The Molecular Basis of Inheritance
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Slide 1
The Griffith experiment demonstrated that there is a molecular basis of heredity, a so-called transforming substance that turned non-virulent bacteria into virulent bacteria. We now know this substance is DNA.

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A bacteriophage is a virus hat invades a bacterium. Bacteriophages are made of a combination of protein and DNA, and they were therefore chosen as an ideal subject in an experiment to answer the question of whether the genetic material is DNA or protein.

Slide 3
In the Hershey-Chase experiment, bacteriophages were radiolabeled with either radioactive sulfur (which labeled the protein) or radioactive phosphorus (which labeled the DNA) to determine whether it is the protein or the DNA that a bacteriophage injects into a bacterium. Since the DNA is what is injected by the virus into the cell, this demonstrates that DNA - and not protein - is the genetic material.

Slide 4
A strand of nucleotides linked end-to-end is called a polynucleotide. Any two adjacent nucleotides within a polynucleotide are held together by a covalent phosphodiester bond that links the sugar of one nucleotide to the phosphate of the adjacent nucleotide. The combination of all the phosphodiester bonds in a polynucleotide is therefore called the sugar-phosphate backbone, and it holds the strand together along its length. The nitrogenous bases are not involved in holding the polynucleotide together; they simply extend to the side of the backbone.

Slide 5
Rosalind Franklin was an expert at x-ray crystallography, a technique in which x-rays are directed at a molecule of unknown shape before exposing a photographic film. Based on the pattern made by the x-rays after being diffracted (bent) by DNA, Franklin was able to help Watson and Crick figure out that the shape of a DNA molecule is a double helix.

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Whereas an RNA molecule occurs as a single strand, a DNA molecule is a double stranded molecule. Two DNA polynucleotides, each with a helical shape, make up one DNA molecule. The two strands have an antiparallel arrangement, with one strand in a 3' to 5' direction, and the other strand in a 5' to 3' direction. The two strands are held together by hydrogen bonds that occur between nitrogenous bases at the same position but on opposite strands. These weak hydrogen bonds allow for easy separation of the two strands for the purposes of replication and transcription.

Slide 7
There are two classes of nitrogenous bases:

- A purines has two rings in its structure. The purines used in DNA are adenine and guanine.
- A pyrimidine has one ring in its structure. The pyrimidines used in DNA are thymine and cytosine.

Chargaff noticed that all organisms have an equal amount of purine and pyrimidine in the DNA. This is because a base pair (which is a nitrogenous base on one strand hydrogen-bonded to a nitrogenous base at the same position on the other strand) must consist of one purine and one pyrimidine. This allows for equal spacing between the two strands at each position.

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Though it is true that a normal base pair consists of one purine and one pyrimidine, an even more stringent requirement is met in DNA. In making a base pair, adenine normally binds only with thymine (and vice versa), and cytosine normally binds only with guanine (and vice versa). This is known as complementarity. Therefore one strand within a DNA molecule is complementary to the other strand.

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The complementary relationship between the two strands of a DNA molecule allow each of the strands in isolation to serve as a template for building the other. Thus, DNA replication occurs by separating the two strands from each other and "reading" the sequence of nucleotides in each strand to assemble a complementary strand. When this is done for each of the original two strands, the result is two identical copies of the original two-stranded molecule. Each copy will end up in one of two daughter cells created when the cell divides.

Slide 10
After learning that DNA replicates by using itself as a template for building new DNA, scientists recognized that there are three possibilities for how this occurs:

- Conservative model - Both original strands are conserved, and a completely new double-stranded molecule is produced.
- Semi-conservative model - One of the original strands is conserved, and a new strand is added to that. The same happens for the other original strand. The result is two molecules, each with one original strand and one new strand.
- Dispersive model - Neither original strand is conserved, and each strand of each new molecule is made of a mixture of original and new material.

