

Concept 13.1: Biologists have learned to Manipulate DNA

(DNA technology has developed new ways to modify organisms.)

I. The Beginnings of DNA Technology

A. **Biotechnology** = use of organisms to perform practical tasks for humans

1. Newest type – applic. that analyze and manipulate the genomes of orgs = **DNA technology**
2. Research with *Escherichia coli* (*E. coli*) bacterium – models for gene manip
 - a. Bacteria can alter DNA in its genome in 3 ways (1940's research)
 - 1.) tunnel like connection with other bacterium, to transfer genes (See Fig. 13.1, p. 266)
 - 2.) Viral injection of DNA from 1 bacterium to another
 - 3.) Bacteria can take up DNA from surrounding enviro. (How Transformation occurred in Pneumonia and mice study)
 - b. **Recombinant DNA technology** = set of laboratory techniques to combine genes from different sources—even different species—into a single DNA molecule.

II. DNA Technology and Frontiers of Research in Biology

A. Sequenced Genomes of: Humans, Fruit fly, baker's yeast, *E. coli* and rice plant

1. Useful applications:
 - a. Ex. rice genome – may help us make it more nutritious
 - b. Ex. research on simpler orgs may be applied to humans w/o as much risk
 - c. Ex. may answer ? about evolutionary patterns

Concept 13.2: Biologists can Engineer Bacteria to Make Useful Products

(Bacteria “workhorses” used to mass produce useful genes and proteins by modern biotech.)

I. Engineering Bacteria: An Introduction

A. **Plasmid** = a small, circular DNA molecule separate from the much larger bacterial chromosome

1. See Fig. 13.3, p. 268
2. may carry a number of genes, and can make copies of itself
 - a. replicated plasmids can be passed from 1 bacterium to another, may ↑ survival w/ adv. genes
 - 1.) Ex. some plasmids carry genes that make them resistant to antibiotics
 - a.) can be replicated, passed to entire pop of bacteria, + even other species of bacteria!!
 - b.) Result – antibiotic resistant bacteria that cause human disease
3. Use of plasmids to Benefit Humans
 - a. Ex. use plasmids to move DNA pieces, such as genes for useful products, into bacteria
 - 1.) See Fig. 13.4, p. 269
 - 2.) Process: (Read p. 269) = **Gene Cloning**
 - a. remove plasmid, insert gene into it, = now recombinant DNA
 - b. recombinant plasmid put back into bacterium
 - c. bacterium replicates plasmid, reproduces itself
 - d. result: pop. of bacteria with new gene!

II. “Cutting and Pasting” DNA

A. How do biologists move a gene to another DNA molecule? (Read p. 269)

1. Cut desired DNA out of longer piece with **restriction enzymes** = enzymes found in bacteria and protect the bacteria against intruding DNA from other organisms and phages. (chop up foreign DNA into small pieces)

- a. Each enzyme is nucleotide sequence specific
- b. Cuts at specific points in longer DNA molecule (See Fig. 13.5, p. 269)
- c. Not clean cuts – leave “sticky ends” on longer DNA = perfect site for recombining!
- d. Any 2 sticky ends can recombine with a complementary sequence – DNA ligase enzyme makes this “pasting” happen

III. Cloning Recombinant DNA

A. Purpose: To set up bacterial “factory” colony to make valuable human protein for human use

1. How? See Fig. 13.6, p. 270

a. Libraries of Cloned Genes - a by-product of Cloning Recombinant DNA

1.) Step 5 – actually results in clones of desired gene, and additional clones of other genes in the DNA fragments = **genomic library** (See Fig. 13.7, p.271)

b. Identifying Specific Genes with Probes

1.) How do you find a gene in the genomic library?

a.) If know part of the genes nucleotide sequence, can build compl. strand w/radioactive isotopes to find it = **nucleic acid probe** (See Fig. 13.8, p. 271)

b.) Rest of process – read p. 271

IV. Useful Products from Genetically Engineered Microorganisms

A. News is full of examples of Recombinant DNA in Action:

1. Bacteria to clean up toxic wastes (oil spills, ex.)

2. Bacteria mass producing useful chemicals: pesticides, useful drugs, etc.

3. Medicines examples

a. hormone insulin for diabetics

1.) Pig insulin – similar, but some people had side effects, + \$\$\$\$

2.) recombinant bacteria w/gene for insulin prod – less \$\$\$\$, no side effects

b. effective vaccines – ex. hepatitis B vaccine cloned using recombinant yeast cells

