D

N

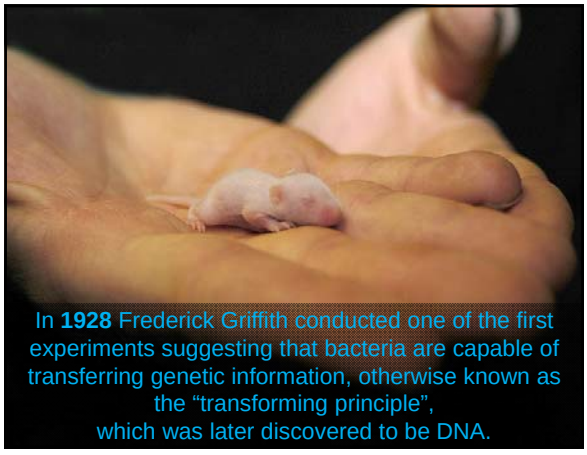
A

Deoxyribonucleic Acid

How do cells know what to do in your body?

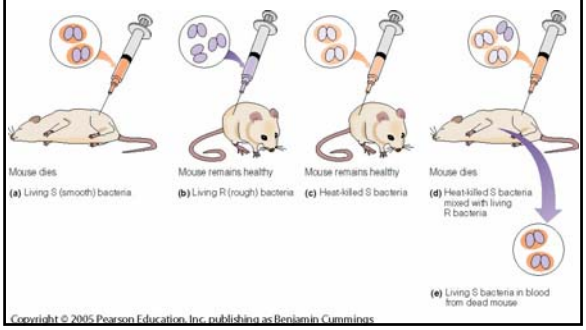
How can DNA determine what you will look like?

Why does DNA testing prove you committed a crime or that you're not my baby's daddy?



In **1928** Frederick Griffith conducted one of the first experiments suggesting that bacteria are capable of transferring genetic information, otherwise known as the "transforming principle", which was later discovered to be DNA.

Griffith's experiment on the transformation of Pneumococcus



But What Causes Transformation?

- Protein? – More subunits (20 amino acids)
  - More likely because it is found throughout the cell's cytoplasm.
  - Cells are constantly making proteins
  - Without the protein coat the bacteria is harmless
- DNA? – Fewer subunits (4 nucleotides)
  - Only found in the nucleus
  - Considered a boring molecule.

What Causes Transformation?

Today, we know that the DNA of the Smooth strain bacteria had survived the heating process, and was taken up by the Rough strain bacteria.

The S strain DNA contains the genes that form the protective polysaccharide capsule.

Equipped with this gene, the former R strain bacteria were now protected from the host's immune system and could kill it.

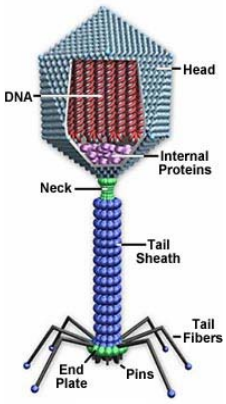
The first bacterial viruses were discovered in 1917 by scientists working independently in London and Paris.

The French scientist, Felix d'Herelle, was studying the feces of patients who had recovered from a bacterial dysentery (diarrhea). This somewhat unpleasant work led him to the discovery of an organism capable of killing bacteria.

He coined the term "bacteriophage," meaning eater of bacteria, to describe his discovery. d'Herelle was hopeful that this discovery would be useful in fighting disease.

The study of a Bacteriophages turned out to be crucial in establishing the identity of DNA as the genetic material of all living things.

Bacteriophage Structure



Meet the Bacteriophage

Certain bacterial viruses, such as the T4 bacteriophage, have evolved an elaborate process of infection.

The virus has a "tail" or landing section which it attaches to the bacterium surface by means of proteinaceous (meaning made of protein) "pins."

The tail contracts and the tail plug penetrates the cell wall and underlying membrane, injecting the viral nucleic acids into the cell.

After the viral nucleic acids are in the cell they takeover the nucleus and begin telling the ribosomes of the host cell to make copies of the bacteriophage's proteins instead of the ones belonging to the host cell.

Eventually the cell becomes so full of bacteriophages that it bursts open allowing them to infect more cells.

