Chapter 5:  
Nucleic Acids, etc.

Voet & Voet: Sections 1 & 3  
Pages 82-84 & 88-93

Any introductory Biochemistry textbook will have an introductory chapter on nucleic acids
Nucleotides and their derivatives participate in nearly all biochemical processes

1. **Monomeric units** of nucleic acids

2. "Energy rich" - Nucleoside triphosphates (ie. ATP) are products of most energy-releasing pathways AND are consumed in energy-requiring processes

3. **Regulators** of metabolic pathways and metabolic processes

4. **Cofactors** - required component of enzymatic reactions (eg. NAD^+, FAD, Coenzyme A)

5. **Catalytic activity** (ie. ribozymes)
Nucleotides are nucleoside phosphates

- **Nucleosides** consist of a nitrogenous base covalently attached via a “β glycosidic bond” to the C1' of a five carbon sugar (pentose)
  - All nucleotides contain a nucleoside
  - Not all nucleosides are nucleotides
Nitrogenous bases are planar, aromatic molecules that are (typically) derivatives of purine or pyrimidine

- Parent compounds for common nucleic acid bases (nucleobases)
**β-D-Ribose**

**Riboses** (or aldopentoses) are five carbon sugars with an aldehyde functional group (linear form only; revisit during energetics)

- Nucleic acids are composed exclusively of the β-D stereoisomer of ribose (or deoxyribose)
- RNA (ribonucleic acid) contains β-D-ribose and DNA (deoxyribonucleic acid) contains β-2'-deoxy-D-ribose
Phosphate

- Phosphate is covalently attached to the D-ribose via phosphate ester bonds
  - Phosphates are typically attached to the C5' (5'-nucleotide)
  - In polymers, the phosphate is attached to both the C5' and C3'
  - Nucleic acids are acidic, polyanions due to the phosphate groups of nucleotides
Nucleic acid polymers have a 5' and 3' end

- Convention: Nucleic Acids are written from 5' to 3'
- Nucleic acids are synthesized from 5'-nucleoside triphosphates in a 5' to 3' direction

Commonly named using a one letter code

eg. DNA tetranucleotide (right)

(a) \(dGdTdAdC\) – assumes 5'-phosphate and 3',5' phosphate link between nucleotides

(b) \(pdGpdTpdpdApdC\) – all phosphates explicitly indicated with 'p'

GUAC represent ribonucleotides
dGdTdAdC represent deoxyribonucleotides

Proper Name

5'-deoxyguanyl-3',5'-deoxythymidyl-3',5'-deoxyadenyl-3',5'-deoxycytidine

(Used for small / uncommon nucleic acid polymers where the explicit location of phosphates is essential)
Nucleic Acid Polymer

- Schematic for (dAdTdCdG)p or d(ApTpCpGp)
- Tetranucleotide deoxyadenyl-3',5'-deoxythymidyl-3',5'-deoxycytidyl-3',5'-deoxyguanyln-3'-phosphate
**Summary**

X refers to:

- H when naming base
- ribose when naming nucleoside
- ribose and phosphate when naming nucleotide

Uracil occurs in RNA and Thymine in DNA

<table>
<thead>
<tr>
<th>Base Formula</th>
<th>Base (X = H)</th>
<th>Nucleoside (X = ribose(^a))</th>
<th>Nucleotide (X = ribose phosphate(^a))</th>
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</thead>
<tbody>
<tr>
<td>NH₂</td>
<td>Adenine</td>
<td>Adenosine</td>
<td>Adenylic acid</td>
</tr>
<tr>
<td></td>
<td>Ade</td>
<td>Ado</td>
<td>Adenosine monophosphate</td>
</tr>
<tr>
<td></td>
<td>A</td>
<td>A</td>
<td>AMP</td>
</tr>
<tr>
<td>H₂N</td>
<td>Guanine</td>
<td>Guanosine</td>
<td>Guanylic acid</td>
</tr>
<tr>
<td></td>
<td>Gua</td>
<td>Guo</td>
<td>Guanosine monophosphate</td>
</tr>
<tr>
<td></td>
<td>G</td>
<td>G</td>
<td>GMP</td>
</tr>
<tr>
<td>NH₂</td>
<td>Cytosine</td>
<td>Cytidine</td>
<td>Cytidylic acid</td>
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<td>Cyd</td>
<td>Cytidine monophosphate</td>
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<tr>
<td></td>
<td>C</td>
<td>C</td>
<td>CMP</td>
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<td>NH</td>
<td>Uracil</td>
<td>Uridine</td>
<td>Uridylic acid</td>
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<td></td>
<td>Ura</td>
<td>Urd</td>
<td>Uracil monophosphate</td>
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<tr>
<td></td>
<td>U</td>
<td>U</td>
<td>UMP</td>
</tr>
<tr>
<td></td>
<td>Thymine</td>
<td>Deoxythymidine</td>
<td>Deoxythymidyllic acid</td>
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<tr>
<td></td>
<td>Thy</td>
<td>dThd</td>
<td>Deoxythymidine monophosphate</td>
</tr>
<tr>
<td></td>
<td>T</td>
<td>dT</td>
<td>dTMP</td>
</tr>
</tbody>
</table>
Modified Bases