Concept 13.3: Biologists can Genetically Engineer Plants and Animals

{Plants can be grown, not just from cuttings, but from a single genetically engineered plant cell!}

I. Producing Genetically Modified Plants

A. **Genetically Modified Organism (GMO)** = any organism that has acquired one or more genes by artificial means

1. **Transgenic** = if the source of the new genetic material is a different species.

B. Genetic Engineering replacing Traditional Plant Breeding

1. Most often used – if a plant’s useful traits determined by 1 – a few genes

2. Ex. ½ the crops of soybeans and corn in U. S. in 2000 were gen. eng. in some way

a. herbicide resistance – can destroy weeds w/o hurting crops

b. others – pesticide and fungi resistance

3. How? – usually use plasmid from a soil bacterium to insert the gene into plant cells

a. See Fig. 13.11, p.275

4. Other Examples: genes for delayed ripening, improved nutritional content, and resistance to spoilage and disease

II. Producing Genetically Modified Animals

A. Much more difficult than GM plants

1. Process: Read p. 275 – must extract egg, fert. w/sperm in test tube, then inject the fert. egg into a surrogate mother, to develop and be born, etc (See Fig. 13.13, p. 276)

(2. This is how “Dolly” the sheep was cloned)

B. Goals for GM animals

1. Often similar to trad. methods of breeding
 - a. Ex. better wool on sheep, pig with leaner meat, fish that mature faster
2. Or, may be to make transgenic animal that makes lg amounts of rare bio. substance
 - a. Ex. Often human hormones, that can then be passed in the orgs milk to be purified

III. Animal Cloning

- A. Read p. 276 for Process
- B. Cloned: sheep, and more recently dogs and cats
- C. Purpose: mass produce animal with a desired set of traits
 1. Much faster than traditional breeding, - may take generations
 2. Esp. useful if cloning a GM animal

IV. The GMO Controversy (Read p. 277)

- A. Concerns
 1. GM Crops pollen may spread to wild types, create “Superweeds”
 2. GM crops and/or animals may have unknown risks to human consumers
- B. U.S. – Govt agencies evaluating safety of GMO projects for public health and consumers’ risks
 1. Should GMO products/foods be labeled? (See Fig. 13.14, p. 277)

Concept 13.4: DNA Technologies have Many Applications

(DNA tech. is rapidly changing the way society solves some problems.)

I. Mass – Producing DNA in a Test Tube

- A. **Polymerase Chain Reaction (PCR)** = a technique that makes many copies of a certain segment of DNA without using living cells.
 1. can generate 100 billion identical molecules in a few hours from 1 DNA molecule
 2. See Fig. 13.15, p. 278 for steps
 3. Adv. – can copy a specific segment, not the whole molecule, do not even need purified DNA
 - a. if using living cells: would need much more time and a lg. amt. of starter DNA
 4. Uses
 1. Further analysis of the sample much easier
 2. rare DNA can be copied – Ex. Clone DNA samples from 5,000 year old human remains, from a woolly mammoth 40,000 yrs old, and a 30 mil yr old plant fossil
 3. medicine – used to detect viral genes infected with HIV

II. Comparing DNA

- A. **Gel Electrophoresis** = technique for sorting molecules or fragments of molecules by length (see Fig 13.16, p. 279)
 1. Steps: read p. 279
 2. Results: a series of colored bands for each DNA fragment in each sample – farther from pocket of DNA = shorter the fragment
(DNA travels across the gel b/c all DNA neg. charged, attracted to pos. charged pole)
- B. **Genetic Markers** = banding pattern produced by specific portions of the genome that are variable among individuals
 1. saves time – each DNA molecule can contain 1000’s of fragments, many which are common to all members of the species
 2. can use radioactively labeled DNA probes to “tag” the bands w/particular genetic markers
 - a. Ex. Can use to ID recessive alleles in people with family history of genetic disease
- C. **DNA Fingerprinting** = a particular banding pattern produced by your restriction fragments
 1. unless identical twin, DNA fingerprint are unique
 2. genetic markers can even show up in noncoding (intron) sections of a person’s genome
 - a. these are the ones used in court cases – least likely to be shared with any other person