In 1952, American biologists Alfred Hershey and Martha Chase set out to determine what composed the genetic material of a bacteriophage.

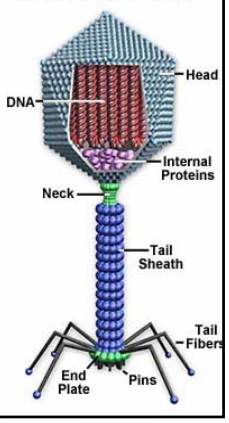
They knew that a bacterial virus was an extremely simple organism, composed only of protein and DNA.

The protein makes up the exterior of the virus, and the DNA is contained within it.

When a bacterium is infected by a bacteriophage, the bacterium's internal machinery falls under the control of the virus, which uses the bacterium to produce more viruses.

What Hershey and Chase wanted to know was: Which substance directed this takeover - DNA or protein?

Bacteriophage Structure

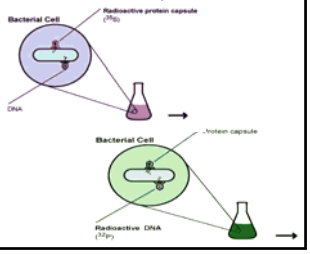


Hershey and Chase added bacteriophage to cultures containing either radioactive sulfur or radioactive phosphorus.

Alfred Hershey and Martha Chase. 1953

Hershey and Chase now had two types of bacteriophages:

1. one with a radioactive external protein coat,
2. the other with highly radioactive DNA.



Each of the two types of radioactive bacteriophage was added to a separate culture of bacteria.

The bacteriophages were allowed to infect the bacteria, then the cultures were whirled in **a kitchen blender**, the spinning blades caused any part of the bacteriophages **that hadn't got inside** the bacteria **to fall off** the cell.

Next the cultures were spun in a centrifuge, which separates materials suspended in liquid according to their weights.

The heavier bacterial cells fell to the bottom and formed a pellet, the lighter bacteriophages and loose phage parts remained in the liquid.

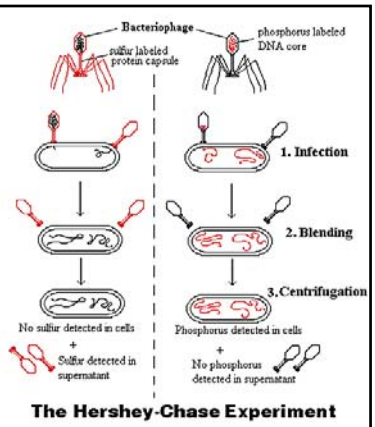
Where was the radioactivity now?

It depended on which radioactive element you looked for.

In the cultures infected by bacteriophages with radioactive sulfur (with labeled protein), most of the radioactivity was in the liquid with the phages.

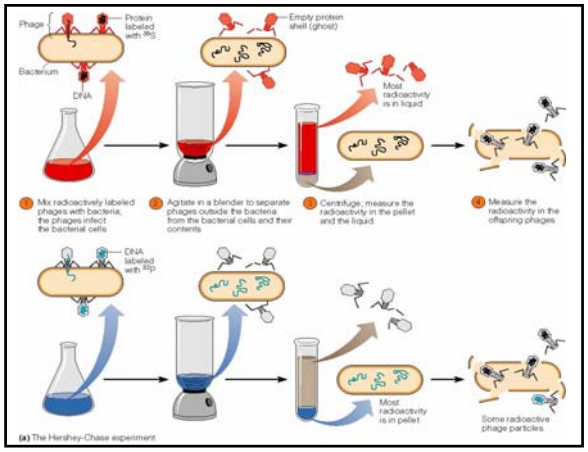
In the cultures infected by bacteriophages with radioactive phosphorus (with most of the label in their DNA), most of the radioactivity was in the pellet of infected bacteria.

The radioactive protein hadn't entered the bacterial cells, but the DNA had.



