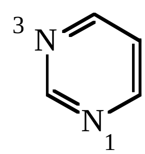
Chapters 7 & 19 - **Nucleosides, Nucleotides and Nucleic Acids**

Nucleosides, nucleotides and nucleic acids function as vitamins and coenzymes, energy carriers, second messengers, catalysts, and as genetic information transmitting and storage materials.

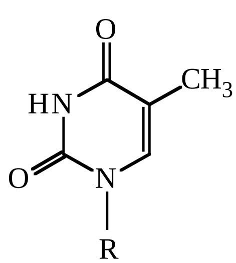
They all contain **nitrogenous bases.**

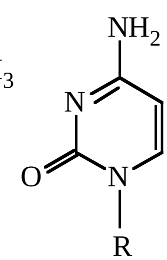
The two Base Families are shown below at pH 7.

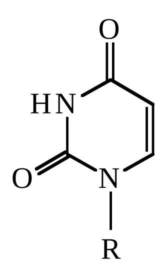
Family 1: Parent base = Pyrimidine:



Thymine T | Cytosine C | Uracil U

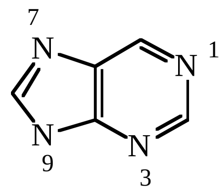




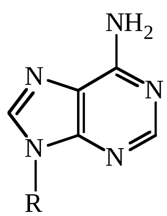


The rings are planar.

Family 2: Parent Base = Purine:

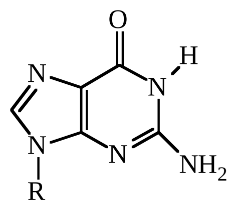


Adenine



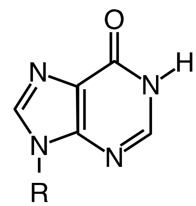
A

Guanine



G

Hypoxanthine



These rings are puckered.

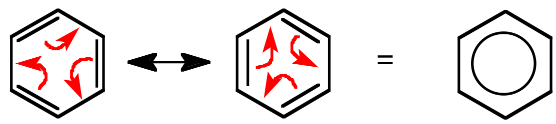
Minor Bases – The most common are methylated bases.
Endocyclic *e.g.* 7-methyl-G

The free bases are **hydrophobic** and have low water solubility at pH 7.

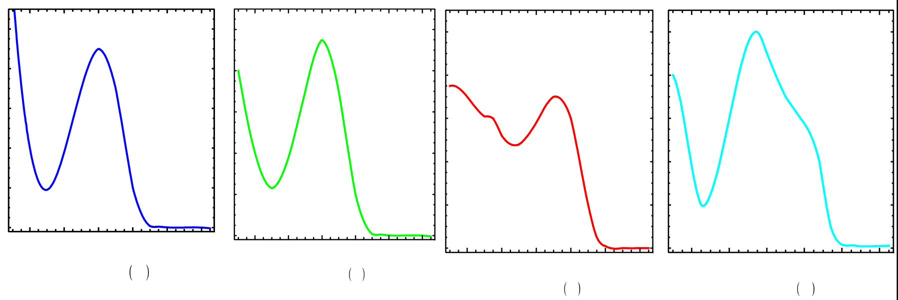
The exocyclic NH_2 are non-ionizable over pH 0 – 14, just like the NH_2 in Asn and Gln. At low and high pH the endocyclic N's ionize and the bases become more soluble.

		pK_a's
C	N^3	4.5
U	N^3	9.5
A	N^1	3.8
G	N^1	9.4
	N^7	2.4

The bases exist as **resonance** structures (electron delocalization) so that each bond has double bond character.

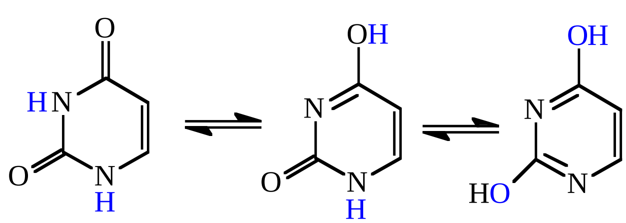


They absorb uv light with a maximum near 260 nm.



