DNA as genetic material?

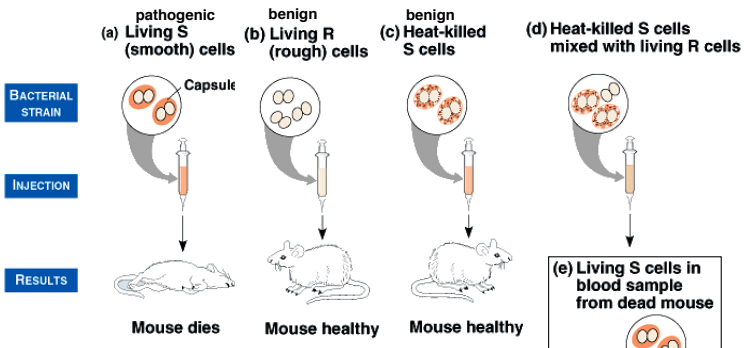
- Enzymes (proteins) correspond to genes(?)
- DNA in nucleus and chromosomes
- DNA vs. protein
- Does DNA carry traits to daughter cells or offspring?
- Does DNA have enough complexity to carry coding information?

Four Classic Experiments that proved DNA is the genetic material

1. Non-living chemical is heritable material in bacteria
2. DNA is heritable material in viruses
3. Nucleotide bases are paired in all organisms.
4. DNA structure is ideal for making copies.

Expt 1. Griffith & Avery

1927, 1944



Conclusion:
heritable material transferred from dead pathogenic to live benign bacteria = transformation

Expt. 1: Griffith and Avery

Live, pathogenic bacteria —mouse→ Dead mouse

Live, benign bacteria —mouse→ Live mouse

Dead, pathogenic bacteria —mouse→ Live mouse

Dead, pathogenic bacteria + Live, benign bacteria —mouse→ Dead mouse

Transformation:

pathogenic DNA → benign bacteria

Expt 2. Hershey & Chase

1952

T2 Bacteriophage: a virus that infects bacteria

Phage infects E. coli and turns E. coli into a phage producing factory

Therefore, phage must have genetic blue prints and uses bacteria to assemble new viruses from genetic plan.

Phage viruses are composed of DNA and protein. Is the genetic material in the protein, or in the DNA?

Phage

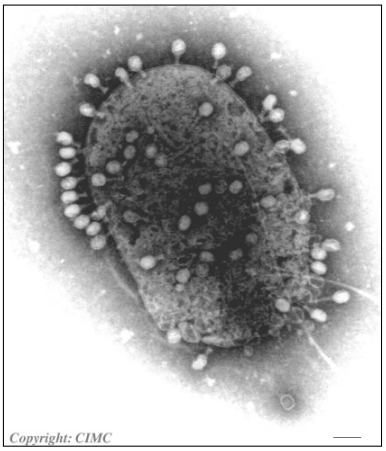
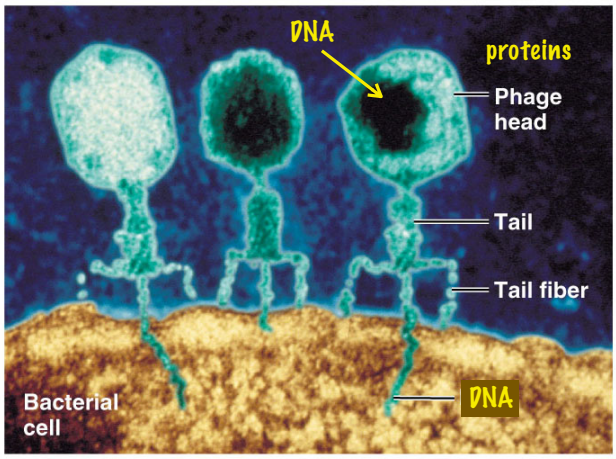


Figure 16.2a The Hershey-Chase experiment: phages



(a) T2 and related phages use their tail pieces to attach to the host cell and inject their genetic material (TEM).

Expt 2. Hershey & Chase

Phage viruses are composed of DNA and protein. Is the genetic material in the protein, or in the DNA?

In other words, does the phage inject the bacteria with protein or DNA?