The Hershey-Chase Experiment

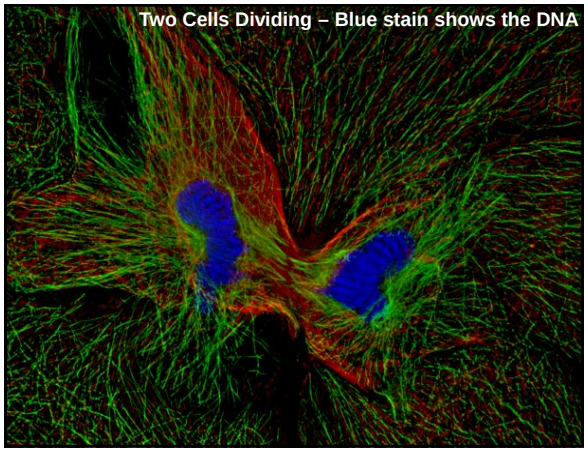
The final proof that DNA, not protein, was the genetic material was provided by the offspring of the phosphorus-labeled bacteriophages.



The final proof that DNA, not protein, was the genetic material was provided by the offspring of the phosphorus-labeled bacteriophages.

The offspring had radioactive DNA, passed down from their parents, but no radioactive protein.

These experiments convinced the scientific community that DNA alone was the material of heredity, and inspired Watson and Crick to begin their efforts to discover its structure.



Your body is made up of trillions of cells. Each of your cells can be very different from another. For example, they can specialize in a particular function, such as carrying oxygen (red blood cells), absorbing food (intestinal cells) or sensing light (cells in your eyes).

In other ways, your cells have a lot in common. For instance, at the center of almost all of your cells is a ball-shaped structure called the nucleus, inside of which are 46 thread-like structures called chromosomes. These chromosomes contain the estimated 25,000 genes that, in many ways, make us who we are.

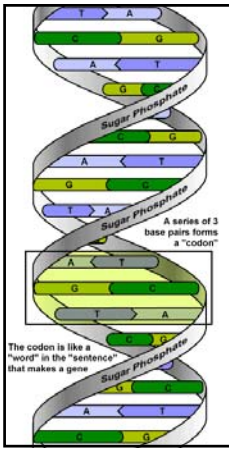
To understand how we end up with a given set of genes, we need to learn more about DNA and the chromosomes.

Instructions that provide almost all of the information necessary for a living organism to grow and function are in the nucleus of every cell.

These instructions tell the cell what role it will play in your body.

The instructions are in the form of a molecule called deoxyribonucleic acid, or DNA.

DNA is the chemical responsible for preserving, copying and transmitting information within cells and from generation to generation.



In humans, the DNA molecule consists of two ribbon-like strands that wrap around each other, resembling a twisted ladder. This is often described as **a double helix.** DNA is contained in tightly coiled packets called chromosomes, found in the nucleus of every cell. **Chromosomes** consist of the double helix of DNA wrapped around proteins.

The twisted ladder is made up of repeating units called **nucleotides**, each of which is a single building block of DNA. Nucleotides are composed of one sugar-phosphate molecule (the linear strands or outer rails of the ladder) and one base. DNA consists of two nucleotide strands joined by weak chemical bonds between the two bases, forming base pairs. A base pair is a rung or step on the ladder of the DNA. The bases are called A (for adenine), C (for cytosine), T (for thymine) and G for guanine.

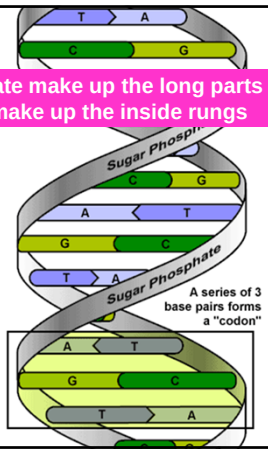
DNA Structure

Made up of a **nucleotide** (DNA molecule)

A nucleotide has 3 parts:

1. **Phosphate group**
2. Deoxyribose, a **5-carbon sugar**
3. **4 Nitrogenous bases**

The Sugar and the phosphate make up the long parts of the ladder; the bases make up the inside rungs

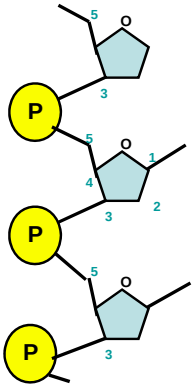


Deoxyribose

A 5-carbon sugar

"ose" = sugar
ribose, glucose, fructose

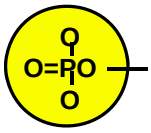
This helps make up the backbone of the ladder along with…..

