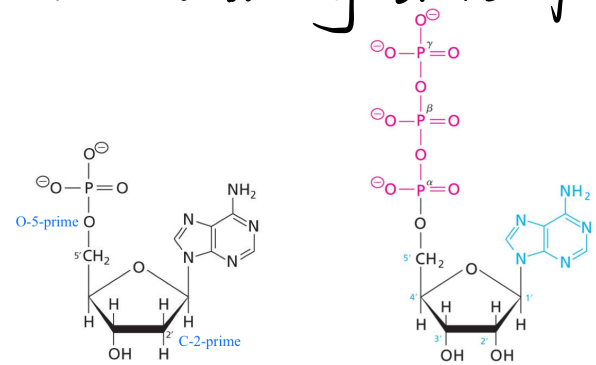
Nucleic Acid Structure and Function (Slide 6)

§1 Nucleotides (核苷酸)

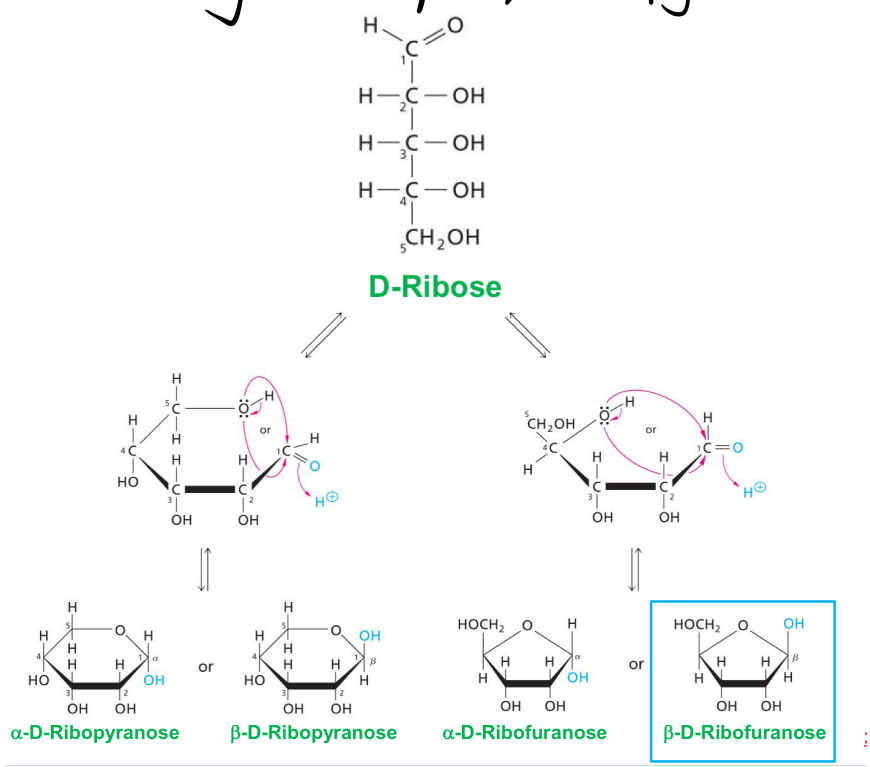
1. Nucleotides are the building blocks of biopolymers - nucleic acid



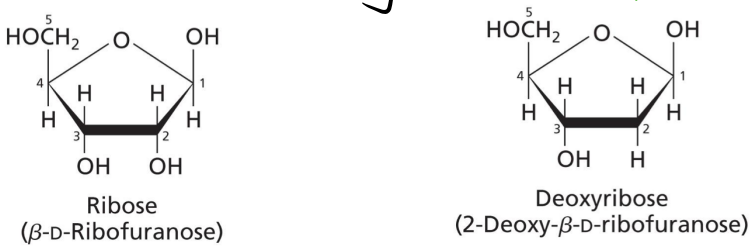
2. Ribose (核糖) structure

1° D-Ribose structure

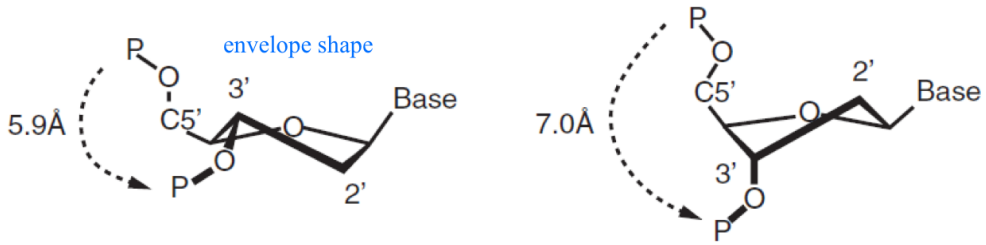
Note: The absolute configuration remain the same, although the designation of R/S configuration may change.



2° Ribose and deoxyribose (脱氧核糖)



3° Two types of sugar puckers (折叠) most commonly found in nucleic acids:

