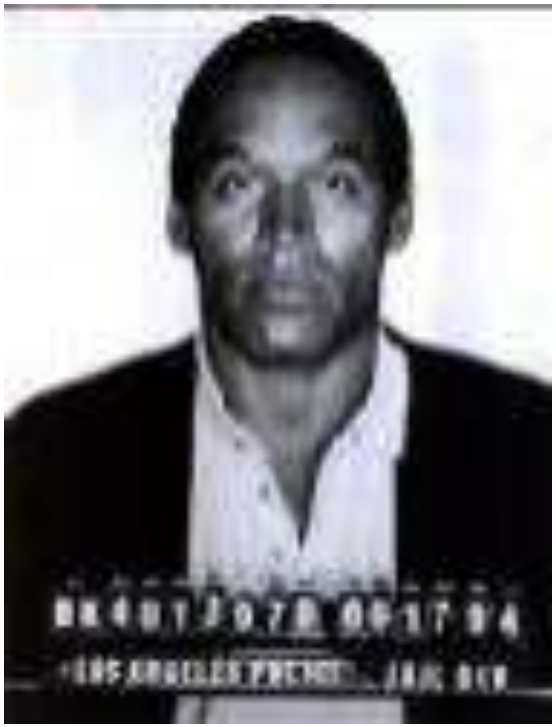
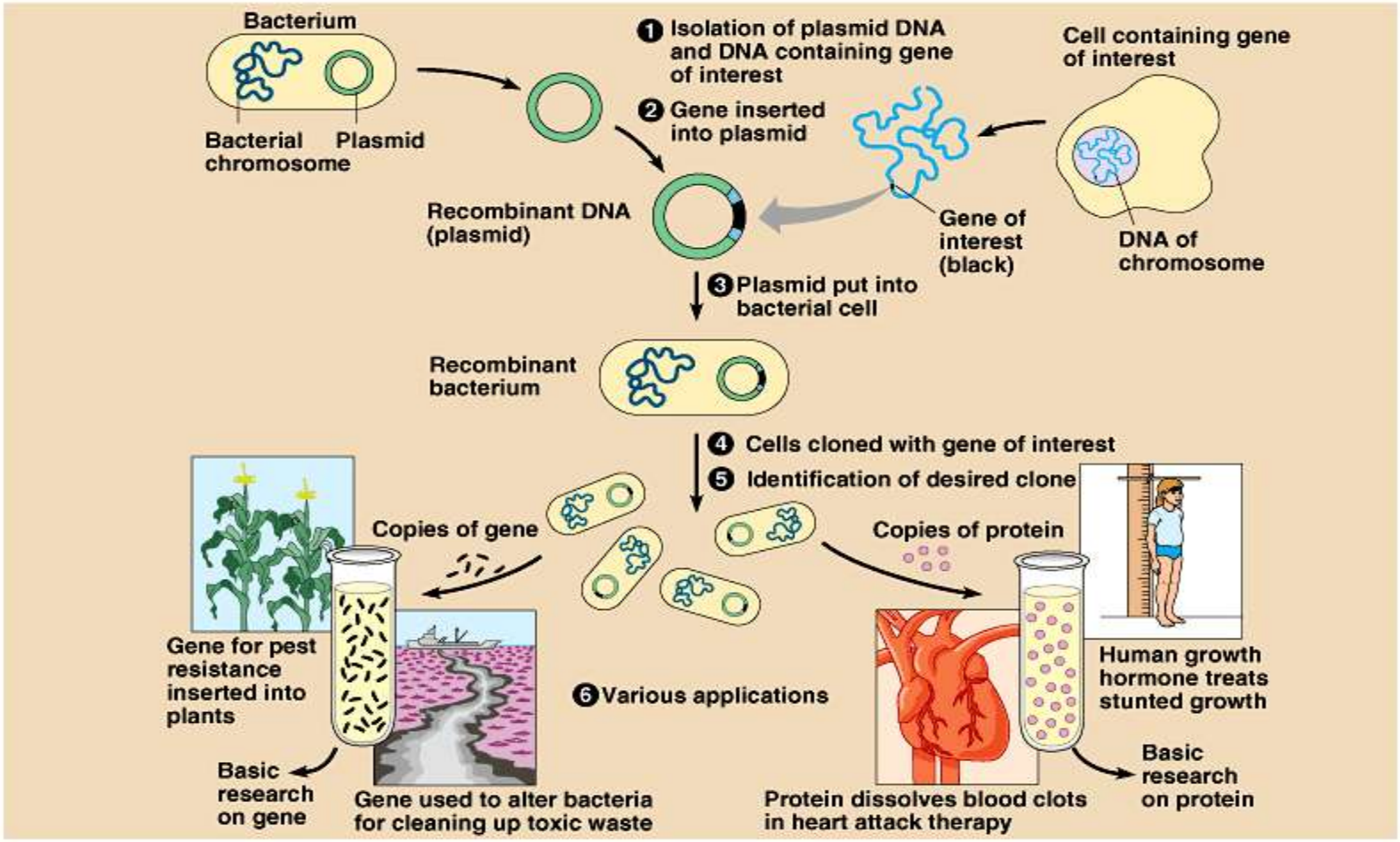