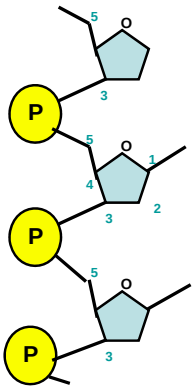


The Phosphate Group

- Phosphate joins with deoxyribose to form **the backbone** of the ladder

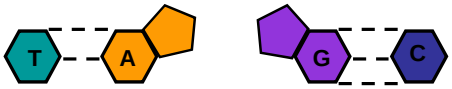
Phosphate Group





Chargaff's Rule

- **Adenine** must pair with **Thymine**
- **Guanine** must pair with **Cytosine**
- Their amounts in a given DNA molecule will be **about the same**.



Base pairing Rule

Adenine **always** pairs with Thymine

A – T

Guanine **always** pairs with Cytosine

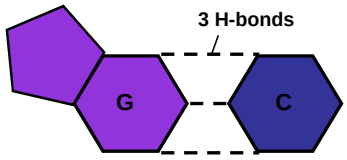
G – C

Always

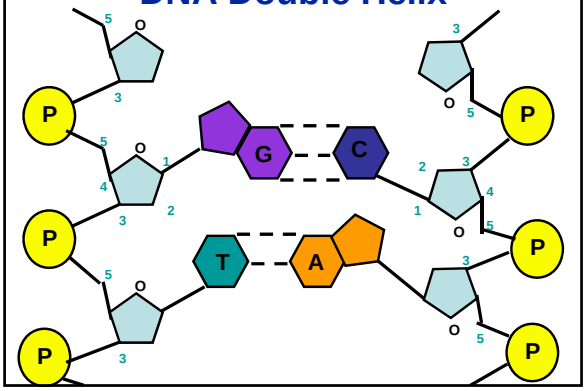
Always Always

BASE-PAIRINGS

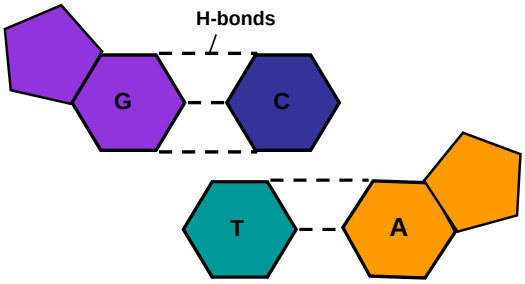
Purines	Pyrimidines	Base Pairs
Adenine (A)	Thymine (T)	A = T
Guanine (G)	Cytosine (C)	C ≡ G

