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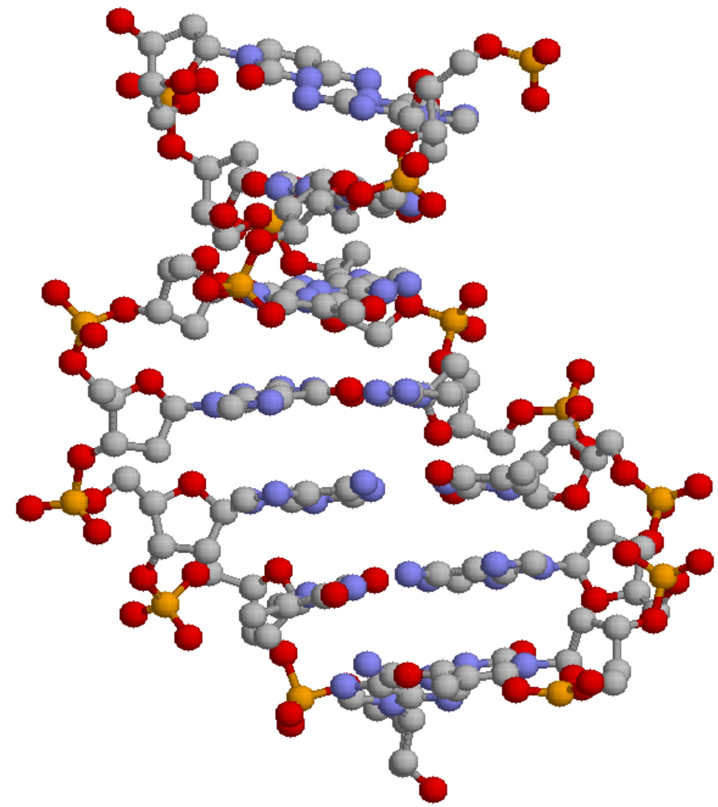


Chapter 5: Nucleic Acids, *etc.*

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

Any introductory Biochemistry textbook will have an introductory chapter on nucleic acids

Nucleotides and Derivatives

Nucleotides and their derivatives participate in nearly all biochemical processes

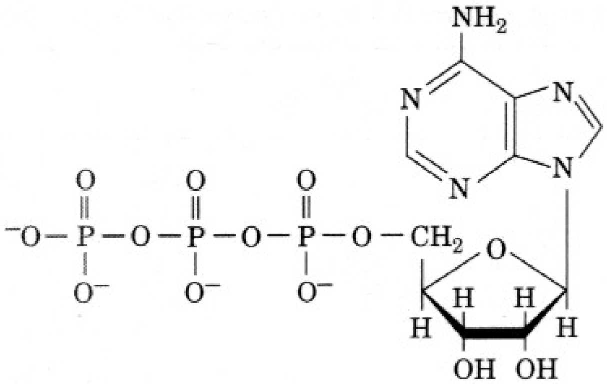
(1) **Monomeric units** of nucleic acids

(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)

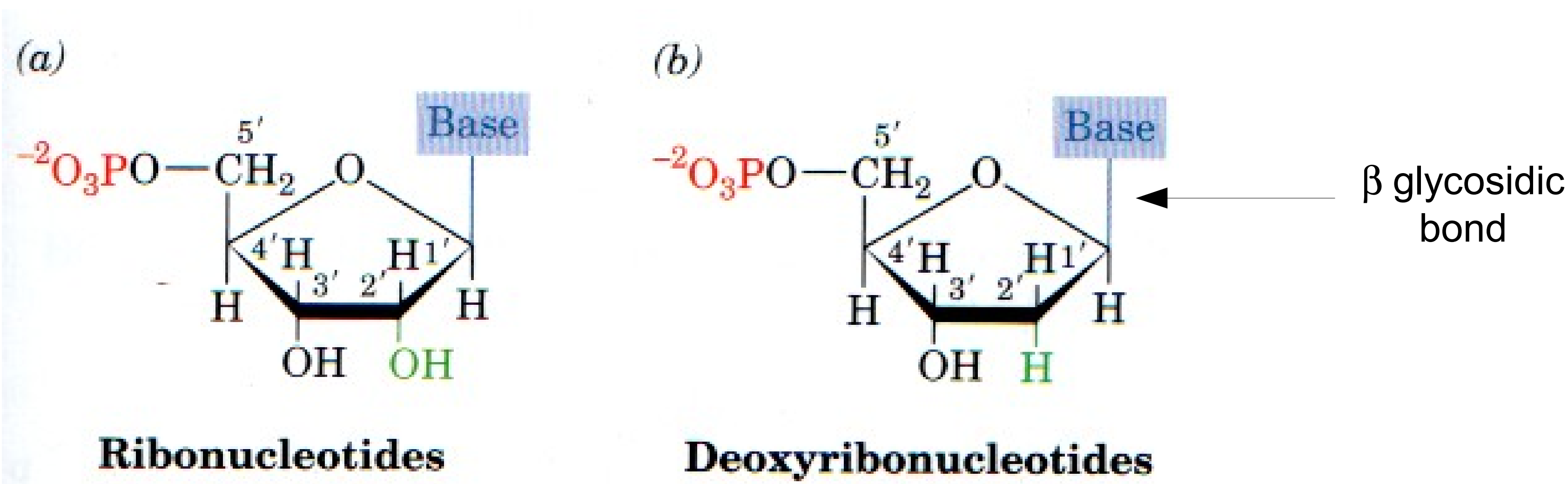


Adenosine triphosphate (ATP)

Nucleic Acids

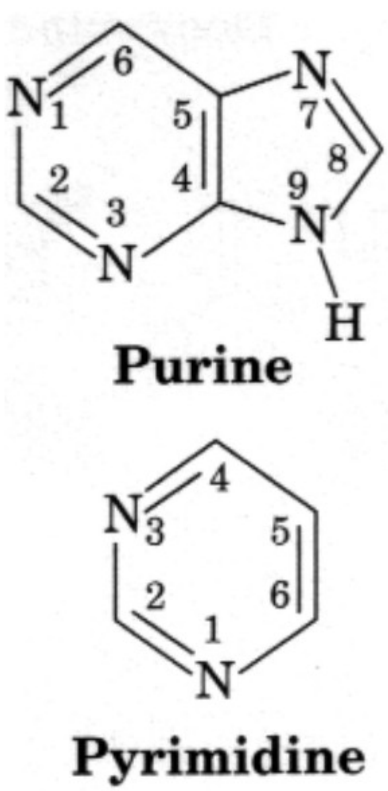
Nucleotides are nucleoside phosphates

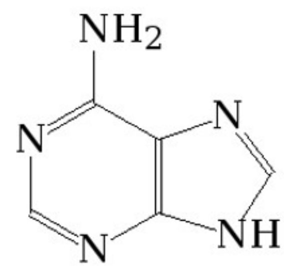
- **Nucleosides consist of a nitrogenous base covalently attached via a "β glycosidic bond" to the C1' of a five carbon sugar (pentose)**
  - **All nucleotides contain a nucleoside**
  - **Not all nucleosides are nucleotides**

