Bacterial Genetics and DNA Technology

I. Bacterial Genetics

A. Circular DNA molecule without histone proteins; found in nucleoid region

B. DNA replication (pg. 340)

1. One origin of replication
2. Proceeds in both directions

C. Plasmids

1. Circular, small piece of DNA
2. Separate from bacterial chromosome – “extra-chromosomal”
3. Contains several traits, i.e. antibiotic resistance

D. Bacterial transformation

1. Uptake of foreign DNA, often via a plasmit
2. Cells must be “competent”
3. Evidenced by a new trait

II. **Restriction enzymes** (endonucleases) pg. 377

A. Make genetic engineering possible

B. Found in bacteria

C. Cut up foreign DNA at specific base pair combinations (restriction sites)

1. Blunt ends

SmaI CCC↓GGG CCC GGG

GGG↓CCC GGG CCC

2. Sticky ends

EcoRI G ↓AATTC G AATTC

CTTAA ↓G CTTAA G

III. **Cloning** (pg. 378)

A. Making copies of genes, cells, or organisms

B. Extract a bacterial plasmid

C. Insert “gene of interest” into plasmid using restriction enzymes

D. Return plasmid to bacteria

E. Bacteria reproduces, making numerous copies of plasmid containing “gene of interest”

IV. Recombinant DNA

A. Combining DNA from 2 organisms

B. Carry foreign DNA into cell by:

1. Plasmids
2. Viral vectors

V. Polymerase chain reaction (PCR) pg. 382

A. Method to make multiple copies of DNA from a very small sample very quickly

B. Amplifies the amount of DNA

VI. Gel Electrophoresis

A. Using electric current passed through a gel matrix to separate macromolecules, like DNA or proteins

B. DNA fragments separate out according to size; agarose gel acts like a sieve

C. Phosphate groups of DNA are negative; attracted to positive end of electric field

VII. DNA sequencing

A. Human genome project complete – entire base sequence of human genome determined

B. How accomplished

1. Cut DNA at specific nucleotides using restriction enzymes
2. Used electrophoresis and radioisotopes to detect distinct patterns and analyze
    a. Radioisotopes allow us to visualize the pieces of cut DNA (or protein) run in a gel