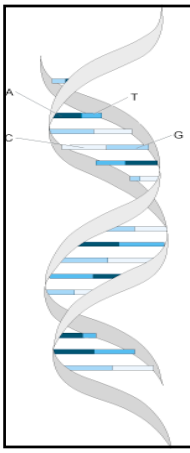


DNA Double Helix



BASE-PAIRINGS



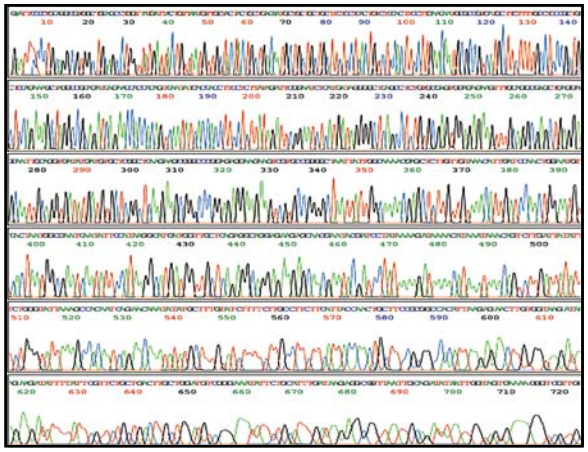


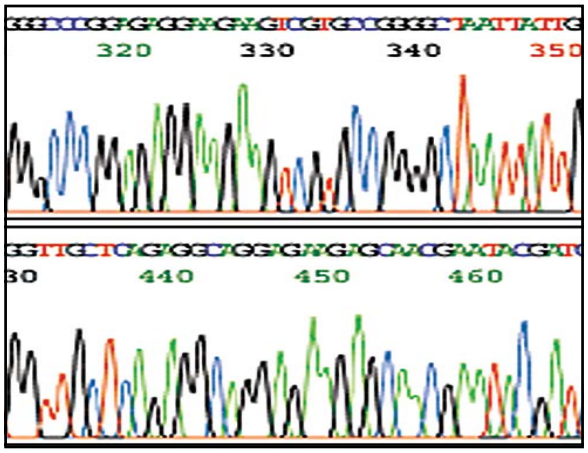
DNA is a large molecule packaged in chromosomes in the nucleus of cells.

The DNA molecule contains genes that direct the production of proteins by the ribosomes.

Proteins are molecules that play a critical role in the structure, function, and regulation of your body's cells, tissues, and organs. Every protein is made up of a chain of building blocks called amino acids.

http://genetics.gsk.com/flash_dna_actual.htm



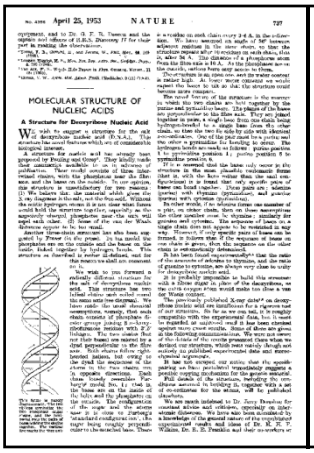

320
330
340
350
440
450
460

DNA is a Polymer

- Polymers like DNA and RNA can store information in their sequence of bases.
- HITS
- THIS
- What some of you are full of when you try to hand in work late even though you could have looked it up on the website or emailed me or… well you get the point.

DNA is a Polymer

- Polymers like DNA and RNA can store information in their sequence of bases.
- HITS vs THIS
- The order of the letters (or in the case of DNA the Nitrogen Bases) determines the meaning of the “word” or gene.



MOLECULAR STRUCTURE OF NUCLEIC ACIDS

A Structure for Deoxyribose Nucleic Acid

Watson and Crick
-The Double Helix
In late 1953, James Watson and Francis Crick presented the correct model of the structure of DNA.

It was already known from chemical studies that DNA was a polymer of nucleotides (sugar, nitrogen bases and phosphate).

X-ray crytallograpy data obtained by Rosalind Franklin, combined with the previous results from Chargaff and other chemists, were fitted together by Watson and Crick into the model published as a one page article in the journal Nature on April 25th 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.

A structure for nucleic acid has already been proposed by Pauling and Corey[1]. They kindly made their manuscript available to us in advance of publication. Their model consists of three inter-twined chains, with the phosphates near the fibre axis, and the bases on the outside. In our opinion, this structure is unsatisfactory for two reasons: (1) We believe that the material which gives the X-ray diagrams is the salt, not the free acid. Without the acidic hydrogen atoms it is not clear what forces would hold the structure together, especially as the negatively charged phosphates near the axis will repel each other. (2) Some of the van der Waals distances appear to be too small.

Another three-chain structure has also been suggested by Fraser (in the press). In his model the phosphates are on the outside and the bases on the inside, linked together by hydrogen bonds. This structure as described is rather ill-defined, and for this reason we shall not comment on it.

According to science historian Victor McElheny of MIT, the date this paper was published was a turning point in a longstanding struggle between two camps of biology, vitalism and reductionism.

While vitalists studied whole organisms and viewed genetics as too complex to understand fully, reductionists saw deciphering fundamental life processes as entirely possible—and critical to curing human diseases.

The discovery of DNA's double-helix structure was a major blow to the vitalist approach and gave momentum to the reductionist field of molecular biology

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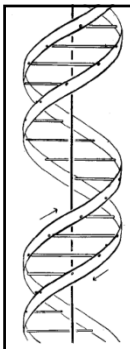
In other words the sugar phosphate backbone was on the inside and the bases to the outside… Watson and Crick are basically saying DUHHHH you got it backwards

A Triple Helix? Are you kidding me? We aren't even going to bother explaining how wrong you are.