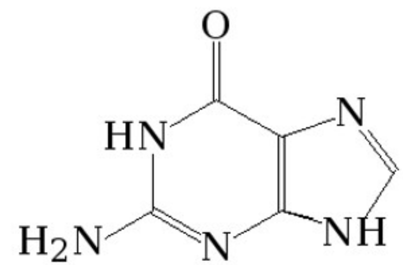


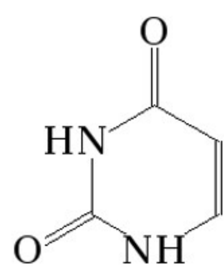
Nitrogenous Base

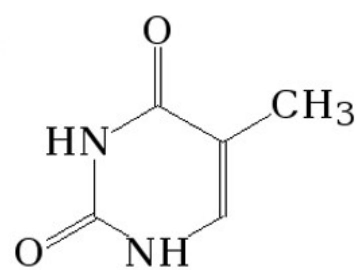
- Nitrogenous bases are planar, aromatic molecules that are (typically) derivatives of purine or pyrimidine
  - Parent compounds for common nucleic acid bases (nucleobases)

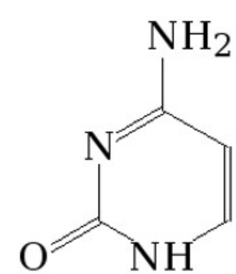



Adenine


Guanine


Uracil

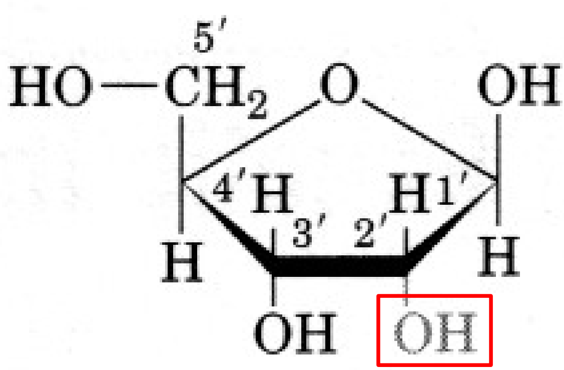

Thymine


Cytosine

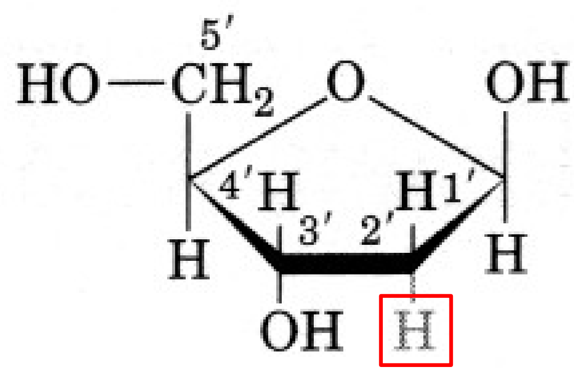
β-D-Ribose

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**



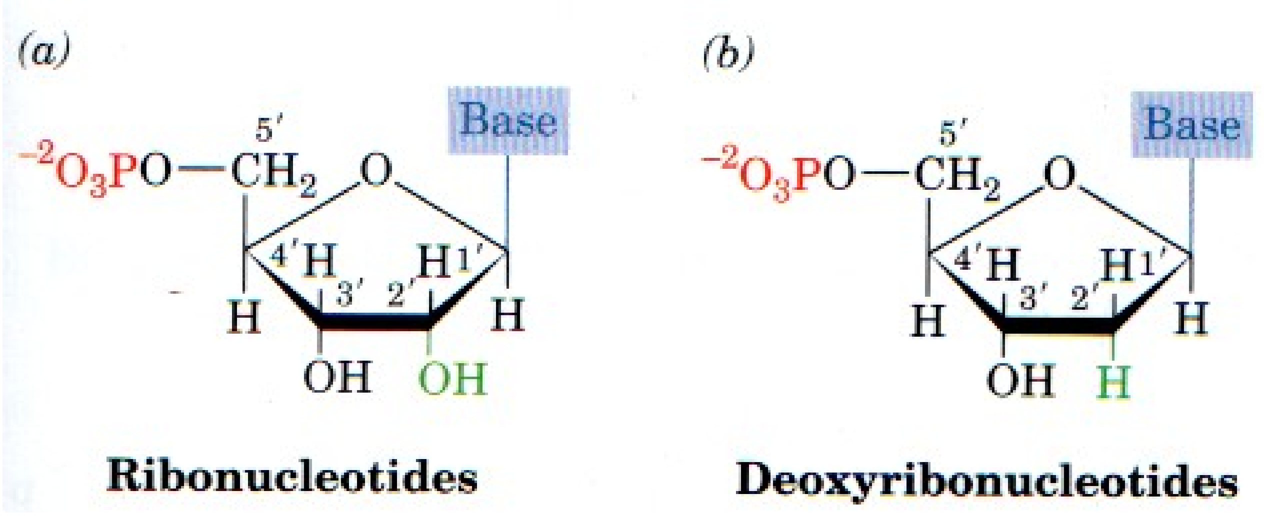
Ribose



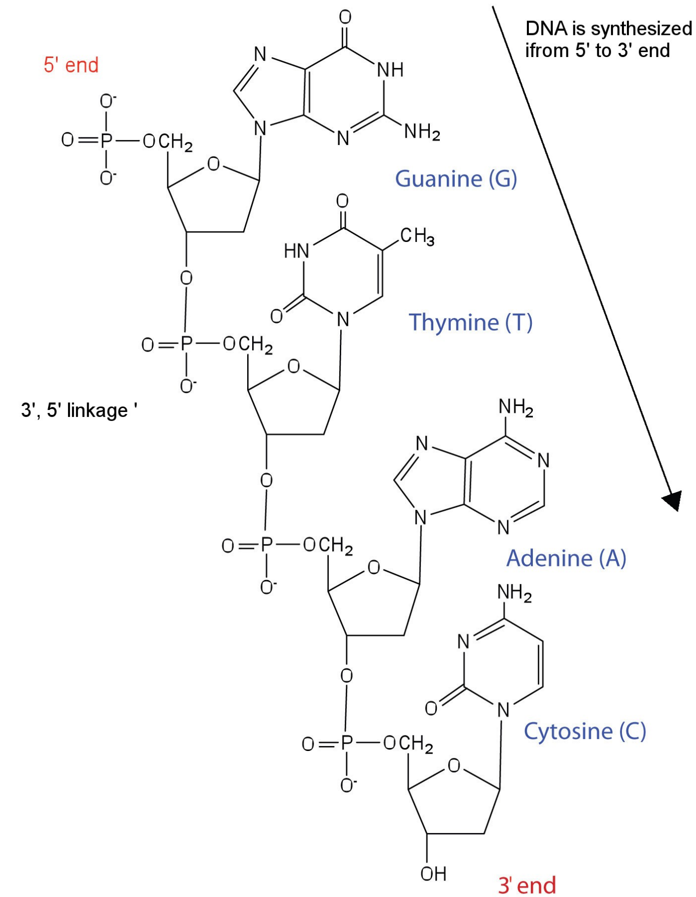
Deoxyribose

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 Polymer



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) pdGpdTpdApdC – all phosphates explicitly indicated with 'p'

GUAC represent ribonucleotides
dGdTdAdC represent deoxyribonucleotides

Proper Name

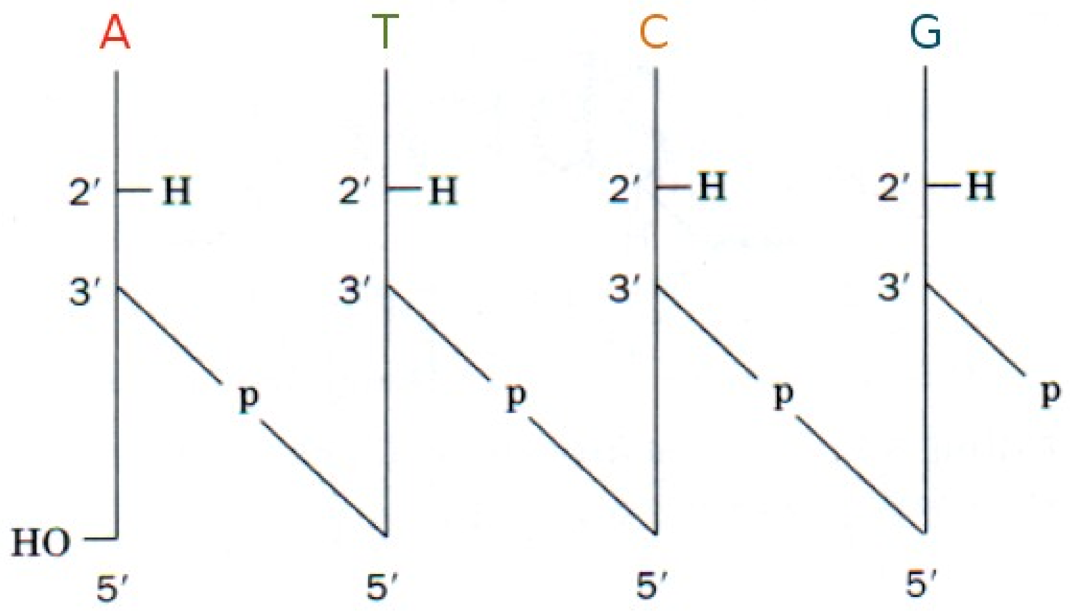
5'-deoxyguanyl-3',5'-deoxythymidyl-3',5'-deoxyadenyl-3',5'-deoxycytidine

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

