

## IN DEPTH FILM GUIDE

### DESCRIPTION

The film *The Double Helix* describes the trail of evidence James Watson and Francis Crick followed to discover the double-helical structure of DNA. Their model's beautiful and simple structure immediately revealed how genetic information is stored and passed from one generation to the next.

### KEY CONCEPTS

- A. DNA is a polymer of nucleotide monomers, each consisting of a phosphate, a deoxyribose sugar, and one of four nitrogenous bases: adenine (A), thymine (T), guanine (G), or cytosine (C).
- B. The relative amounts of A, T, G, and C bases vary from one species to another; however, in the DNA of any cell from organisms within a single species, the amount of A is equal to the amount of T and the amount of G is equal to the amount of C. This finding can be explained by the fact that in the DNA double helix, A pairs with T and G with C.
- C. Even before the structure of DNA was solved, studies indicated that the genetic material must be able to store information; be faithfully replicated and be passed on from generation to generation; and allow for changes, and thus evolution, to occur. The structure of the double helix immediately showed that DNA had these properties.
- D. Scientists use different techniques to measure things that are too large or too small to see. The structure of DNA was determined by combining mathematical interpretations of x-ray crystallography data and chemical data.
- E. Scientists build models based on what they know from previous research to derive testable hypotheses. Data from experiments are used to revise models and ask additional research questions. The ultimate goal is to find a model that is valid in all or most of the observations.
- F. The process of scientific discovery involves brainstorming and evaluating ideas, making mistakes, and rethinking those ideas based on evidence. Failure is an important aspect of scientific discovery.
- G. Communication among scientists plays a crucial role in scientific discoveries. To unlock the structure of DNA, Watson and Crick also relied on observations made by other scientists.

### CURRICULUM AND TEXTBOOK CONNECTIONS

Curriculum	Standards
NGSS (April 2013)	MS-LS3.A, MS-LS3.B, MS-ETS1.B, MS-PS1.B, HS-PS2.B, HS-PS4.C, HS-LS1.A, HS.LS3.A, HS.LS3.B
AP (2012–13)	3.A.1, 4.A.1
IB (2009)	3.3, 3.4, 7.1, 7.2

Textbook	Chapter Sections
Miller and Levine, <i>Biology</i> (2010 ed.)	12.1, 12.2, 12.3
Reese <i>et al.</i> , <i>Campbell Biology</i> (9th ed.)	5.5, 16.1, 16.3

### KEY TERMS

chemical bonds, chromosomes, DNA, genes, helix, hydrogen bonds, mutation, nucleotides, nucleus, proteins, structure, replication

### PRIOR KNOWLEDGE

It may be helpful for students to

- know that biological molecules are composed of different types of atoms, including carbon, oxygen, nitrogen, and hydrogen atoms;

- know that the shapes of biological molecules depend on the arrangement of the component atoms and their chemical bonds, which constrain the distances between atoms;
- know that genes are made of DNA, that they are inherited from one generation to the next, and that mutations are changes in the DNA sequence;
- have a basic understanding of DNA replication and the central dogma that DNA is transcribed to RNA and RNA is translated into proteins; and
- be familiar with the scientific process of testing ideas with evidence.

## PAUSE POINTS

	Begin	End	Content description	Review Questions	Standards
1	0:00	4:08	<ul style="list-style-type: none"> <li>• In the early 20<sup>th</sup> century several scientists were trying to solve the mystery of the structure of DNA in order to better understand inheritance of traits.</li> <li>• James Watson &amp; Frances Crick met in Cambridge in 1951. Both were interested in finding the structure of the gene.</li> <li>• In the 1920s, genes had been located in the nucleus and associated with chromosomes. Scientists needed to determine if genes were in proteins or DNA.</li> <li>• DNA is a polymer of nucleotide monomers, each consisting of a phosphate, a deoxyribose sugar, and one of four nitrogenous bases: adenine (A), thymine (T), guanine (G), or cytosine (C).</li> <li>• Oswald Avery demonstrated that DNA could carry genetic information.</li> </ul>	<ul style="list-style-type: none"> <li>• What are chromosomes made of?</li> <li>• Where are genes found?</li> <li>• What is the structure of DNA?</li> <li>• Why was Oswald Avery's work significant to Watson and Crick?</li> </ul>	<p><a href="#">NGSS (April 2013)</a> MS-LS3.A, MS-LS3.B, MS-ETS1.B, MS-PS1.A, HS-LS1.A, HS-LS3.A, HS-LS3.B</p> <p><a href="#">AP Biology (2013)</a> 3.A.1, 4.A.1</p> <p><a href="#">IB Biology (2009)</a> 3.3, 3.4, 7.1, 7.2</p>
2	4:09	9:05	<ul style="list-style-type: none"> <li>• X-ray crystallography is a technique for determining molecular structure. It can determine the location of atoms within a molecule.</li> <li>• Maurice Wilkins, a physicist at Kings College, was using X-ray crystallography to determine the structure of DNA.</li> <li>• Rosalind Franklin was a colleague of Wilkins, but she and Wilkins worked separately.</li> <li>• Linus Pauling was also searching for DNA's structure.</li> <li>• Pauling, Watson and Crick believed DNA was a helical molecule. Watson and Crick's first model of DNA was incorrect. They relied on information from other scientists.</li> </ul>	<ul style="list-style-type: none"> <li>• Why was x-ray crystallography being used to determine the structure of DNA?</li> </ul>	<p><a href="#">NGSS (April 2013)</a> MS-LS3.A, MS-ETS1.B, MS-PS1.A, HS-PS4.C, HS-LS1.A, HS-LS3.A,</p> <p><a href="#">AP Biology (2013)</a> 3.A.1, 4.A.1</p> <p><a href="#">IB Biology (2009)</a> 3.3, 7.1</p>
3	9:06	13:50	<ul style="list-style-type: none"> <li>• The structure of DNA was determined by combining mathematical interpretations of x-ray crystallography data and chemical data.</li> <li>• Wilkins showed Watson an x-ray crystallography picture taken by Franklin (Photo 51). The X-shaped diffraction pattern is characteristic of a helical molecule.</li> <li>• Chargaff had reported that base ratios were always the same in all organisms.</li> <li>• Watson and Crick built a model of DNA as a double helix, with the bases arranged inside.</li> </ul>	<ul style="list-style-type: none"> <li>• What is the structure of DNA?</li> <li>• What were the key pieces of evidence that led Watson and Crick to determine that structure?</li> </ul>	<p><a href="#">NGSS (April 2013)</a> MS-LS3.A, MS-ETS1.B, MS-PS1.A, HS-PS2.B, HS-PS4.C, HS-LS1.A, HS-LS3.A</p> <p><a href="#">AP Biology (2013)</a> 3.A.1, 4.A.1</p> <p><a href="#">IB Biology (2009)</a> 3.3, 3.4, 7.1, 7.2</p>
4	13:51	16:53	<ul style="list-style-type: none"> <li>• Solving the structure had far-reaching implications for biology.</li> </ul>	<ul style="list-style-type: none"> <li>• How does DNA's structure explain the</li> </ul>	<p><a href="#">NGSS (April 2013)</a></p>

