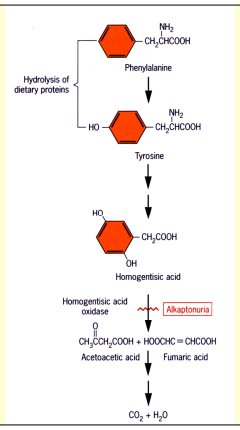
Outline

- The search for the genetic material
- The structure of DNA
- How DNA is replicated
- DNA repair
- How do you replicate the ends?

What do "genes" actually do?

- Garrod (1908) studied the physiology of alkaptonuria, an inherited disorder
- Symptoms
- Causes

The tyrosine and phenylalanine metabolic pathway



Conclusion: one "defective gene" = absence of one enzyme

"one gene, one protein"

How do we know DNA is the genetic material?

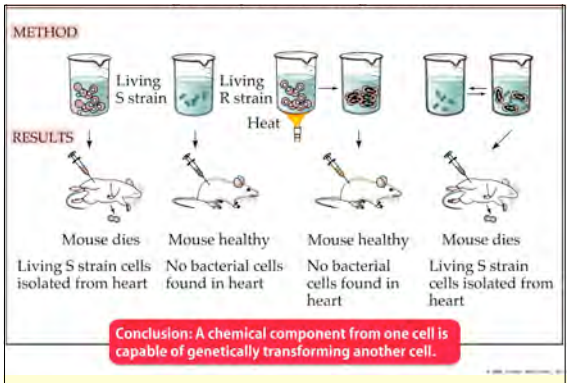
- Morgan (1920s and 1930s) showed that chromosomes contain the genetic material
- Chromosomes are made of Protein (60%), DNA (35%) and RNA (5%)
- What part of the chromosome carries genetic information, protein or nucleic acid?

Protein or DNA?

- Protein
  - Made from 20 different amino acids
  - Up to 10,000 units
  - Enzymes are involved in every internal process
- 4 nucleotides
- Structure not known
- No known function

Griffith and Bacterial Transformation

- In the first key experiment, Griffith showed that molecules from dead cells could "transform" the genes of living cells

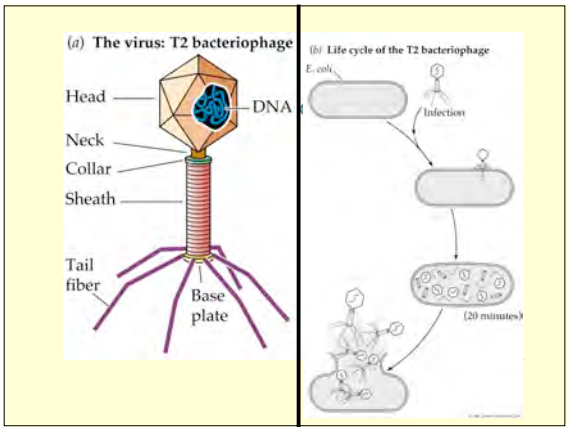


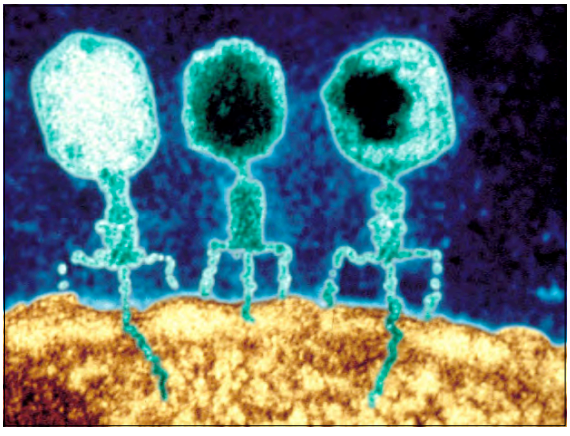
Avery built on Griffith's results

- Avery purified various chemicals from the transforming cells.
- The only chemical that worked alone in transformation was DNA
- Conclude: DNA is the transforming substance - DNA carries information
- This was not widely accepted.