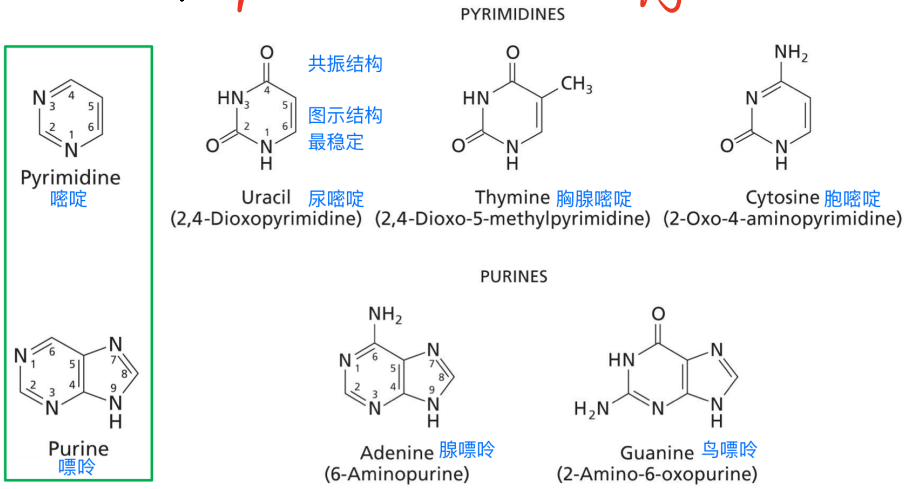


C3'-endo
Prevalent in A-form DNA and RNA duplexes

C2'-endo
Characteristic of B-form DNA duplexes

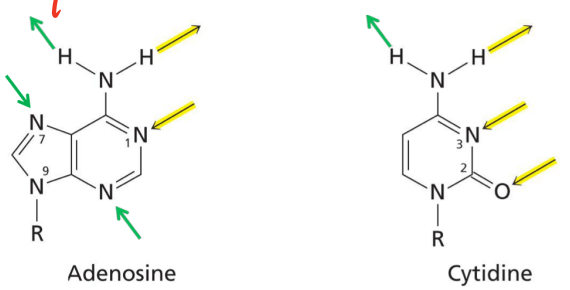
3. Base (碱基) structures

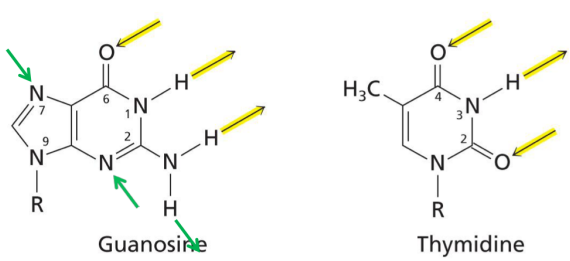
1° The two types of heterocyclic bases (杂环碱) are derivatives of purine (嘌呤) and pyrimidine (嘧啶)



2° Hydrogen bond donors and acceptors

An exocyclic (环外) amine group can't be a hydrogen bond acceptor.

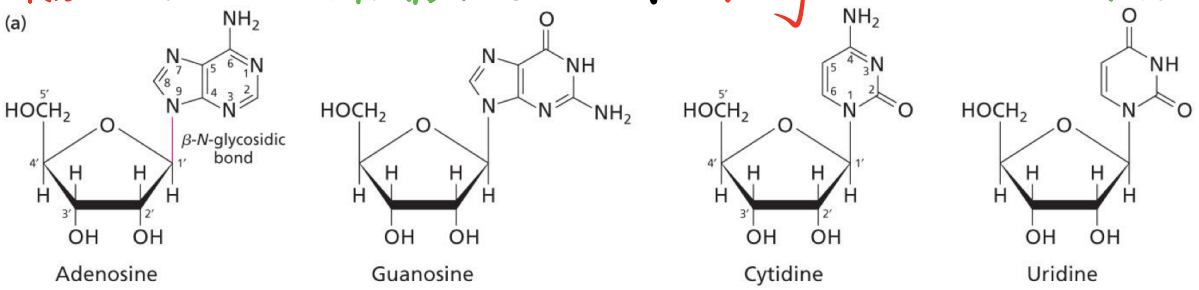


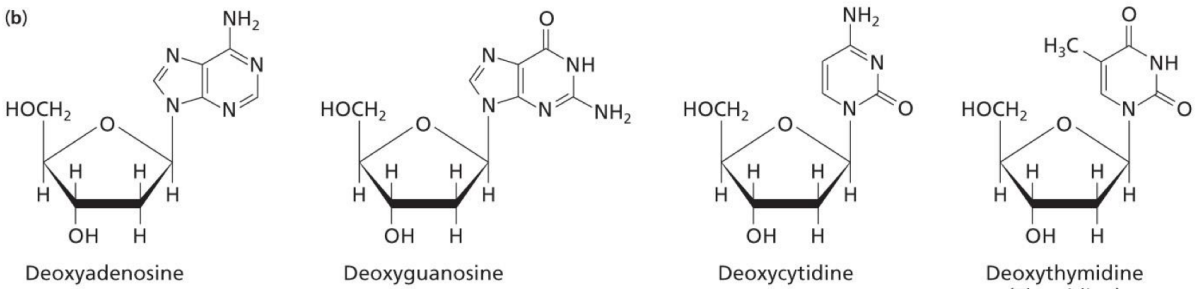


4. Nucleoside (核苷) structure

1° A nitrogenous base with a ribose

2° Ribonucleoside (核糖核苷) and deoxyribonucleoside (脱氧核糖核苷)





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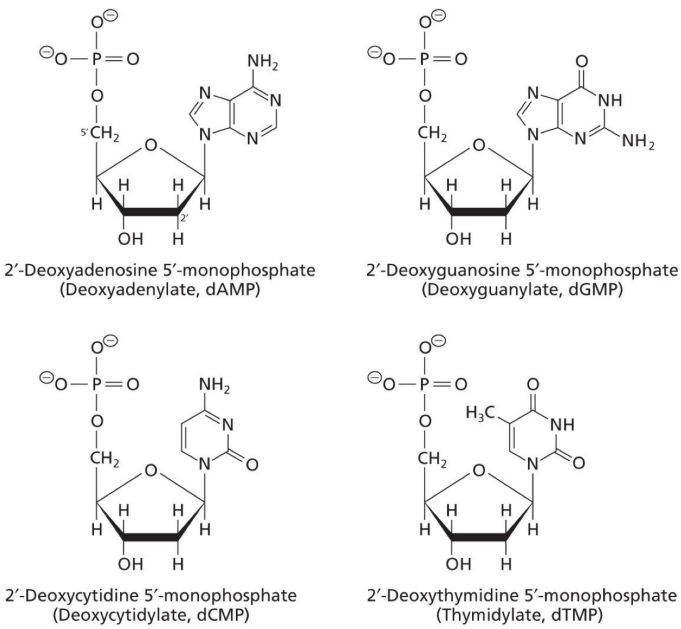
5. Nucleotide (核苷酸) structure

