Chem 431A-L06-F'07
3 Oct; Wed
admin:

Reminding you to do online quizzes.
How to go there:
www. whfreeman.com/lehninger/

register. Enter my email address:
gsantil@calstatela. edu

For chapt 1 AND 2 Deadline is
Wednesday 10/10/2007
11:59 pm PDT

Last time: buffers; thermodynamics;
started DNA
Looked at ΔG in more detail as applied to chem rxns.
ATP as energy currency - coupling agent to drive
nonspontaneous rxns. ATP structure.

(1) Terms comparing genes:

Homologous genes = genes similar in sequence;
the proteins they encode =homologs

2 homologous genes in <u>same</u> species:
the species are "paralogous"
their protein products are *paralogs*

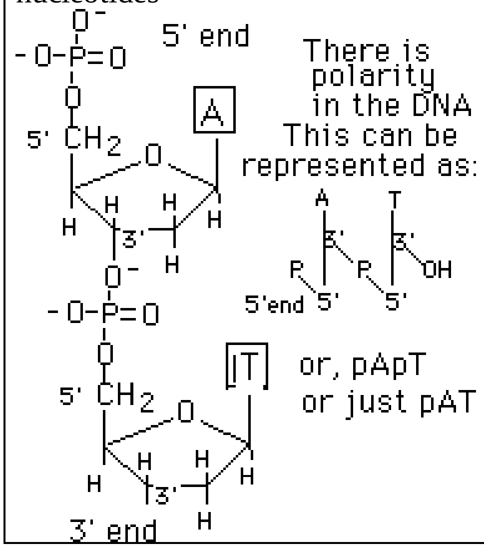
(probably due to gene duplication and these 2 genes
diverge gradually from mutations)

2 homologous genes or proteins found in different
species : orthologous, orthologs

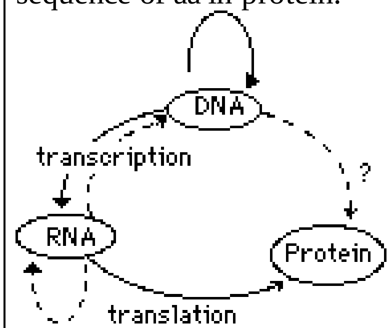
the variation between homologous genes is a
measure of the divergence of the species

Nucleic acids: Now we look at the structure of major classes of molecules:

2) STRUCTURE: There are 2 kinds of m
a) DNA structure: very long molecules, thousands of nucleotides



1) Why is it important?
Central dogma of molecular genetics:
3 steps in the processing of genetic information:
a) replicatn: parent dna -> daughter DNA
b) transcription: DNA's genetic message rewritten in RNA
c) translation: RNAs genetic message transl'd into a sequence of aa in protein.



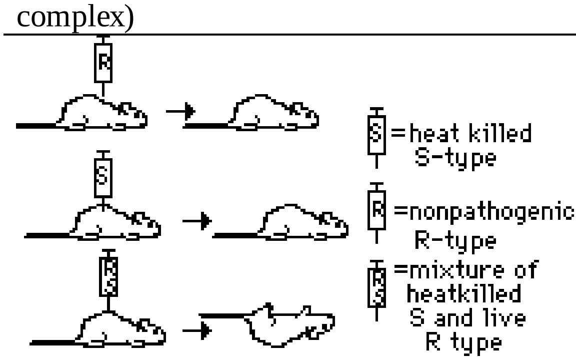
Note that there are 2 kinds of Nucleic Acids: DNA and RNA

4 types of deoxynucleotides used. What differs from organism to organism is the sequence of the bases. all organisms use the same 4 bases.ATGC (note thymine); sugars: deoxyribose
eg chromosome of procaryotes is made up of a single dna molec
chromosome of eukaryotes is made up of a single DNA molecule + some proteins

eucaryotic cells typically have many chromosomes.
structure of DNA linkage: has polarity: there's the 5' and the 3' end: In denoting the sequence, start w/ 5' end --> 3' end.
b) RNA structure: long molecs but not as long as dna. more abundant than dna : also have 4 bases: AUGC. (uracil instead of thymine); sugars ribose

History:
Experimental evidence for DNA being the genetic material:
As early as 1868 DNA (called nuclein) was isolated from cell nuclei. Although many suspected DNA to be the genetic substance, most people believed protein to be the real hereditary material because NA only had 4 kinds of bases. (they expected something more complex)