Tautomers are rapidly interconverting isomers that exist in equilibrium. This is a sort of H "*delocalization*".

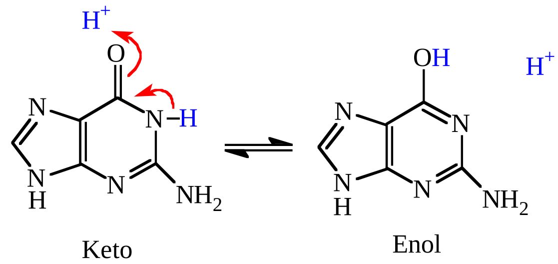
For Uracil 3 tautomers exist.



pH 7 Low pH

Lactam Lactim Double Lactim

Guanine Tautomers:

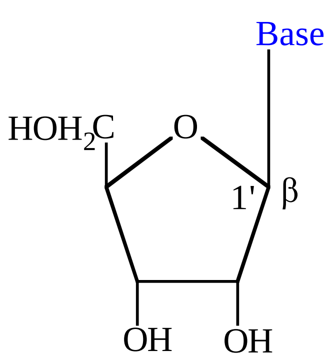


Tautomerization can lead to **mutations**; changes in DNA sequence that may lead to a change in function.

http://www.amazon.ca/exec/obidos/tg/detail/-/dvd/B000087F7J/similarities

Nucleosides are riboses with a 1'-nitrogenous base

The sugars exist only in the ring forms.



Pentose C numbers are given a prime ' to distinguish them from the base numbers.

Nucleoside Naming:

Ribose + Adenine = Adenosine
Ribose + Guanine = Guanosine
Ribose + Cytosine = Cytidine
Ribose + Uracil = Uridine
Ribose + Hypoxanthine = Inosine

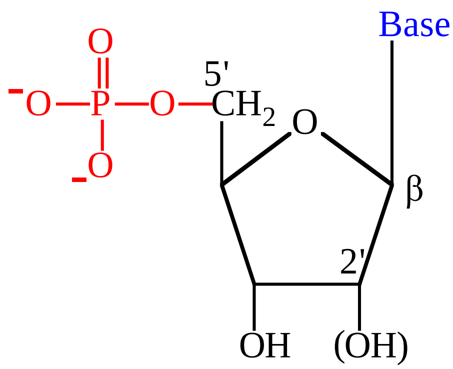
deoxyRibose + Thymine = Thymidine
deoxyRibose + Adenine = deoxyAdenosine …..etc

Adenosine is a local hormone and neuromodulator. Other nucleosides are mainly functional as components of nucleotides.

Nucleotides = Nucleosides + phosphate

Structure: Ribose-5'-**phosphate + Nitrogenous base**:

The phosphate oxygens have pK_a's of about 1.0 and 6.0 so are ionized at pH 7.



Nucleotide and Deoxynucleotide Naming:

Adenosine 5'-**m**ono**p**hosphate, Adenylate, **AMP**

Guanosine 5'-**m**ono**p**hosphate, Guanylate, **GMP**

Cytidine 5'-**m**ono**p**hosphate, Cytidylate, **CMP**

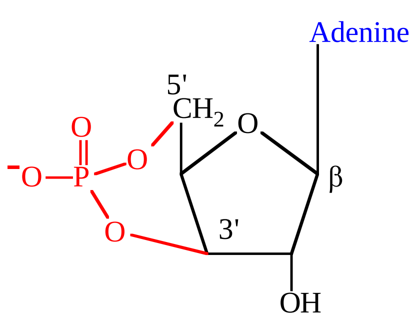
Uridine 5'-**m**ono**p**hosphate, Uridylate, **UMP**

Inosine 5’-**m**ono**p**hosphate, Inosinate, **IMP**

deoxy**t**hymidine 5' **m**ono**p**hosphate, Deoxythymidylate, **dTMP**
deoxy**a**denosine 5' **m**ono**p**hosphate, Deoxyadenylate, **dAMP**
…….. etc.