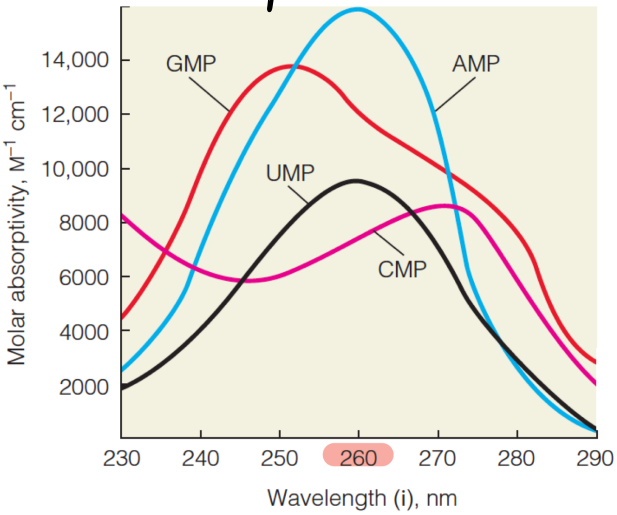
1° A nitrogenous base, a ribose and a phosphate (磷酸基团)

Shown below are deoxyribonucleotide structures:



6. Ultraviolet absorption spectra (紫外吸收光谱) of ribonucleotides

This strong absorbance is often used for quantitative determination of nucleic acid concentration.



7. Nomenclature

Nomenclature of **bases**, **nucleosides**, and **nucleotides**

Base	**Ribonucleoside**	**Ribonucleotide (5′-monophosphate)**
Adenine (A)	Adenosine	Adenosine 5′-monophoshate (AMP); adenylate[a]
Guanine (G)	Guanosine	Guanosine 5′-monophosphate (GMP); guanylate[a]
Cytosine (C)	Cytidine	Cytidine 5′-monophosphate (CMP); cytidylate[a]
Uracil (U)	Uridine	Uridine 5′-monophosphate (UMP); uridylate[a]
Base	**Deoxyribonucleoside**	**Deoxyribonucleotide (5′-monophosphate)**
Adenine (A)	Deoxyadenosine	Deoxyadenosine 5′-monophosphate (dAMP); deoxyadenylate[a]
Guanine (G)	Deoxyguanosine	Deoxyguanosine 5′-monophosphate (dGMP); deoxyguanylate[a]
Cytosine (C)	Deoxycytidine	Deoxycytidine 5′-monophosphate (dCMP); deoxycytidylate[a]
Thymine (T)	Deoxythymidine or thymidine	Deoxythymidine 5′-monophosphate (dTMP); deoxythymidylate[a] or thymidylate[a]

[a]Anionic forms of phosphate esters predominant at pH 7.4.

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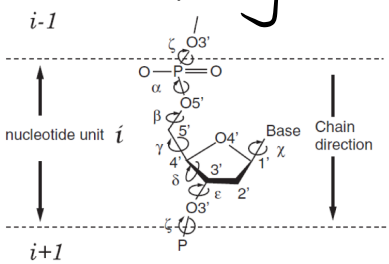
§2 DNA Structure: Double Helix

1. Dihedral angle (二面角)

1° Dihedral angles on sugar-phosphate backbone



α = $O3'_{i-1}$—P—O5'—C5'

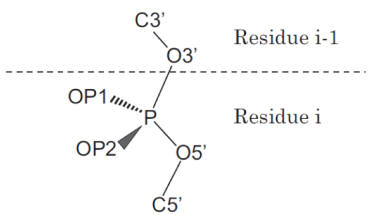
β = P—O5'—C5'—C4'

γ = O5'—C5'—C4'-C3'

δ = C5'—C4'—C3'—C2'

ε = C4'—C3'—O3'—P_{i+1}

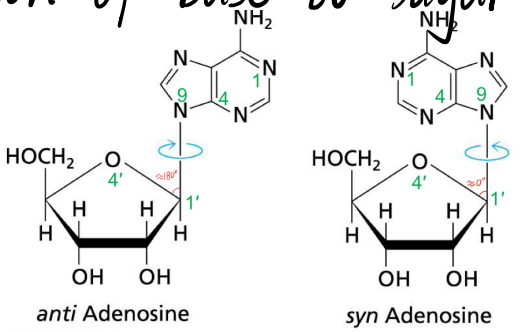
ζ = C3'—O3'—P_{i+1}-$O3'_{i+1}$

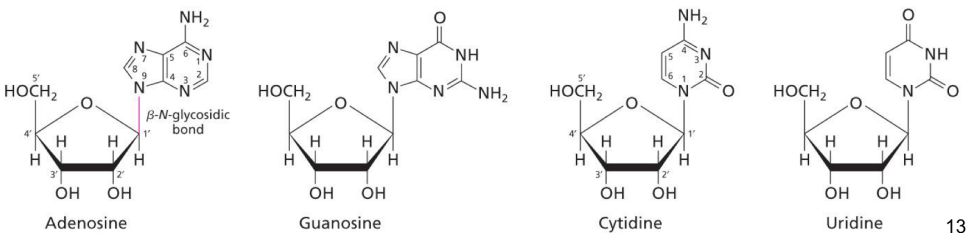


Online link:
http://en.wikipedia.org/wiki/Greek_alphabet

2° Dihedral angles of glycosidic bond (糖苷鍵)
(relative orientation of base to sugar)

- χ O4′ - C1′ - N1 – C2 (**pyrimidines**)
- χ O4′ - C1′ - N9 – C4 (**purines**)