This figure is purely diagrammatic. The two ribbons symbolize the two phosphate—sugar chains, and the horizontal rods the pairs of bases holding the chains together. The vertical line marks the fibre axis

We wish to put forward a radically different structure for the salt of deoxyribose nucleic acid. This structure has two helical chains each coiled round the same axis (see diagram). We have made the usual chemical assumptions, namely, that each chain consists of phosphate di-ester groups joining β-D-deoxy-ribofuranose residues with 3′,5′ linkages. The two chains (but not their bases) are related by a dyad perpendicular to the fibre axis. Both chains follow right-handed helices, but owing to the dyad the sequences of the atoms in the two chains run in opposite directions. Each chain loosely resembles Furberg's[2] model No. 1; that is, the bases are on the inside of the helix and the phosphates on the outside. The configuration of the sugar and the atoms near it is close to Furberg's 'standard configuration', the sugar being roughly perpendicular to the attached base. There

is a residue on each chain every 3·4 A. in the *z*-direction. We have assumed an angle of 36° between adjacent residues in the same chain, so that the structure repeats after 10 residues on each chain, that is, after 34 A. The distance of a phosphorus atom from the fibre axis is 10 A. As the phosphates are on the outside, cations have easy access to them.

The structure is an open one, and its water content is rather high. At lower water contents we would expect the bases to tilt so that the structure could become more compact.

The novel feature of the structure is the manner in which the two chains are held together by the purine and pyrimidine bases. The planes of the bases are perpendicular to the fibre axis. They are joined together in pairs, a single base from one chain being hydrogen-bonded to a single base from the other chain, so that the two lie side by side with identical *z*-co-ordinates. One of the pair must be a purine and the other a pyrimidine for bonding to occur. The hydrogen bonds are made as follows: purine position 1 to pyrimidine position 1; purine position 6 to pyrimidine position 6.

If it is assumed that the bases only occur in the structure in the most plausible tautomeric forms (that is, with the keto rather than the enol configurations) it is found that only specific pairs of bases can bond together. These pairs are: adenine (purine) with thymine (pyrimidine), and guanine (purine) with cytosine (pyrimidine).

In other words, if an adenine forms one member of a pair, on either chain, then on these assumptions the other member must be thymine; similarly for guanine and cytosine. The sequence of bases on a single chain does not appear to be restricted in any way. However, if only specific pairs of bases can be formed, it follows that if the sequence of bases on one chain is given, then the sequence on the other chain is automatically determined.

It has been found experimentally[3,4] that the ratio of the amounts of adenine to thymine, and the ratio of guanine to cytosine, are always very close to unity for deoxyribose nucleic acid.

It is probably impossible to build this structure with a ribose sugar in place of the deoxyribose, as the extra oxygen atom would make too close a van der Waals contact.

The previously published X-ray data[5,6] on deoxyribose nucleic acid are insufficient for a rigorous test of our structure. So far as we can tell, it is roughly compatible with the experimental data, but it must be regarded as unproved until it has been checked against more exact results. Some of these are given in the following communications. We were not aware of the details of the results presented there when we devised our structure, which rests mainly though not entirely on published experimental data and stereochemical arguments.

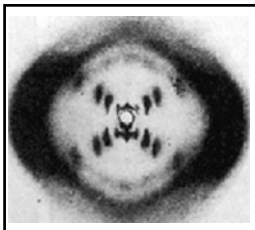
It has not escaped our notice that the specific pairing we have postulated immediately suggests a possible copying mechanism for the genetic material.

Full details of the structure, including the conditions assumed in building it, together with a set of co-ordinates for the atoms, will be published elsewhere.

We are much indebted to Dr. Jerry Donohue for constant advice and criticism, especially on interatomic distances. 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

Franklin was frustrated with an inhospitable environment at King's an institution that barred women from the dining room and other social venues, she was denied access to the informal discourse that is essential to any scientist's work.

Seeing no chance for a tolerable professional life at King's, Franklin decided to take another job. As she was preparing to leave, she turned her X-ray photographs over to her colleague Maurice Wilkins (a longtime friend of Crick).



