Biotechnology:

New Terms Today:
Genome
Genetic engineering, transgenic organisms, GM food,
Reproductive and therapeutic cloning
Stem cells, plouripotent, totipotent
Gene therapy

Genomics: field that compares the entire DNA content of different organisms

the **genome:** the full complement of genetic information of an organism (i.e., all of its genes and other DNA)

DNA sequencing: a process that allows scientists to read each nucleotide in a strand of DNA

What's going on in DNA sequencing?

- ❖ Identifying a chain of nucleotides so the order of their nitrogenous bases is documented.
- ❖ DNA fingerprinting

The publication of the sequence of the ***entire human genome*** occurred on June 26, 2000

- ❖ the human genome contains more than 3 billion base pairs
- ❖ It is estimated that there are between 20K and 30K protein-encoding genes

Genetic engineering: moving genes from one organism to another

- 1st: chop up the source DNA and obtain a copy of the gene you want to transfer
- **restriction enzymes** bind to specific short sequences on the DNA and make a specific cut
- the sequence is symmetrical
- the cut generates DNA fragments that are "sticky"
- DNA from another source that is cut with the same restriction enzyme will have the same sticky ends
- these ends can be joined together by the enzyme ligase

Restriction enzymes are the basic tools of genetic engineering

DNA library: collection of DNA fragments representing all of the DNA from an organism

How do you get the DNA into the host cell?

- You need a vehicle to carry the source DNA into the host cell

4 stages of a gene transfer experiment

1. **Cleaving DNA:** cut the source & vector DNA

2. **Producing recombinant DNA:** place DNA fragments into vectors and then transferring the DNA into the target cells

3. **Cloning:** introduce DNA-bearing vectors into target cells. Then allow target cells to reproduce

4. **Screening:** select the particular infected cells that have received the gene of interest

Genetic Engineering and Medicine

- potential to improve medicine, to aid in curing and preventing illness

- Advances have been made in the following areas

    - the production of proteins used to treat illness
    - the creation of new vaccines to combat infection
      the replacement of defective genes (i.e., **gene therapy**)

      Many genetic defects occur because our cells fail to make critical proteins

    - Ie: diabetes
      Insulin (protein) transports glucose from blood across cell membranes.
      ↓ insulin → ↑blood glucose level.

      A diabetic can receive a donation of protein made by another body

      genes encoding insulin have been introduced in bacteria, which can cheaply produce lg quantities of protein

Genetically Engineered Drugs

Subunit Vaccines aka Piggyback

- ❖ produced from specific protein subunits of a virus
- ❖ has less risk of adverse reactions than whole virus vaccines.
- ❖ Used to treat herpes and hepatitis
- ❖ engineers splice genes from the coat of the virus into a fragment of cowpox (vaccinia) virus genome
- ❖ the smallpox virus is the vector
- ❖ carry the viral coat genes into cultured mammalian cells
- ❖ where the immune system can develop an immunity to the virus prior to being exposed to a fully active virus

Genetic Engineering and Agriculture

- ❖ Genetic engineering of crop plants has successfully
- ❖ made plants more resistant to disease
- ❖ improved nutritional content (?) and yield
- ❖ made crops hardier and better able to resist environmental stresses

Engineering crops to be resistant to insect pests

- ❖ reduces the need to add insecticides to the environment
- ❖ There is a soil bacteria: *Bacillus thuringiensis* (Bt),
- ❖ it contains a gene that produces a protein that is toxic when eaten by crop pests
- ❖ This gene has been inserted into the chromosomes of tomatoes
- ❖ because the plants can synthesis Bt protein, they are toxic to pests, such as the tomato hornworm

Herbicide resistance has also been genetically engineered

- ❖ **Glyphosphate:** powerful herbicide that kills most actively growing plants and is used to control weeds
- ❖ using a **gene gun**, engineers inserted an isolated gene from a bacterium that is resistant to glyphosphate into crop plants
- ❖ the glyphosphate can now be widely applied to fields and orchards where it retards weed growth but not crop growth
- ❖ Gold particles coated w/ DNA fired into plant cells where it is incorporated into cells DNA

How do you feel about this?

- ❖ Worldwide micronutrient deficiency of vitamin A and iron: genetic engineers created GM "golden" rice
- ❖ this transgenic rice contains genes from a bean, a fungus, wild rice, and a daffodil to increase its nutritional value
- ❖ Makes rice enriched with iron, and the body better able to absorb the iron in the small intestine

Bioengineers claim modify crops in two major ways

- ❖ makes the crop easier to grow
- ❖ *improves the food itself (argued)*

Is eating GM food dangerous?

- does adding genes introduce novel proteins that maybe potentially harmful when consumed?
- could introduced proteins become allergens?

Those concerned about the widespread use of GM crops raise 3 concerns

1. Poss of unintentional harm to other organisms

- ❖ Are weeds important source of food and shelter for non-pest insects?

2. potential for new resistance

- ❖ pests might become resistant to engineered proteins.
- ❖ farmers required to plant some non-GM crop alongside GM crop to slow the selection pressure for resistance

3. gene flow:

- ❖ modified genes may spread to non-GM species due to interbreeding

Reproductive Cloning

Theory of irreversible determination:

- ❖ animal cells become irreversibly committed after the first cell divisions of the developing embryo
- ❖ **nuclear transplants:** transplanting a nucleus from an animal cell into the an enucleated egg and seeing if it develops
- ❖ only cells extracted from early embryos (no further than the 16-cell stage) will develop into an adult
- ❖ we now know that this theory is WRONG

Keith Campbell, a geneticist, proposed that, in order for a successful nuclear transplant to take place, both the egg and the donated nucleus need to be in the same stage of the cell cycle

- Starve the cells so that they pause at the G_1 checkpoint

- the nuclear transfers succeed in producing cloned farm animals

Reproductive biologist Ian Wilmut worked with Campbell to clone a sheep using the mammary cells of an adult

- Despite the success of "Dolly the Sheep," only a small fraction of transplanted embryos survive to term

- most embryos will die late in pregnancy

- many exhibit **large offspring syndrome** or lateral developmental problems as they become adults

- almost none survive to a normal lifespan

Embryonic stem cells: form early in development and each has the capacity to develop into a healthy individual

Totipotent is the ability of cells, such as stem cells, to have the ability to form any body tissue, and even an adult animal

- As development proceeds, some of the embryonic stem cells begin to become committed to forming a certain type of tissue only

- every major tissue is formed from its own tissue-specific **adult stem cell**

- possibility of restoring damaged tissues

- Using embryonic stem cells to restored damaged tissue

Therapeutic Cloning

- Aka **somatic cell nuclear transfer**
  - address the issue of immune acceptance of a transplanted cell used for therapy
  - **therapeutic cloning:** cloned embryo is destroyed to harvest embryonic stem cells, which will be automatically tolerated by the recipient of the therapy
  - **Reproductive cloning**, the cloned embryo is allowed to develop into adults

many ethical issues

Gene Therapy

Gene transfer therapy involves introducing "healthy" genes into cells that lack them

- early work with a cold virus vector, called an adenovirus, was unsuccessful in humans because of immune attack
- a new vector, **adeno-associated virus** (**AAV**) does not elicit a strong immune response and seems promising