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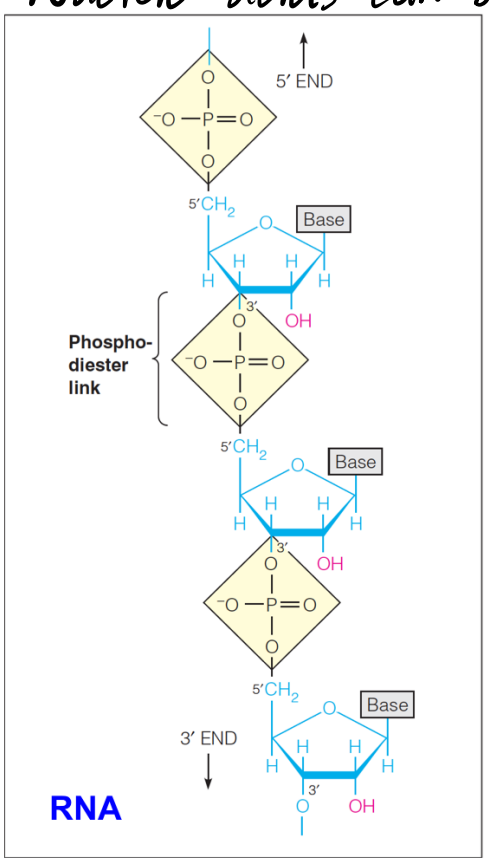
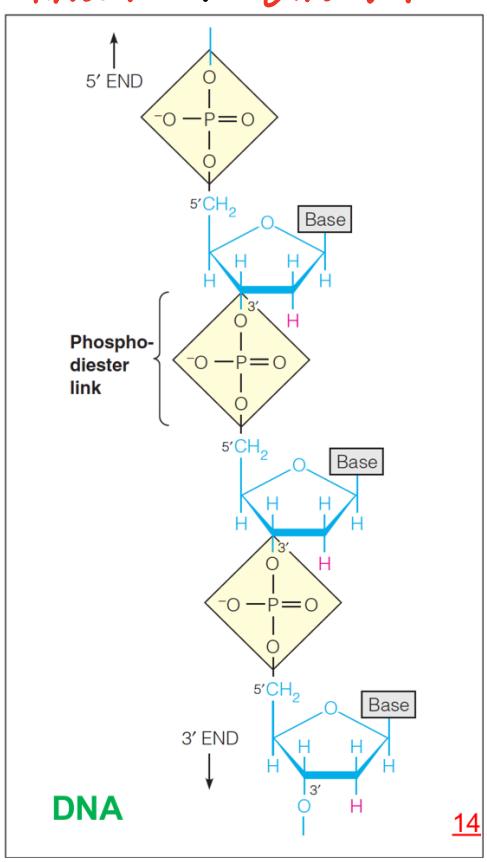
2. Primary structure

1° RNA contains the sugar ribose, DNA has the deoxyribose

2° Directionality: 5′ → 3′

3° Nucleic acids can be linear or circular





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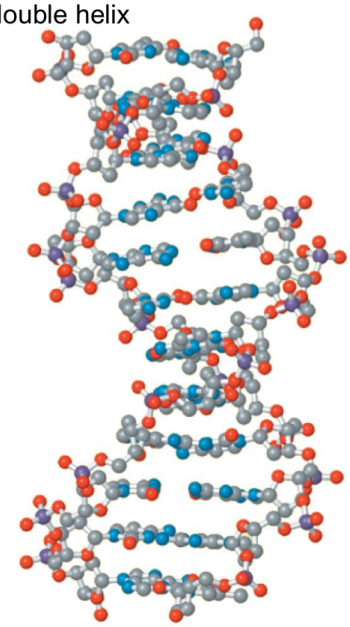
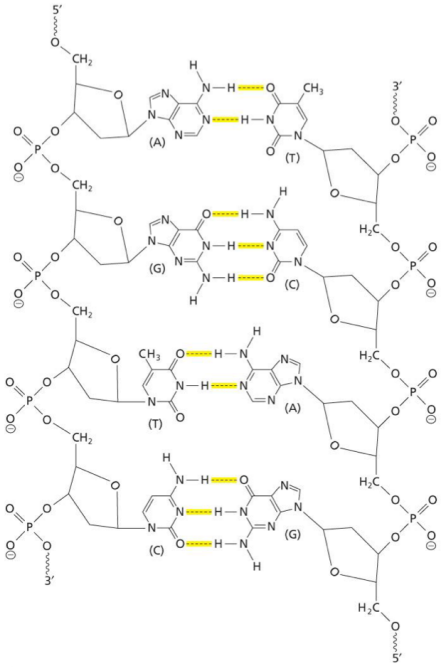
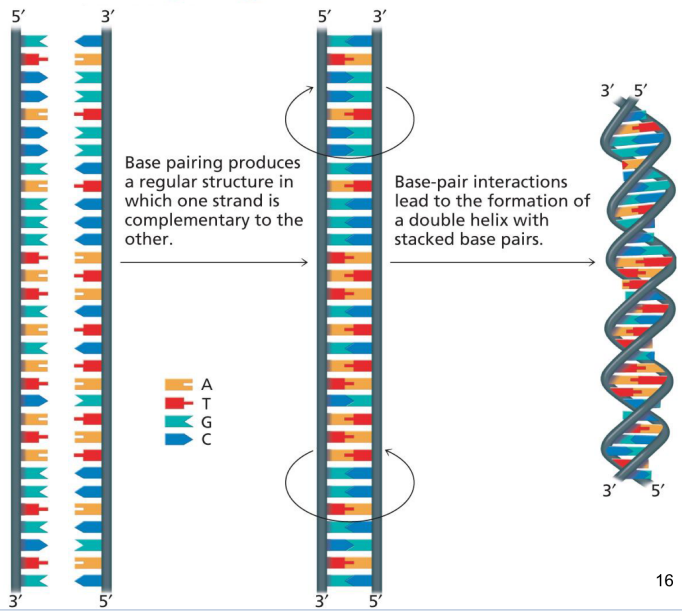
3. Secondary structure

DNA double helix





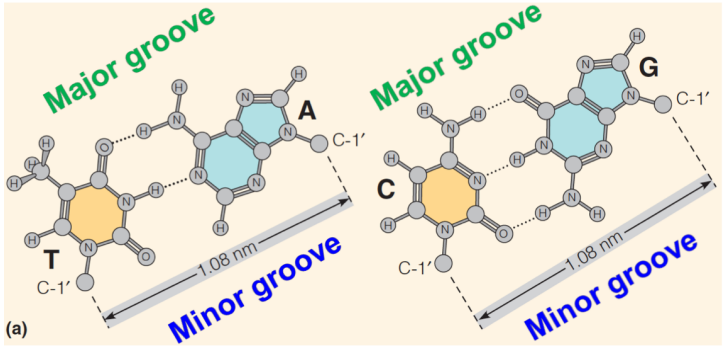


1° A-T (2 hydrogen bonds) and G-C (3 hydrogen bonds) are the base pairs in the Watson-Crick model of DNA.