Cells also contain nucleotides with phosphates
at the 2'OH, 3'OH, and 3',5' cyclic monophosphates *etc*.

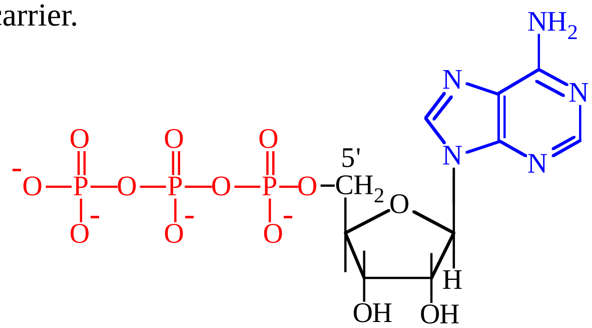
cAMP



cAMP & cGMP are **second messengers** important in intracellular **“signal transduction”**.

Nucleotides as Energy Currency

Adenosine-5'-Triphosphate – ATP – is the most common energy carrier.



CTP, GTP, UTP, are also used.

Nucleic Acids are linear polymers of nucleotides.

Deoxyribo**n**ucleic **a**cid - **DNA** - Functions in the storage of genetic information for *most* cells.

The figure shows loops of DNA attached to the protein-RNA scaffold of a **chromosome**.

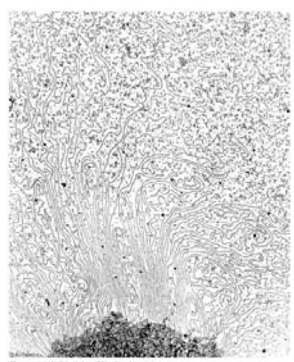


Figure 19-26b Principles of Biochemistry, 4/e

DNA - contains 2'deoxy-*D*-Ribose and the bases adenine thymine, cytosine and guanine.

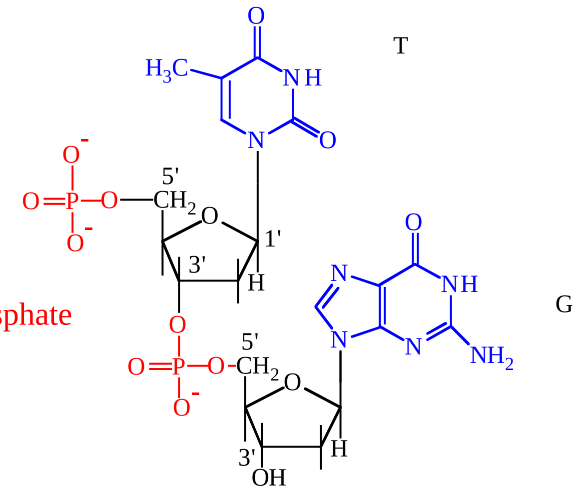
Ribo**n**ucleic **a**cid - RNA – functions in the storage of genetic information for *some* viruses *e.g.* influenza and HIV.

RNA is primarily a carrier of genetic information and is involved in some catalysis.

RNA - contains *D*-Ribose and uridine instead of thymine.

Structure of Nucleic Acids

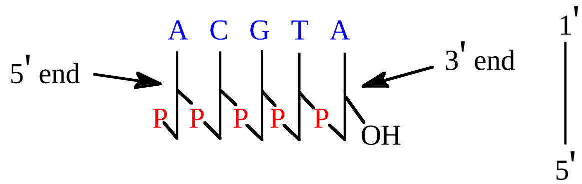
Here is the DNA dinucleotide T-G. **Phosphodiester bonds** link 3'OH to 5'OH.



Backbone = sugar-phosphate
Side Chains = bases

The sequences of nucleotides is always written 5' to 3', left to right.

pApCpGpTpA = ACGTA



Fewer than ~ 50 is an **oligonucleotide**.

Greater than ~ 50 is a **polynucleotide = nucleic acid**.

DNA 3D Structure In 1953, it was known that:

1. DNA is the component of chromosomes that carries genetic information.

2. # dA = # dT & # dC = # dG

Watson & Crick determined the 3D structure of **B-DNA** in 1953 by **X-ray diffraction**.