- DN*A Technology & Genomics*

Recombinant DNA

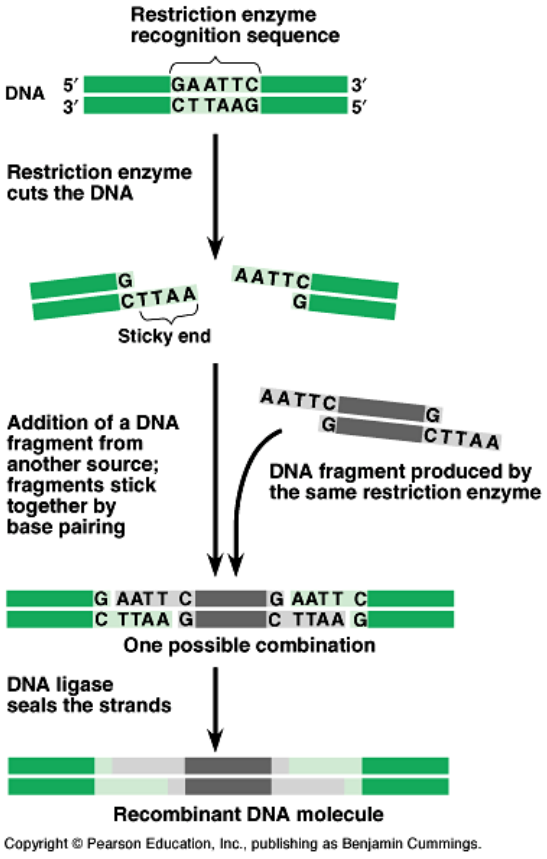
- **<u>Def:</u> DNA in which genes from 2 different sources are linked**
- **<u>Genetic engineering:</u> direct manipulation of genes for practical purposes**
- **<u>Biotechnology:</u> manipulation of organisms or their components to perform practical tasks or provide useful products**