Hershey and Chase and Bacteriophages

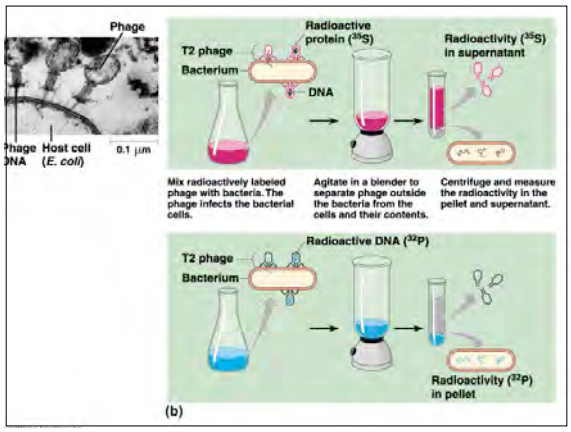
- In the second key experiment, Hershey and Chase infected bacteria with phages (viruses)
- The viruses, made of protein and DNA, cause the bacteria to produce more phages.





Hershey and Chase

- Use radioactive isotopes to "label" different parts of the virus
- P^{32} is incorporated into DNA
- S^{35} is incorporated into protein
- Grow, infect, disrupt, centrifuge.
- Where is the radioactivity?



Conclude: DNA is sufficient to oversee production of new viral particles

One gene, one protein - Garrod

"Transforming" substance is DNA - Griffith & Avery

DNA directs production of virus - Hershey & Chase

DNA is the genetic material

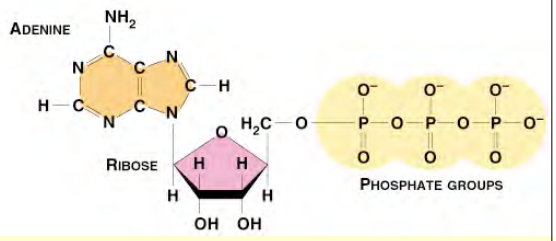
The Double Helix

- If DNA contains the genetic information, how does it work?
- Watson and Crick discovered the structure of the DNA molecule
- The structure they discovered immediately suggested how information is stored and replicated.

DNA Structure

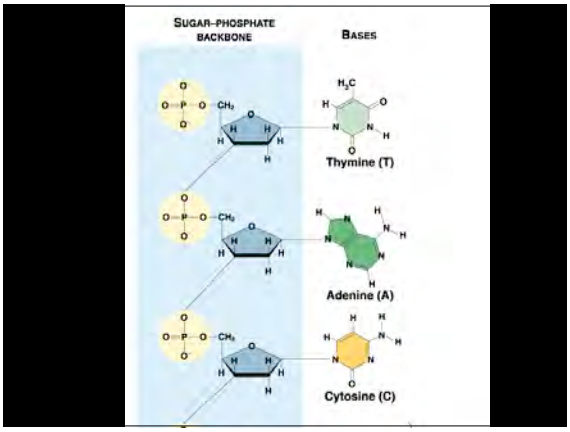
- DNA is a polymer of nucleotides
- Each nucleotide consists of a sugar (deoxyribose), a nitrogenous base, and a phosphate group
- There are four different nitrogenous bases: Adenine, Thymine, Guanine, Cytosine (A,T,G,C)

ATP: a nucleoside triphosphate



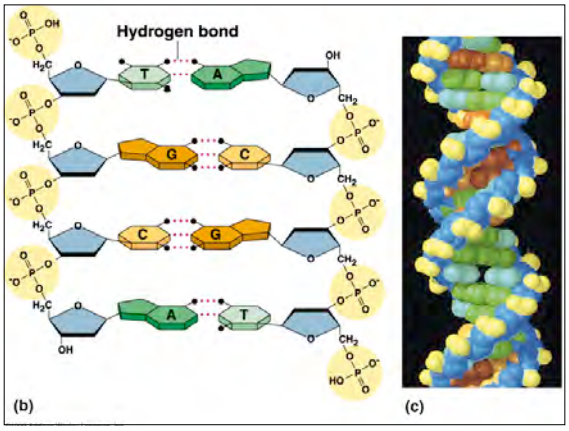
DNA structure, cont.

