic acid	$CH_3(CH_2)_{16}COOH$
Arachidic acid	$CH_3(CH_2)_{18}COOH$
Behenic acid	$CH_3(CH_2)_{20}COOH$
Lignoceric acid	$CH_3(CH_2)_{22}COOH$
Monoenoic fatty acids	
Oleic acid	$CH_3(CH_2)_7CH\overset{cis}{=}CH(CH_2)_7COOH$
Dienoic fatty acid	
Linoleic acid	$CH_3(CH_2)_4(CH\overset{cis}{=}CHCH_2)_2(CH_2)_6COOH$
Trienoic fatty acids	
α-Linolenic acid	$CH_3CH_2(CH\overset{cis}{=}CHCH_2)_3(CH_2)_6COOH$
γ-Linolenic acid	$CH_3(CH_2)_4(CH\overset{cis}{=}CHCH_2)_3(CH_2)_3COOH$
Tetraenoic fatty acid	
Arachidonic acid	$CH_3(CH_2)_4(CH\overset{cis}{=}CHCH_2)_4(CH_2)_2COOH$
Unusual fatty acids	
Tariric acid	$CH_3(CH_2)_{10}C\equiv C(CH_2)_4COOH$
Lactobacillic acid	$CH_3(CH_2)_5\overset{CH_2}{\overbrace{CH—CH}}(CH_2)_9COOH$
Prostaglandin (PGE_2)	

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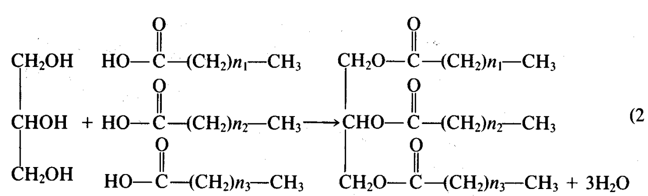
Fats

Fats are esters of fatty acids with *glycerol*.
Phosphoglycerides ,found in membranes are similar to fats, but contain only two fatty acids and a phosphate esterified to one of the glycerol carbons.



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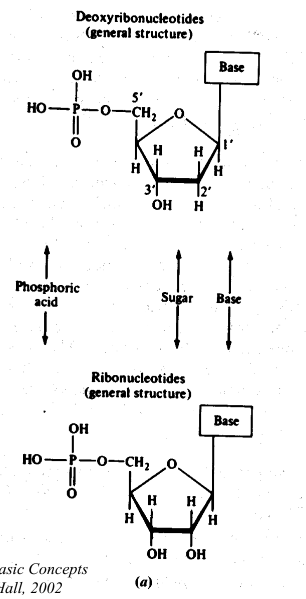
Nucleic Acids (DNA and RNA)

Nucleotides are the building blocks of DNA and RNA. They are composed of a sugar (ribose or deoxyribose), phosphate, and a base.



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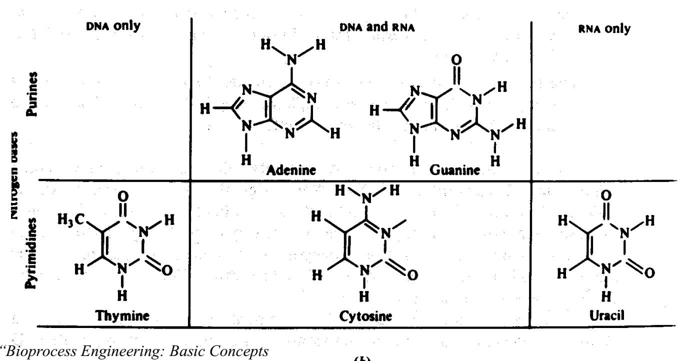
Bases Found in Nucleotides

Bases are various nitrogen containing compounds, as shown by these molecules for DNA and RNA.



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David Figure 2.15. (a) General structure of ribonucleotides and deoxyribonucleotides. (b) Five

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Structure of DNA and RNA

DNA and RNA are polymers composed of nucleotides linked by phosphodiester bonds between the 3' and 5' carbons on adjacent sugar units.

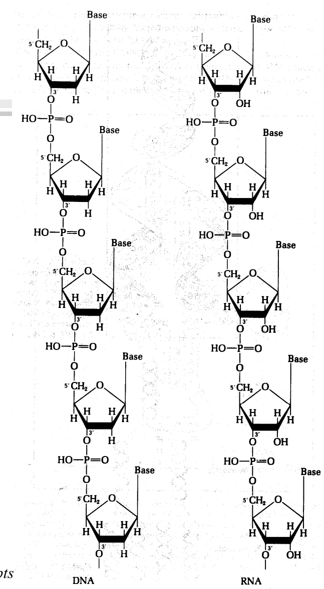

Figure 2.16. Structure of DNA and RNA chains. Phosphodiester bonds are

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Double Helix structure of DNA

DNA is always found in complimentary strands in a double helix having A matched with T and G matched with C. On *RNA*, U replaces T.