Bacterial plasmids in gene cloning



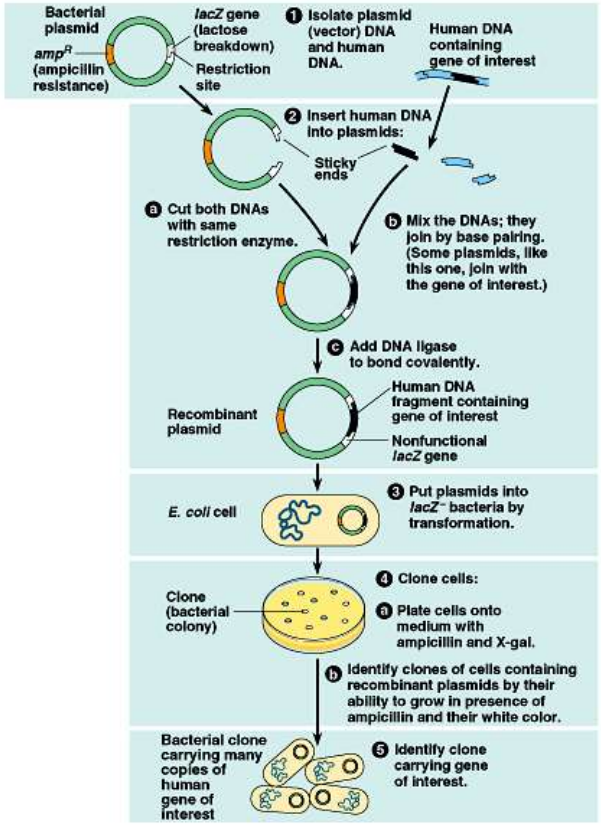
DNA Cloning

- **Restriction enzymes (endonucleases): in nature, these enzymes protect bacteria from intruding DNA; they cut up the DNA (restriction); very specific**
- **Restriction site: recognition sequence for a particular restriction enzyme**
- **Restriction fragments: segments of DNA cut by restriction enzymes in a reproducable way**
- **Sticky end: short extensions of restriction fragments**
- **DNA ligase: enzyme that can join the sticky ends of DNA fragments**
- **Cloning vector: DNA molecule that can carry foreign DNA into a cell and replicate there (usually bacterial plasmids)**



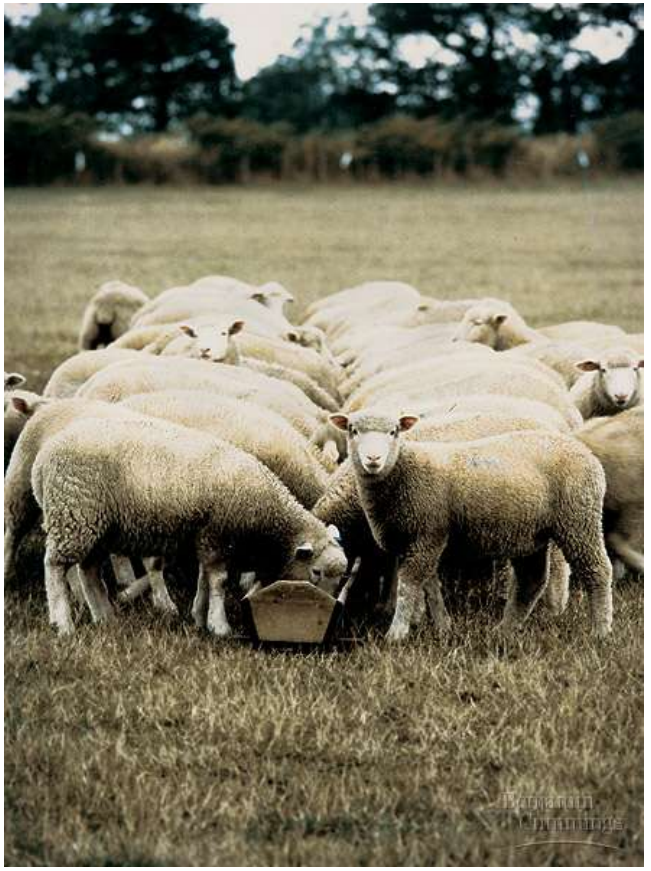
Steps for eukaryotic gene cloning

- Isolation of cloning vector (bacterial plasmid) & gene-source DNA (gene of interest)
- Insertion of gene-source DNA into the cloning vector using the same restriction enzyme; bind the fragmented DNA with DNA ligase
- Introduction of cloning vector into cells (transformation by bacterial cells)
- Cloning of cells (and foreign genes)
- Identification of cell clones carrying the gene of interest