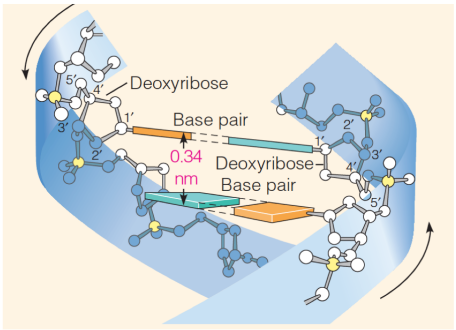
2° The complementary (互补的), two strand structure of DNA explains how the genetic material (遗传物质) can be replicated (复制).

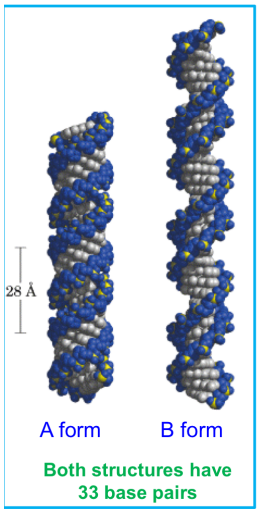
The bases are closely packed within the helix through van der Waals interactions.

The functional groups are exposed in the major and minor grooves.

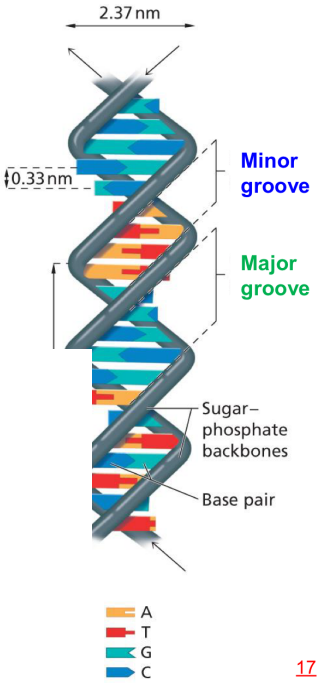


2. Double helix





	A Form	B Form
Direction of helix rotation	Right	Right
Number of residues per turn (n)	11	10
Rotation per residue ($= 360°/n$)	33°	36°
Rise[a] in helix per residue (h)	0.255 nm	0.34 nm
Pitch[a] of helix ($= nh$)	2.8 nm	3.4 nm



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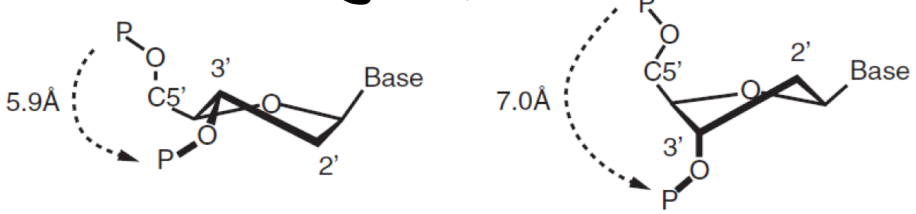
3° The two major forms of polynucleotide secondary structure are called A and B forms.

① Most DNA Watson-Crick duplex (双链) is in the B form.

② Most RNA Watson-Crick duplex is in the A form

③ The 2′ hydroxyl of RNA lies too close to the phosphate for RNA to adopt a B form with a C2′-endo sugar pucker.



C3′-endo
Prevalent in A-form DNA and RNA duplexes

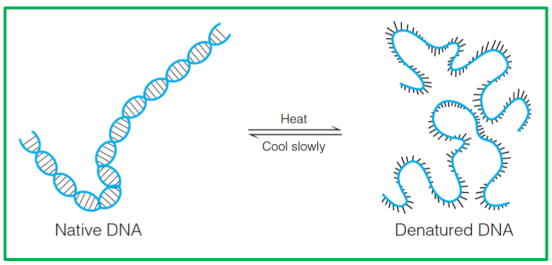
C2′-endo
Characteristic of B-form DNA duplexes

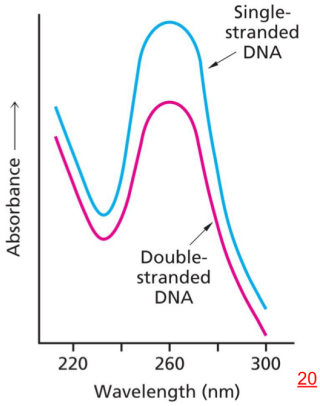
4. Denaturation of DNA

1° Absorption spectra (吸收光谱)

Absorption spectra of native and denatured DNA show that native DNA absorbs less light than denatured DNA, with the maximum difference occurring at a wavelength of 260 nm. This **hypochromicity** of double-stranded DNA can be used to distinguish between native and denatured forms.

The change in absorbance can be used to follow the denaturation of DNA as temperature increases.





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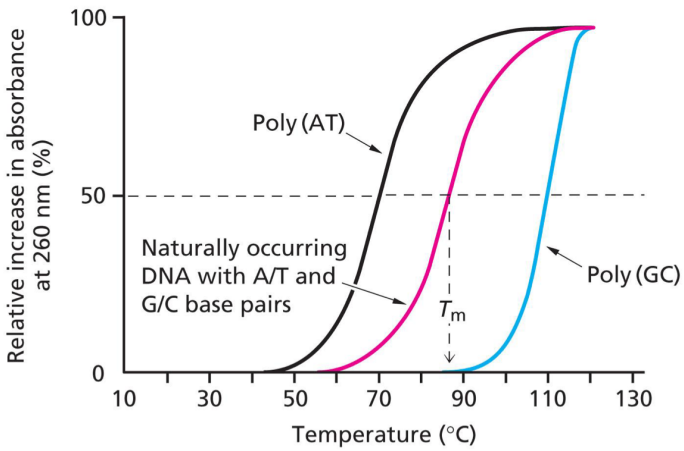
2° G-C pairs are more stable than A-T pairs.

