Zoo-352 Principles of genetics
Lecture 10

Genetic engineering

Outlines:

- An overview of genetic engineering
- Definition of gene cloning
- Steps of recombinant DNA cloning
- Examples of using genetic engineering

Introduction

- Genetic engineering is the key technology to develop several recombinants on the DNA, RNA and protein levels.
- These recombinants are useful to explore gene function, gene expression and in genetic modification.
- Genetic engineering is used in medicine, agriculture, livestock, related research fields and industries.
- For instance, bacteria can be used to produce human insulin by inserting an engineered DNA sequence into a plasmid, allowing the bacteria to replicate and synthesize the insulin.
- In the 1970s, gene cloning became a reality.

Using DNA cloning for producing human insulin

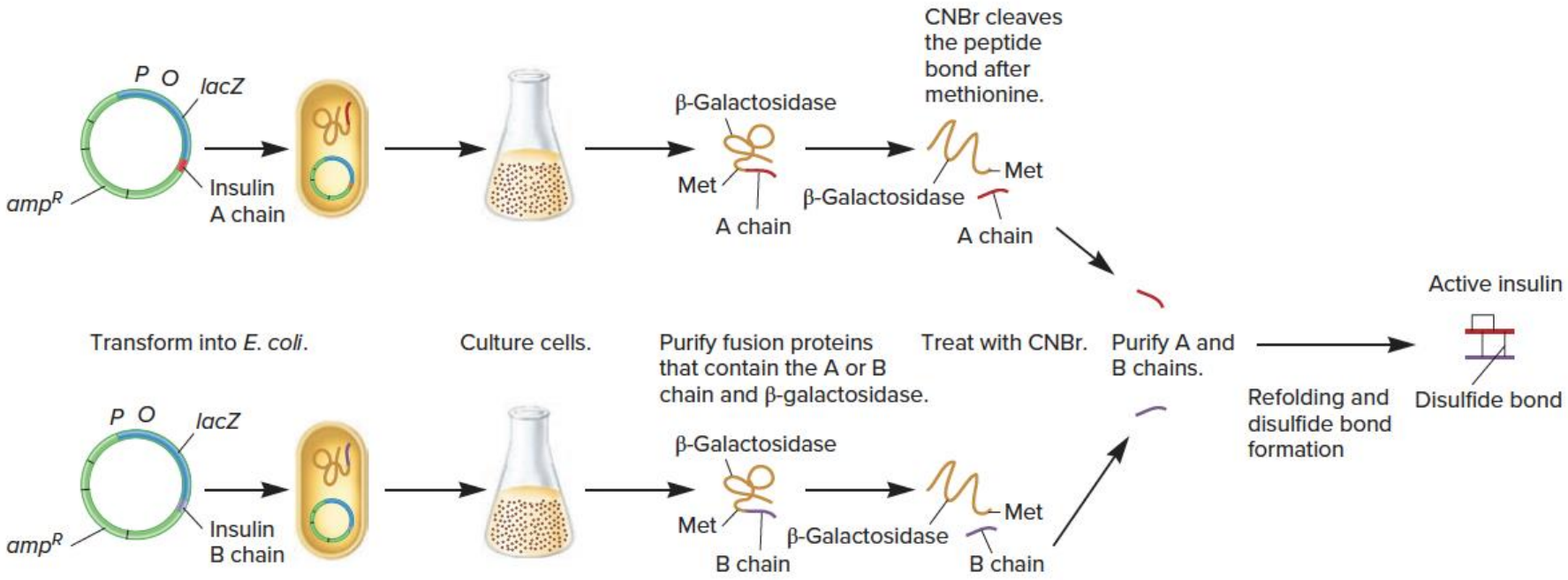


FIGURE 21.1 The use of bacteria to make human insulin. In recent forms of manufactured insulin, slight changes have been made to the insulin amino acid sequence. These changes prevent insulin molecules from clumping together, and thereby improve the manufactured insulin's biological properties.