1. Two DNA strands coil around the same axis to form a right-handed **double helix**. The DNA strands are **antiparallel**.

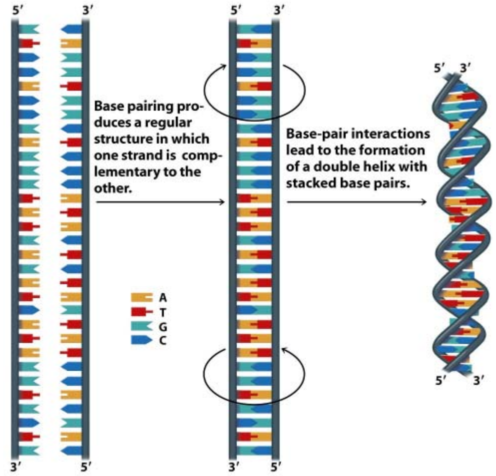


Figure 19-13 Principles of Biochemistry, 4/e
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2. The sugar-phosphates face the water.

3. The **hydrophobic** bases **stack** one on top of the other forming a hydrophobic interior. They interact *via* VdW and dipole:dipole weak interactions. This arrangement raises **water entropy**. Base stacking makes the largest contribution to the stability of the double helix.

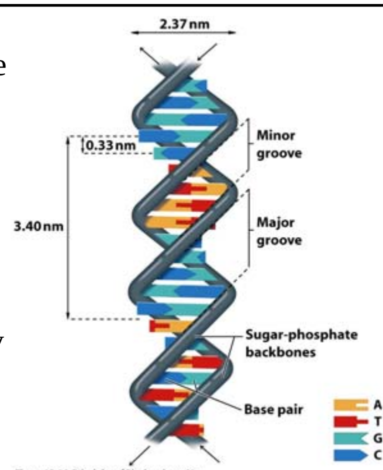

Figure 19-14 Principles of Biochemistry, 4/e
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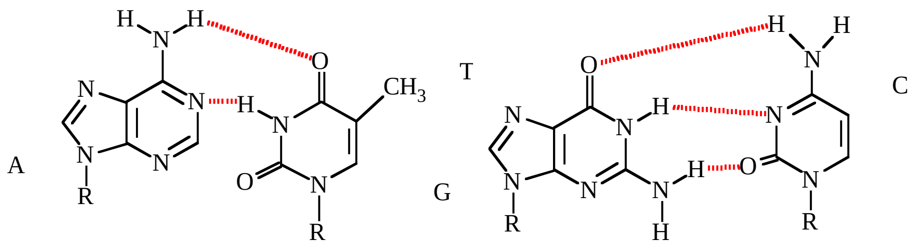
Base stacking reduces the uv absorptivity at 260 nm. This is called the **hypochromic effect**.

4. The bases are H-bonded. In DNA A:T G:C

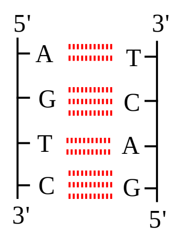
In RNA A:U G:C

The following show Watson:Crick H-bonding.



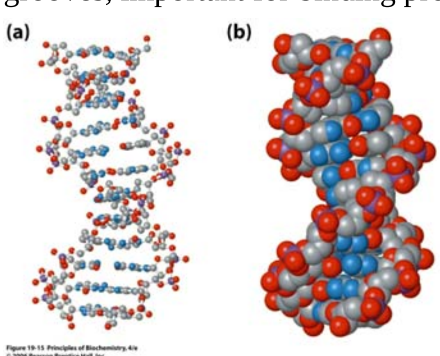
Pairing of A-T, G-C means the 2 strands have different but **complementary** sequences.

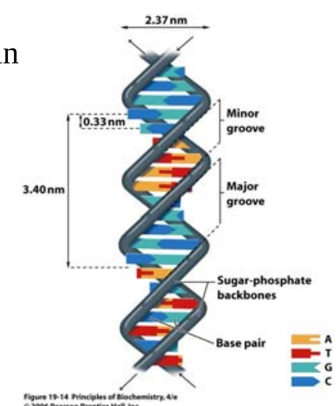
The bases are 0.33 nm apart.