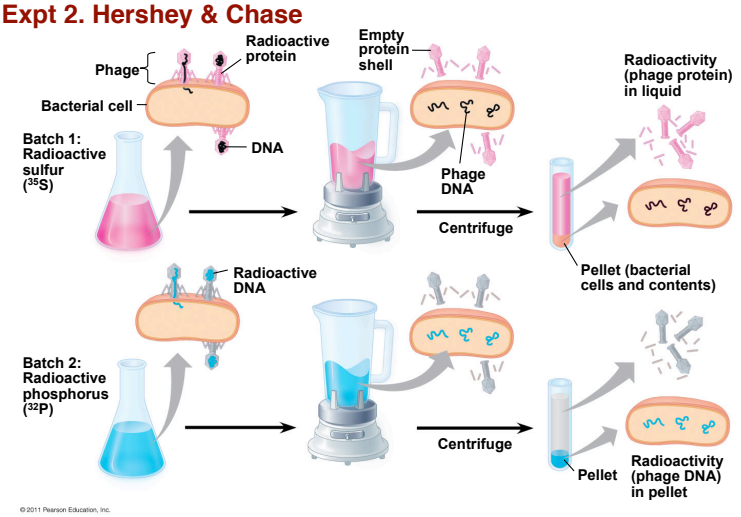
Use radioactive isotopes to label protein or label DNA.

Sulfur is an element only in protein: so use ^{35}S as label.

Phosophorus is an element in DNA: so use ^{32}P as label.

Figure 16.4-1

Expt 2. Hershey & Chase

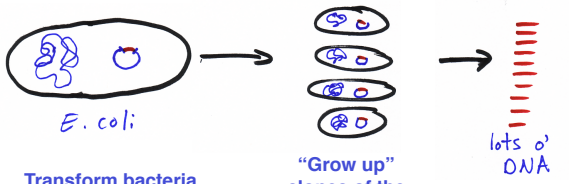


Biotech: Subcloning and Bacterial/Phage Amplification of DNA

Circular Plasmid DNA

Linearized plasmid DNA

DNA inserted into plasmid



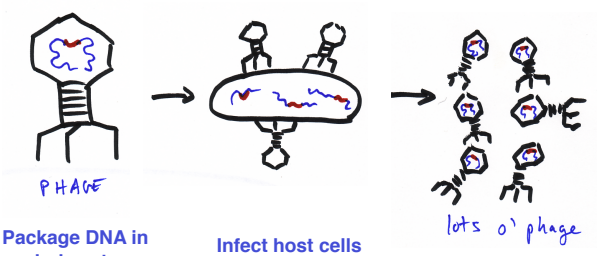
Transform bacteria

"Grow up" clones of the bacteria

or, lots o' Protein

e.g., human insulin

Phage Amplification

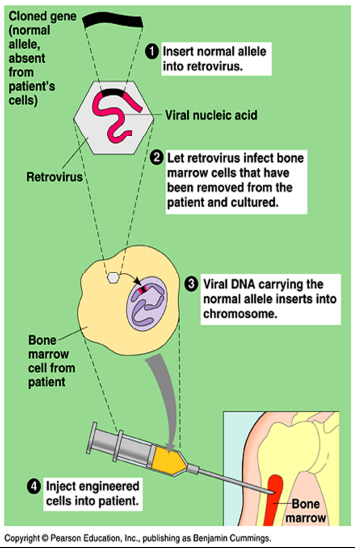


Package DNA in viral vector

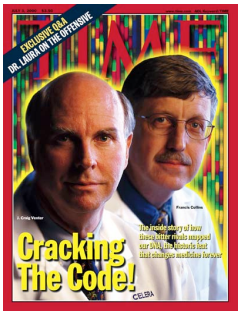
Infect host cells

But doesn't have to be a bacteriophage...