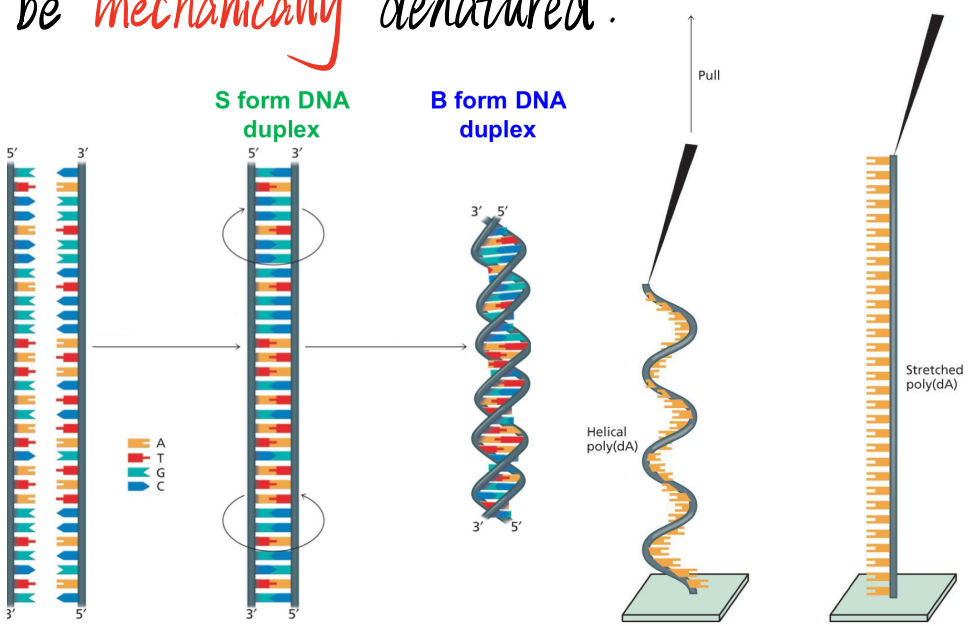
When **native** (double-stranded) DNA is heated above its "melting" temperature, it is **denatured** (separates into single strands). An abrupt increase in absorbance, corresponding to the sudden "melting" of DNA, is seen at T_m.

The midpoint of the curve marks the "melting" temperature, T_m, of DNA.

G-C pairs are more stable than A-T pairs



3° DNA duplexes are elastic (有弹性的) and can be mechanically denatured.



§3 DNA Structure: Nucleosome (核小体) and Chromatin (染色质)

1. Nucleosome and chromatin

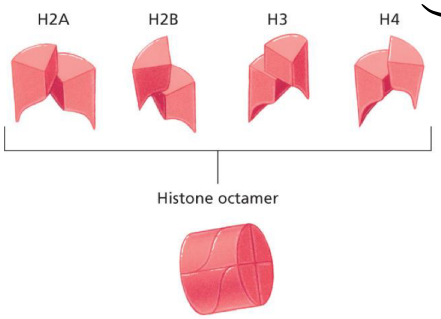
1° Histone octamer (组蛋白八聚体): Two copies of H_2A, H_2B, H_3, H_4

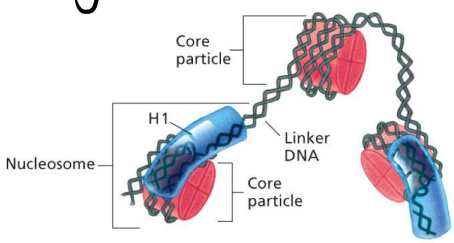
2° Nucleosome core particle (核小体核心颗粒): Histone octamer and 146 bp of DNA.

3° Nucleosome: A nucleosome core particle phuse histone H1 and linker DNA (about 54 bp),

4° Histone H1: binds to the core particle and to linker DNA

5° On the right are two extended chromatins showing the "beads-on-a-string" organization.





2. Structure of nucleosome core particle

1° The histone octamer is shaped like a flat disk (with the width of about 5 nm), with positively charged grooves to accommodate the negatively charged sugar-phosphate backbone of DNA.

2° DNA (about 146 bp) wraps around the histone octamer. Formation of nucleosome core particle reduces the DNA end-to-end distance.

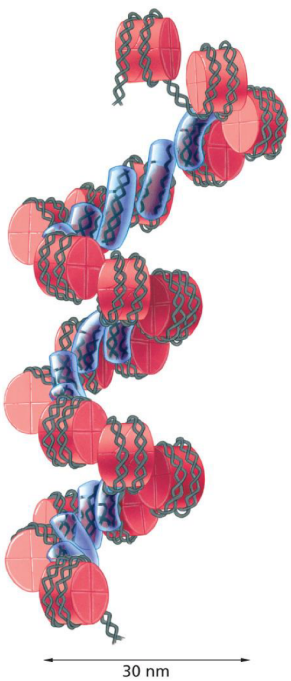
3° Histones have a high content of basic amino acids.

(碱性蛋白质: pI > 7.0，在pH=7状态下带正电荷)
(pI > pH: 带正电；pI < pH: 带负电)

3. Structure of 30-nm chromatin fiber

1° The nucleosomes associate through contacts between adjacent (邻近的) histone H1 molecules

2° The fiber forms a left-handed helix and achieves a further 4-fold reduction in chromosome end-to-end distance

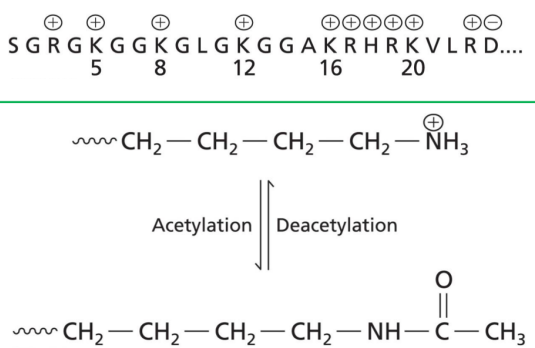


4. Histone acetylation (乙酰化) and deacetylation (脱乙酰化)

1° Specific lysine (赖氨酸) residues can be acetylated. Acetylation decreases the net positive charge of the histone.

