CHAPTER 13 DNA: STRUCTURE AND FUNCTIONS

Chapter Outline

13.1 The Genetic Material

A. Early researchers knew that the genetic material must be:
   1. able to store information used to control both the development and the metabolic activities of cells;
   2. stable so it can be replicated accurately during cell division and be transmitted for generations; and
   3. able to undergo mutations providing genetic variability required for evolution.

B. Previous Knowledge About DNA
   1. Knowing the chemistry of DNA was essential to discovery that DNA is genetic material.
   2. In 1869, Swiss chemist Friedrich Miescher removed nuclei from pus cells and isolated DNA "nuclein"; it was rich in phosphorus and lacked sulfur.
   3. Nuclein was analyzed by other scientists who found that it contained an acid: **nucleic acid.**
   4. Two types of nucleic acids were soon discovered: **DNA** (**deoxyribonucleic acid**) and **RNA** (**ribonucleic acid**).
   5. In the early twentieth century, discovery that nucleic acids contain four types of **nucleotides**.
      a. DNA was composed of repeating units, each of which always had just one of each of four different nucleotides (A, T, G, or C).
      b. In this model, DNA could not vary between species and therefore could not be the genetic material; therefore some other protein component was expected to be the genetic material.

C. Transformation of Bacteria
   1. In 1931, bacteriologist Frederick Griffith experimented with *Streptococcus pneumoniae* (a pneumococcus) that causes pneumonia in mammals.
   2. Griffith injected mice with two strains of pneumococcus: an encapsulated (S) strain and a non-encapsulated (R) strain.
      a. The S strain is virulent (the mice died); it has a mucous capsule and forms shiny colonies.
      b. The R strain is not virulent (the mice lived); it has no capsule and forms dull colonies.
   3. In an effort to determine if the capsule alone was responsible for the virulence of the S strain, he injected mice with heat-killed S strain bacteria; the mice lived.
   4. Finally, he injected mice with a mixture of heat-killed S strain and live R strain bacteria.
      a. The mice died and living S strain pneumococcus were recovered from their bodies.
      b. Griffith concluded some substance necessary to synthesis of the capsule and, therefore, virulence must pass from dead S strain bacteria to living R strain bacteria so the R strain were *transformed*.
      c. This change in phenotype of the R strain bacteria must be due to a change in their genotype, which suggested that the transforming substance may have passed from S strain to R strain.

D. DNA: The Transforming Substance
   1. Oswald Avery and his coworkers reported that the transforming substance was DNA.
   2. Purified DNA is capable of bringing about the transformation; their evidence included the following:
      a. DNA from S strain pneumococcus causes R strain bacteria to be transformed.
      b. Enzymes that degrade proteins cannot prevent transformation, nor do enzymes that digest RNA.
      c. Digestion of the transforming substance with enzyme that digests DNA prevents transformation.
      d. Molecular weight of the transforming substance is great enough for some genetic variability
   3. Their experimental results demonstrated DNA is genetic material and DNA controls biosynthetic properties of a cell.

E. Transformation Experiments Today
   1. Transformation experiments today are common in high schools and research labs.
   2. Transformation occurs whenever organisms receive foreign DNA and receive a new trait.
   3. Modern experiments with bacteria show some can take up DNA to gain penicillin resistance.

F. Reproduction of Viruses
   1. **Bacteriophage** is a virus that infects bacteria; it consists only of a protein coat surrounding a nucleic acid core.
   2. **Bacteriophage T2** is a virus that infects the bacterium ***Escherichia coli*** (*E. coli*), a species of intensely

studied bacteria that normally lives within the human gut.

3. In 1952, Alfred Hershey and Martha Chase used bacteriophage T2 in their experiments.
    a. The purpose of their experiments was to see which of the bacteriophage components—the protein coat or the DNA—entered bacterial cells and directed reproduction of the virus.
    b. In two separate experiments, they labeled the protein coat with ^{35}S and the DNA with ^{32}P.
    c. Viral coats are sheared away from bacterial cells; they are separated by centrifugation.
    d. Results: radioactive ^{32}P alone is taken up by bacterial host and incorporated in virus reproduction.
    e. This result reinforced the notion that DNA (and not the protein) is the genetic material.

