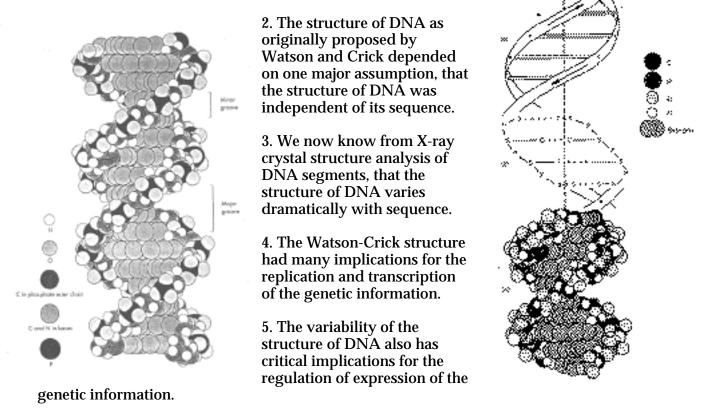# DNA Structures

Biochemistry 201
Molecular Biology
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The Structural Conformations of DNA

1. The principle message of this lecture is that the structure of DNA is much more flexible than previously conceived. DNA is a highly flexible molecule that can undergo a series of transformations leading to many conformations with different biological functions.

2. The structure of DNA as originally proposed by Watson and Crick depended on one major assumption, that the structure of DNA was independent of its sequence.

3. We now know from X-ray crystal structure analysis of DNA segments, that the structure of DNA varies dramatically with sequence.

4. The Watson-Crick structure had many implications for the replication and transcription of the genetic information.

5. The variability of the structure of DNA also has critical implications for the regulation of expression of the genetic information.

    A. The first important consequence of the Watson-Crick model of DNA was that the molecule was double helical and that the two strands contained complementary base sequences. This meant that the genetic information is redundant. This redundancy allows repair of damaged DNA and simplifies replication of the DNA via strand separation.

DNA Strucures

B. Another important consequence of the Watson-Crick structure was the anti-parallel nature of the DNA chains. Anti-parallel chains cause considerable difficulty for replication and transcription.

C. The similarity of the DNA base pairs to each other also makes faithful replication with less than one mistake in a hundred million a formidable task.

D. The structural similarity of diverse sequence makes recognition of genetic control sites difficult.

Watson-Crick B-Form DNA

1. A review of the origin and the experimental support for Watson and Crick's structure for the B-form of DNA.

2. From basic chemical analysis, Watson and Crick knew:

A. DNA contained long polymeric chains.

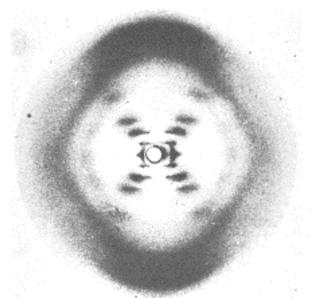B. DNA contained deoxyribonucleosides in 5'   3' phosphodiester linkage.

C. X-ray diffraction suggested a helical structure.

FIGURE 1-2
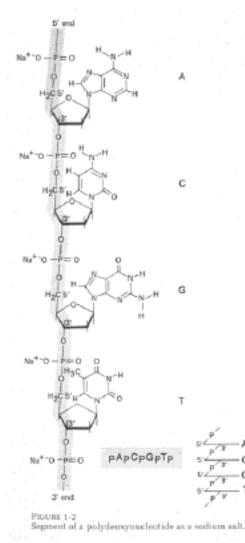Segment of a polydeoxynucleotide as a sodium salt.

- The cross pattern suggested a helical pitch angle about 45°.

- Axial reflections gave repeating units of 3.4 and 34 Å.

- Radial reflections gave a fiber width of 20 Å.

D. Chargaff's work showed that the base composition of DNA varied from organism to organism but certain relationships between the amounts of various bases always held. These relationships are called Chargaff's rules:

DNA Strucures

Source	Adenine to Guanine	Thymine to Cytosine	Adenine to Thymine	Guanine to Cytosine	Purines to Pyrimidines
Ox	1.29	1.43	1.04	1.00	1.1
Human	1.56	1.75	1.00	1.00	1.0
Hen	1.45	1.29	1.06	0.91	0.99
Salmon	1.43	1.43	1.02	1.02	1.02
Wheat	1.22	1.18	1.00	0.97	0.99
Yeast	1.67	1.92	1.03	1.20	1.0
Hemophilus influenzae	1.74	1.54	1.07	0.91	1.0
E-coli K2	1.05	0.95	1.09	0.99	1.0
Avian tubercle bacillus	0.4	0.4	1.09	1.08	1.1
Serratia marcescens	0.7	0.7	0.95	0.86	0.9
Bacillus schatz	0.7	0.6	1.12	0.89	1.0

- The amount of adenine equals the amount of thymine.