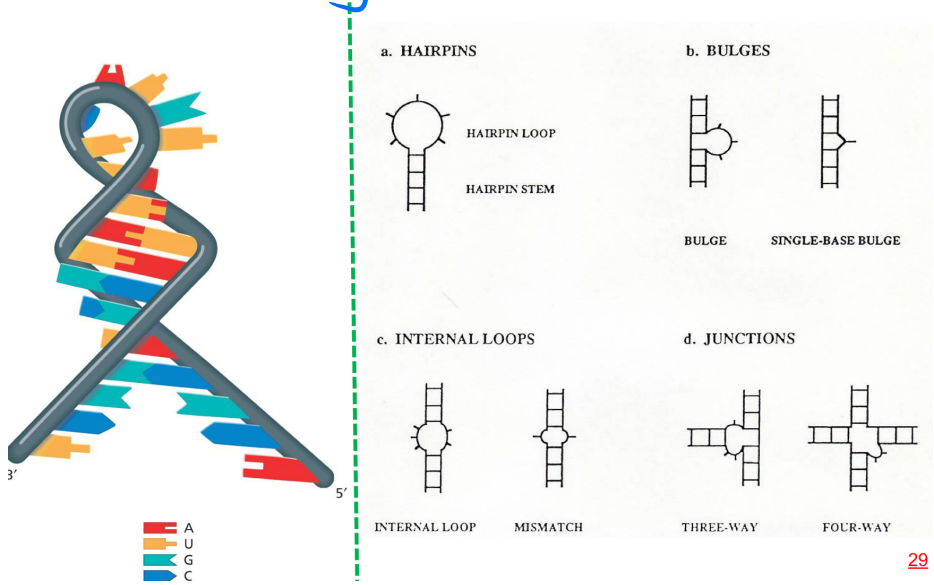
2° Acetylation results in loosing up of chromatin structure.

Below shows that residues 5, 8, 12, 16, and 20 in histone 4 can be modified by acetylation:



§4 RNA Structures

1. RNA secondary structure motifs



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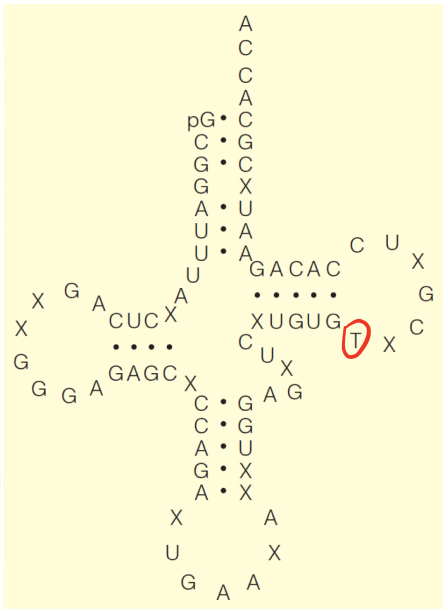


Self-complementarity in a base sequence allows a chain to fold back on itself and form RNA secondary structure containing stems (with Watson-Crick C-G and A-U pairs), harpins and junctions ...

2. Properties of naturally occurring RNA molecule

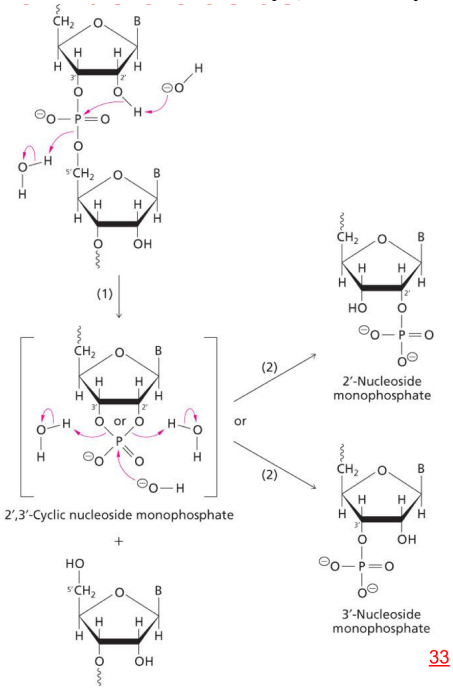
Source (Organism)	Designation	Function	Size (b or bp)
tRNA (transfer RNA)			
E. coli	$tRNA^{Leu}$	Transfers leucine in protein synthesis	87 b
Yeast	$tRNA^{Phe}$	Transfers phenylalanine in protein synthesis	76 b
Rat	$tRNA^{Ser}$	Transfers serine in protein synthesis	85 b
rRNA (ribosomal RNA)			
E. coli	5S RNA	Part of ribosome structure	120 b
	16S RNA	Part of ribosome structure	1542 b
	23S RNA	Part of ribosome	2904 b
mRNA (messenger RNA)			
Chicken	$mRNA_{LYS}$	Messenger RNA for protein lysozyme	584 b
Rat	$mRNA_{SA}$	Messenger RNA for protein serum albumin	~2030 b
vRNA (viral RNA)			
Polio virus	Polio RNA	Genome of the virus	7440 b
Cytoplasmic polyhedrosis virus of tussock moth	CPV RNA	Genome of the virus	Ten double-stranded molecules, ~890 to ~5150 bp
miRNA (microRNA), siRNA (small interfering RNA)			
All or most eukaryotes	miRNA, siRNA	Control of gene expression	21–24 b

§5 Nucleases and hydrolysis of nucleic acids

1. Alkaline hydrolysis (碱性水解) of RNA

1° RNA treated with 0.1 M NaOH at room temperature is degraded (分解) to a mixture of 2'- and 3'- nucleoside monophospate within a few hours.

2° DNA is stable under the same condition.