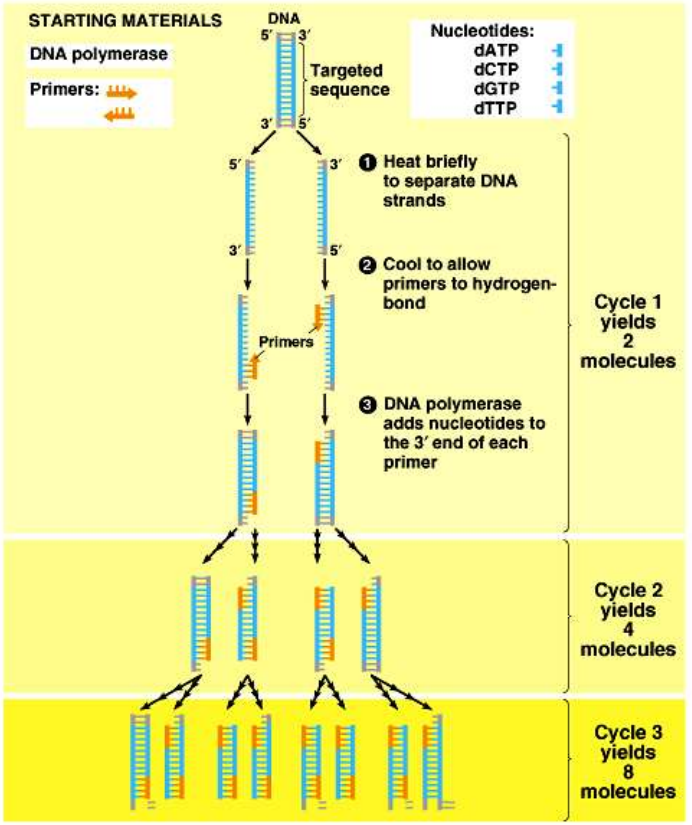
DNA Analysis & Genomics

- PCR (polymerase chain reaction)
- Gel electrophoresis
- Restriction fragment analysis (RFLPs)
- Southern blotting
- DNA sequencing
- Human genome project



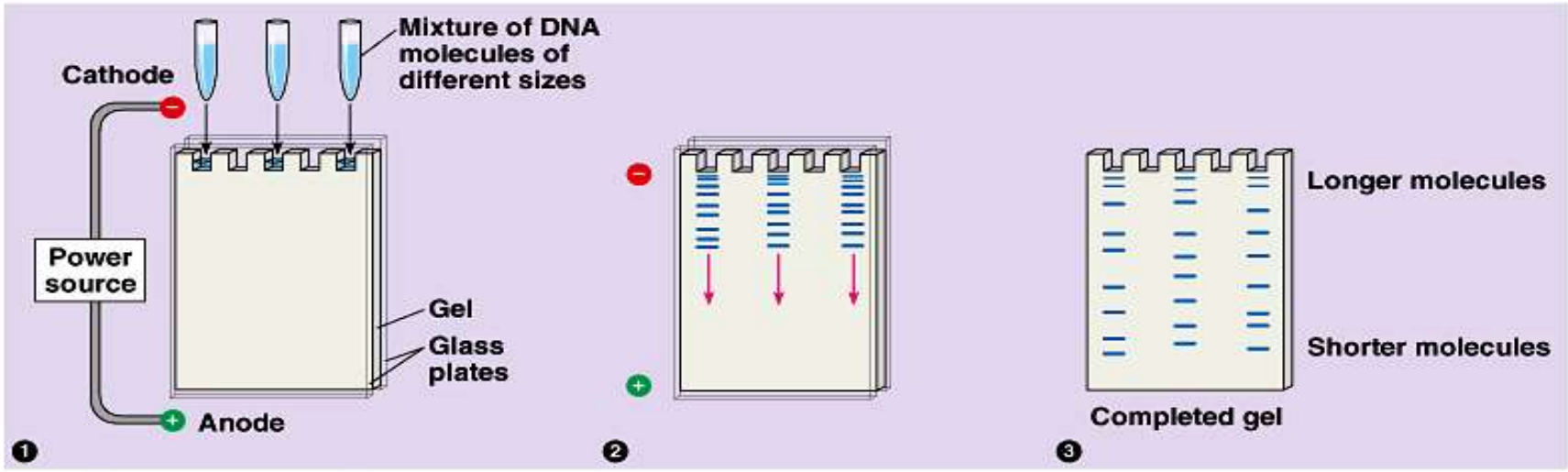
Polymerase chain reaction (PCR)

- Amplification of any piece of DNA without cells (in vitro)
- Materials: heat, DNA polymerase, nucleotides, single-stranded DNA primers
- Applications: fossils, forensics, prenatal diagnosis, etc.