3. can be made from a single drop of blood or a hair follicle – using both PCR and gel electr.
4. 2 fingerprints alike? Less than 1 in 100,000 up to less than 1 in 1 billion (if more markers compared)

Concept 13.5: Control Mechanisms switch Genes On and Off

(In multicellular orgs, all genes have same DNA, but cells look and function differently – How?)

I. Regulation of Genes in Prokaryotes

- A. Do not turn genes “on or off” like eukaryotes
 1. Can change its functions in response to envir. Changes
 - a. Ex. *E. coli* ‘s response to milk in your dig. sys.(produces 3 enzymes to digest it)
vs. no milk (does not produce the enzymes)
- B. **Operon** = a cluster of genes, along with its control sequences (see Fig. 13.18, p. 281)
 1. Ex. *Lac* operon for lactose digestion
 2. Consists of: Promoter, operator and genes to be activated
 3. how it works – read p. 281 – 282
 4. **Repressor** = a protein that functions by binding to the operator and blocking the attachment of RNA polymerase to the promoter, active when substance (ex. Lactose) missing in diet.
 - a. keeps genes for enzymes “turned off” (see Fig. 13.19, p. 282)
 - b. when Lactose present, it binds to the repressor, so it cannot stop the operator, enzymes get made
- C. Many diff. operons in prokaryotes – can adjust cell chemistry to suit their envir.

II. Regulation of Genes in Eukaryotes

- A. More complicated than Prokaryotes
 1. do have promoter sequences
 2. **Transcription factors** = proteins that regulate transcription by binding to those promoters or to RNA polymerases
 - a. activated/deactivated by chem. signals in cell
 - b. ex. Hormones attach to TF’s, signal cells to express certain genes
 - 1.) **gene expression** = transcription and translation of genes into proteins

III. From Egg to Organism

- A. Gene regulation begins w/fertilized egg
 1. positions of cells within embryo determine which genes get expressed – read p. 283
 2. **cellular differentiation** = process of individual cells becoming increasingly specialized in structure and function
 - a. each cell only expresses genes coding for proteins for functions of that cell
 - 1.) Ex. See Fig. 13.21 p, 283
 3. **DNA “chip”** = used to track changes in DNA activity during devel.
Or, can be used to reveal genetic disorders (Text does not say what a “chip” consists of)

II. Stem Cells

- A. **Stem cells** = cells that remain undifferentiated throughout an orgs life, potential to be any cell
 1. 7 day old embryo – ball of 100 cells with stem cells in middle (see fig. 13.22, p. 284)
 2. after birth – clusters of stem cells stay in several organs
 - a. Ex. bone marrow - can make dozens of diff. types of blood cells
 - b. adult stem cells cannot make all types of tissues as easily as embryonic stem cells
 - c. there are no stem cells for nervous tissue and heart muscle in adults
 3. Hope: can use embryonic stem cells to help people with disabling diseases of heart and brain, etc. – **but is it ethical?**
 4. New research – bone marrow stem cells may be used to generate new nervous tissue in self!

- poor success rate so far, though

III. **Homeotic Genes** = master control genes that direct development of body parts in specific locations in many orgs.

A. Ex. Master gene in fruit fly for eye location on head

1 Mutation – causes eyes to be in wrong places with up to 14 eyes total!
(on antenna, legs + wings) (see fig. 13.23, p. 284)

2. **Homeobox** = 180 nucleotide sequence in a homeotic gene that codes for a protein that promotes transcription of genes involved in the development of specific body parts
a. common in multicellular orgs.

b. Ex. Similar homeobox in humans and other animals to fruit fly to determine eye location

c See Fig. 13.24, p. 285)

IV. Summary:

A. Mechanisms of development very similar in many diff. orgs.

1. Evidence that supports the hypo that certain genes in diverse species were inherited from common ancestors to these species