Note: When it is understood that DNA is being considered, the 'deoxy' prefix (ie. GTAC) is often omitted …

Nucleic Acid Polymer

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



Schematic representation of DNA structure

Summary

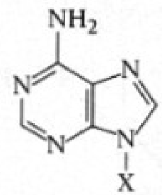
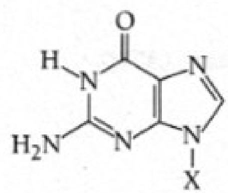
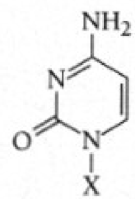
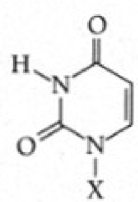
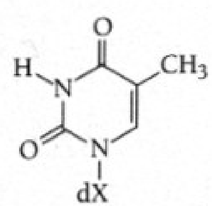
X refers to :

H
when naming base

ribose
when naming nucleoside

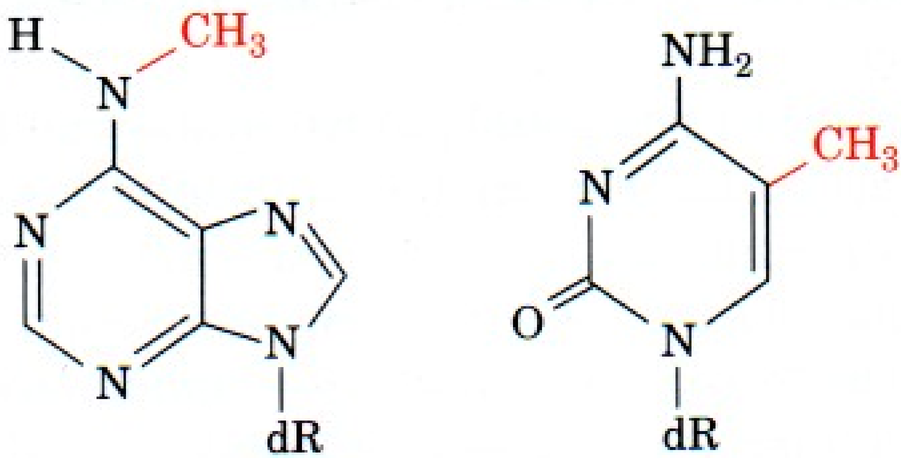
ribose and phospate
when naming nucleotide

Uracil occurs in RNA and Thymine in DNA

Base Formula	Base (X = H)	Nucleoside (X = ribose[a])	Nucleotide[b] (X = ribose phosphate[a])
	Adenine Ade A	Adenosine Ado A	Adenylic acid Adenosine monophosphate AMP
	Guanine Gua G	Guanosine Guo G	Guanylic acid Guanosine monophosphate GMP
	Cytosine Cyt C	Cytidine Cyd C	Cytidylic acid Cytidine monophosphate CMP
	Uracil Ura U	Uridine Urd U	Uridylic acid Uridine monophosphate UMP
	Thymine Thy T	Deoxythymidine dThd dT	Deoxythymidylic acid Deoxythymidine monophosphate dTMP