- The amount of guanine equals the amount of cytidine.

- The amount of adenine plus guanine equals 50% of the total implying that 50% of the bases in DNA are purines.

- The amount of thymine plus cytosine equals 50% of the total implying that 50% bases in DNA are pyrimidines.
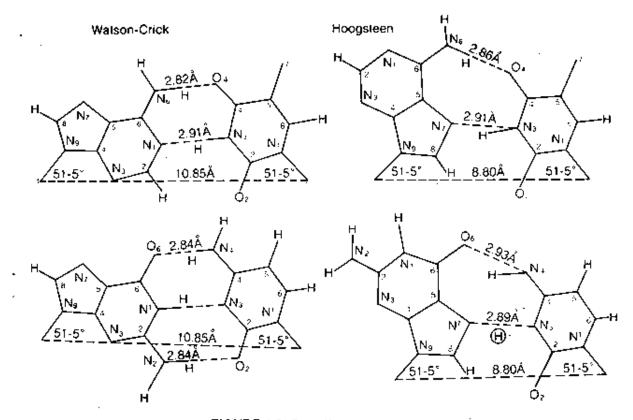

E. Watson and Crick proposed base pairing rules to explain Chargaff's equalities.

- They chose base pairs connected by hydrogen bonds.

- The bases were in their normal tautomeric forms (uncharged) at pH 7.0.

- They picked an AT pair and a GC pair that gave superimposable locations of the glycosylic linkages. A consequence of this is that the structure of the DNA would be sequence independent.

- The first base pairs observed in X-ray crystallography experiments were the Hoogstein base pairs and not the Watson and Crick base pairs.

FIGURE 4.2. Coordinates of Watson—Crick and Hoogsteen pairs.

DNA Strucures

F. The deoxyribose of each base pair are attached in opposite orientation.
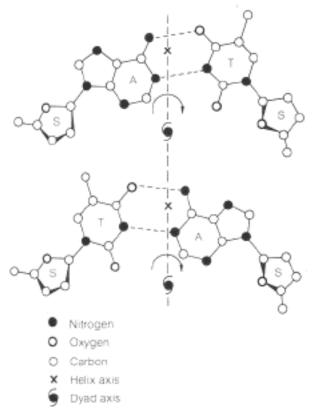
G. Each base was in the anti-conformation.

H. This conformation of nucleosides resulted in the opposite polarity of DNA chains in the resulting helix.

I. This conformation and resulting antiparallel chains generate an axis of dyad symmetry (axis of two-fold rotational symmetry) at each base pair. Dyad axes are very important for proteins that bind to DNA. Most DNA-binding proteins possess an axis of symmetry and bind to symmetric DNA sequences.

J. This structure has the base-pair as the primary repeating unit and results in an additional axis of symmetry between each base pair.

K. Watson and Crick then connected base pairs with phosphodiester bonds that spaced the bases 3.4 Å apart and rotated each subsequent base pair by 36°. This rotation generates a right-handed double helix with 10 base per turn and repeating elements every 3.4 and 34 Å.

L. The obtuse angle of the glycosylic linkages leads to major and minor grooves in helix with specific groups in each group.

M. The bases were perpendicular to the helix axis.

Crystal Structure of B-DNA

● Nitrogen
O Oxygen
O Carbon
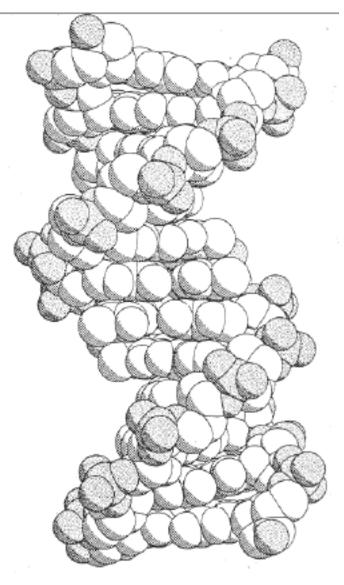✗ Helix axis
⚲ Dyad axis

DNA Strucures

1. Thirty years after the Watson-Crick proposal, Dickerson and Rich determined the complete structure of crystalline DNAs. Advances in the synthesis of large quantities of short synthetic segments of DNA allowed each of them to crystallize a unique DNA sequence.