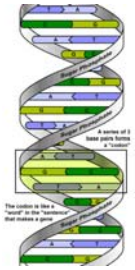
Then, in perhaps the most pivotal moment in the search for DNA's structure, Wilkins showed Watson one of Franklin's photographs without Franklin's permission. As Watson recalled, "The instant I saw the picture my mouth fell open and my pulse began to race." To Watson, the cross-shaped pattern of spots in the photo meant that DNA had to be a double helix.

Was it unethical for Wilkins to reveal the photographs?

Should Watson and Crick have recognized Franklin for her contribution to this paper? Why didn't they?

Would Watson and Crick have been able to make their discovery without Franklin's data?

DNA is copied using a process called **Replication**



Replication is the process of making two new identical copies of the original DNA molecule.

As Watson and Crick so cleverly alluded to, DNA acts as a template for its own replication.

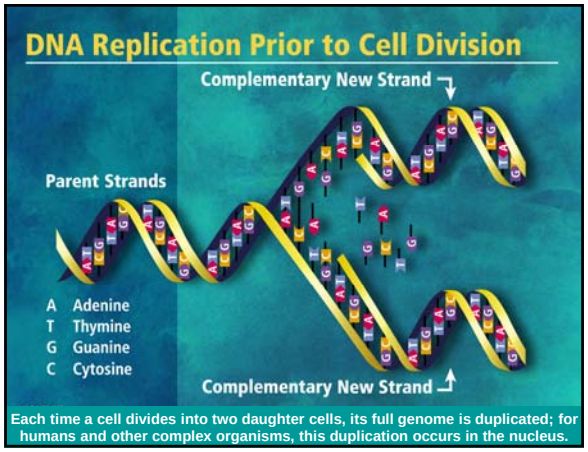
Since DNA strands are antiparallel and complementary, each strand can serve as a template for the reproduction of the opposite strand.

Translation:

Since the DNA molecule has two matching opposite sides (for every A on the left there is a T on the right) that obey the base pairing rule you can split the chain in half down the center and use each side as a guide for rebuilding the missing nucleotides.

DNA Replication

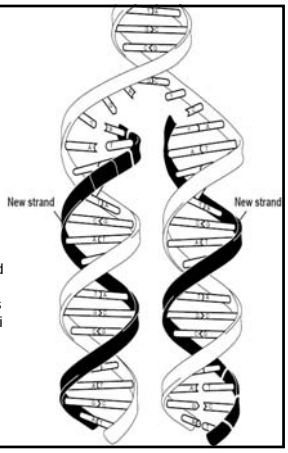
- The template strand is preserved as a whole piece and the new strand is assembled from spare nucleotides stored in the nucleoplasm.
- This process is called **semiconservative** replication. (semiconservative because ½ of the original molecule remains in the "new" DNA)
- **The two resulting strands should be identical,** although in reality there are always a few errors, though proofreading and error-checking mechanisms exist to ensure a very high level of fidelity.



Each time a cell divides into two daughter cells, its full genome is duplicated; for humans and other complex organisms, this duplication occurs in the nucleus.

DNA Replication

1. In the first step, the double helix is unwound by an enzyme called **Helicase**.
2. Next, a different enzyme called **DNA polymerase** attaches to one side of the split strand of the DNA. It moves along the strand, using it as a template for assembling a leading strand of new nucleotides and reforming a double helix.
3. A second DNA polymerase molecule is used to bind to the other template strand as the double helix opens. This molecule must synthesize discontinuous segments of nucleotides (called Okazaki fragments).
4. Lastly another group of enzymes check to make sure that there were no mistakes made… we will call these "**proofreading enzymes**"



Replicate the Following Strand of DNA

A-T-C-G-C-G-T-A-T-G-C-A-T-A-C-T-A-G

T-A-G-C-G-C-A-T-A-C-G-T-A-T-G-A-T-C

A-T-C-G-C-G-T-A-T-G-C-A-T-A-C-T-A-G

Step 1 – the enzyme Helicase unzips base pairs

Step 2 – the enzyme DNA Polymerase adds missing bases

Step 3 – Proofreading enzymes correct mistakes (to avoid mutations)