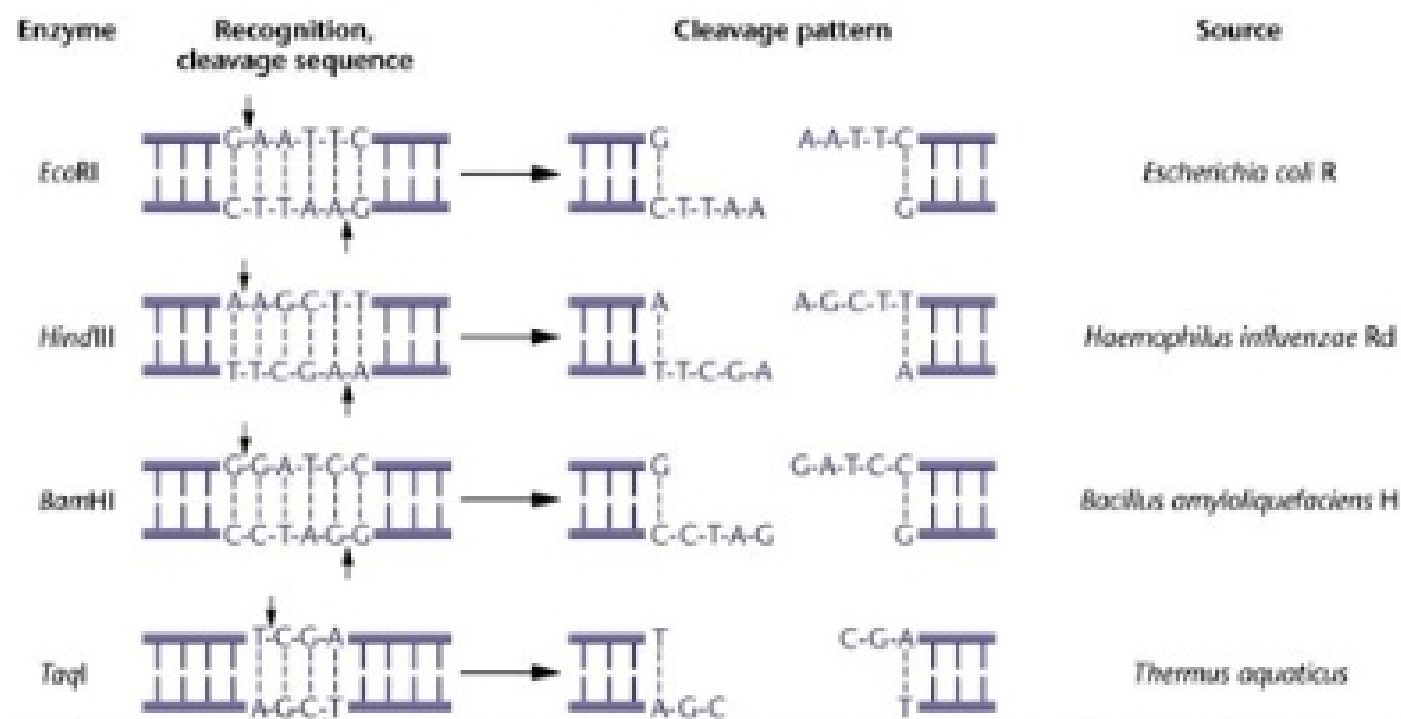


## Recombinant DNA Technology

- Human Experimentation with DNA
  - Modern roses are the result of centuries of breeding between wild roses
  - All dogs belong to a single species
  - Variation in animals teaches us about variation amongst ourselves
- Why Recombinant DNA Technology?
  - Cloning of the Human Growth Hormone (HGH) or somatotropin gene permits the production of HGH
  - It is a 191 aa protein found at it's highest levels in children and young adults
  - By cloning the human somatotropin gene and expressing it in bacteria young children with dwarfism can achieve close to normal size
    - before cloning this was almost impossible
- Biotech Industry in the U.S.
  - 1. The US is the largest market and leading consumer of biotech products in the work and home to more than 1300 firms
  - 2. Between 2001-2010 the US bioscience industry grew by 6.4% adding more the 96000 jobs
    - in that time total employment for all private sector industries in the US fell by 2.9%
  - 3. more than 5.5 million scientists, engineers and technicians in the US
  - 4. Research and development in the US biotech sector drives the commercialization of products for domestic consumption and international trade
  - **Industry Subsectors Medical Biotechnology**
    - The largest component of the biotech industry
      - Key product areas: biological drugs, vaccines, and invitro diagnostics
    - In the past 10 years research testing, and medical lab jobs have increased in the US by 24%
  - **Agricultural Biotech**
    - Key products: biotech crops, crop seeds, related products
    - the US accounts for more than ½ of all biotech crops in the world
  - **Industrial Biotech**
    - Key products: industrial application that cut across other industry sectors
    - primary product categories: nanotech, enzymes, and bio fuels
    - one of the main drivers of demand for biofuels in the US is the RFS
- Why do we clone genes?
  - **Organismal Cloning**- asexual reproduction in eukaryotes
  - **Molecular Cloning**- replication of DNA with aid of plasmid or viral vectors (a small self-replicating genetic element) in appropriate host cells
  - After coming a gene it's function can be investigated by many new molecular techniques
    - 1. The nucleotide sequence can be determined