2. Dickerson's B form crystals confirmed in most part the Watson-Crick model. DNA was double helical with antiparallel strands. The bases associated in Watson and Crick base-pairs with hydrogen bonds in the center.

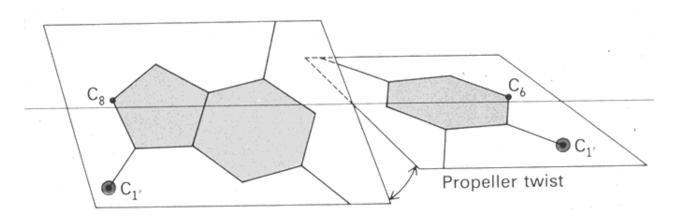3. There were also two major differences.

    A. First, the base pairs were not flat, but were twisted with respect to each other. This was called a propeller twist.

    B. The rotation from one base pair to the next was not a constant 36° as predicted, but instead varied from 27° to 40°. This variation in twist angle was extremely important because it implied that the structure of B-DNA was sequence dependent.

B-DNA Wide Groove: CGCGAATTCGCG
Wing, Drew et al., 7/80

DNA Strucures

4. The propeller twist of the base pairs results in purine-purine clash in the center of the helix. Because the purines are larger than the pyrimidine rings, they extend beyond the helical axis of DNA.
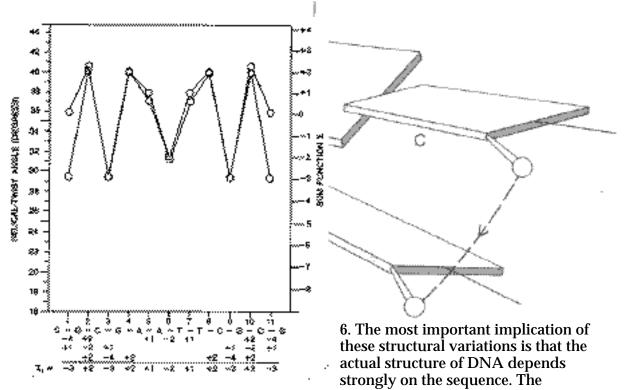

Propeller twist

5. DNA attempts to reduce purine-purine clash in several ways:

> A. The base pairs rotate less along the helix axis in the purine-pyrimidine sequences (lower average helical twist). They tend to rotate less in the pyrimidine-purine sequences (lower than average helical twist). The average helical twist was still very close to the 36° proposed by Watson and Crick.

> B. Another way DNA minimizes the purine-purine clash is that it bends toward the minor grove or major groove to reduce the interaction.

> C. Finally clashing base pairs could slide left or right toward the phosphodiester backbones to minimize the purine-purine interaction.

DNA Strucures

6. The most important implication of these structural variations is that the actual structure of DNA depends strongly on the sequence. The positions of the phosphate groups, the positions of the amino and keto groups in DNA reflect the sequence in a predictable way. Current research is aimed at understanding this structural code and to determine if regulatory sequence-specific DNA binding proteins make use of this variation in recognizing DNA.
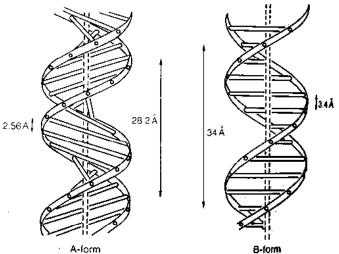
A-form of DNA

1. There are also many other forms of DNA more distinct from the B form that are biologically important. Most well known is the A-form which DNA assumes during dehydration or in RNA-DNA hybrid helices.

2. The base-pairs are not perpendicular to the helical axis but instead they are tilted at a steep angle.

DNA Strucures

3. In the A form, the base pairs are also closer together along the helical axis; 2.55 Å center-to-center distance.

4. The helical pitch of A-form DNA is closer to 11 base pairs per turn in 28 Å rather that 34 Å. As a result, the A-form is 25% shorter than the B-form. DNA shrinks when it dries.