	<ul style="list-style-type: none"> <li>The complementary nature of the bases (A-T and G-C) provided a method for replicating a new complementary copy.</li> <li>DNA's structure revealed how genetic information is stored in the sequence of the bases and how mutations can happen.</li> </ul>	<p>stability of life?</p> <ul style="list-style-type: none"> <li>How does it explain the mutability of life?</li> </ul>	<p>MS-LS3.A, MS-LS3.B, MS-PS1.A, HS-PS2.B, HS-LS1.A, HS-LS3.A, HS-LS3.B</p> <p><u>AP Biology (2013)</u> 3.A.1, 4.A.1</p> <p><u>IB Biology (2009)</u> 3.3, 3.4, 7.1, 7.2</p>
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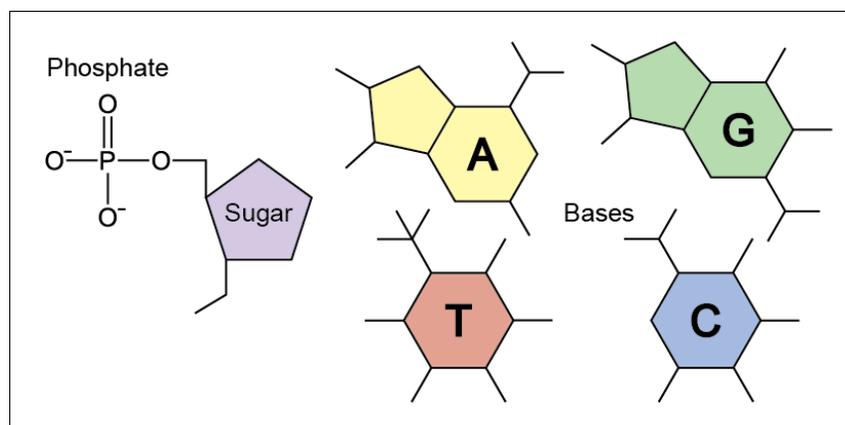
## BACKGROUND

The discovery of the three-dimensional structure of DNA was made possible by earlier work of many scientists who had uncovered evidence about heredity, genes, and DNA. The film *The Double Helix* mentions many of these findings, which are listed in more detail below.

The trail of evidence begins in the 19<sup>th</sup> century, when Austrian monk **Gregor Mendel** discovered patterns in the way characteristics, or traits, are inherited from one generation to the next. Doing experiments using garden pea plants, Mendel found that traits like pea shape and color are passed from parent to offspring as discrete units in a predictable way.

In the early 1900s, American geneticist **Thomas Hunt Morgan** demonstrated that these discrete units of heredity—or genes as they were by now called—were located on chromosomes. Chromosomes were known to be composed of DNA and protein, but it was unclear which of the two types of molecules was the source of genetic information.

Most researchers favored protein as the genetic material. Proteins are built from 20 distinct amino acid components and show great structural diversity and specificity. In comparison, DNA seemed monotonous. **Frederich Miescher**, a Swiss physician, had first isolated DNA from white blood cells in 1871. Shortly after that, American biochemist **Phoebus Levene** identified the components of DNA: deoxyribose sugar, phosphate, and one of four different nitrogenous bases.



**The nucleotide structure of DNA.** DNA consists of chains of nucleotides, which are made of a sugar, phosphate, and one of four bases. DNA seemed like a boring substance compared to proteins.

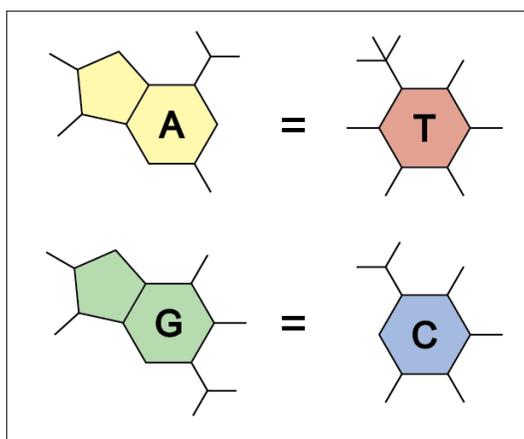
In 1938, British physicist **William Astbury** took the first x-ray diffraction images of DNA. He used these images to build a model of the structure of DNA using metal plates and rods. While his model was very tentative and contained errors, Astbury correctly positioned the bases lying flat, stacked like a pile of pennies, 0.34 nm apart.

A series of experiments set the stage for establishing that genes were made of DNA and not proteins. **Frederick Griffith's** 1928 experiments showed that pneumococcal bacteria could transfer genetic information between different strains through a process he called transformation. **Oswald Avery, Colin MacLeod, and Maclyn McCarty** determined that the

molecule responsible for this transformation was DNA and not protein. Avery and colleagues' 1944 paper was initially met with skepticism, as many scientists continued to believe that proteins were the genetic material.

In the meantime, more information was emerging about the structure of DNA. American biochemist **Erwin Chargaff** reported in 1949 that the proportions of the four nucleotides in a DNA molecule varied between species. However, within a species, the percentages of adenine (A) and thymine (T) bases were always equal, as were the percentages of guanine (G) and cytosine (C). The significance of this finding was not appreciated until Watson had the insight to use it to inform the model of DNA he was building.