• Similar to the case with amino acids, nucleic acid bases also occur as modified forms of the standard bases
  – inevitably modified bases have functional consequences

Modified bases occur in DNA and more commonly in RNA
Base Tautomers

Each base has a tautomer

- T and G tautomers are shown

Tautomeric forms of base display altered hydrogen bonding patterns

- Mechanism that can lead to spontaneous mutation during transcription and replication (simple organisms)
- C and A tautomers are between amine and imine forms (not shown)
Chapter 5 & 29: Deoxyribonucleic Acid.

Voet & Voet:
Chapter 5 - Sections 1 & 3
pages 82-84 & 88-93
Chapter 29 – pages 1145-1153 & 1158-1159
DNA (Watson-Crick)

- Double helix
- Deduced from X-ray diffraction pattern (R. Franklin & M. Wilkins)

Franklin's X-ray diffraction pattern from crystalline DNA
Late 1940's, Chargaff showed

1. DNA has equal amounts of purines and pyrimidines \((G+A = C+T)\)
2. Amount of A = T and amount of G = C

Result is due to complementary base pairing

Indirectly led to discovery of “Double Helix”

- Ultimately led to the discovery of the mechanism of DNA replication

Figures: Deoxyribose C1' is shown as 1' – note that both deoxyribose sugars are on the same side of interacting bases
Denaturation of DNA

Above a characteristic critical temperature, DNA double helix structure is lost (denatures) and the two strands dissociate

- Detectable by ultraviolet spectroscopic measurements
  - significant increase in UV-absorption as dsDNA denatures to ssDNA
- Process is reversible if heat is removed slowly
Denaturation is Cooperative

Denaturation of one region of DNA duplex destabilizes the remainder of the duplex

- This phenomenon is known as a cooperative process and is characterized by a sigmoidal curve

$T_m$ is the melting temperature of DNA and varies linearly with the G+C content of the DNA

Note: For very short DNA species (oligonucleotides) size has significant effect on $T_m$
# Size of DNA

<table>
<thead>
<tr>
<th>Organism</th>
<th>Number of base pairs (kb)</th>
<th>Contour length (μm)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Viruses</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Polyoma, SV40</td>
<td>5.2</td>
<td>1.7</td>
</tr>
<tr>
<td>λ Bacteriophage</td>
<td>48.6</td>
<td>17</td>
</tr>
<tr>
<td>T2, T4, T6 bacteriophage</td>
<td>166</td>
<td>55</td>
</tr>
<tr>
<td>Fowlpox</td>
<td>280</td>
<td>193</td>
</tr>
<tr>
<td><strong>Bacteria</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Mycoplasma hominis</em></td>
<td>760</td>
<td>260</td>
</tr>
<tr>
<td><em>Escherichia coli</em></td>
<td>4,600</td>
<td>1,600</td>
</tr>
<tr>
<td><strong>Eukaryotes</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yeast (in 17 haploid chromosomes)</td>
<td>12,000</td>
<td>4,100</td>
</tr>
<tr>
<td><em>Drosophila</em> (in 4 haploid chromosomes)</td>
<td>180,000</td>
<td>61,000</td>
</tr>
<tr>
<td>Human (in 23 haploid chromosomes)</td>
<td>3,200,000</td>
<td>1,100,000</td>
</tr>
<tr>
<td>Lungfish (in 19 haploid chromosomes)</td>
<td>102,000,000</td>
<td>35,000,000</td>
</tr>
</tbody>
</table>
dsDNA Structures