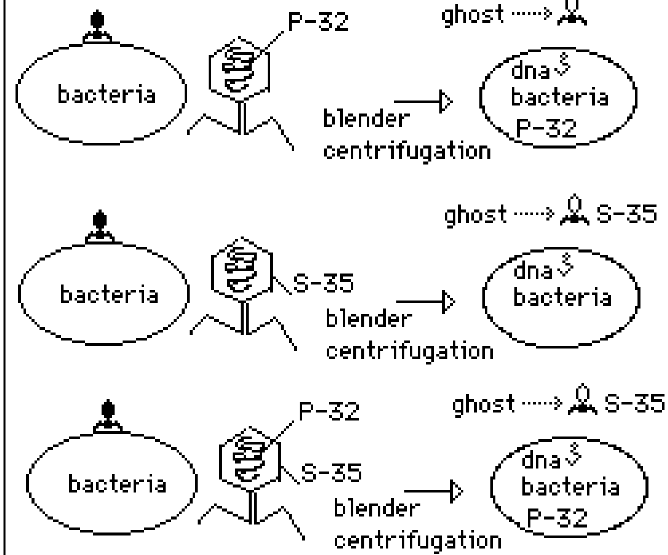
Evidence for DNA (1943):
Fred Griffith (1928): discovered that if inject mice with heat-killed S-type (encapsulated, virulent strain,smooth) pneumococcus bacteria mixed w/ live R-type (nonencap'd, nonpathogenic,rough) bacteria, the mice died and live S-type bacteria were obtained from the mice. But what was nature of "transforming principle"?



Avery, Macleod & McCarty:(Rockefeller Inst.,1944): interested in cell wall carbohyds

Maybe: it was the cell wall "primer carbohydrates" that contained the cell wall. Conclusion after 10 years: reported that the transforming principle was DNA. since the transforming principle was unaffected by proteases, lipid extraction, or ribonucleases but was affected by DNAses. Skeptical critics claimed that trace levels of protein were still present and could not be excluded as the transforming principle. (idea was not yet ready for acceptance)

Hershey-Chase experiment: Alfred Hershey & Martha Chases (1952) - Did experiment using bacteriophage T2 (phages are viruses that infect bacteria). Phages are made up of only two types of biomolecules: proteins and DNA. Of the two (or both), it was not clear what was the "transforming principle" that allowed the phages to multiply inside the bacteria. They used phages grown in either P-32 media or S-35 media allowing either only the DNA or the protein to be radiolabelled. P-32 labels only DNA since only DNA has phosphorus (as part of phosphate) and only proteins contain S-containing Amino acids. They found that only the P-32 is transmitted into the infected bacteria & in viral progeny.



Hershey-Chase Expt clinched the role of DNA as carrier of genetic info in phages and by extension, all other living orgs.

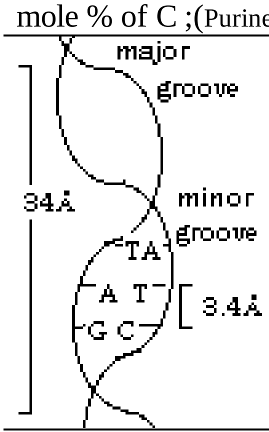
Other relevant evidence: Chargaff's rules: (Chargaff & coworkers, late 1940's, Columbia U.made observations on the nature and structure of DNA:

1) The base composition of a species is constant: %A,G,C& T is same for members of a species. A/T=1, C/G=1
2) Different species have different base compositions.
3) In a given species, the base composition does not vary with the age of the organism, its nutritional state, or changes in the environment
4) The mole % of A = mole % of T ; mole % of G= mole % of C ;(Purine/pyrimidine= 1)

There were other structural clues as to its structure: Xray diffraction studies showed that DNA fibers have two periodicities alont their long axis. 3.4 Å and 34 Å.

The X-ray diffraction data was collected by Rosalind Franklin, This was interpreted as being the diffraction coming from a helical molecule. Most of the elucidation of the structure of DNA came from model building studies. now known as B-DNA (ond of the various forms that DNA can take.

This is the 2° structure of DNA.