Relative Proportions (%) of Bases in DNA				
Organism	A	T	G	C
Human	30.9	29.4	19.9	19.8
Chicken	28.8	29.2	20.5	21.5
Grasshopper	29.3	29.3	20.5	20.7
Sea Urchin	32.8	32.1	17.7	17.3
<i>E. coli</i>	24.7	23.6	26.0	25.7



**Chargaff's rule.** Erwin Chargaff discovered that in a DNA molecule, the proportion of adenine (A) always equals that of thymine (T) and the proportion of guanine (G) always equals that of cytosine (C).

The most convincing evidence that DNA was the molecule of heredity came from **Alfred Hershey** and **Martha Chase** in a paper published in 1952. Working at Cold Spring Harbor Laboratory on Long Island, NY, they used radioactive isotopes of sulfur and potassium to label proteins and DNA, respectively, in bacteriophages—which are viruses that infect bacteria. The experiment showed that the bacteriophage DNA, and not the proteins, entered bacteria for infection.

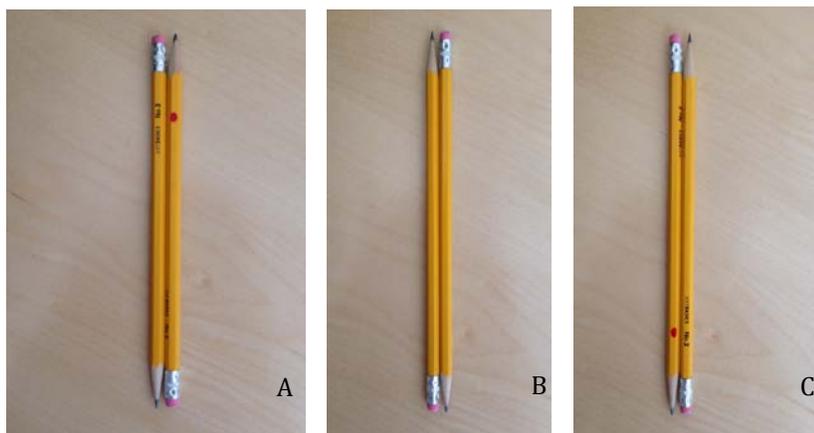
At the time of the Hershey-Chase experiment, a number of groups had started working to determine the molecular structure of DNA. Among them was **Linus Pauling** of Cal Tech, famous for having solved the structure of several proteins by building models based on chemical bonding principles and biochemical evidence. He was an inspiration to Watson and Crick as well as the person most likely to solve the structure before them. In 1951, Pauling had proposed that the polypeptide chains of proteins fold in  $\alpha$ -helical structures. Today, the  $\alpha$ -helix is known to form the backbone of tens of thousands of proteins.

Pauling had turned his model-building skills to the structure of DNA. In England, **Maurice Wilkins** and **Rosalind Franklin** at King's College London were using x-ray crystallography to analyze DNA's structure. Despite a few confusing blurry spots, the initial images they obtained hinted that DNA might come in the form of a twisted spiral—or helix. However, it was not clear how the phosphates, sugars, and bases were arrayed within that helix.

Shortly after Wilkins and Franklin began their experiments, Watson and Crick decided to work on DNA as well. Inspired by Pauling's work, they started building models of DNA molecules. Unlike Wilkins and Franklin's, their approach was to formulate a possible structure of DNA and then determine whether it fit experimental observations. In one of their first attempts, they created a helix with three sugar-phosphate chains held together by chemical bonds facilitated by magnesium ions, with the bases projecting outward from this central backbone. Franklin saw that the model did not fit the x-ray evidence. Based on her measurements, DNA fibers contained at least 10 times as much water as Watson and

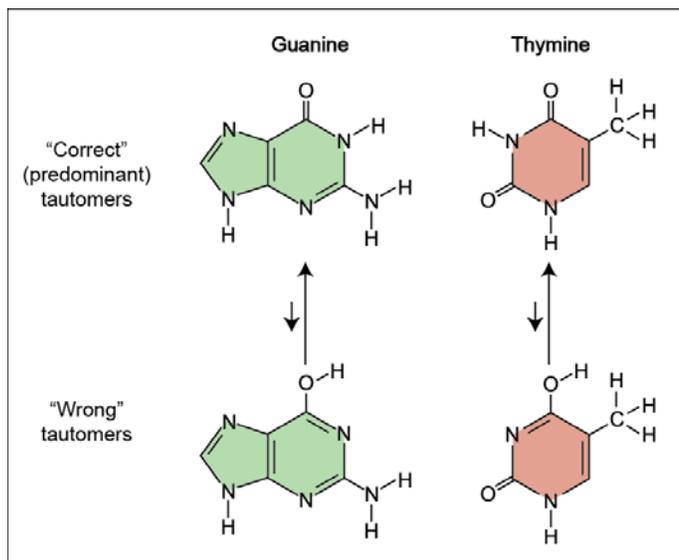
Crick's model allowed for, and there was no evidence that DNA was associated with magnesium ions. As constructed, the model could not explain how the three phosphate backbones could be held together at the center of the molecule. (Shortly before Watson and Crick produced their successful double-helix model, Pauling produced his own flawed triple-helix model, again with the phosphates in the center of the molecule, but without magnesium ions. Pauling had hydrogen bonds holding the phosphate chains together, but based on what was known about chemical bonds that model was not valid.)

Franklin's measurements of the water associated with DNA suggested that the phosphate groups would be located in an aqueous environment on the exterior of the DNA molecule. Ever-better x-ray images, including Franklin's famous photo B51, provided information about the dimensions of the repeating subunits in a DNA molecule. In addition, her images indicated that DNA molecules look the same when they are turned upside down and front to back. When Crick found out about DNA's dyad symmetry, he inferred that the phosphate chains must run in opposite orientations, or antiparallel, to one another—a brilliant insight that Franklin and others had missed.



**DNA molecules have dyad symmetry.** One way to explain dyad symmetry is to pretend that the two pencils in (A) are a DNA crystal. When flipped upside down (B) and front to back (C), it looks the same as the original (A).