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



N^6-Methyl-dA   5-Methyl-dC

Modified bases occur in DNA and more commonly in RNA

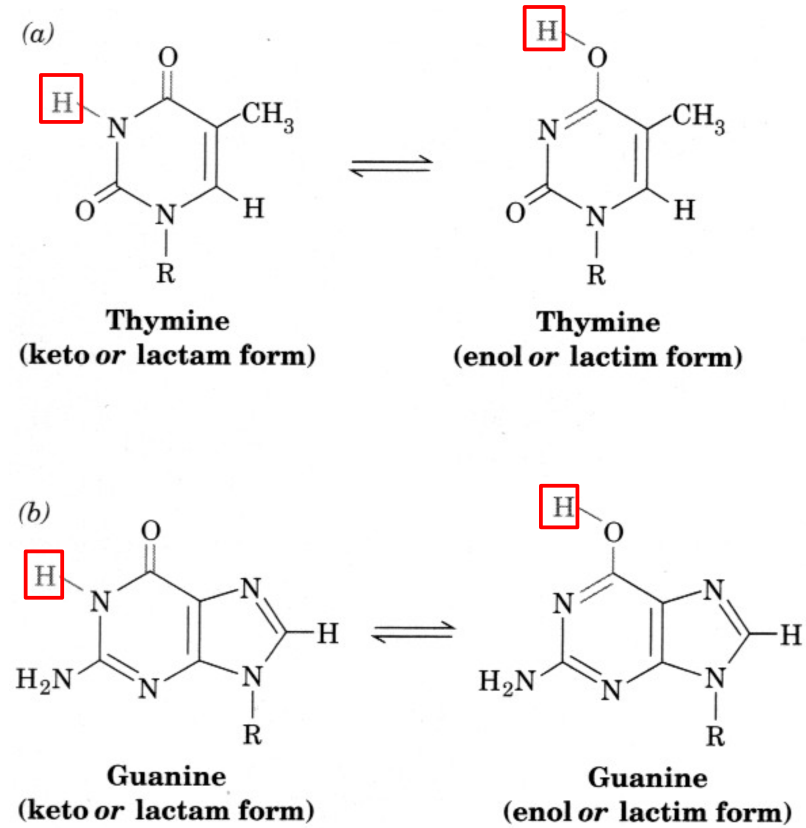
Base Tautomers

Each base has a tautomer

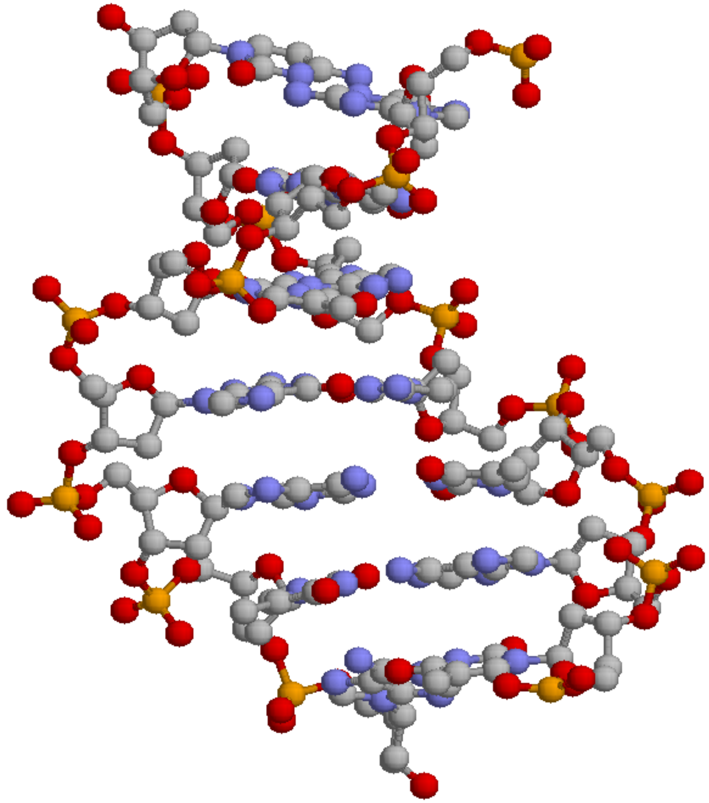
- **T and G tautomers are shown**

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

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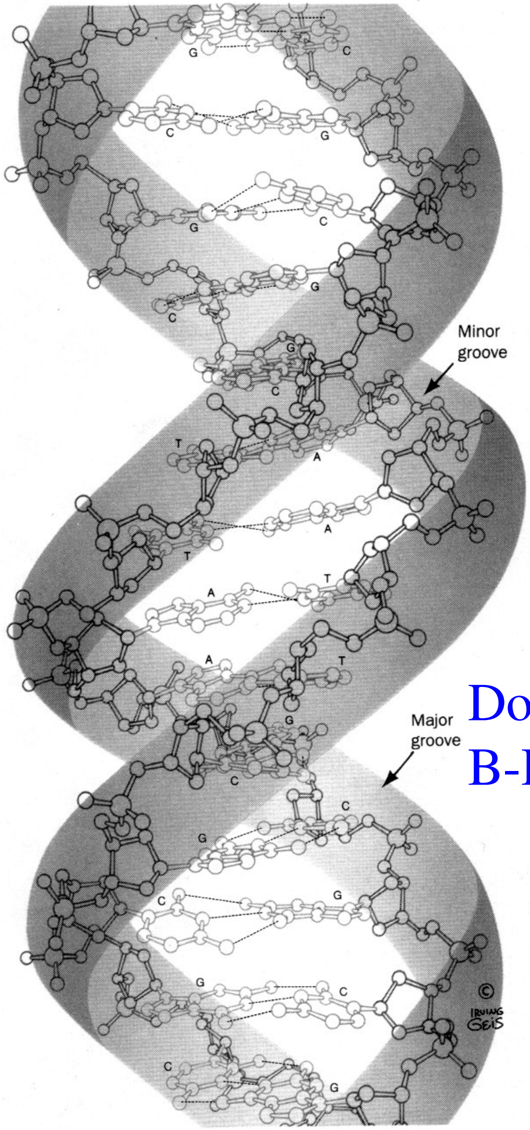
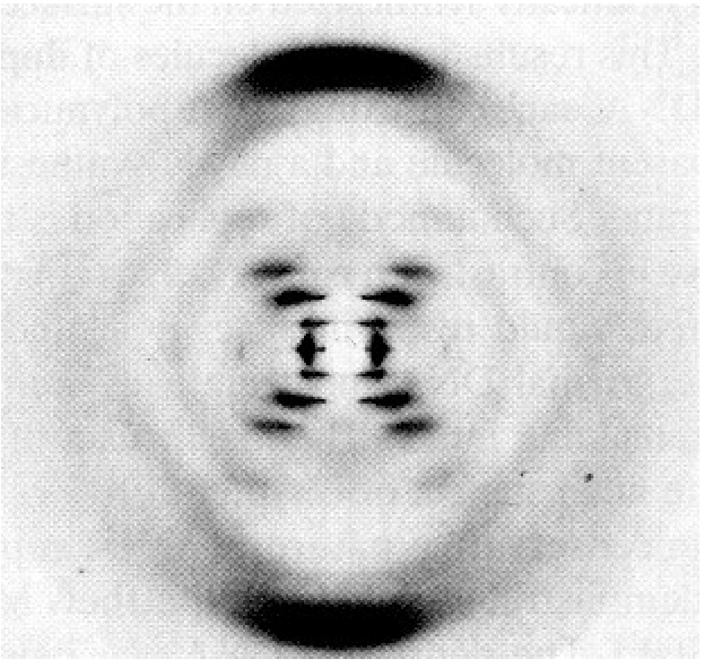
DNA (Watson-Crick)

