DNA and Biotechnology

OUTLINE:

- Form of DNA
- Replication of DNA
- Gene Expression
- Mutations
- Regulating Gene Activity
- Genetic Engineering
- Genomics

Form of DNA

- DNA (deoxyribonucleic acid)
  - Double-stranded molecule twisted to form a double helix
  - Resembles a ladder
    - Each side and half of each rung is made from a string of repeating units called nucleotides
- Nucleotide
  - One sugar (deoxyribose in DNA)
  - One phosphate
  - One nitrogenous (nitrogen-containing) base

Form of DNA

- DNA has four nitrogenous bases
  - Adenine (A)
  - Guanine (G)
  - Thymine (T)
  - Cytosine (C)

Form of DNA

- Rungs of ladder consist of paired nitrogenous bases attached according to complementary base pairing rules
  - Adenine (A) pairs only with thymine (T)
  - Cytosine (C) pairs only with guanine (G)
  - Each pair of bases held together by hydrogen bonds
- Genetic information is encoded in the exact sequence of bases

Form of DNA

Replication of DNA

- DNA must be exactly copied (replicated) before both mitotic and meiotic cell divisions
  - Begins when an enzyme breaks the hydrogen bonds that hold together the paired bases of the parent molecule
    - Two strands of the double helix separate and unwind
    - Nitrogenous bases on the separated regions are temporarily exposed
    - Free nucleotide bases in the nucleus attach to exposed complementary bases

Replication of DNA

- DNA polymerases link the sugars and phosphates
- As each of the new double-stranded DNA molecules forms, it twists into a double helix
- DNA replication is described as semiconservative
  - In each of the new double-stranded:
    - One original (parent) strand is saved (conserved)
    - Other (daughter) strand is new
- Complementary base pairing creates two new DNA molecules that are identical to the parent molecule
  - Genetic information is passed accurately from a parent cell to daughter cells

Gene Expression

- How does DNA direct cellular activities?
  - DNA codes for RNA, which codes for proteins
    - Proteins play structural or functional roles in cells
- Gene
  - Segment of DNA with instructions for producing a particular protein (or polypeptide)
  - Expressed when the protein it codes for is produced

RNA Synthesis

- RNA (ribonucleic acid) differs from DNA
  - Single-stranded
  - Contains the base uracil (U), and not thymine (T)
  - Contains the sugar ribose, not deoxyribose

RNA Synthesis

- Occurs in two steps
  - Transcription
  - Translation
- Transcription
  - Synthesis of messenger RNA (mRNA) using DNA as a template
    - mRNA carries DNA's instructions for synthesizing a protein from the nucleus to the cytoplasm

RNA Synthesis

- Steps in transcription
  - DNA unzips along hydrogen bonds
  - Free RNA nucleotides pair with the complementary DNA bases (C-G and U-A) along the exposed DNA strand, forming an RNA transcript
  - RNA transcript released from the DNA
  - DNA closes again
  - Promoter: DNA sequence that starts transcription
  - RNA polymerase: enzyme that binds with promoter, opens up DNA helix, and aligns and links together the RNA nucleotides
  - Another sequence of bases is the stop signal

RNA Synthesis

- Modifications to mRNA before it leaves the nucleus
  - Removal of introns
    - Introns = intervening sequences
    - Regions of DNA between promoter and stop signal that do not have codes that will be translated into a protein
    - Regions of mRNA corresponding to introns are snipped out by enzymes
  - Exons = expressed sequences
    - Left to direct protein synthesis

RNA Synthesis

Protein Synthesis

- Translation
  - Converts the nucleotide language of mRNA into the amino acid language of a protein
- Genetic code
  - Converts the DNA base sequence into an amino acid sequence
  - Codon
    - "Words" in the genetic code
    - A three-base sequence that translates into one amino acid
    - Example: UUC on mRNA specifies the amino acid phenylalanine

Protein Synthesis

- Stop codons
  - UAA, UAG, and UGA
  - Signal the end of a protein
  - Do not code for an amino acid

Protein Synthesis

Transfer RNA (tRNA)

- Converts genetic message carried by mRNA into particular sequence of amino aids
- Recognizes both the codon on mRNA and the amino acid specified by the codon
- Enzymes ensure tRNA binds with correct amino acid
- Ferries amino acid to the correct location along the mRNA strand