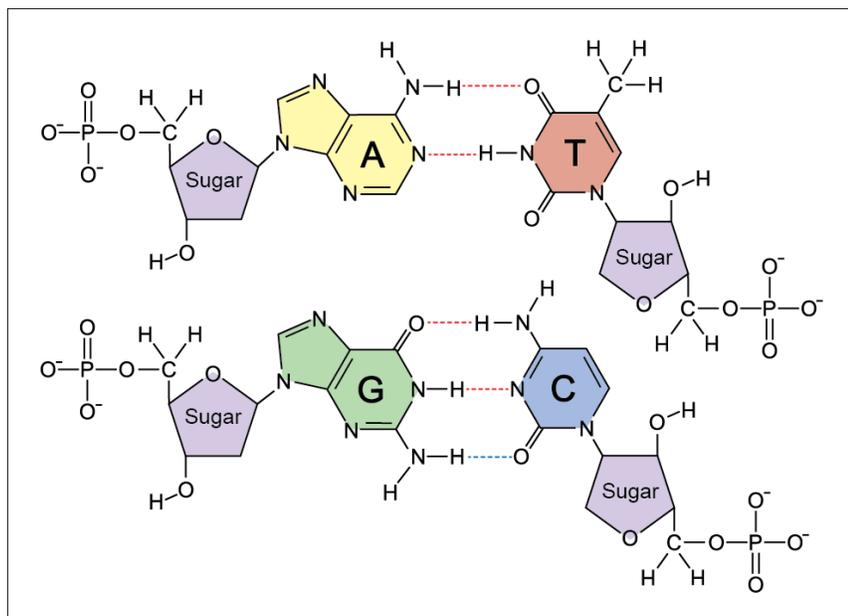
Building on these clues and Wilkins and Franklin's measurements, Watson and Crick once again turned to models to test their hypotheses of DNA structure. This time they tried building models with two antiparallel phosphate chains on the outside of the molecule. In this arrangement, the chains would have to be held together by the bases on the inside, but Watson and Crick were not sure how these bases might pair up. That's when they remembered Chargaff's ratios. Crick reasoned that A must always pair with T and G with C. But where were the bonds between these bases? They consulted J. N. Davidson's *The Biochemistry of Nucleic Acids*, published in 1950. However, as with other books of that time, it contained incorrect forms, or tautomers, of guanine and thymine (see figure). No matter how they tried, the bases did not form a nice hydrogen-bonding pattern, as for example in the protein backbone of an  $\alpha$ -helix. A visiting American chemist, **Jerry Donohue**, looked at their base structures and realized that these were wrong.



**The textbooks were wrong.** Guanine and thymine can have alternate molecular structures based on different locations of a particular hydrogen atom. These two types of structures are known as tautomers. The tautomeric forms of each base exist in equilibrium, but one form is more stable and therefore predominates under the conditions found inside most cells. In the early 1950s, chemistry textbooks had drawings of the “wrong” tautomers of guanine and thymine. Jerry Donohue told James Watson what the correct structures were. This piece of the puzzle allowed Watson to build pairs of A’s and T’s and of G’s and C’s with accurate hydrogen bonds.

Once Watson incorporated the new, correct shapes of the bases into his model, he saw where the hydrogen bonds would form and his model of DNA immediately fell into place!

The A-T and G-C pairings were consistent with the measurements of the molecule from x-ray images. The hydrogen bonds between the base pairs make the molecule structurally stable. Today we know that there are two hydrogen bonds in an A-T pair and three hydrogen bonds in a G-C pair. However, at the time Watson and Crick built their model they had initially identified two hydrogen bonds between both A-T pairings and G-C pairings.



**Base pairings used to construct the double helix.** Watson and Crick rejected the idea of a third hydrogen bond (shown by the dotted lines) between guanine and cytosine because data hinted that such a bond would be weak. Later evidence showed that there are in fact three strong hydrogen bonds in a guanine-cytosine pair. (The third bond is shown in blue.)

### Key Evidence Came from X-ray Diffraction

Developed early in the 20<sup>th</sup> century, x-ray crystallography allows the indirect observation of molecular structures too small to be seen or photographed. The father-son team of William H. and William L. Bragg shared the Nobel Prize in 1915 for using x-rays to reveal how the repeating structures of crystals form. Table salt was the first crystal structure solved by x-ray crystallography in 1914, soon followed by the repeating carbon structure of diamond. Dorothy Hodgkin and Max Perutz were pioneers in solving the structures of organic molecules containing more complex atomic arrangements, including cholesterol, penicillin, vitamin B12, insulin, and hemoglobin.

William L. Bragg was director of the Cavendish Laboratory at the time that Watson and Crick were there. He had been striving to determine the structures of protein components, but Linus Pauling got there first by discovering the structure of the  $\alpha$ -helix.

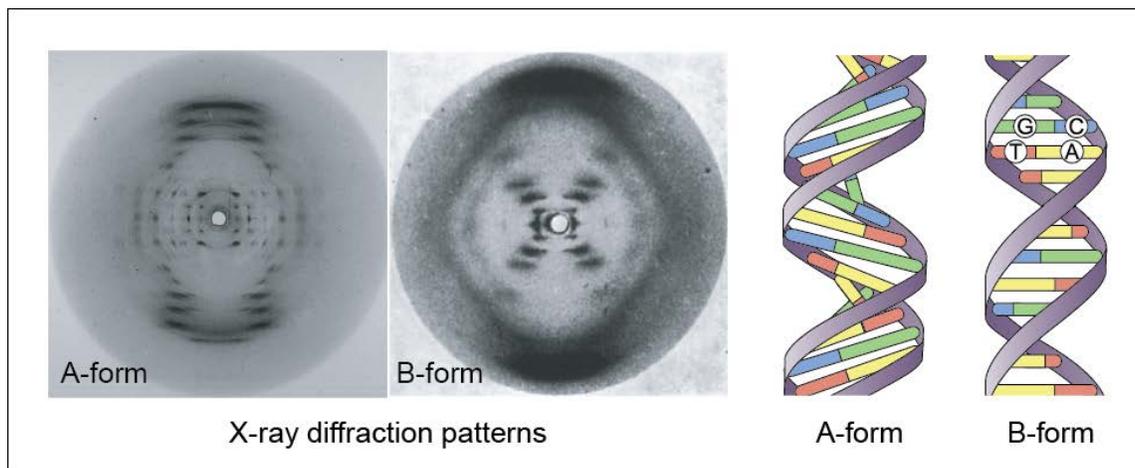
X-ray crystallography involves mounting a molecule on a stage and bombarding it with a beam of x-rays. The wavelengths of x-rays are so short that they bounce off atoms within the molecule, scattering at specific angles that depend on the distances between atoms of various sizes. The scattered x-rays produce patterns that can be captured on photographic film or digitally. To interpret these patterns, crystallographers must determine when x-rays scattered from different atoms overlap. This overlap changes the intensity of spots in the x-ray pattern.

