**Chapter Review**

**Chapter 4: Fundamentals of Molecular Biology**

4.1

Genes are carried on chromosomes, with DNA as the genetic material. DNA is a double helix in which hydrogen bonds form between purines and pyrimidines on opposite chains. Because base pairing is specific—A with T and G with C—the two stands of DNA are complementary. DNA replicates by semiconservative replication in which the two strands separate and each serves as template for synthesis of a new progeny strand.

4.2

The order of nucleotides in DNA specifies the order of amino acids in proteins. DNA serves as a template for synthesis of mRNA (transcription) and mRNA serves as a template for protein synthesis on ribosomes (translation). Thus the central dogma states that information flows from DNA to RNA to protein. Transfer RNAs serve as adaptors between amino acids and mRNA, with each amino acid specified by a codon consisting of three nucleotides. DNA can also be synthesized from RNA by reverse transcription, first discovered in retroviruses.

4.3

Restriction endonucleases cleave specific DNA sequences, yielding defined fragments of DNA molecules. Either genomic DNA fragments or cDNAs synthesized by reverse transcriptase can be ligated to a vector that is able to replicate in an appropriate host cell and isolated as molecular clones. The nucleotide sequences of cloned DNA fragments can be readily determined, and proteins encoded by cloned genes can be expressed at high levels in either bacteria or eukaryotic cells.

4.4

Amplification of DNA by PCR is a sensitive method for detecting and isolating small amounts of specific DNA or RNA molecules. Nucleic acid hybridization allows the detection of specific DNA or RNA sequences either after separation by gel electrophoresis or within cells. Antibodies can be used to detect specific proteins either in cells or cell extracts.

4.5

Cloned genes can be introduced into complex eukaryotic cells and multicellular organisms for functional analysis. The effect of engineered mutations can be studied by *in vitro* mutagenesis of cloned DNAs, which can be introduced into chromosomal gene copies by homologous recombination. The CRISPR/Cas system uses homologous guide RNAs to efficiently target desired cellular genes. Gene expression at the level of mRNA can also be blocked by antisense nucleic acids or RNA interference.