The double helix rises 3.4 nm in 1 turn = 10.5 bases.

A space-filling model shows the major and minor grooves, important for binding protein





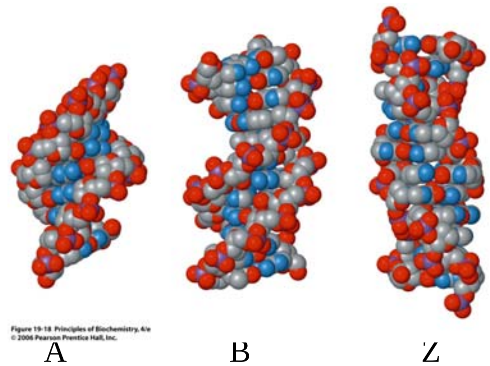
The sugar-phosphate backbone has many flexible bonds so different conformations are possible.

In the absence of water a more compact **A-DNA** is formed.

The double helix is right-handed, complementary, antiparallel.

Z-DNA named after Zig-Zag.

It forms a left-handed, complementary, double helix.



Palindromes read the same forward and backward:

ROTATOR NURSES RUN

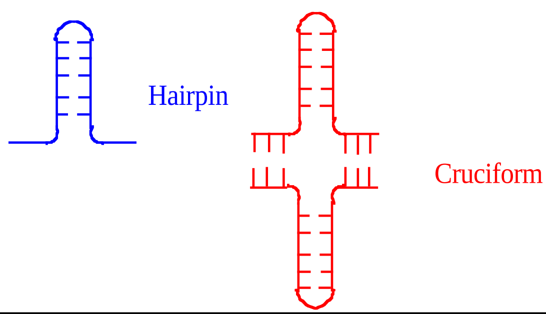
A Nucleic Acid "*Palindrome*" is:

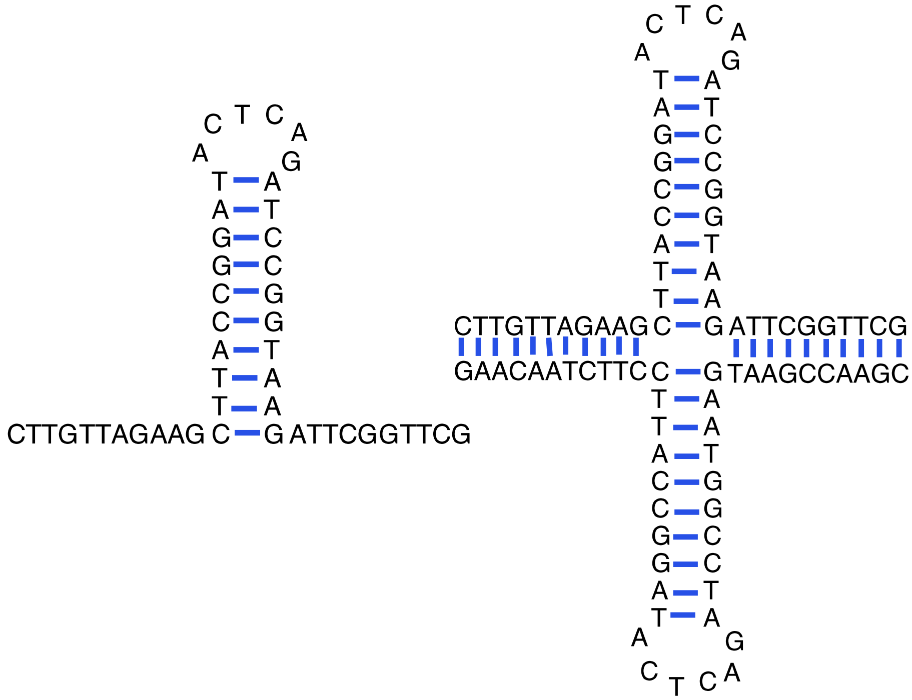
→

5'-T-T-A-G-C-A-C-G-T-G-C-T-A-A-3'
3'-A-A-T-C-G-T-G-C-A-C-G-A-T-T-5'

←

There is self-complementarity **within** the strands. Hairpins and cruciforms can form.