Two-dimensional images taken at different angles are converted into a three-dimensional model of the molecule using mathematical calculations termed Fourier transformations, which allow the positions of atoms within the molecule to be determined. When averaged over many observations, these measurements can be accurate to just a fraction of an ångstrom (or one 10-billionth of a meter).

The number of x-rays diffracted by a single molecule would simply be too small to be observed. Therefore, x-ray crystallography examines many identical molecules packed into a highly regular three-dimensional array rather than single molecules. Because the x-rays travel through many layers of atoms, it's important that the atoms always occur in the same arrangement in all the molecules in a sample. If they don't, the x-rays are bent into overlapping patterns, resulting in blurry or completely smeared diffraction patterns that cannot be interpreted or provide very poor resolution. Because crystals have a repeating arrangement of atoms in identical orientation, they leave a pattern of sharp, clear spots. For this reason, biological molecules are typically turned into crystals, or crystallized, before they are analyzed with x-rays. In fact much time and effort may be spent in "growing" crystals of a particular molecule of interest, and this can be the rate-limiting step in a research effort to solve a molecular structure.

Franklin took x-rays not of crystals but of DNA fibers that, although thin, consisted of thousands of individual DNA molecules tightly arrayed, somewhat like the individual strands of thick rope, or hair gathered into a pony tail. While not in crystal form, the DNA had a sufficiently regular arrangement to diffract x-rays in a way that produced decipherable patterns.

By exposing DNA to varying levels of atmospheric humidity, Franklin and her graduate student Raymond Gosling demonstrated that DNA existed in two forms, which they called A and B. The "dry" A form occurs when the relative humidity is less than 75% and produces a scattered x-ray diffraction pattern consisting of many distinct spots. Water molecules cling to the "wet" B-form DNA, causing the strands to elongate, producing an X-shaped diffraction pattern. Because all molecules in cells are immersed in liquid, the B-form DNA is the form that exists primarily inside cells. It was not until 1980 that an actual crystal structure of more than a complete turn of B-DNA, in which individual atoms of the DNA could be distinguished, was published (Wing, R. *et al.* 1980. *Nature* 287:755-8).

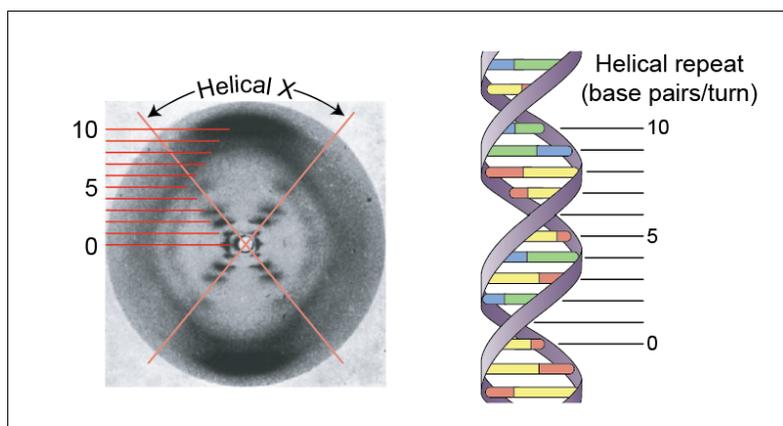


**The A and B forms of DNA.** The “dry” A form of DNA and the “wet” B form of DNA have distinct structures as shown by their x-ray diffraction patterns on the left.

Franklin had initially focused her attention on A-form DNA because she thought those images contained more information. It was in fact one of the A-form photos that had revealed that the two strands of DNA ran in opposite directions, although neither Franklin nor the others had been able to interpret the evidence to make this conclusion—it was Francis Crick who realized its significance.

The famous photo B51, taken by Franklin and Gosling in May 1952, was of B-form DNA. (As it was the 51<sup>st</sup> photo taken, Franklin labeled the image B51.) The X-shaped diffraction pattern is characteristic of a helical molecule.

Independent lines of evidence have confirmed that the diamond shapes formed by the arms and legs of the “x” indicate the repetition of the helical pattern as well as the placement of the phosphate sugar backbone on the exterior of the molecule and the bases in the interior. Analysis of the blurry smears composing the “x” of the photo allows the calculation of the dimensions of the DNA molecule: a radius of 1.0 nm, 0.34 nm between base pairs, and 3.4 nm (10 base pairs) within a complete turn of the helix. Although the model built by Watson and Crick was based on fiber diffraction, their knowledge of the chemical nature of the components of DNA allowed them to build a model that included the positions of atoms and the chemical bonds between them, which was later confirmed by high-resolution x-ray crystallography.



**Photo B51.** Photo B51 was taken by Franklin and Gosling. It revealed that B-form DNA was a double helix with 10 nucleotide base pairs within a complete turn of the helix. The cross indicates a helix. The dark patches indicate the bases.

### The Discovery of the Structure of DNA Led to Key Insights

On the morning of February 28, 1953, Watson constructed the model of DNA that unlocked the secret of life. Two months later, Watson and Crick published their findings in *Nature* in a paper titled “A structure for deoxyribose nucleic acid.” In the same issue, immediately following Watson and Crick’s paper, was a paper written by Wilkins and his colleagues Alec Stokes and Herbert Wilson. The third paper in the series was written by Franklin and Gosling. Wilkins’s and Franklin’s papers presented the evidence they had obtained, corroborating Watson and Crick’s proposed structure of DNA.

An understanding of the structure of DNA provided an explanation of how DNA functioned as the hereditary material. Watson and Crick noted this in their *Nature* paper. They wrote: “It has not escaped our notice that the specific pairing we have postulated immediately suggests a possible copying mechanism for the genetic material.” Because A is always paired with T and G with C, the order of bases on one strand determines the order on the other. Thus, if a DNA molecule were unwound, each strand could be copied into a complementary strand, producing an exact replica of the original molecule. Errors in the copying mechanism could result in mutations, or changes in the DNA sequence, that could be inherited by future generations.