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



Double stranded B-DNA



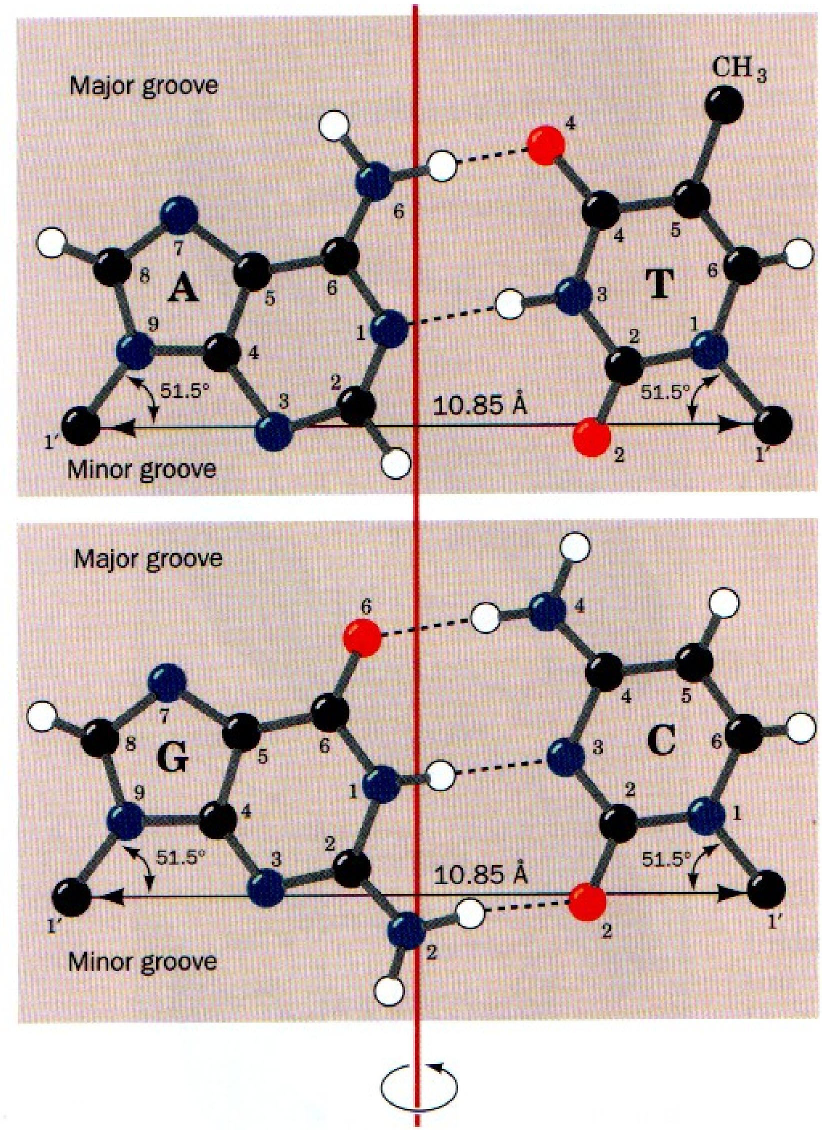
Franklin's X-ray diffraction pattern from crystalline DNA

Base Pairing (Chargaff's Rules)

- 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

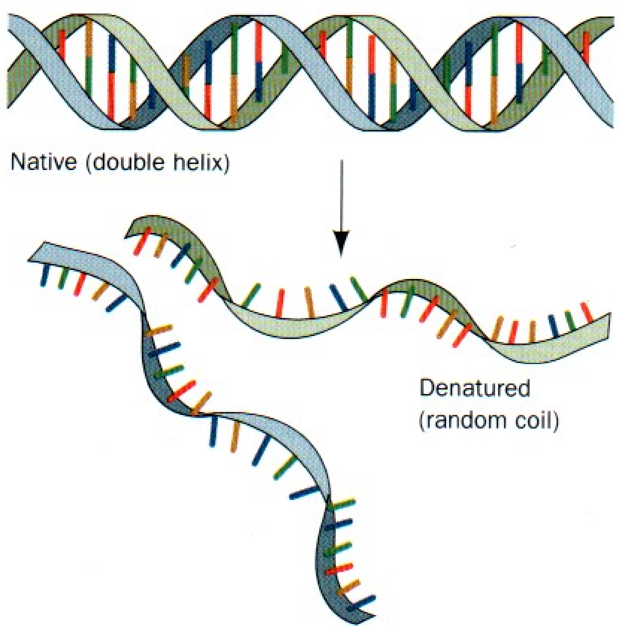
Result is due to complementary base pairing

- Indirectly led to discovery of "Double Helix"
  - Ultimately led to the discovery of the mechanism of DNA replication