Protein Synthesis

- Anticodon
  - A three-base sequence on tRNA that is complementary to the mRNA codon
  - Binds to codon on mRNA and specific amino acid is added to growing polypeptide chain

Protein Synthesis

- Ribosomes
  - Structure
    - Two subunits, each composed of ribosomal RNA (rRNA) and protein
  - Function

        - Bring tRNA bearing an amino acid close enough to mRNA to interact
            - Permit alignment of anticodon and codon

Protein Synthesis

- Three stages of translation
    1. Initiation
        - Major players in protein synthesis come together (mRNA, tRNA, and ribosomal subunits)

Protein Synthesis

2. Elongation
    - Occurs as enzymes in the ribosome link the amino acids, forming a growing chain of amino acids
    - Polysome
        - Cluster of ribosomes simultaneously translating the same mRNA

Protein Synthesis

3. Termination
    - Occurs when a stop codon moves into the ribosome

Protein Synthesis

Mutations

- Changes in DNA
- Passed to offspring only when they occur in a gamete
- Types
    - Chromosomal
        - Occur when whole sections of chromosomes are duplicated or deleted
    - Gene
        - Changes in the order of nucleotides in DNA
        - Those involving insertion or deletion of nucleotides can greatly change resulting protein

Regulating Gene Activity

- Only certain genes are active in a certain type of cell and most genes are turned off in any given cell
    - This leads to specialization
    - Active genes produce specific proteins that determine the structure and function of a particular cell

Regulating Gene Activity

- Gene activity is regulated simultaneously at several levels
    - Chromosomal level: coiling and uncoiling of DNA (when DNA is tightly coiled, genes are not expressed)
    - Transcription of genes: enhancers increase rate of transcription of certain genes
    - Steroid hormones turn on specific genes

Genetic Engineering

- Biotechnology
  - Field in which scientists make controlled use of living cells to perform specific tasks
- Genetic engineering
  - A subset of biotechnology involving the manipulation of DNA for human purposes
  - Some uses
    - Produce drugs and hormones
    - Improve treatments of human diseases
    - Increase food production from plants and animals

Recombinant DNA

- DNA made from two or more sources
- Created when a gene of interest (one that produces a useful protein or trait) is put into another piece of DNA

Recombinant DNA

Recombinant DNA

- Steps in creating recombinant DNA
  1. Restriction enzymes used to cut gene of interest out of original DNA and splice it into vector DNA
  2. Plasmids are common vectors (circular pieces of DNA that exist separately from bacterial chromosome)
  3. Vector (recombined plasmids) taken up by bacteria (new host cells)
  4. Bacteria with gene of interest identified and isolated
  5. Produce numerous copies of the gene (amplify the gene)
     - Bacterial cloning
     - Polymerase chain reaction (PCR)

Recombinant DNA

Applications of Genetic Engineering

- **Environmental**—example: oil-eating microbes that can withstand ocean conditions
- **Livestock**—example: cows that produce more milk
- **Pharmaceuticals**—example: gene pharming
  - Create transgenic animals that produce a protein with medicinal value in their milk, eggs, or blood
  - Collect and purify protein
- **Raw materials**—example: goats and bacteria genetically engineered to produce spider silk proteins
- **Agriculture**—example: crops genetically engineered to be resistant to pests

Gene Therapy

- Put functional genes into body cells affected by defective (mutant) gene
  - Viruses used to deliver healthy gene
- Potential to cure inherited diseases
  - Some success with severe combined immunodeficiency disease (SCID) and X-SCID (mutant gene on X chromosome)

Genomics

- Genome
  - Entire set of genes carried by one member of a species
- Genomics
  - Study of entire genomes

Genomics

- Human Genome Project
  - Sequenced all of the DNA in a representative human cell
  - Human genome consists of 20,000 to 25,000 genes
  - Humans are identical in 99.9% of the sequences of their genes

Genomics

- Microarray
  - Tool used to analyze gene activity under different conditions
  - Thousands of DNA sequences stamped onto a single glass slide (DNA chip)
  - Can be used to identify genes that are active in cancerous cells, but not in healthy cells

Genomics

- Comparisons of genomes of different species provide evidence of common evolutionary past
  - We share 50% of our genes with fruit flies
  - We share 90% of our genes with mice

You Should Now Be Able To:

- Know the form of DNA
- Know the process of DNA replication
- Know all the steps of gene expression
- Understand what are mutations
- Understand the regulation of gene activity
- Understand genetic engineering and genomics