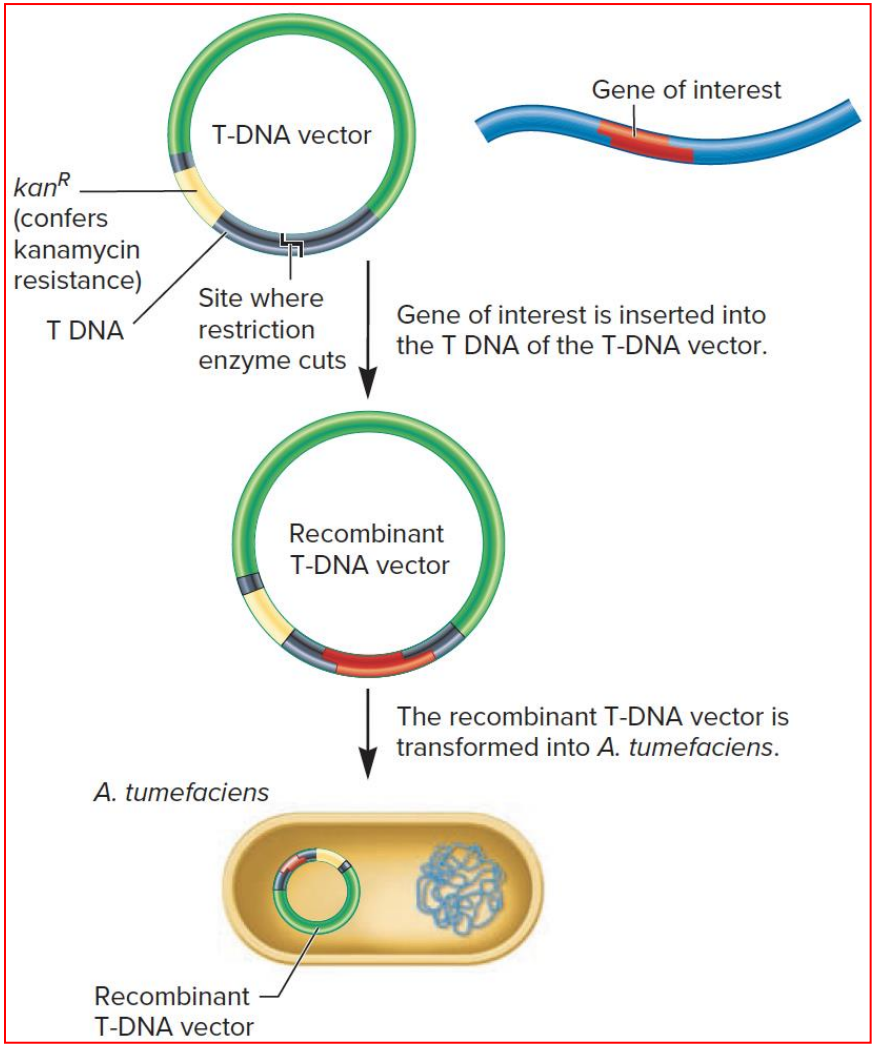
Genes→Traits The synthesis of human insulin is not a trait that bacteria normally possess. However, genetic engineers can introduce the genetic sequences that encode the A and B chains of human insulin via recombinant DNA technology, yielding bacteria that make these polypeptides in the form of fusion proteins that also contain β-galactosidase.

- **Gene cloning** is the process of producing multiple copies of a gene using molecular techniques, such as PCR or by introducing the gene into a vector that replicates in a host cell.

Steps of recombinant DNA cloning

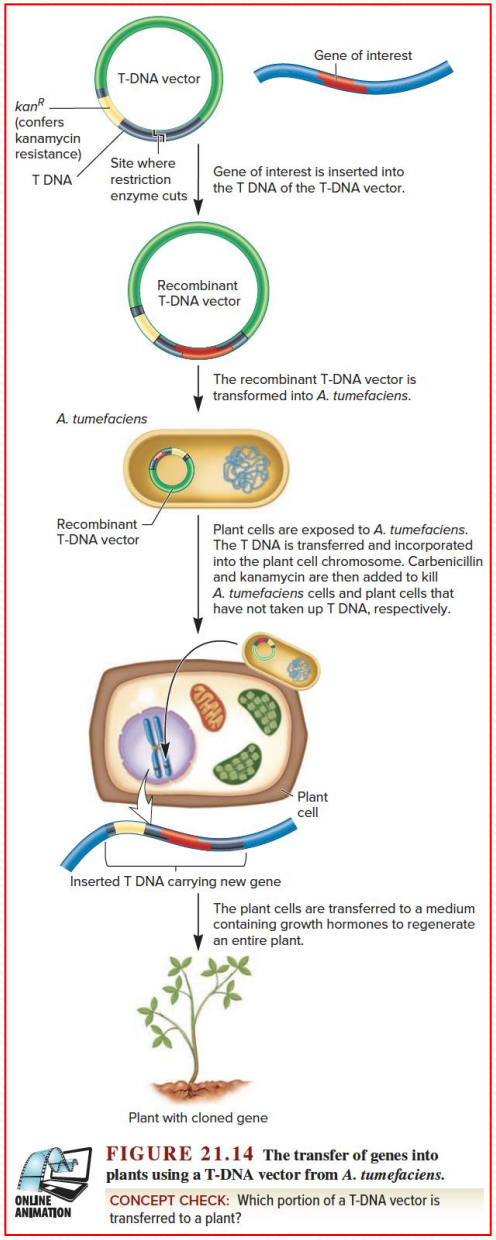
1. Isolating a specific gene or DNA sequence.
2. Prepare a plasmid DNA vector and the engineered DNA sequence using restriction enzymes
3. Ligating the engineered DNA sequence into the plasmid DNA vector to generate a circular plasmid DNA using DNA ligase.
4. Transforming bacteria with the engineered plasmid allows for the cloning of the DNA. Competent strains of *E. coli* are commonly used as biological control agents in this process.
5. Depending on the purpose, either:
   a. the recombinant RNA or proteins will be obtained from the transformed bacterial lysate, or
   b. the cloned plasmid DNA will be used for further cloning by transfection into other cell types, e.g. to generate a genetically modified organism (GMO).