- **A DNA**
  - grooves are more equal in size
  - larger diameter

- **B DNA**
  - pronounced major groove
  - narrow diameter

- **Z DNA**
  - shallow grooves
  - narrowest diameter

The red line lies within major groove
dsDNA Backbone

- DNA backbone has 6 rotatable bonds or torsion angles
  - 5 are freely rotatable and 1 is a pseudorotatable bond (ribose)
- More theoretical conformers than the polypeptide backbone
- Highly constrained by double helix
  - results in far fewer conformers than even the smallest protein
Ribose Conformations

- Ribose adopts **C2'-endo** or **C3'-endo** in normal dsDNA structures
  - endo ≡ atom out of plane on same side as C5'
- Ribose conformation affects phosphate separation
  - A-DNA (large diameter) adopts a C3'-endo ribose conformation (left) with phosphate groups near to one another
  - B-DNA (narrow diameter) adopts a C2'-endo ribose conformation (right) with phosphate groups relatively farther apart
**Base conformations**

- Conformation of planar base is dependent upon 'glycosidic bond' torsion angle
  - **anti conformation** (favored) places bulky groups away from ribose
    - H over ribose ring
  - **syn conformation** (disfavored) places bulky groups over the ribose
    - 6 member ring (purine) or O (pyrimidine) over ribose ring

![Chemical structures](image)

- syn-Adenosine
- anti-Adenosine
- anti-Cytidine
## dsDNA: Structural Parameter

<table>
<thead>
<tr>
<th></th>
<th>A - DNA</th>
<th>B - DNA</th>
<th>Z - DNA</th>
</tr>
</thead>
<tbody>
<tr>
<td>Helical Sense</td>
<td>right</td>
<td>right</td>
<td>left</td>
</tr>
<tr>
<td>Diameter</td>
<td>2.6 nm</td>
<td>2.0 nm</td>
<td>1.8 nm</td>
</tr>
<tr>
<td>Base pairs / turn</td>
<td>11.6</td>
<td>10</td>
<td>12</td>
</tr>
<tr>
<td>Twist / base pair</td>
<td>31°</td>
<td>36°</td>
<td>60°</td>
</tr>
<tr>
<td>Helix Pitch</td>
<td>3.4 nm</td>
<td>3.4 nm</td>
<td>4.4 nm</td>
</tr>
<tr>
<td>Helix Rise</td>
<td>0.29 nm</td>
<td>0.34 nm</td>
<td>0.74 nm</td>
</tr>
<tr>
<td>Major Groove</td>
<td>Narrow &amp; Deep</td>
<td>Wide &amp; Deep</td>
<td>Flat</td>
</tr>
<tr>
<td>Minor Groove</td>
<td>Wide &amp; Shallow</td>
<td>Narrow &amp; Deep</td>
<td>Narrow &amp; Deep</td>
</tr>
<tr>
<td>Sugar Pucker</td>
<td>C3'-endo</td>
<td>C2'-endo</td>
<td>C2'-endo/C3'-endo</td>
</tr>
<tr>
<td>Glycosidic Bond</td>
<td>Anti</td>
<td>Anti</td>
<td>Anti &amp; Syn</td>
</tr>
</tbody>
</table>

**dsDNA is typically in the B-form under physiological conditions**

Note: Learn material in red boxes.
Other Base Pairs

There are many **non Watson-Crick base pairing** interactions between nucleotides

- occur in unusual dsDNA structures including 'mismatches'

Rare in dsDNA but **common in ssRNA**

- often involve N7 of a purine (eg. Hoogsteen Base Pair – middle structure)
Supercoiling in dsDNA

- **Helix pitch** can vary in the presence of proteins or in closed-circular dsDNA
  - replication, transcription, chromatin formation and gene regulation all require local changes to the helix pitch

  - supercoiling is defined by the *writhe* and *twist*
    - writhe = number of helix coils about self
    - twist = length (bp) / pitch (bp/turn)