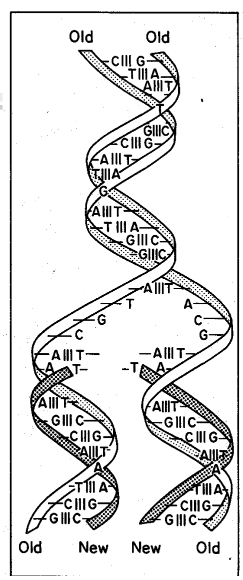

Figure 2.17. of DNA, sho replication by pairing. (Witl Brock, D. W gan, *Biology* ed., Prentice NJ, 1984, p

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Plasmids

Plasmids are nonchromosomal, autonomous, self replicating double-stranded DNA found in some microorganisms. They are easily moved into and out of cells and are useful in genetic engineering. In nature, they encode for factors (proteins) that protect the cell from antibiotics or other harmful chemicals. In genetically engineered cells, the plasmids code for some desired protein product.

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Cell Composition

TABLE 2.7 Chemical Analyses, Dry Weights, and the Populations of Different Microorganisms Obtained in Culture

Organism	Composition (% dry weight) Protein	Nucleic acid	Lipid	Typical population in culture (numbers/ml)	Typical dry weight of this culture (g/100 ml)	Comments
Viruses	50–90	5–50	<1	10^8–10^9	0.0005[a]	Viruses with a lipoprotein sheath may contain 25% lipid.
Bacteria	40–70	13–34	10–15	2×10^8–2×10^{11}	0.02–2.9	PHB content may reach 90%
Filamentous fungi	10–25	1–3	2–7		3–5	Some *Aspergillus* and *Penicillium* sp. contain 50% lipid.
Yeast	40–50	4–10	1–6	1–4×10^8	1–5	Some *Rhodotorula* and *Candida* sp. contain 50% lipid.
Small unicellular algae	10–60 (50)	1–5 (3)	4–80 (10)	4–8×10^7	0.4–0.9	Figure in ( ) is a commonly found value but the composition varies with the growth conditions.

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Cell Culture Macronutrient Elements

TABLE 2.9 The Eight Macronutrient Elements and Some Physiological Functions and Growth Requirements

Element	Physiological function	Required concentration (mol l^{-1})
Carbon	Constituent of organic cellular material. Often the energy source.	$>10^{-2}$
Nitrogen	Constituent of proteins, nucleic acids, and coenzymes.	10^{-3}
Hydrogen	Organic cellular material and water.	—
Oxygen	Organic cellular material and water. Required for aerobic respiration.	—
Sulfur	Constituent of proteins and certain coenzymes.	10^{-4}
Phosphorus	Constituent of nucleic acids, phospholipids, nucleotides, and certain coenzymes.	10^{-4} to 10^{-3}
Potassium	Principal inorganic cation in the cell and cofactor for some enzymes.	10^{-4} to 10^{-3}
Magnesium	Cofactor for many enzymes and chlorophylls (photosynthetic microbes) and present in cell walls and membranes.	10^{-4} to 10^{-3}

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Cell Culture Media

TABLE 2.10 Compositions of Typical Defined and Complex Media

Defined medium		
Constituent	**Purpose**	**Concn (g/liter)**
Group A		
Glucose	C, energy	30
KH_2PO_4	K, P	1.5
$MgSO_4 \cdot 7H_2O$	Mg, S	0.6
$CaCl_2$	Ca	0.05
$Fe_2(SO_4)_3$	Fe	15×10^{-4}
$ZnSO_4 \cdot 7H_2O$	Zn	6×10^{-4}
$CuSO_4 \cdot 5H_2O$	Cu	6×10^{-4}
$MnSO_4 \cdot H_2O$	Mn	6×10^{-4}
Group B		
$(NH_4)_2HPO_4$	N	6
$(NH_4)H_2PO_4$	N	5
Group C		
$C_6H_5Na_3O_7 \cdot 2H_2O$	Chelator	4
Group D		
Na_2HPO_4	Buffer	20
KH_2PO_4	Buffer	10

Complex medium used in a penicillin fermentation	
Glucose or molasses (by continuous feed)	10% of total
Corn-steep liquor	1–5% of total
Phenylacetic acid (by continuous feed)	0.5–0.8% of total
Lard oil (or vegetable oil) antifoam by continuous addition	0.5% of total
pH to 6.5 to 7.5 by acid or alkali addition	

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