5. If binding of protein to DNA removes water it may result in altered conformation of the DNA thus stabilizing the interaction.
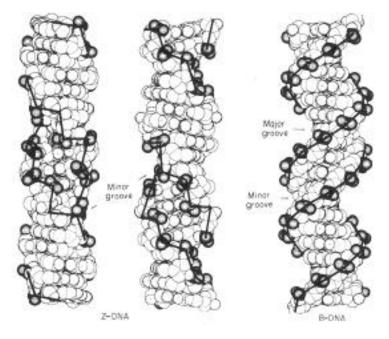

FIGURE 4.8. Schemes of the A and B forms of DNA.

6. The tilted base pairs allow room for the 2' oxygen present in RNA chains and therefore all double helices containing at least one RNA strand are present in the A-form.

7. Duplex RNA (such as found in the replication intermediates of many viral RNAs such as polio virus RNA) is always in the A-form.

Z-form of DNA

1. When the self-complementary polymer (CG)$_3$ was crystallized in high ionic strength conditions, a very unusual form of DNA called the Z-form was discovered.
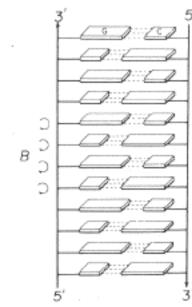
2. The Z-form differs from the B-form in several ways:
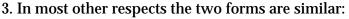

A. The helix was left-handed instead of right-handed.

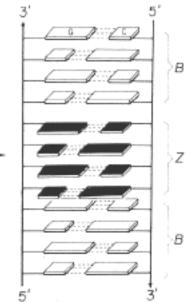B. The helix showed only a single groove rather than two.

C. The nucleotides along one strand alternate between the syn- and anti-conformation. The guanosines are all in the syn conformation while the cytidines are all in the anti conformation like the B-form.

DNA Strucures

3. In most other respects the two forms are similar:

A. Both forms are double helical and both have two chains of opposite chemical polarity.

B. Watson and Crick hydrogen bonds hold the chains together.

4. Since conformations between the purine and the pyrimidines in Z-DNA alternate, the basic repeating unit is no longer the base pair, but is a dinucleotide. This implies that there is no axis dyad symmetry at each base pair, only between base pairs.

	A DNA	B DNA	Z DNA
HANDEDNESS	RIGHT	RIGHT	LEFT
HELICAL TWIST (DEGREES)			
MEAN AND STANDARD DEVIATION	33.1 ± 5.9	35.9 ± 4.3	G–C: −51.3 ± 1.6 C–G: −8.5 ± 1.1
OBSERVED RANGE	16.1 to 44.1	27.7 to 42.0	
BASE PAIRS PER TURN	10.9	10.0	12.0
HELIX RISE PER BASE PAIR (ANGSTROM UNITS)	2.92 ± .39	3.36 ± .42	G–C: 3.52 ± .22 C–G: 4.13 ± .18
BASE INCLINATION (DEGREES)	13.0 ± 1.9	−2.0 ± 4.6	8.8 ± .7
PROPELLER TWIST (DEGREES)	15.4 ± 6.2	11.7 ± 4.8	4.4 ± 2.8
BASE ROLL (DEGREES)	5.9 ± 4.7	−1.0 ± 5.5	3.4 ± 2.1

5. Even the best evidence for Z-DNA in nature is controversial. Some of the best evidence comes from experiments involving antibodies directed specifically at the Z-DNA structure. Some authors have demonstrated that the presence of the Z-form of the DNA in these cytological preparations is an artifact of the preparation and if one prepares chromosomes carefully, no or very little Z-DNA antibody will bind.

DNA Strucures

6. The most one can say is that Z-DNA can form under physiological conditions in natural DNA sequences in which purines alternate with pyrimidines. Whether Z-DNA does form in cells and whether nature takes advantage of this unusual form is still speculation.

7. The possibility that DNA can assume two structures as distinct as the Z-form and the B-form shows that the chains are capable of much more flexibility than many had considered possible before.
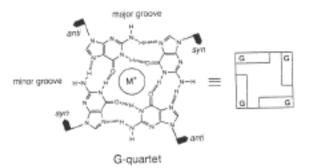
Novel DNA Structures

1. Several novel forms of DNA involving the pairing of more than two strands and also forms involving parallel chains have been described. These structures generally form with specific DNA sequences and may have profound biological consequences.