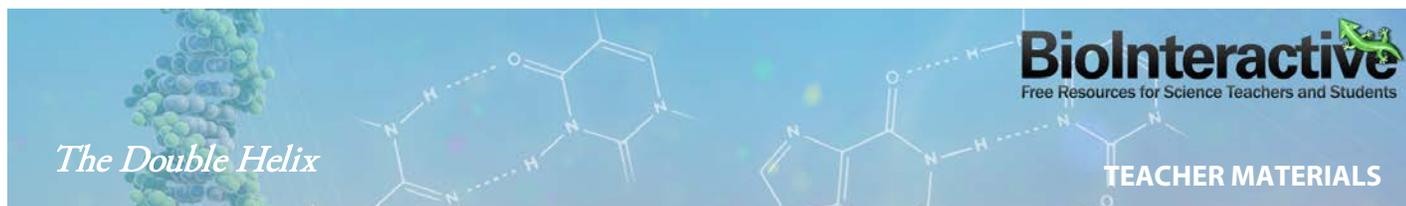
In 1962, Watson, Crick, and Wilkins were awarded the Nobel Prize in Physiology or Medicine “for their discoveries concerning the molecular structure of nucleic acids and its significance for information transfer in living material.” Franklin’s death in 1958 from ovarian cancer precluded her from receiving many of the honors for the discovery of DNA’s structure, including the possibility of sharing in the Nobel Prize, which cannot be awarded posthumously.

It took several years of subsequent study, including a classic 1958 experiment by American geneticists Matthew Meselson and Franklin Stahl, before the exact relationship between DNA structure and replication was understood (Meselson, M., and Stahl, F. 1958. The replication of DNA in *Escherichia coli*. *Proceedings of the National Academy of Sciences* **44**, 671–682).

### DISCUSSION POINTS

- The way the story unfolds in this short film may give students the impression that each piece of evidence fell into place one after the other, in a somewhat linear path. In reality most of the pieces were in play and moving around in Watson and Crick’s heads and in their discussions as they almost continuously thought about the problem of DNA over many months. The process was one of trial and error, and circling back repeatedly. After seeing photo B51, Watson suspected that DNA might be a double helix, but he continued to consider both a double or triple helix until he built a double-helical structure that fit all the known evidence. Similarly, Franklin’s x-ray data had suggested that the phosphate chains were on the outside, but that piece of evidence did not fit until Watson and Crick figured out how the nitrogenous bases might pair together at the center.
- At the beginning of the film, Sean Carroll sets the stage for the unfolding of the story by saying, “The three-dimensional arrangement of atoms in those molecules had to explain the stability of life, so that traits were passed faithfully from generation to generation, and also the mutability of life.” The notion that heredity could be explained by the “arrangement of atoms” of molecules was first raised by world-renowned physicist Erwin Schrödinger. In 1944, Schrödinger published the book *What Is Life?* in which he argued that living things should be considered in terms of molecular and atomic structure, as they obey the same laws of chemistry and physics. According to Schrödinger, genes are passed from generation to generation because the genetic code was a result of the arrangement of atoms within a molecule. These thoughts inspired a whole generation of researchers, including Watson and Crick. Before your students watch the film, you may want to discuss with them the importance of understanding three-dimensional molecular structures. Ask them “How does knowing the structure of any object tell you about its function?”
- There are many opportunities throughout the film to discuss the nature of scientific inquiry. For example, science tries to answer questions about the natural world. In the early 1950s, the principles of genetics were known, but no one knew what the genetic material was, let alone how any physical or chemical structure related to the consistency of inheritance and also the capacity to evolve new traits. Lead students in a discussion of the questions scientists were trying to answer by determining the structure of DNA.

- Individual scientists take different approaches in trying to understand a process or solve a problem, due in part to differences in training, the tools available where they work, and their personalities. Franklin and Wilkins wanted to solve the structure of DNA by obtaining x-ray diffraction data; they relied on experiments and observations. Watson and Crick built theoretical models which allowed them to see whether those models agreed with what was known about chemical bonding and x-ray data. Many scientists use a combination of data-gathering and hypothesis-testing approaches.
- Hypotheses must be tested and evaluated against evidence. Earlier researchers gathered evidence that DNA was the genetic material. Evidence from x-ray diffraction patterns and Chargaff's base-pairing ratios supported Watson and Crick's ball-and-stick model of the structure of DNA. Ask students to identify the key pieces of evidence that Watson and Crick used to construct their model of DNA. Some of the key evidence presented in the film includes the structure of the nucleotide (a phosphate linked to a sugar linked to one of four nitrogenous bases); Chargaff's ratios ( $A = T$  and  $G = C$ ); and x-ray diffraction images (showing that DNA is a helix and the molecule's dimensions).
- People do science, and make intuitive leaps of genius, while also sometimes missing the obvious and making outright mistakes. As Dr. Karolin Luger, a structural biologist and HHMI investigator at Colorado State University, points out, scientists cannot allow themselves to be paralyzed by fear of making mistakes, and although most scientists want to prove that their hypotheses are correct, formally speaking, refuting a hypothesis is also useful. Point out to students that both Pauling and Watson initially hypothesized a triple helix with the phosphates on the interior and the bases pointing out, and why that was based on good logic that did not however fit the data. As new evidence emerges, models are modified and sometimes rejected.
- The film contains many illustrations and animations of structures of DNA molecules and base pairings. Some of the DNA animations are based on historical models that are wrong; you might use these animations to test your students' knowledge of molecular structures. In the animation in which James Watson is building the DNA molecule and the bases start coming together, only two hydrogen bonds are shown between bases. Today we know that there are two hydrogen bonds in an A-T pair and three hydrogen bonds in a G-C pair. In another animation showing the DNA molecule replicating, both strands are being replicated in the same direction. Today we know that the two DNA strands are replicated in antiparallel directions, but the mechanism of DNA replication was unknown at the time of the double-helix discovery.
- Students might be interested in knowing who the people featured in the film are. **James Watson** is currently at Cold Spring Harbor Laboratory in NY; he is the only scientist who was involved in the original research who was interviewed in this film. The interviews with Francis Crick consist of historical footage obtained when Crick was at the Salk Institute in California. After leaving the Cavendish Laboratory, Watson joined the department of biology at Harvard University, where he contributed to an understanding of RNA's role in the transfer of genetic information. He went on to lead the Cold Spring Harbor Laboratory in NY, where he continues to conduct research focusing on the genetic basis of cancer and to write about science in his emeritus role. During the Clinton administration, Watson spearheaded the National Institutes of Health's Human Genome Project, an effort that ultimately involved numerous scientific leaders and sequencing centers, as well as hundreds of individual research labs. Watson published several leading textbooks, most notably *The Molecular Biology of the Gene*, as well as numerous popular books including the classic science discovery story *The Double Helix*. The film narrator, **Olivia Judson**, received her doctorate from Oxford and is an evolutionary biologist based at Imperial College London. She is well known for her 2002 book, *Dr. Tatiana's Sex Advice to All Creation*. In addition, she was featured in the NOVA documentary, *What Darwin Never Knew*, and wrote a weekly blog on evolutionary biology for the *New York Times* website. The three commentators in the film, who provided insights into the discovery and its significance, include Sean B. Carroll, Karolin Luger, and Robert Olby. **Sean B. Carroll** is a Howard Hughes Medical Institute investigator at the University of Wisconsin-Madison and HHMI vice president for science education. Carroll is an internationally recognized evolutionary biologist. His research focuses on the way new animal forms have evolved. His studies of a wide variety of animal species are revealing how changes in the genes that control animal development shape the evolution of body parts and body patterns. In addition to his research, Carroll is well known for his books *Making of the Fittest*, *Endless Forms Most Beautiful*, *Remarkable Creatures*, and *Into the Jungle*. He is also a co-author of the genetics textbook *Introduction to Genetic Analysis*. His most recent book, published in 2013, is entitled *Brave Genius*. **Karolin Luger** is an HHMI investigator at Colorado State University. Luger's research interests include the structural biology of chromatin, the complex of DNA and proteins that forms chromosomes in the nucleus of eukaryotic cells. In 1997, she determined the structure of the basic unit of DNA