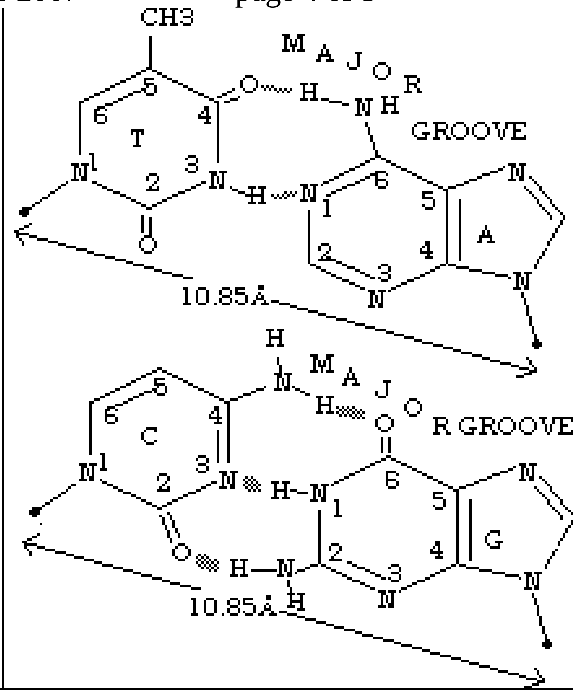


The features of the double stranded (duplex) DNA are that it is helical with two DNA strands coiled around the same axis to form a right handed double helix in which the 2 chains are antiparallel (5',3' phosphodiester bridges running in the opposite direction). The strands are held together by hydrogen bonds between the bases (separated by 3.4Å) and by hydrophobic interactions which put the polar phosphate groups and sugars on the outside interacting with the aqeuous environment. The H-bonds are specific between A & T and G & C. Thus the molar equivalence of these groups observed by Chargaff. It is a <u>double helix</u> in that it has a pitch of 34Å,ie 10 bp per turn - *on the average*..(actually varies as the sequence).

Show the overheads:
These paired base structures profoundly influenced the course of biology: immeidately suggested how genetic information is stored.

Essential features:
a) 2 polynucleotide chains running in opposite directions around a common axis to form a right-handed double helix.
b) Bases are on the inside, phosphate and deoxyribose sugars outside (slightly tilted)
c) Pairing is by precisely directed H-bonds. A=T (2 bonds) , G∫ C (3 bonds)

Helix has 2 grooves which are not the



same. It is not symmetric. Groove in which C(1')-helix axis-C(1') <180° is the *minor* groove. Other groove is *major* groove.

We note that GA pairs are too big to fit into the helix. TC pairs are too small to fit into the helix. How about the AC pair? It would not fit. If you want to practice it yourself, try it and see what you'll get. Real DNA apparently deviates from B-DNA. It was found by Dickerson and Drew by X-ray crystallography of dodecamer, d(CGCGAATTCGCG). that indeed the dodecamer crystallizes in the B-DNA form with an average helical rise per residue of 3.4 Å and an average of 10.1 bp per residue. The average twist per base pair is 35.5°. The deviation from the ideal double helix makes DNA irregular *in a sequence specific manner*: the helical twist per base pair ranges from 28-42°. There is also the presence of the "propeller twisting" (twisting in opposite directions about the base pair's axis. And also some base pair roll (tilting of the bp as a whole about its axis). It appears that this deviation is sequence-specific and may be very important in the ability of some DNA-binding proteins to bind in a sequence-specific manner.

Another type of change occurs when the humidity of a DNA prep is lowered to 75% humidity. It shifts to A-DNA. Segments of DNA in vivo may assume the A conformation. Another more different form has been shown to be possible: Z-DNA (stabilized at high salt for the double helix of the self complementary segment: d(CGCGCG). The presence of Z-DNA has been shown in Ecoli. Its function is not known.

We summarize the various forms of DNA in the table below:

	A	B	Z
Helix sense	right-handed	right-handed	left-handed
residues/turn	11	10	12
rise per residue	2.55	3.4	3.7Å

RNA can't assume B-DNA conformation because of steric hindrance involving the -OH at 2' position. But it can take conformation of A-DNA (ie "A-RNA" or RNA-11). rRNA and tRNA contain complementary sequences that form double helical stems. Hybrid double helices, which consist of one strand each of RNA and DNA, also have an A-DNA-like conformation.

most fundamental constituents of the cell
polymeric, containing monomers with phosphate, sugar and base. sugar=ribose/ribose derived.
base is either a purine or a pyrimidine ring.

2 types of nucleic acids:
RNA and DNA : show the overhead - polymeric
difference is in the ribose group of the molecule

their linkage is via phosphate group = phosphodiester linkage

diff types of purine and pyrimidine bases (ring systems)

* structure numbering system

adenine, guanine, thymine, cytosine, uracil

nucleosides (base + ribose)
nucleotides (base + ribose + phosphate) eg AMP

bases are capable of tautomerization.