    A. Wells and others have evidence showing that oligopurine-oligopyrimidine sequences can fold back on themselves to form an internal region containing one triple stranded region and one single-stranded region. The third strand is base-paired in the major groove of a normal DNA duplex using hydrogen bonds similar to those found in the Hoogstein base pairs.
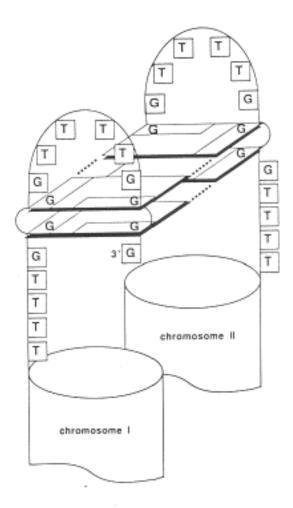
DNA Strucures

B. Sen and Gilbert have reported that DNA helices containing specific guanine rich sequences can self-associate to form four-stranded structures. The two helices are hydrogen bonded together by Hoogstein pairing.

C. Tom Cech and Aaron Klug's laboratories have demonstrated that sequences found at the ends of eukaryotic chromosomes can also form specific tetranucleotide base pairs, referred to as

G-quartet

G-quartets that may be involved in holding chromosome ends together during mitosis.

chromosome II

chromosome I

D. Englund and others noticed that certain DNA sequences had an unusual migration during electrophoresis. Analyses of such DNAs have shown that they often contain runs of 3-4 As or Ts in a row and that these runs are repeated every 10 base pairs. Such runs result in bending of the DNA towards the minor groove and the repeating nature makes the DNA helix as a whole bend in one direction.

E. Tom Jovin and Johan van de Sande have demonstrated that specific AT rich DNA sequences can base pair to form a parallel double-helical structure. These structures are physically very similar to the B-form of DNA but they are ineffective as substrates for many enzymes.

References

Dickerson, R. E. (1992). DNA structure from A to Z. Methods Enzymology 211, 67-111.

Hagerman, P. J. (1990). Sequence-Directed Curvature of DNA *Annu. Rev. Biochemistry* **59**, 755-781.

DNA Strucures

Herbert, A., & Rich, A. (1996). The biology of left-handed Z-DNA. *J Biol Chem*, 271(20), 11595-8.

Johnston, B. H. (1992). Generation and detection of Z-DNA. *Methods Enzymol* 211, 127-58.

Rich, A., Nordheim, A. and Wang, A. H. J. (1984). The Chemistry and Biology of Left-Handed Z-DNA. *Annual Review of Biochemistry*, **53**, 791-846.

Rich, A. (1993). DNA comes in many forms. *Gene*, **135**(1-2), 99-109.

Rich, A. (1994). Speculation on the biological roles of left-handed Z-DNA. *Ann N Y Acad Sci*, **726**, 1-16; discussion 16-7.

Rippe, K. and Jovin, T. M. (1992). Parallel-stranded duplex DNA. *Methods Enzymol* **211**, 199-220.

Travers, A. A. (1990). Why bend DNA? *Cell*, **60**(2), 177-80.

Watson, J. D. and Crick, F. H. C. (1953a). Molecular structure of nucleic acids. *Nature* 171, 737-738.

Watson, J. D. and Crick, F. H. C. (1953b). The structure of DNA. *Cold Spring Harbor Symp. Quant. Biol.* 18, 123-131.

Wells, R. D. (1988). Minireview: Unusual DNA structures. J. Biol. Chem. 263, 1095-1098.

Williamson, J. R. (1994). G-quartet structures in telomeric DNA. *Annu Rev Biophys Biomol Struct*, **23**, 703-30.

Internet Resources

Structure Database   http://www.ndb.bnl.gov/

Molecules R US   http://molbio.info.nih.gov/cgi-bin/pdb

RasMol Distribution   http://www.umass.edu/microbio/rasmol/

Kinemage Distribution
    http://www.faseb.org/protein/kinemages/MageSoftware.html

Protein/DNA Kinemages   http://www.faseb.org/protein/ProTeach/