packaging, or the nucleosome, which consists of a segment of DNA wrapped around histone proteins. Using this structure as a starting point, Luger's work has shed light on how the nucleosome changes shape, how chromatin interacts with the cell's transcription machinery, and how subtle changes in histones can affect overall nucleosome structure. **Robert Olby** is a science historian at the University of Pittsburgh. He is the author of *The Path to the Double Helix* and a biography of Francis Crick entitled *Francis Crick: Hunter of Life's Secrets*.

## RELATED BIOINTERACTIVE RESOURCES

**Teacher Guide: DNA** (<http://www.hhmi.org/biointeractive/teacher-guide-dna>)

This curriculum guide assists in filtering through the vast available resources from BioInteractive and HHMI, and organizes the material according to various topics related to DNA, including DNA structure.

**Building Blocks of DNA** (<http://www.hhmi.org/biointeractive/building-blocks-dna>)

This animation shows the four nitrogenous bases, adenine (A), thymine (T), guanine (G), and cytosine (C), that make up DNA.

**Paired DNA Strands** (<http://www.hhmi.org/biointeractive/paired-dna-strands>)

This animation shows that DNA has a double-helix structure. If untwisted, DNA would look like two parallel strands. Each strand has a linear sequence of A, T, G, and C bases. The precise order of the letters carries the coded instructions. One strand is a complementary image of the other: A always pairs with T, and G always pairs with C.

**Chargaff's Ratio** (<http://www.hhmi.org/biointeractive/chargaffs-ratio>)

This animation features Erwin Chargaff's 1950 paper stating that in the DNA of any given species, the ratio of adenine to thymine is equal, as is the ratio of guanine to cytosine. This became known as Chargaff's ratio, and it was an important clue for solving the structure of DNA.

**Pauling's Triple Helix Model** (<http://www.hhmi.org/biointeractive/pauling-triple-helix-model>)

This animation shows Linus Pauling's triple-helix model, one of the failed hypothetical models of DNA. This structure would be unstable under normal cellular conditions.

**DNA Replication** (<http://www.hhmi.org/biointeractive/dna-replication-schematic>)

The structure of DNA, discovered by James Watson and Francis Crick, suggests a mechanism of replication. This animation shows that as the double helix unwinds, each strand acts as a template for the construction of the new DNA molecule.

**The Chemical Structure of DNA** (<http://www.hhmi.org/biointeractive/chemical-structure-dna>)

This animation shows how DNA's chemical properties can be harnessed for a variety of biotechnology applications.

**Watson Constructing Base-Pair Models** (<http://www.hhmi.org/biointeractive/watson-constructing-base-pair-models>)

This video shows Jim Watson explaining how, during the process of trying to elucidate the structure of DNA, he made some cardboard models to understand how DNA nucleotides are paired. The models helped him visualize how hydrogen atoms of paired nucleotides interact with each other to form a symmetrical structure that fits the double-helix model.

## USING THE QUIZ

The quiz is designed as a summative assessment that probes student understanding of the key concepts addressed in the film. However, some teachers use the quiz before and during the film to assess students' prior knowledge and to guide students as they watch the film. Teachers are encouraged to choose the use that best fits their learning objectives and their students' needs. Teachers are encouraged to modify the quiz (e.g. only ask some of the questions, explain complicated vocabulary for ELL students) as needed.

## QUIZ QUESTIONS AND ANSWERS

- (Key Concept A) What are the chemical components of a DNA nucleotide?
  - a. a phosphate, a sugar, and a nitrogenous base**
  - a phosphate, a nitrogenous base, and an amino acid
  - a nitrogenous base, a sugar, and an amino acid
  - a nitrogenous base, ATP, and a sugar
- (Key Concept C) The instructions for the traits of an organism are determined by
  - the proportions of A,T, C, and G in DNA molecules
  - b. the order of nucleotides in DNA molecules**
  - the length of DNA molecules
  - the way nucleotides are paired in the two strands of a DNA molecule
- (Key Concept B) The two strands of a DNA molecule are held together by hydrogen bonds between the
  - phosphate groups on each strand
  - b. nitrogenous bases on each strand**
  - bases and the phosphate-sugar backbone
  - carbon atoms in the sugars
- (Key Concept B) In the diagram below, strands I and II represent the two complementary strands of a portion of a DNA double helix. The sequence of strand I is indicated below. What is the sequence of strand II?