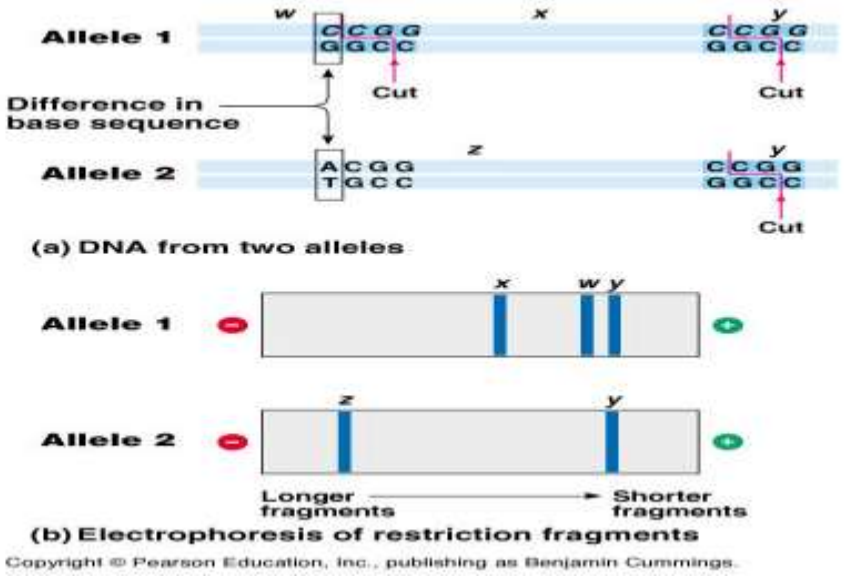
DNA Analysis

- **<u>Gel electrophoresis</u>: separates nucleic acids or proteins on the basis of size or electrical charge creating DNA bands of the same length**



Restriction fragment analysis

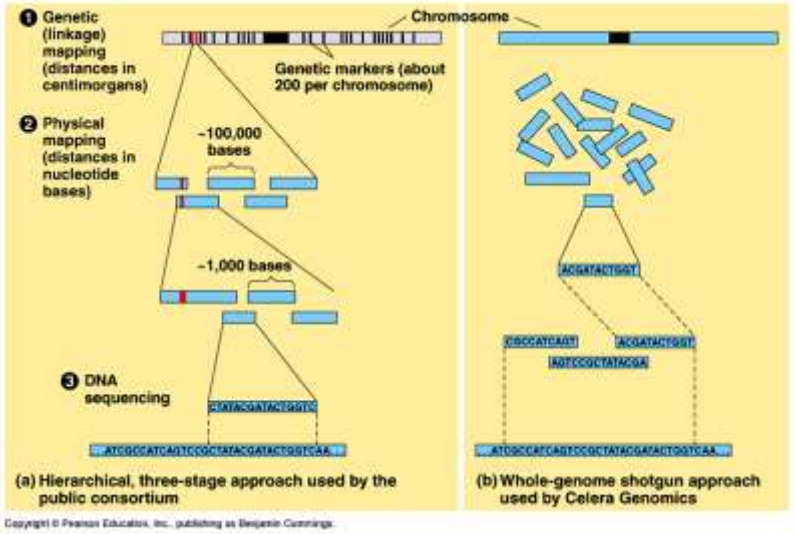
- Restriction fragment length polymorphisms (RFLPs)
- Southern blotting: process that reveals sequences and the RFLPs in a DNA sequence
- DNA Fingerprinting



(c) Completed gel

DNA Sequencing

- Determination of nucleotide sequences (Sanger method, sequencing machine)
- Genomics: the study of genomes based on DN sequences
- Human Genome Project



Practical DNA Technology Uses

- **Diagnosis of disease**
- **Human gene therapy**
- **Pharmaceutical products (vaccines)**
- **Forensics**
- **Animal husbandry (transgenic organisms)**
- **Genetic engineering in plants**
- **Ethical concerns?**