"We ran a series of DNA tests, Jerry, and the results are conclusive. *You* are the one who took the last cup of coffee and forgot to make a new pot!"
the last cup of coffee and forgot to make a new pot!"

RNA Structure RNA is usually single-stranded and usually a right-handed helix.

http://www.not1.xpg.com.br/dna-e-rna-diferencas-funcoes-codigo-genetico-propriedades/

Base-stacking maintains the structure.

Since Pu-Pu stacking is strongest it can disrupt regular stacking.

Self-complementary sequences can also alter the structure *via* Watson-Crick H-bonds + G:U.

RNA can also base pair with RNA and DNA.

Usually A-form, right-handed, antiparallel, double helices are formed.

A rich variety of structures is possible.

Connection between Structure and Function:

Before a cell divides, the double helix unfolds and each strand serves as a template for the **replication** of a new complementary strand, resulting in 1 double helix for each new cell.

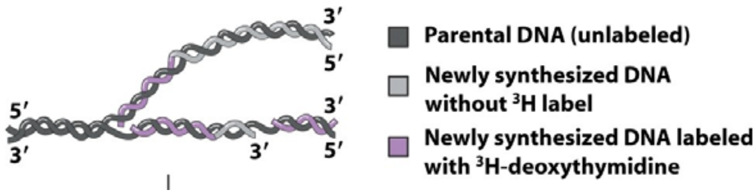
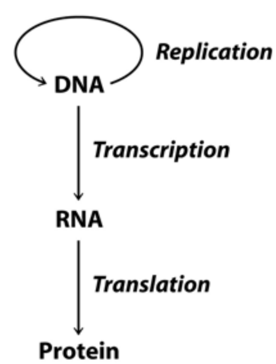


Figure 20-10 Principles of Biochemistry, 4/e

In eukaryotes, DNA is made and stored in the cell nucleus.

Information contained in the DNA is **transcribed** into RNA in the nucleus.

Proteins are made in the cytoplasm of eukaryotes according to information stored in the DNA.



1. **Messenger RNA** carries the genetic information from the nucleus to the cytoplasm, where it is **translated** into protein.

mRNA makes up ~5% of total cellular RNA – it is short-lived, seconds to minutes.

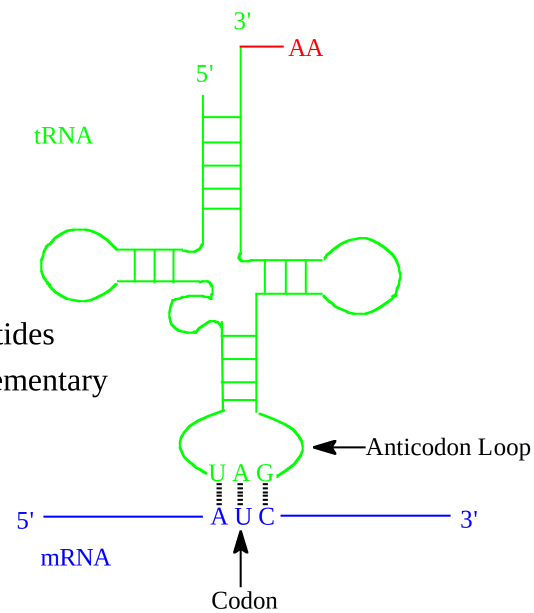
The length depends on the gene that it encodes, usually 100's – 1000's of nucleotides.

Monocistronic: RNA that encodes 1 polypeptide.

Polycistronic: RNA that encodes 2 or more polypeptides. This is more common in prokaryotes and viruses.

2. **Transfer RNA** carries the amino acids to the mRNA for protein assembly. ~15% of cellular RNA. It is the "*translator*".