Though these are the three possibilities that had to be tested, the semiconservative model is the only one that actually happens in cells.

Slide 11
Experimental evidence indicates that DNA replicates by a semi-conservative model (and not by a conservative or a dispersive model). Bacterial DNA was labeled with different isotopes of nitrogen so newly made DNA could be separated from original DNA by centrifugation. The predictions for the semiconservative model were met, while the predictions for the other models were not.

Slide 12
DNA replication begins at the origin of replication, where the two strands are separated, thus forming a replication bubble with two replication forks. Each strand is used as a template for replacing the other strand. At completion of replication, two identical copies of the double-stranded DNA molecule replace the original one. A prokaryotic cell has a single circular chromosome, and it replicates by making one replication bubble that grows. A eukaryotic cell has multiple linear chromosomes, each of which replicates by forming multiple replication bubbles, thereby increasing the speed of replication.

Slide 13
Helicase separates the two DNA strands at the origin of replication to form a replication bubble and initiate replication. Single-strand binding proteins temporarily attach to the separated strands to keep them from bonding back together. Primase lays down a short sequence of RNA nucleotides known as a primer. The primer is required, because new DNA nucleotides can be laid down only by adding them to existing nucleotides, whereas RNA nucleotides can be laid down without attaching them to existing nucleotides. As helicase further separates the two strands at the replication fork, the DNA experiences over-twisting. That over-twisting is relieved by topoisomerase.

Slide 14
The raw material for a new nucleotide to be added to a growing strand starts out as a nucleoside triphosphate, which has two additional phosphates compared to a nucleotide. When the two extra phosphates are removed to produce the nucleotide, the energy released is used for the endergonic reaction required to form a phosphodiester bond between the new nucleotide and the previous nucleotide. The enzyme that catalyzes this reaction is DNA polymerase III. Each new nucleotide must be complementary to the nucleotide at the same position on the template strand. Also, any new nucleotide must be added to he 3' end of the growing strand.

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Each replication bubble features two leading strands that are being produced and two lagging strands that are being produced. Each leading strand requires just one primer, and the strand is produced continuously, because the direction of production is the same as the direction in which the corresponding replication fork is opening.

Slide 16
Each lagging strand requires multiple primers and is produced discontinuously (piece by piece), because the direction of production is opposite to the direction in which the corresponding replication fork is opening.

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Each piece of the lagging strand is called an Okazaki fragment. For each Okazaki fragment, primase lays down a primer, DNA polymerase adds DNA nucleotides to the primer until the gap is filled between the current fragment and the previously constructed fragment. DNA polymerase I replaces the nucleotides of RNA primers (in both the leading and the lagging strands) with DNA nucleotides. The fragments are then joined together with final phosphodiester bonds with the action of DNA ligase.

Slide 19
When errors are made in the DNA replication process, the cell's proofreading mechanisms nearly always detect and correct the error. For instance, when a dimer forms between two adjacent thymine bases, a nuclease cuts out the erroneous fragment, DNA polymerase I replaces the nucleotides that were removed, and DNA ligase bonds the replacement fragment to the strand. This is called excision repair.

Side 20
Only eukaryotic chromosomes (because they are linear) feature telomeres, which are repeating sequences of nucleotides at the ends of the chromosomes. Each time a linear chromosome is replicated, it gets a bit shorter, because no DNA can be laid down to replace the RNA primer at the very end of the new strand. After repeated replication (many cell cycles), this would result in the destruction (erosion) of genes, which would be fatal to the cell. Eukaryotic cells avoid this problem by producing an enzyme called telomerase, whose job it is to add fresh telomeres to the ends of chromosomes, allowing for shortening of the chromosome without eroding into genes.

Slides 21 & 22
The enormously long DNA molecule in a chromosome is able to fit into a tiny cell nucleus, because the DNA is spooled around complexes of histone proteins. These complexes are then extensively looped, resulting in the chromatin condensing into a compact chromosome.