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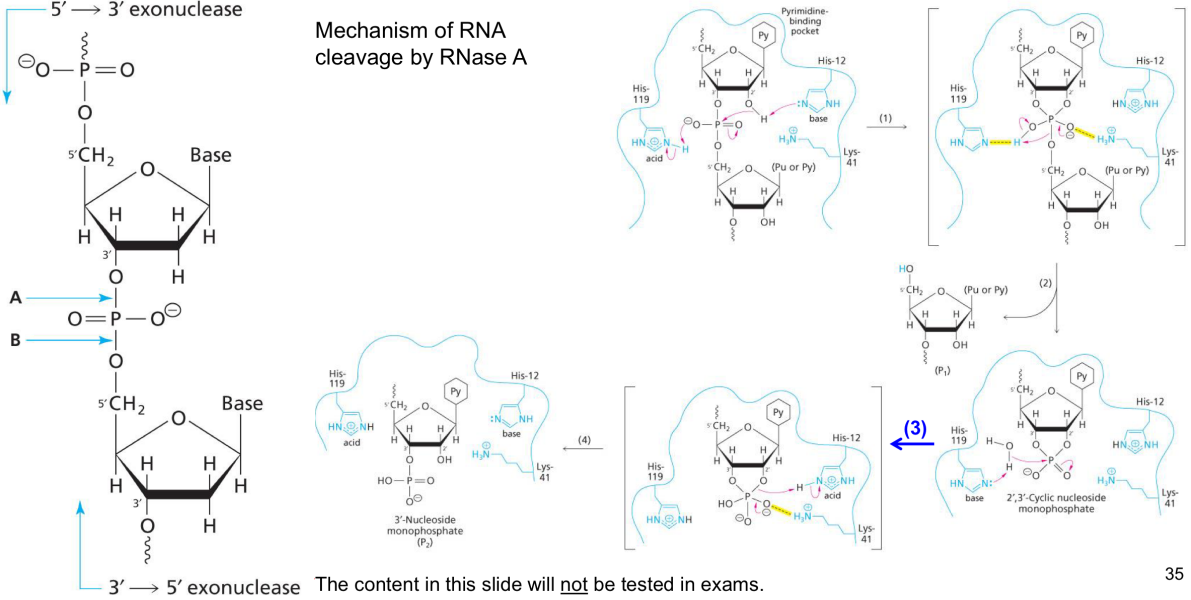
2. Nuclease cleavage (核酸酶切) of nucleic acids

1° Ribonuclease (核糖核酸酶) (RNase) acts on RNA
Deoxyribonuclease (脱氧核糖核酸酶) (DNase) acts on DNA

2° Exonucleases (核酸外切酶) catalyze the hydrolysis of phosphodiester linkages (磷酸二酯键) to release nucleotide residues from only the terminal end (5' or 3' end) of a polynucleotide chain.

3° Endonucleases (核酸内切酶) catalyze the hydrolysis of phosphodiester linkages (sites A and B as shown in the figure) at various site within a polynucleotide chain.

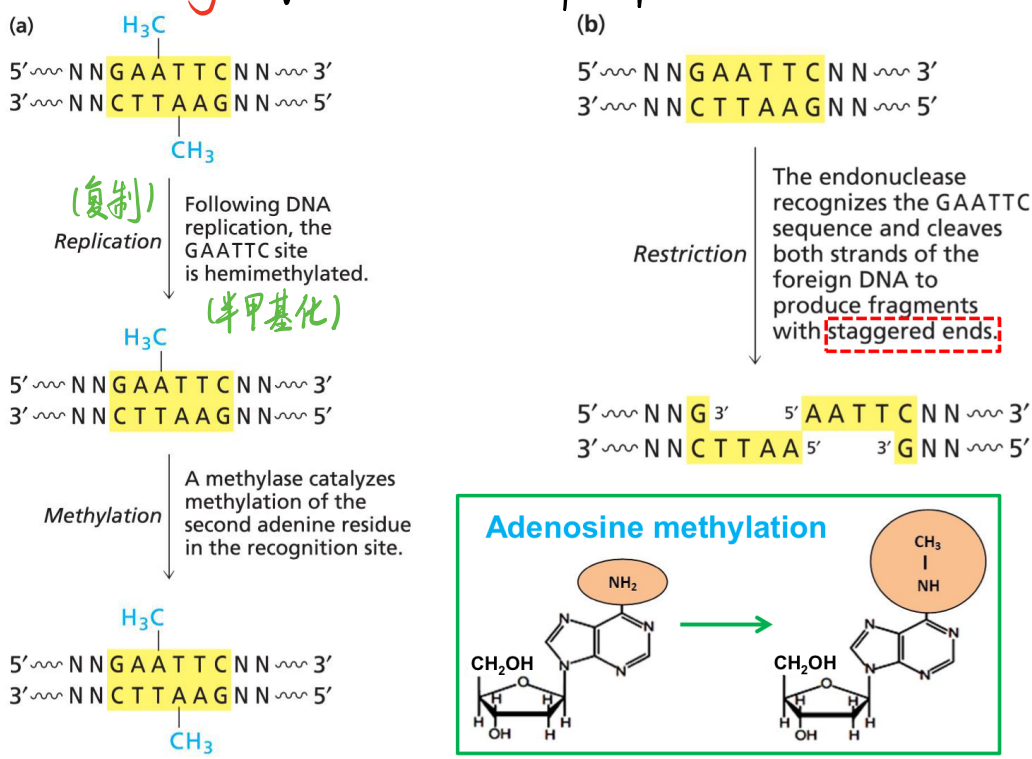


The content in this slide will not be tested in exams.

4° Restriction endonucleases: a class of bacterial enzymes that catalyze

① methylation (甲基化) of DNA bases

② cleavage of DNA at specific sites



EcoRI is composed of two subunits (purple and blue).

EcoRI binds tightly to specific palindromic (回文的) DNA sequence (GAATTC)

EcoRI recognizes the specific nucleotide sequence by contacting base pairs in the major groove.

The minor groove is exposed to the aqueous environment.