Viral Gene Therapy

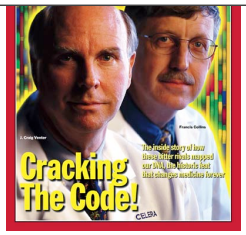


Genome Transplant by Craig Venter 2010



Genome Transplant by Craig Venter 2010

- **Sequence genome of bacteria**
- **Rearrange and alter sequence on computer (“in silico” vs. “in vivo” or “in vitro”)**
- **Make artificial genome in using DNA synthesis machines**
- **Insert new genome into host bacterium**
- **bacteria reproduces with artificial genome**



“Watermarks” in artificial genome

- **CRAIGVENTER** coded as:
  TTAACTAGCTAATGTCGTGCAATTGGAGTAGAGAACACAGAACGATTAACTAGCTAA
- **VENTERINSTITVTE** coded as:
  TTAACTAGCTAAGTAGAAAACACCGAACGAATTAATTCTACGATTACCGTGACTGAG
  TTAACTAGCTAA
- **HAMSMITH** coded as:
  TTAACTAGCTAACATGCAATGTCGATGATTACCCACTTAACTAGCTAA
- **CINDIANDCLYDE** coded as:
  TTAACTAGCTAATGCATAAACGACATCGCTAATGACTGTCTTTATGATGAATTAACTA
  GCTAATGGGTCGATGTTTGATGTTATGGAGCAGCAACGATGTTACGCAGCAGGGCAGT
  CGCCCTAAAACAAAGTTAAACATCATG
- **GLASSANDCLYDE** coded as:
  TTAACTAGCTAAGGTCTAGCTAGTAGCGCGAATGACTGCCTATACGATGAG
  TTAACTAGCTAA

For their latest research coup, the team went a bit further, adding not only their names but three other hidden messages within the code. One is an explanation of the coding system itself. The code also includes a handful of famous quotes ("TO LIVE, TO ERR, TO FALL, TO TRIUMPH, TO RECREATE LIFE OUT OF LIFE" from James Joyce's *A Portrait of the Artist as a Young Man* is just one of the appropriate selections) and a URL that ambitious genome geeks can decipher and visit as proof that they cracked the code.

http://www.popsci.com/science/article/2010-05/venter-institutes-synthetic-cell-genome-contains-hidden-messages-watermarks

1951

Expt 3. Chargaff's Rule

Chargaff analyzed base compostion of DNA from several species (bacteria to humans):

A: 30.9%

T: 29.4%

G: 19.9%

C: 19.8%

Number of A's = number of T's

Number of G's = number of C's

What could this mean?

Figure 16.UN04

Source	Adenine	Guanine	Cytosine	Thymine
E. coli	24.7%	26.0%	25.7%	23.6%
Wheat	28.1	21.8	22.7	27.4
Sea urchin	32.8	17.7	17.3	32.1
Salmon	29.7	20.8	20.4	29.1
Human	30.4	19.6	19.9	30.1
Ox	29.0	21.2	21.2	28.7

Expt 1. Griffith & Avery

Heritable material can be transferred from dead pathogenic to live benign bacteria = transformation

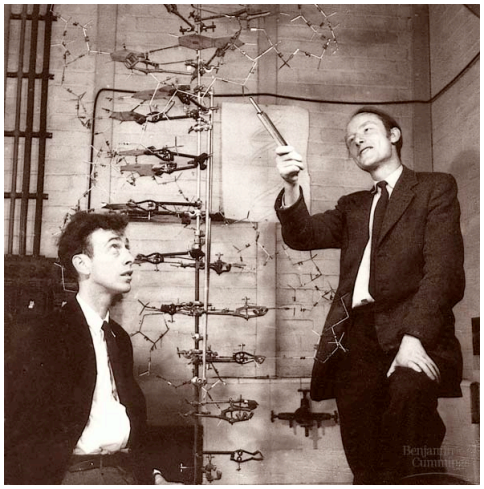
Expt 2. Hershey & Chase

Phage viruses are composed of DNA and protein. Using radioactive isotopes to label either protein or DNA, showed that DNA was heritable material of phage transfected into bacterial host.