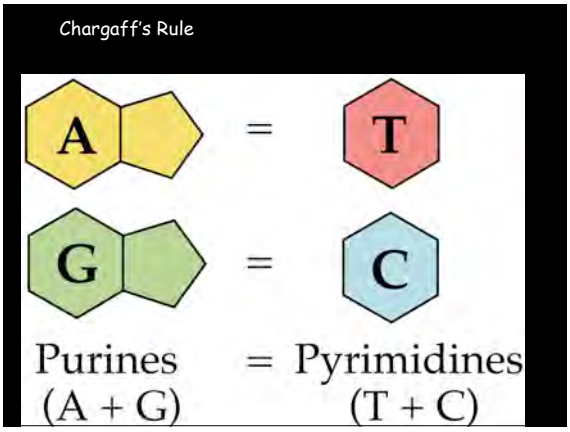
- The backbone of each DNA strand is composed of covalently bonded sugars and phosphates
- The two strands are antiparallel

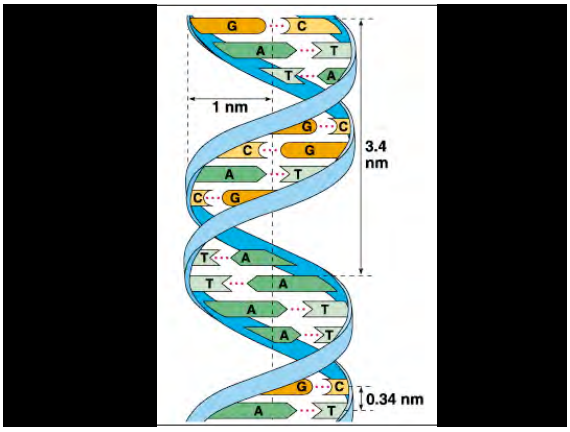


DNA Structure

- Bases project from each strand into the center of the helix, where they form hydrogen bonds with complementary bases
- *A* pairs with *T*
- *C* pairs with *G*





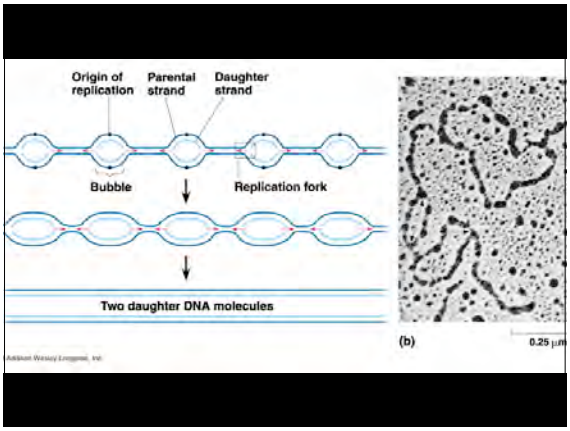


The structure of DNA allows for information storage and copying

- Information is contained in the order of the bases
- Note that complementary strands have the same information

DNA Replication

- DNA replication is semi-conservative
- Each strand serves as template for a new complementary strand
- Replication happens at replication forks and is bidirectional
- Replication is carried out by a team of enzymes