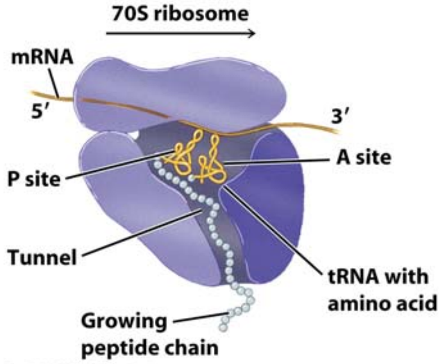
tRNA



Usually about 100 nucleotides in length with self-complementary sequences that form a "*clover-leaf*" structure.

3. **Ribosomal RNA** is the catalytic component of the **Ribosome**, a machine for assembling proteins.

About 80% of cellular RNA and typically made of molecules from 100-5000 nucleotides in length.



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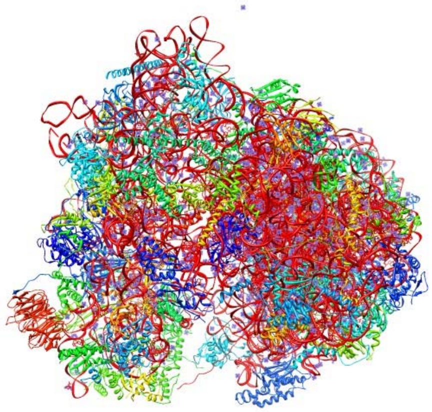
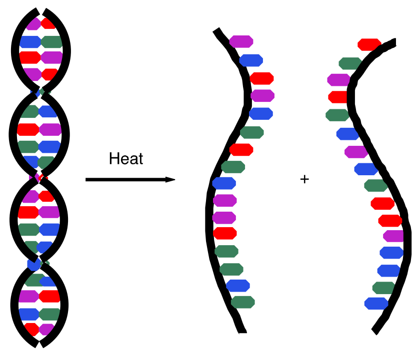


PDB ID: 3U5C

4. **MicroRNA:** About 22 base pair double stranded RNA molecules that regulate the expression of mRNA. They show potential as new types of drugs.

Physical Properties of Nucleic Acids

70 – 90^{o}C the DNA double helix **denatures**, H-bonds are broken, bases unstack, and the strands separate.

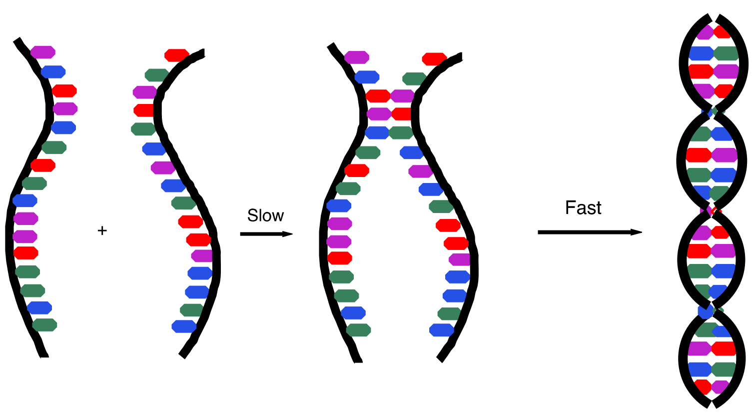


Extremes of pH can also unfold DNA.

Renaturation at lower temperatures – it occurs in 2 steps.

1. Complementary bases pair.

2. The rest of the structure forms **cooperatively**; it "*zips-up*".



Denaturation is detected by an **increase** in uv absorption of the single strands; **hyperchromicity**. Renaturation causes **hypochromicity**.

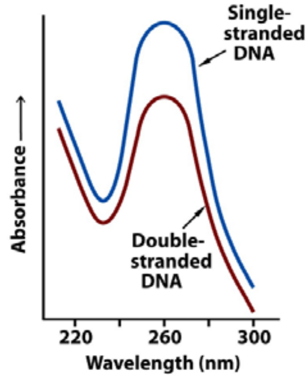


Figure 19-16 Principles of Biochemistry, 4/e

DNA with high G:C content denatures @ a higher temperature than A:T-rich DNA.