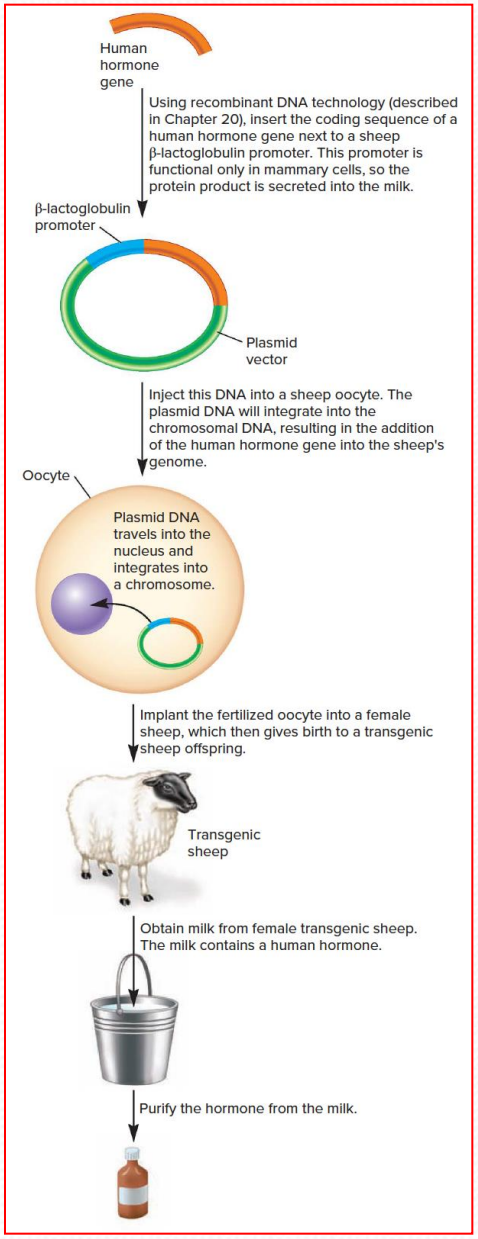
- Gene editing is experimentally altering the sequence of a gene.
- Endonuclease is an enzyme that can cleave a bond between adjacent nucleotides within a DNA or RNA strand.
- Restriction enzyme is an endonuclease that cleaves DNA.
- The restriction enzymes used in cloning experiments bind to specific base sequences and then cleave the DNA backbone at two defined locations, one in each strand.



Examples of using genetic engineering

- The techniques employed in genetic engineering have led to the production of important products, including:
  - human insulin,
  - human growth hormone,
  - hepatitis B vaccine,
  - the production of cancer therapies,
  - the development of genetically modified organisms such as disease-resistant plants and livestock.





Quiz: Genetic engineering

1. What is commonly used to clone recombinant DNA?
   - A segment of bacterial DNA from the nucleoid
   - Plasmids from a bacteria
   - DNA from a recombinant bacterium
   - DNA from a eukaryotic cell

2. How do plasmids enter bacterial cells?
   - Restriction enzymes
   - DNA ligase
   - DNA vector
   - Transformation

3. Which enzyme assists in joining the recombinant plasmid together after the action of restriction enzymes?
   - Restriction enzymes
   - DNA polymerase
   - DNA ligase
   - RNA polymerase

4. What are applications of DNA cloning?
   - genes for pest resistance
   - human growth hormones
   - genes used to treat cancer
   - all of the above