Strand I -----C-T-A-C-----  
Strand II -----?-?-?-?-----

  - AGCA
  - CTAC
  - TCGT
  - d. GATG**
- (Key Concept F) In the 1950s when Watson and Crick were working on their model of DNA, many scientists did not think that DNA carried the genetic code.
  - What was the other type of molecule that some scientists thought might carry genetic information? **Proteins**
  - Why did this other type of molecule seem like a likely candidate? **Proteins were favored by many scientists because there are many proteins and they come in different shapes and have many different functions. Also, proteins are made of 20 different amino acid components, while DNA is composed of only four basic subunits.**
- (Key Concept F) In 1928, Frederick Griffith conducted an experiment in which he injected mice with different kinds of bacteria. When bacteria that cause disease (pathogenic) were injected in healthy mice, these mice got sick and died. Other types of bacteria (nonpathogenic) did not cause the mice to die. Griffith took the DNA from dead pathogenic bacteria and transferred it into living nonpathogenic bacteria. These altered bacteria were then injected into healthy mice. The mice died of the same disease caused by the pathogenic bacteria. Based on this information, which statement would be a valid conclusion?
  - When an organism dies, the DNA changes; it no longer provides the same genetic information.
  - When DNA from one organism is transferred to another organism, the DNA no longer functions.
  - DNA in different types of bacteria carries exactly the same type of information.
  - d. When DNA from one organism is transferred to another organism, it can give new traits to the second organism.**
- (Key Concept F) The following table is a sample of the data Erwin Chargaff published in 1952.

**Proportions\* of Nitrogenous Bases in the DNA of Different Organisms**

Organism	Tissue	% Adenine	% Guanine	% Cytosine	% Thymine
Yeast		31.3	18.7	17.1	32.9
Sea urchin	Sperm	32.8	17.7	18.4	32.1
Rat	Bone marrow	28.6	21.4	21.5	28.4
Human	Thymus	30.9	19.9	19.8	29.4
Human	Sperm	30.3	19.5	19.9	30.3

(\*Defined as moles of nitrogenous constituents per 100 g of atoms of phosphate.  
 Source: E. Chargaff and J. Davidson, Eds. *The Nucleic Acids*. Academic Press, 1955)

a. Which of the following observations can be supported by the data in the table? (Place a check mark in the box next to the correct statements.)

**All organisms contain about the same amounts of adenine and thymine in their DNA.**

The proportions of adenine + thymine and guanine + cytosine are the same in all organisms.

Larger organisms have greater amounts of each nitrogenous base than smaller organisms have.

The total length of DNA molecules in all organisms is about the same.

b. In one or two sentences, explain how these observations helped Watson and Crick develop their model of DNA.

**It suggests that in the double helix, adenine always pairs with thymine and guanine always pairs with cytosine.**

c. In one or two sentences, explain why the proportions of nitrogenous bases in the DNA of two different human tissues (thymus and sperm) are about the same.

**All body cells and tissues in a particular organism contain the same DNA.**

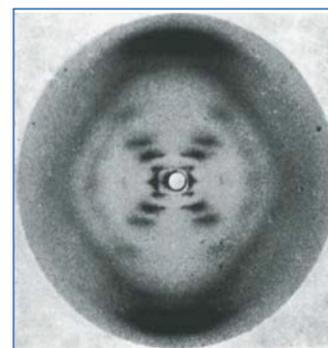
8. The image on the right shows the famous photo B51 taken in May 1952 by Rosalind Franklin and her student Raymond Gosling. This x-ray diffraction pattern provided information about the positions of atoms in a DNA molecule.

a. (Key Concept G) Identify the clue in this photo that revealed that DNA is a helix.

**The clue is the X shape in the image.**

b. (Key Concept D) Measurements revealed that the distance between the two strands was always equal. Explain how this information helped Watson and Crick build a successful model of DNA. **Watson and Crick concluded that a large purine molecule (adenine or guanine) always bonds with a smaller pyrimidine molecule (cytosine or thymine)—that way, the distance between the two strands of DNA is always the same.**

c. (Key Concept E) Was this information consistent with the data obtained by Chargaff (question 7)? Explain your answer. **Yes. Chargaff discovered that in the DNA of an organism, the proportion of adenine (a purine) is the same as the proportion of thymine (a pyrimidine). The same is true for guanine (a purine) and cytosine (a pyrimidine). These data are consistent with a model in which a purine on one strand always bonds with a pyrimidine on the other strand.**



9. (Key Concepts E and F) Scientists build models based on what they know from previous research to derive testable hypotheses. Independently, both Watson and Crick and their competitor Linus Pauling constructed an incorrect triple-helix model with the nitrogenous bases arranged so they were on the exterior of the molecule and the phosphate groups on the interior.

a. Although their model was wrong, what assumption made it reasonable to build a model with the bases projecting to the outside?

**The bases contain the genetic information as they vary in amount between species and in their arrangement within the molecule; based on this information it was reasonable to assume that the bases would be on the outside of the DNA molecule.**

b. What evidence caused Watson and Crick to revise their model?

**Based on the film, students should realize that information about the dimensions of the double helix from Franklin's images, as well as Chargaff's pairing rules for the bases, led them to a revised model. The film says that Watson had misremembered some key measurements from Franklin's work. Her images showed there was much more water in DNA than the triple-helix model allowed. Further, the triple-helix model could not explain how the three phosphate backbones could be held together. (Students may not know these last two points as they are not explicitly stated in the film.)**

10. (Key Concept C) Even before the structure of DNA was solved, studies indicated that the genetic material must have the following properties:

- be able to store information;
- be faithfully replicated and be passed on from generation to generation; and
- allow for changes, and thus evolution, to occur.

Explain how the structure of the double helix showed that DNA had these properties. Write one or two sentences per point.

**The order of the bases, A, T, G, and C, contained information. Because A is always paired with T and G with C, the order of bases on one strand determines the order on the other strand. Thus, if a DNA molecule were unwound, each strand could be copied into a complementary strand, producing an exact replica of the original molecule. Errors in the copying mechanism could result in mutations, or changes in the DNA sequence, that could be inherited by future generations.**

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