- 2. The amino acid (protein) sequence can be predicted from the DNA sequence
  - 3. The protein function can be explored by comparing its amino acid sequence with those of other proteins stored in large databases
  - 4. The cloned gene can be studied using molecular biology techniques
  - 5. Recombinant genes (and proteins there from) can be used in medical and other applicants
- Steps in Constructing Recombinant DNA molecules
  - 1. purify DNA from organism
  - 2. Cut DNA with 'restriction endonucleases' that recognize DNA at specific sequences
  - 3. Join restricted fragments into carrier molecules to make 'recombinant DNA (rDNA) molecules'
  - 4. Introduce rDNA molecules into host cell by transformation
  - 5. rDNA molecules replicate (are cloned) in host cell
  - 6. Cloned DNA purified in large amount for study
  - 7. Cloned genes can expressed to make proteins for application
  - **KEY POINT: Natural existing enzymes are the main tool of rDNA tech**
- Different restriction enzymes recognize different palindromes



Palindromes are nucleotide pair sequences that read the same forward

- **palindrome**- nucleotide pair sequence that reads the same forward or backward from a central axis of symmetry
- Restriction enzymes cut DNA at specific sites

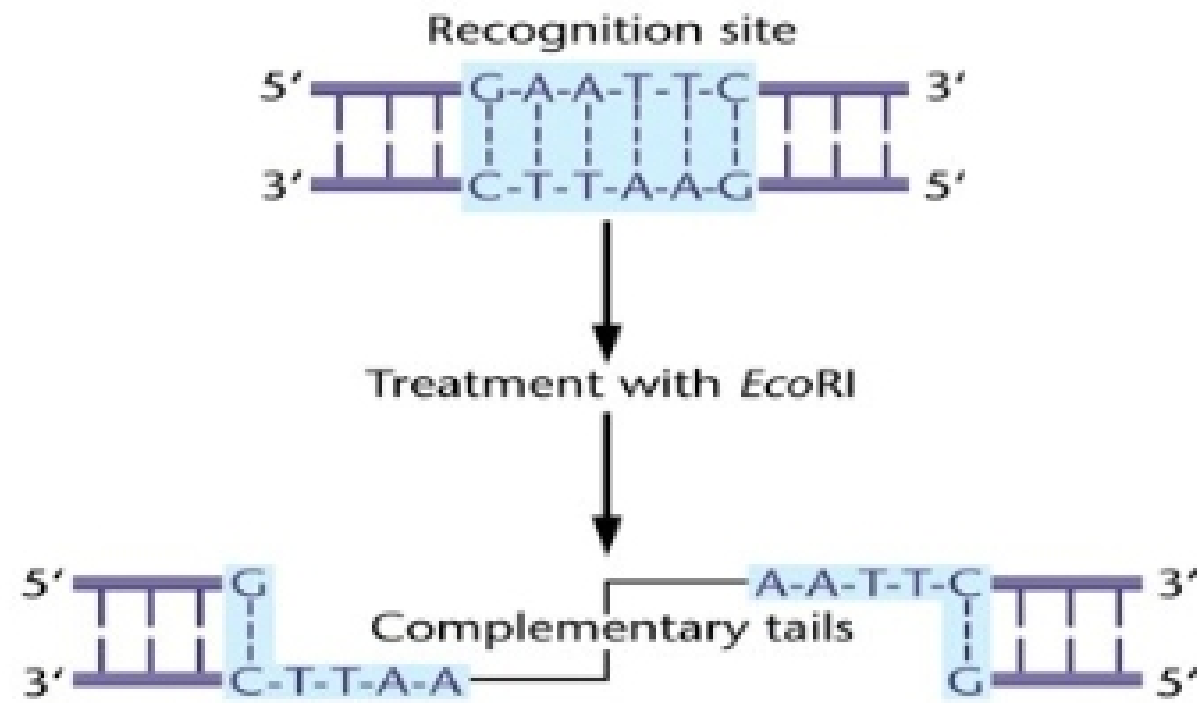
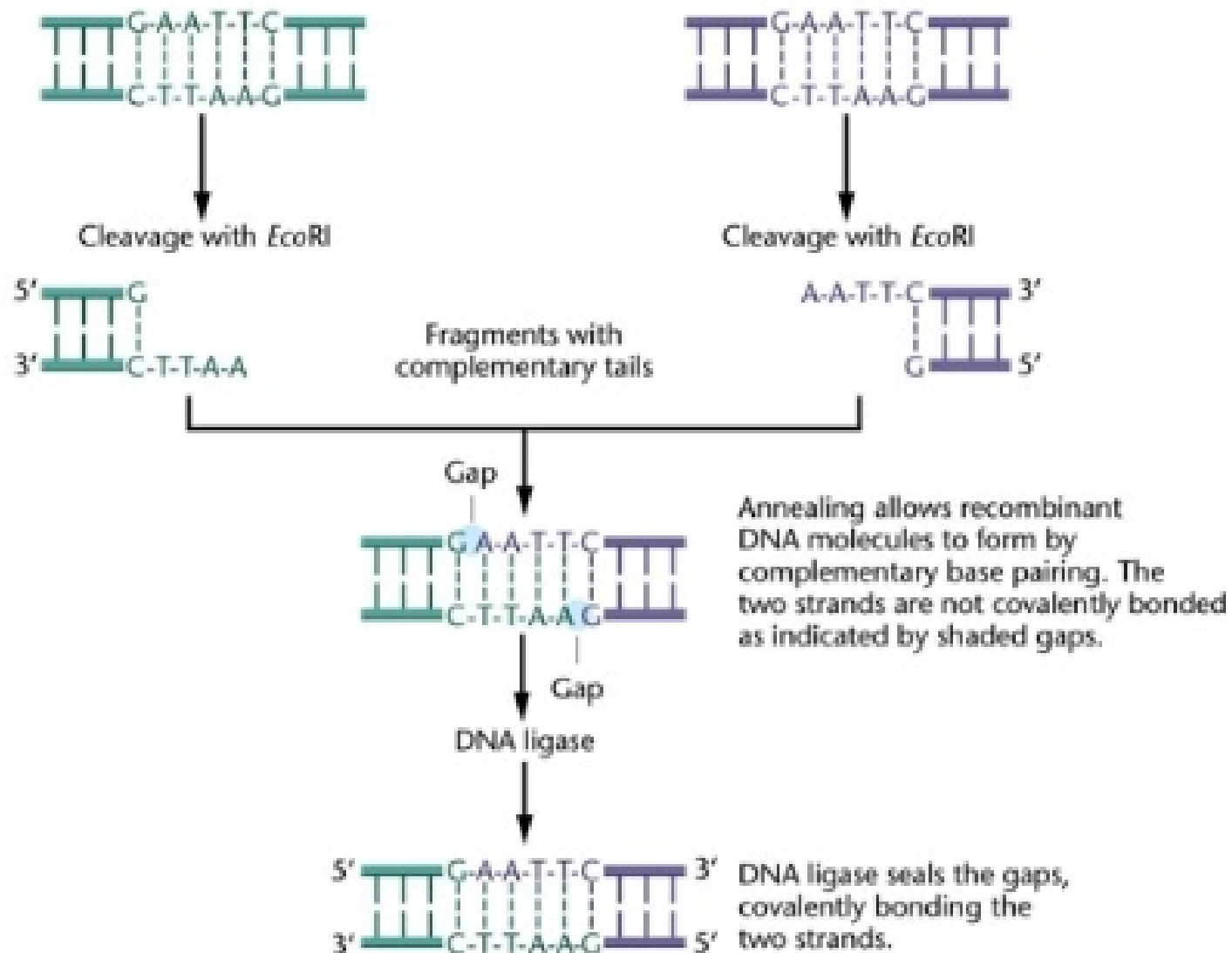


Figure 17-1 *Essentials of Genetics*, 6th  
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- As long as the single stranded tails are complementary they can be ligated back together



- Electrophoresis allows DNA molecules to be separated by length
  - Sugar phosphate backbone of DNA is negatively charged and will migrate toward the positive pole (anode) in an electric field
- Bacterial Cloning Vectors
  - Essential Features of a Cloning Vector
    - 1. Origin of replication for bacteria (*ori*R) Centromeres and telomeres for yeast