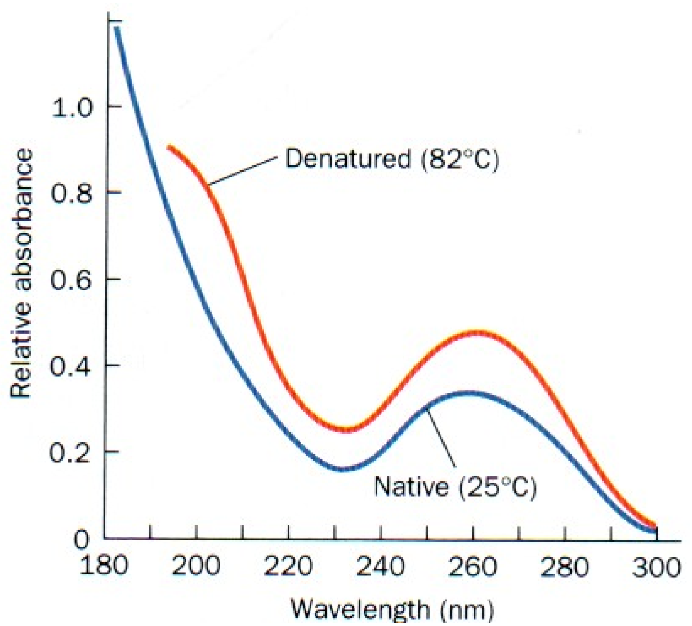
Figures: Deoxyribose C1' is shown as 1' – note that both deoxyribose sugars are on the same side of interacting bases

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

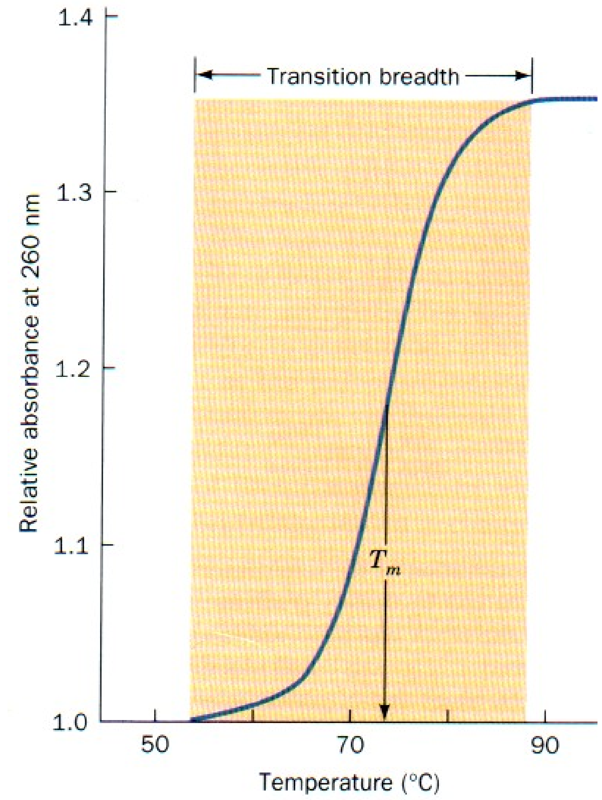


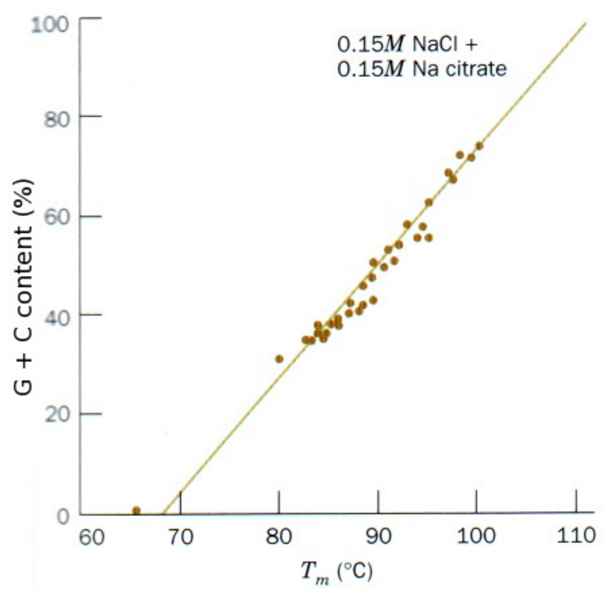
Denaturation is Cooperative

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





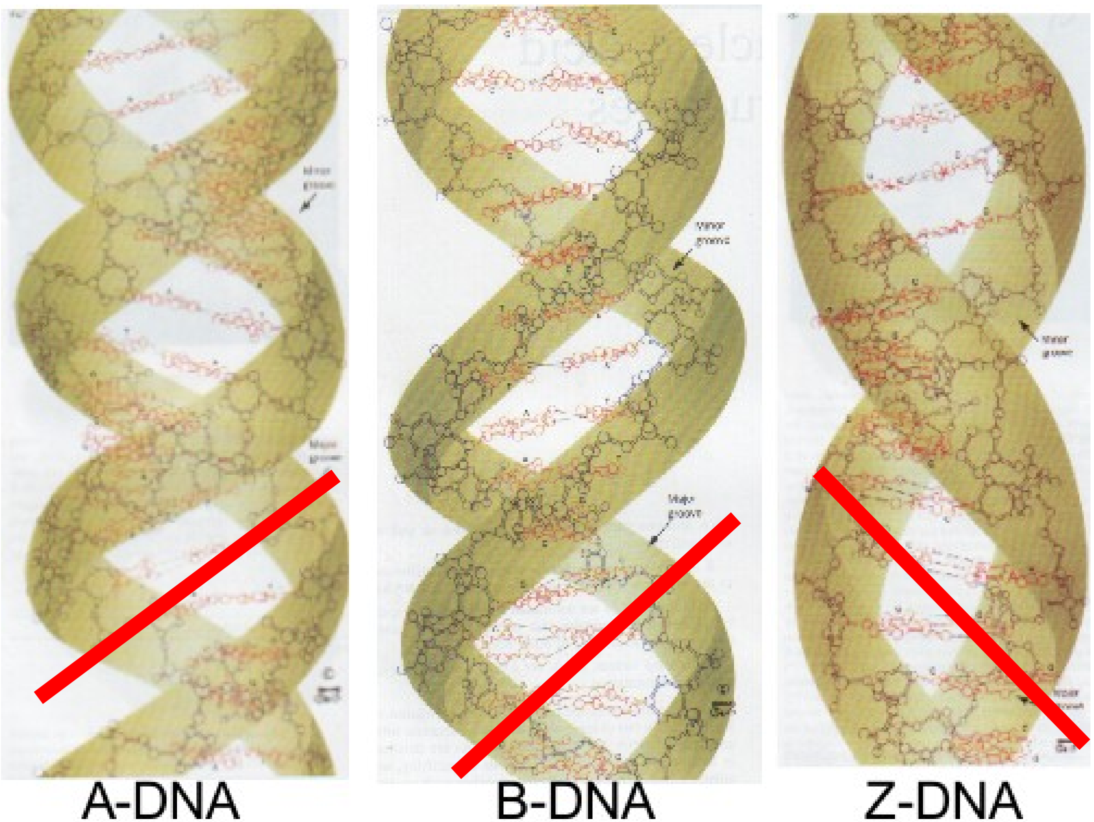
Note: For very short DNA species (oligonucleotides) size has significant effect on T_m

