



Evidence for DNA as Genetic Material

Griffith's Experiment (1928)

Objective: To understand the difference between virulent and non-virulent strains of <i>Streptococcus pneumoniae</i> .
S Strain (Virulent): Causes pneumonia and kills mice.
R Strain (Non-Virulent): Does not cause pneumonia and does not kill mice.
Experiment 1: Live S strain → Mouse dies.
Experiment 2: Live R strain → Mouse lives.
Experiment 3: Heat-killed S strain → Mouse lives.
Experiment 4: Heat-killed S strain + Live R strain → Mouse dies.
Conclusion: Transformation occurred, where the R strain acquired virulence from the dead S strain. Griffith did not identify the transforming principle.

Avery, MacLeod, & McCarty (1944)

Objective: To identify the molecule responsible for transformation in Griffith's experiment.
Experiment: Repeated Griffith's experiment using purified cell extracts from S strain.
Procedure: 1. Removed proteins - Transformation still occurred. 2. Removed RNA - Transformation still occurred. 3. Removed DNA - Transformation did NOT occur.
Conclusion: DNA is the genetic material responsible for transformation, at least in bacteria.

Hershey & Chase Experiment (1952)

Objective: To determine whether DNA or protein is the genetic material in bacteriophages.
Bacteriophages: Viruses that infect bacteria, composed of DNA and protein.
Experiment: 1. Labeled phage DNA with radioactive phosphorus (³²P). 2. Labeled phage protein with radioactive sulfur (³⁵S).
Procedure: 1. Infected bacteria with labeled phages. 2. Separated phages from bacteria. 3. Measured radioactivity inside the bacteria.
Results: Radioactive phosphorus (³² P) was found inside the bacteria, while radioactive sulfur (³⁵ S) remained outside.
Conclusion: DNA is the genetic material that is injected into the bacteria and used to produce more bacteriophages. Protein is not the genetic material.

DNA Structure and Components

Nucleotide Components

DNA is a nucleic acid composed of nucleotides:
Deoxyribose: A 5-carbon sugar.
Phosphate Group (PO₄): Attached to the 5' carbon of the sugar.
Nitrogenous Base: Adenine (A), Thymine (T), Cytosine (C), Guanine (G).
Hydroxyl Group (-OH): Attached at the 3' carbon of the sugar.

Purines vs. Pyrimidines

Purines:	Pyrimidines:
Two-ringed structures (Adenine and Guanine).	Single-ringed structures (Cytosine and Thymine).

DNA vs. RNA Nucleotides

DNA:	RNA:
Contains deoxyribose sugar.	Contains ribose sugar.
Uses Thymine (T) as a base.	Uses Uracil (U) instead of Thymine.

Phosphodiester Bonds

Phosphodiester Bond: Bond between adjacent nucleotides.
Formed between the phosphate group of one nucleotide and the 3' -OH of the next nucleotide.
Creates a chain of nucleotides with a 5'-to-3' orientation.

Chargaff's Rules

Chargaff's Rules:
The amount of Adenine (A) equals the amount of Thymine (T).
The amount of Cytosine (C) equals the amount of Guanine (G).
The ratio of A-T and G-C varies by species.

DNA Structure and Replication

Watson and Crick Model

Watson and Crick (1953):
Deduced the structure of DNA using evidence from Chargaff, Franklin, and others.
DNA molecule is made of two intertwined chains of nucleotides, forming a double helix structure.

Double Helix Structure

Double Helix: Two strands arranged as a double helix.
Forms two grooves: major groove and minor groove.
Strands connected via hydrogen bonds between bases on opposite strands.
Base-Pairing: A-T (2 hydrogen bonds), G-C (3 hydrogen bonds).
Consistent diameter and stability due to thousands of low-energy hydrogen bonds.

Antiparallel Configuration

Antiparallel: Each phosphodiester strand has inherent polarity based on the orientation of the sugar-phosphate backbone.
One end terminates in 3' OH, and the other in 5' PO ₄ .
Strands have either 5'-to-3' or 3'-to-5' polarity.
The two strands of a single DNA molecule have opposite polarity to one another.

DNA Replication Models

Conservative Model: Both strands of parental DNA remain intact; new DNA copies consist of all new molecules.
Semiconservative Model: Daughter strands each consist of one parental strand and one new strand (Correct model).
Dispersive Model: New DNA is dispersed throughout each strand of both daughter molecules after replication.

DNA Replication Process

Requirements for DNA Replication

1. Template: Parental DNA molecule to copy.
2. Enzymes: Proteins to do the copying.
3. Building Blocks: Nucleotide triphosphates to make the copy.

Stages of DNA Replication

1. Initiation: Replication begins at specific sites called origins of replication.
2. Elongation: New strands of DNA are synthesized by DNA polymerase.
3. Termination: Replication is terminated, often at specific termination sites or when replication forks meet.

DNA Polymerase

DNA Polymerase: Matches existing DNA bases with complementary nucleotides and links them to build new DNA strands.
Features: - Adds new bases to the 3’ end of existing strands. - Synthesizes in the 5’-to-3’ direction. - Requires a primer of RNA to initiate synthesis.

Semi-Discontinuous Replication

Semi-Discontinuous: DNA polymerase can only synthesise in the 5’-to-3’ direction.
Leading Strand: Synthesized continuously from an initial primer.
Lagging Strand: Synthesized discontinuously with multiple priming events, creating Okazaki fragments.

Enzymes Involved in Lagging-Strand Synthesis

DNA Pol III: Synthesizes Okazaki fragments.
Primase: Makes RNA primer for each Okazaki fragment.
DNA Pol I: Removes all RNA primers and replaces them with DNA.
DNA Ligase: Joins Okazaki fragments to form complete strands.
DNA Gyrase (Topoisomerase): Unlinks two copies of DNA at the termination site.