Expt 3. Chargaff's Rule

Number of A nucleotides = number of T nucleotides
Number of C nucleotides = number of G nucleotides

Expt 4. Watson & Crick: The Double Helix



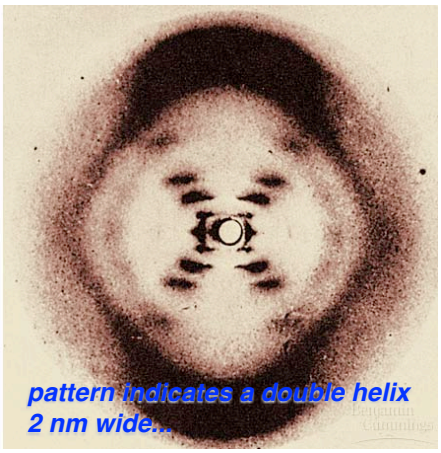
Expt 4. Watson & Crick: The Double Helix

Established key features of DNA:

1. **DNA is a polymer**
2. **Rosalind Franklin had X-ray crystallography data that DNA forms helix 2 nm wide**
3. **DNA is a double helix (based on width)**
4. **Sugar-phosphate backbone on the outside, with bases facing inside like the rungs of a ladder**
5. **Base-pairing explains Chargaff's Rule: A-T, C-G**

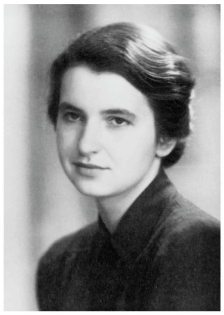
Figure 16.4 Rosalind Franklin and her X-ray diffraction photo of DNA

double helix (not single, not triple)
but how to put bases together?

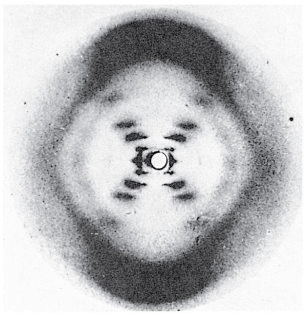


pattern indicates a double helix
2 nm wide...