T_m is the **Melting Temperature**, the mid-point of the transition where the strands are 50% melted, 50% duplex.

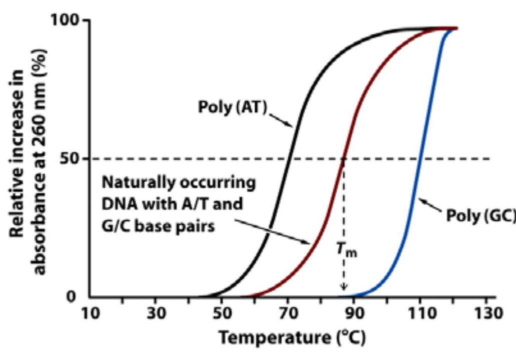


Figure 19-17 Principles of Biochemistry, 4/e

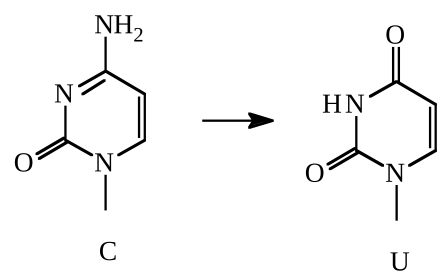
Stability of Duplexes:

RNA:RNA > DNA:RNA > DNA:DNA

Mutations are changes in the DNA structure that lead to changes in the genetic information carried by the cell.

This is important in aging and cancer. A single mutation can lead to the death of a cell or organism.

1. Deamination of C → U



Occurs in 1 / 10^7 Cytosines per 24 h.

This can be repaired by **enzymes**. They recognize that U does not belong in DNA.

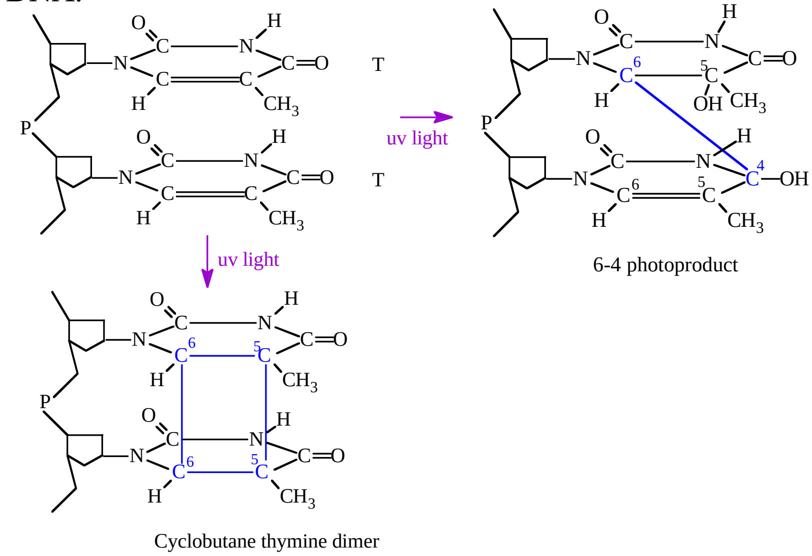
Imagine if DNA contained U instead of T. How would enzymes recognize correct *vs.* incorrect U? Thus, DNA uses T and not U.

Other deaminations occur in 1 / 10^9 bases per 24 h.

2. Depurination occurs in 1 /10^5 purines per 24 h.

Release of the base allows formation of the linear aldehyde of ribose. This can also be repaired.

3. **UV light** induces formation of a cyclobutane thymine dimer or 6-4 photoproduct if two T's are stacked. This causes kinking in DNA.



4. X-rays and other radiation can also damage DNA.

e.g. Potassium-40 is a long-lived naturally-occuring radioactive isotopes present at about 0.01%. It's half life is 1.2 billion years.

5. Oxidative Damage by H_2O_2 and free radicals: $OH^{\cdot}$, $O^{\cdot}$

6. Chemicals - nitrous acid,
- alkylating agents
- base analogs