13.2 The Structure of DNA

A. Nucleotide Data
1. In the 1940s, Erwin Chargaff analyzed the base content of DNA using new chemical techniques.
2. It was known DNA contained four different nucleotides:
    a. two with *purine* bases, **adenine (A)** and **guanine (G)**; a **purine** is a type of nitrogen-containing base having a double-ring structure.
    b. two with *pyrimidine* bases, **thymine (T)** and cytosine **(C)**; a **pyrimidine** is a type of nitrogen-containing base having a single-ring structure.
3. The results of his analysis proved DNA does have the variability necessary to code genetic material.
4. Chargaff discovered that for a species, DNA has the *constancy* required of genetic material.
5. This constancy is given in Chargaff's rules:
    a. **The amount of A, T, G, and C in DNA varies from species to species.**
    b. **In each species, the amount of A = T and the amount of G = C.**
6. The tetranucleotide hypothesis (proposing DNA was repeating units of one of four bases) was disproved; each species had its own constant base composition.

B. Variation in Base Sequence
1. The variability is staggering; a human chromosome contains about 140 million base pairs.
2. Since any of the four possible nucleotides can be present at each nucleotide position, the total number of possible nucleotide sequences is $4^{140 \times 10^6} = 4^{140,000,000}$.

C. Diffraction Data
1. Rosalind Franklin, a student at King's College, produced X-ray diffraction photographs.
2. Franklin's work provided evidence that DNA had the following features:
    a. DNA is a helix.
    b. One part of the helix is repeated.

C. The Watson and Crick Model
1. American James Watson joined with Francis H. C. Crick in England to work on structure of DNA.
2. Watson and Crick received the Nobel Prize in 1962 for their model of DNA.
3. Using information generated by Chargaff and Franklin, Watson and Crick built a model of DNA as a double helix; sugar-phosphate molecules were on the outside, paired bases were on the inside.
4. Their model was consistent with both Chargaff's rules and the dimensions of the DNA polymer provided by Franklin's photograph of X-ray diffraction of DNA.
5. **Complementary base pairing** is the paired relationship between purines and pyrimidines in DNA, such that A is hydrogen-bonded to T and G is hydrogen-bonded to C.

13.3 Replication of DNA

A. DNA replication is the process of copying a DNA molecule.
1. **Unwinding:** old strands of the parent DNA molecule are unwound as weak hydrogen bonds between the paired bases are unzipped and broken by the enzyme **helicase.**
2. **Complementary base pairing:** free nucleotides present in the nucleus bind with complementary bases on unzipped portions of the two strands of DNA; this process is catalyzed by DNA polymerase.
3. **Joining:** complimentary nucleotides bond to each other to form new strands; each daughter DNA molecule contains an old strand and a new strand; this process is also catalyzed by DNA polymerase.
4. DNA replication must occur before a cell can divide; in cancer, drugs with molecules similar to the four nucleotides are used to stop replication.

B. Replication is Semiconservative
1. DNA replication is semiconservative because each daughter double helix has one parental strand and one new strand.
2. In 1958, Matthew Meselson and Franklin Stahl confirmed a model of DNA replication.

a. They grew bacteria in a medium with heavy nitrogen (^{15}N), then switched to light nitrogen (^{14}N).
b. The density of DNA following replication is intermediate as measured by centrifugation of molecules.
c. After one division, only hybrid DNA molecules were in the cells.
d. After two divisions, half the DNA molecules were light and half were hybrid.

3. These were exactly the results to be expected if DNA replication is semiconservative.

C. Prokaryotic Versus Eukaryotic Replication

1. Prokaryotic Replication
    a. Bacteria have a single loop of DNA that must replicate before the cell divides.
    b. Replication in prokaryotes may be bidirectional from one point of origin or in only one direction.
    c. Replication only proceeds in one direction, from 5' to 3'.
    d. Bacterial cells are able to replicate their DNA at a rate of about 10^6 base pairs per minute.
    e. Bacterial cells can complete DNA replication in 40 minutes; eukaryotes take hours.
2. Eukaryotic Replication
    a. Replication in eukaryotes starts at many points of origin and spreads with many replication bubbles—places where the DNA strands are separating and replication is occurring.
    b. **Replication forks** are the V-shape ends of the replication bubbles; the sites of DNA replication.
    c. Eukaryotes replicate their DNA at a slower 500–5,000 base pairs per minute.
    d. Eukaryotes take hours to complete DNA replication.

D. Replication Errors

1. A genetic mutation is a permanent change in the sequence of bases.
2. Base changes during replication are one way mutations occur.
3. A mismatched nucleotide may occur once per 100,000 base pairs, causing a pause in replication.
5. **DNA repair enzymes** perform a proofreading function and reduce the error rate to one per billion base pairs.
6. Incorrect base pairs that survive the proofreading process contribute to gene mutations.