Figure 16.6

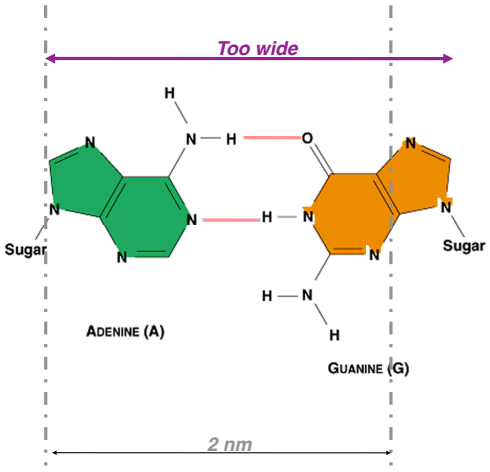


(a) Rosalind Franklin

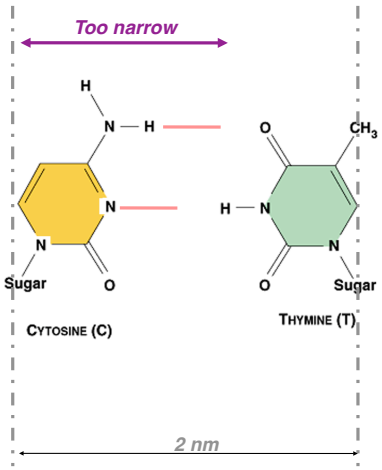


(b) Franklin's X-ray diffraction photograph of DNA

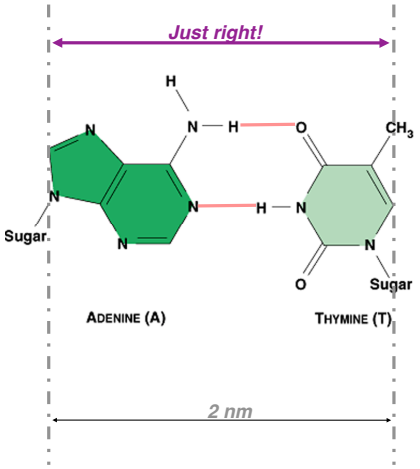
Base Pairing in Double Helix



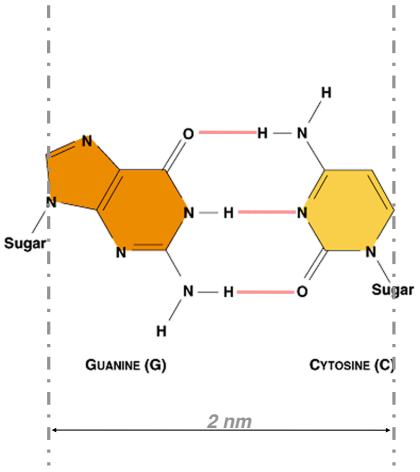
Base Pairing in Double Helix



Base Pairing in Double Helix



Base Pairing in Double Helix



Purine + purine: too wide

Pyrimidine + pyrimidine: too narrow

Purine + pyrimidine: width consistent with X-ray data

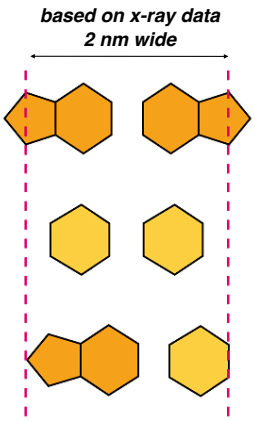
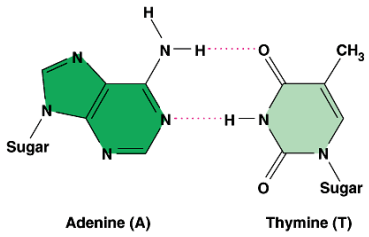
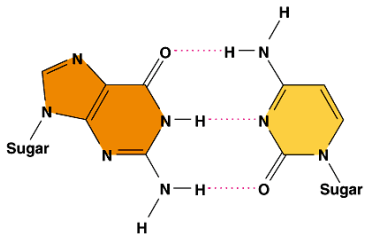
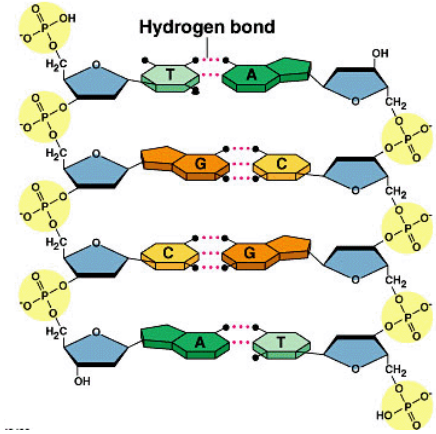


Figure 16.6 Base pairing in DNA

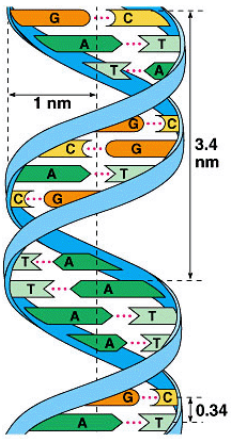


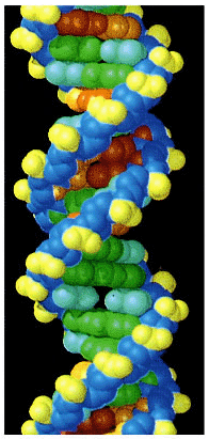


Expt 4. Watson & Crick: The Double Helix



Expt 4. Watson & Crick: The Double Helix





Put the bases on the inside of the helix

Watson & Crick

(*Reprinted from Nature, Vol.* 171, *p.* 737, *April* 25, 1953)

MOLECULAR STRUCTURE OF NUCLEIC ACIDS

A Structure for Deoxyribose Nucleic Acid

WE wish to suggest a structure for the salt of deoxyribose nucleic acid (D.N.A.). This structure has novel features which are of considerable biological interest.

●●●

It has not escaped our notice that the specific pairing we have postulated immediately suggests a possible copying mechanism for the genetic material.

Molecular Structure of Deoxypentose Nucleic Acids

WHILE the biological properties of deoxypentose nucleic acid suggest a molecular structure containing great complexity, X-ray diffraction studies described here (cf. Astbury[1]) show the basic molecular configuration has great simplicity. The purpose of this communication is to describe, in a preliminary way, some of the experimental evidence for the polynucleotide chain configuration being helical, and existing in this form when in the natural state. A fuller account of the work will be published shortly.

Wilkins and Franklin, same issue of Nature.

We have also been stimulated by a knowledge of the general nature of the unpublished experimental results and ideas of Dr. M. H. F. Wilkins, Dr. R. E. Franklin and their co-workers at King's College, London. One of us (J. D. W.) has been aided by a fellowship from the National Foundation for Infantile Paralysis.

J. D. WATSON
F. H. C. CRICK