Size of DNA

TABLE 5-2 Sizes of Some DNA Molecules

Organism	Number of base pairs (kb)[a]	Contour length (μm)
Viruses		
Polyoma, SV40	5.2	1.7
λ Bacteriophage	48.6	17
T2, T4, T6 bacteriophage	166	55
Fowlpox	280	193
Bacteria		
Mycoplasma hominis	760	260
Escherichia coli	4,600	1,600
Eukaryotes		
Yeast (in 17 haploid chromosomes)	12,000	4,100
Drosophila (in 4 haploid chromosomes)	180,000	61,000
Human (in 23 haploid chromosomes)	3,200,000	1,100,000
Lungfish (in 19 haploid chromosomes)	102,000,000	35,000,000

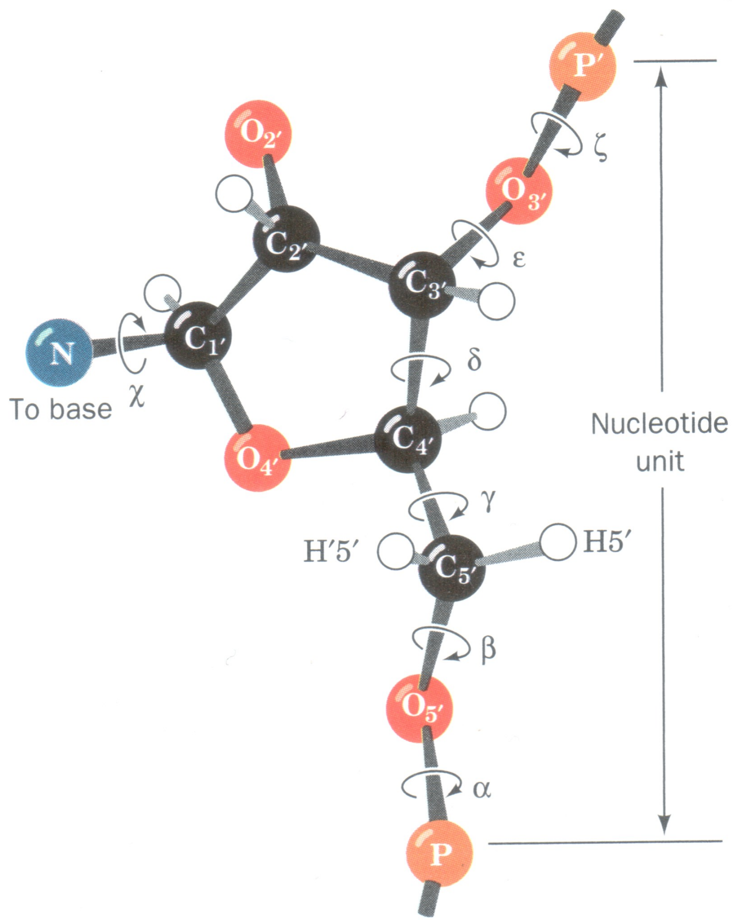
dsDNA Structures



The red line lies within major groove

- **A DNA**
  - **grooves are more equal in size**
  - **larger diameter**
- **B DNA**
  - **pronounced major groove**
  - **narrow diameter**
- **Z DNA**
  - **shallow grooves**
  - **narrowest diameter**