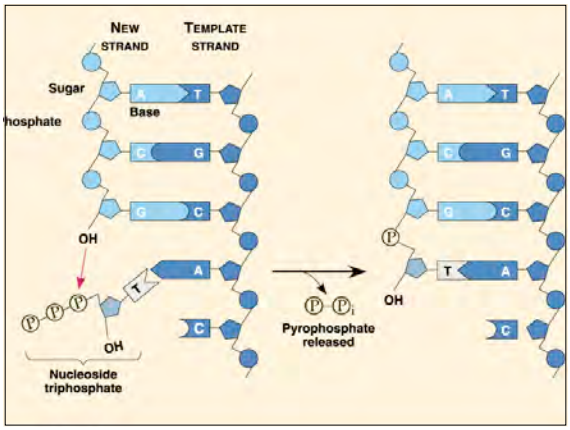


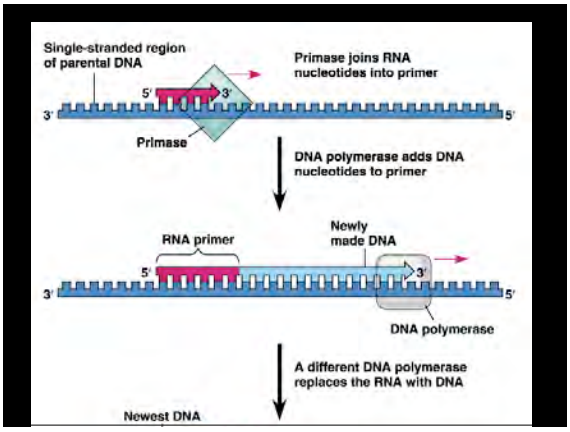
DNA replication enzymes

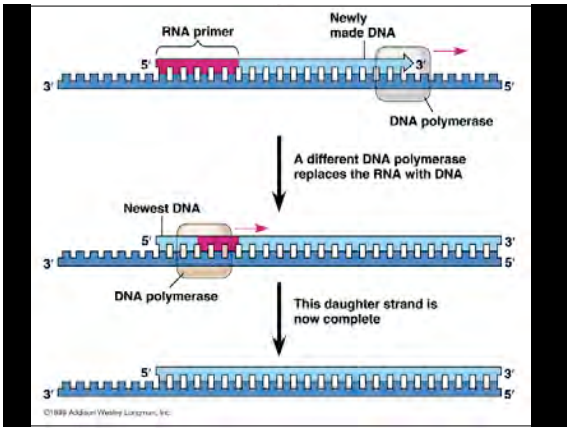
- DNA polymerases build new DNA
- Primase forms RNA primers
- Helicase unwinds DNA
- DNA Binding proteins hold strands apart
- Ligases seal nicks
- Topoisomerases prevent twisting

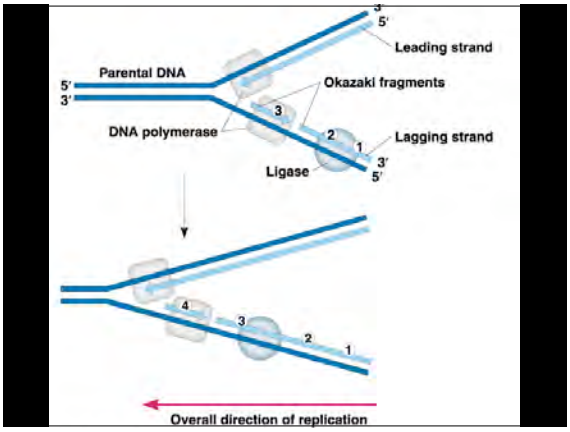
The actions (and limitations) of DNA polymerase

- Adds nucleoside triphosphates to a free 3-prime end of a DNA strand
- Inserts appropriate nucleotide (complementary to base on opposite strand)
- Can only lengthen an existing strand
- Only lengthens strands in 5'-3' direction









DNA repair

- During synthesis polymerases incorporate the wrong nucleotide once in 10^6 cases
- Proofreading enzymes cut out errant bases and repair the strand, reducing the overall error rate to one in 10^9
- Expensive but essential