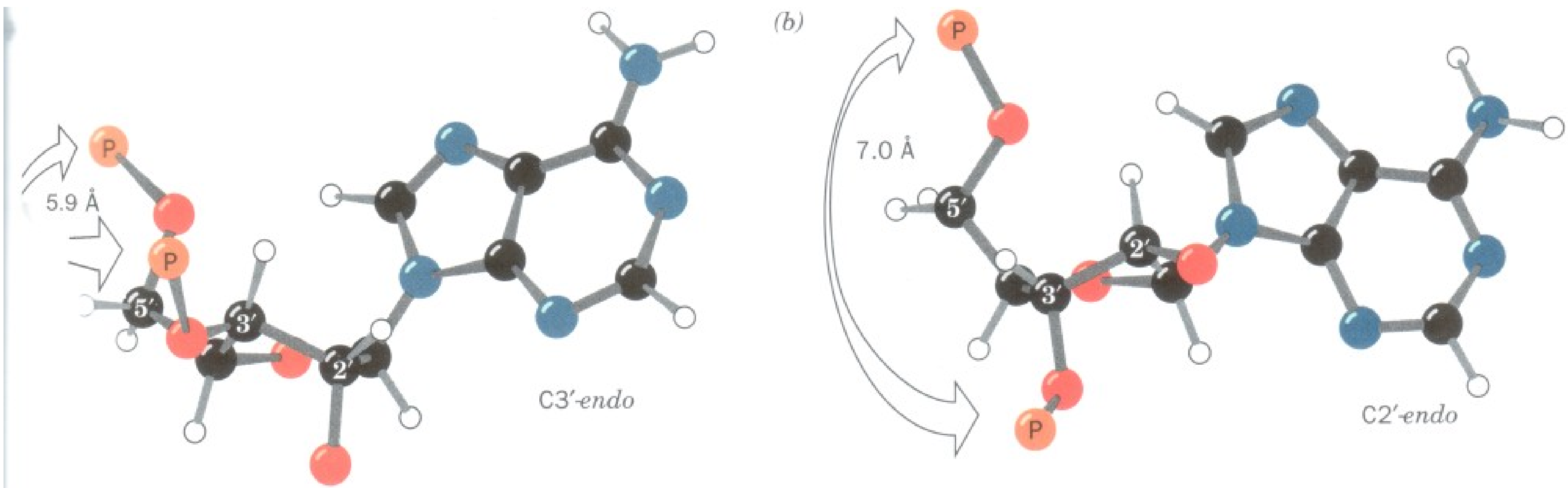
dsDNA Backbone



- 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

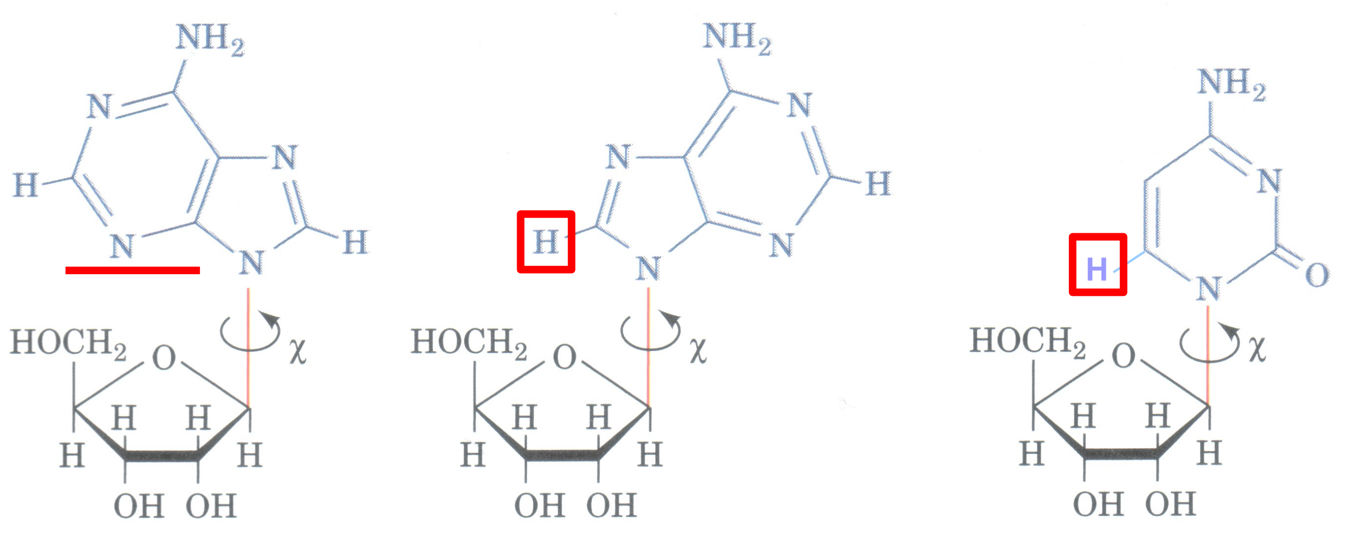
Ribose Conformations

- 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



dsDNA: Structural Parameter

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

dsDNA is typically in the B-form under physiological conditions

Note: Learn material in red boxes.

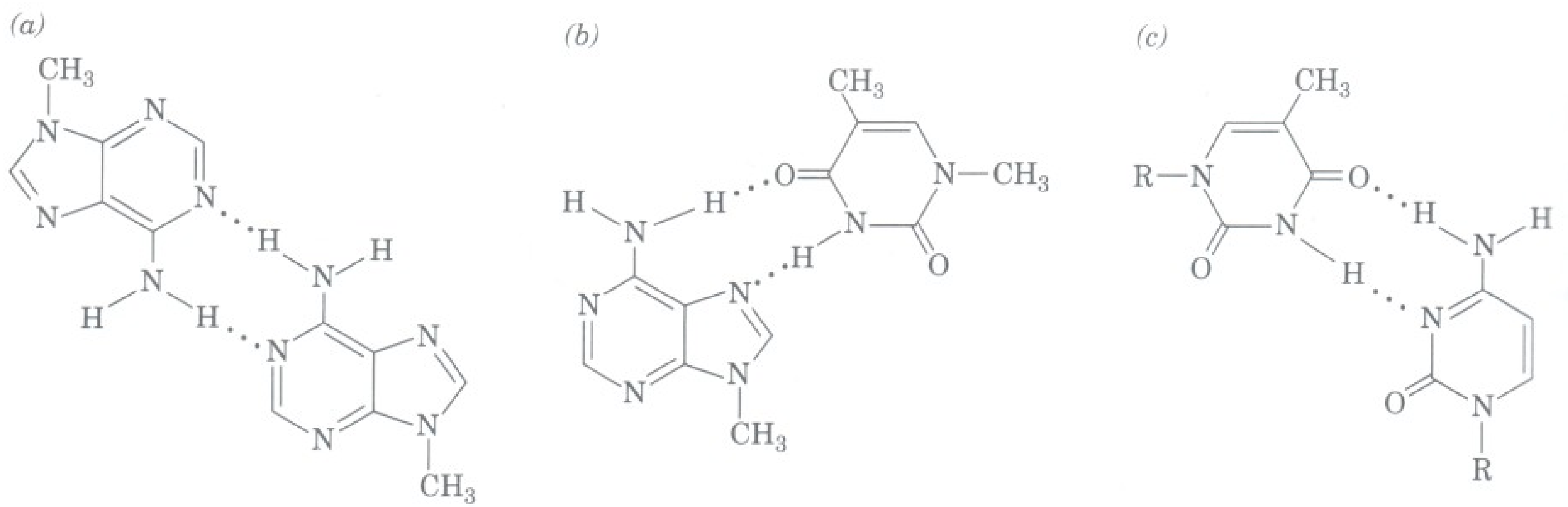
Other Base Pairs

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

- **occur in unusual dsDNA structures including 'mismatches'**

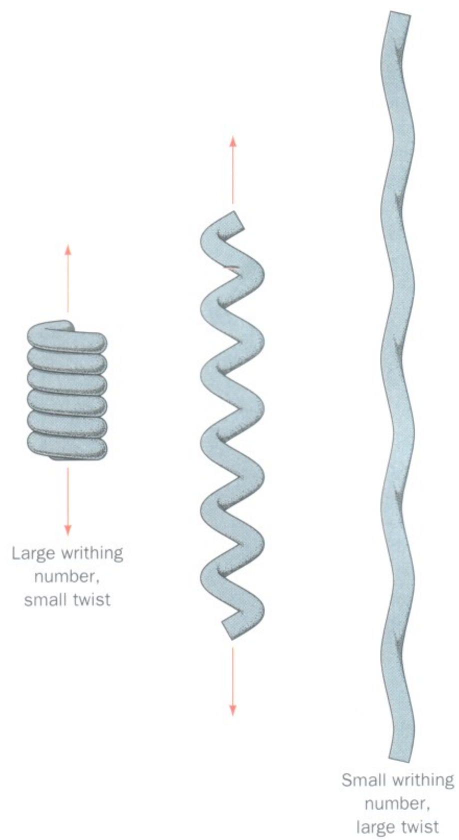
Rare in dsDNA but common in ssRNA

- **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)