Please do not write on these and Return them in good condition. Write your answers on a separate piece of paper

10

Biotechnology and Genetic Engineering

Key Terms

Biotechnology
Cloning
Dolly
Founder
Gene enhancement
Gene mapping
Gene therapy
Genetic testing
Genetically modified organism
Micropropagation
Nutraceuticals
Pharmacogenomics
Polymerase chain reaction
Recombinant bovine somatotropin
Pharming
Recombinant DNA
Restriction enzyme
Somatic cell
Transgenic
Vector
Xenotransplantation

Learning Objectives

After you have studied this chapter, you should be able to:

- Describe the magnitude of the biotechnology industry in the United States.
- Define *biotechnology* and explain how genetic engineering is a part of biotechnology.
- Describe the recombinant DNA technology: how DNA can be fragmented and amplified, how specific genes can be cloned and mapped.
- Describe current and future uses of genetic engineering as it applies to field crop, food crop, and livestock production.
- Describe developing uses of rDNA organisms as disease models, for organ donation, as bioreactors for pharmaceuticals, as nutraceutical producers, and as waste managers.
- Explain the regulatory mechanism in place to control genetically engineered organisms.
- Identify some of society's concerns about genetic engineering.

INTRODUCTION

Biotechnology has and will continue to revolutionize modern life. The past and future contributions of biotechnology touch all areas of modern life, including medicine, agriculture, the environment, manufacturing, bioprocessing, and almost anything else one would care to mention. For this reason, this chapter includes discussions of a broad range of applications in areas other than just animal science. Any other approach would not do the science—or you—justice. Even so, this chapter is only meant to whet your appetite for a science that is producing miracles every day.

Biotechnology is the development of products by a biological process. This may be done by using intact organisms, such as yeasts and bacteria, or by using natural substances (e.g., enzymes) from organisms. Biotechnology can also involve the use of plant and animal cells to produce products, especially products that could not previously be produced. A variety of biological processes are used, ranging from traditional fermentation to modern **transgenic** mammals that produce vaccines in their milk. The products of biotechnology include bread, cheese, wine, penicillin, plant and animal health diagnostic kits, vaccines, biopesticides, herbicide-resistant crops, nutrient-enhanced rice, cloned animals of several species, and even a tomato engineered to delay ripening, allowing for improved flavor and reduced spoilage (Flavr Savr).

Biotechnology The application to industry of advances made in the techniques and instruments from the biological sciences.

Transgenic An animal or plant that has had DNA from an external source inserted into its genetic code.

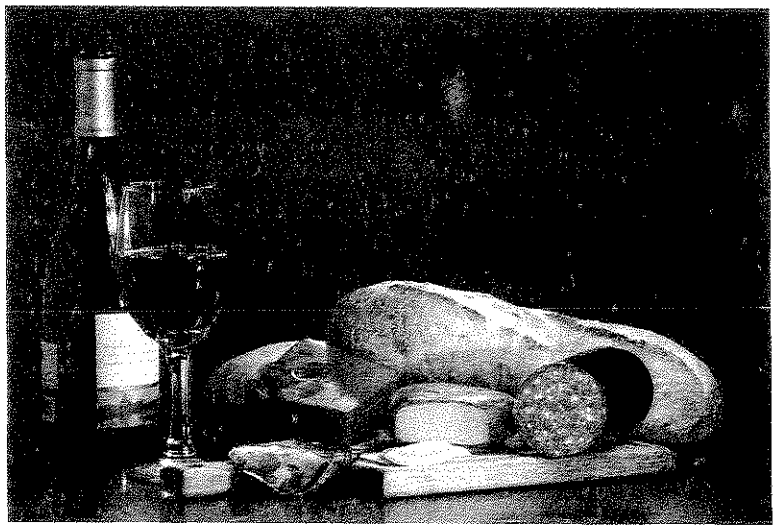

Figure 10–1
Biotechnology has been used for millennia to produce wine, cheese, bread, and fermented sausages.
(Oklahoma State University. Photo by Todd Johnson. Used with permission.)

Biotechnology integrates many disciplines, including agriculture, biology, genetics, molecular biology, biophysics, biochemistry, chemical engineering, and computer science and applies them to bring about the development of a practical and beneficial product. This may be a new product or it may be an enhancement of a traditional product. Those products may be used in disease research, food production, waste management, or a myriad of other areas of need.

Even though the word *biotechnology* is now part of the common vocabulary, biotechnology itself isn't really anything new. People have used it for millennia in traditional applications such as the production of beer, cheese, and bread (Figure 10–1). However, a new approach to biotechnology is causing a biotechnological revolution in the way that life can be lived. The modern era of biotechnology began in 1953, when James Watson and Francis Crick proposed the double-helix structure of DNA. New biotechnology and its products are changing the way we function on this planet. New biotechnology owes its existence to the understanding of the cell and its components, and especially the genetic code. It is this new biotechnology that is stirring imaginations. It is new biotechnology that most people are referring to when they use the word *biotechnology*. Compare the previously mentioned products of alcohol, bread, and cheese as products of traditional biotechnology to new biotechnology, which brings us tomatoes with an extended shelf life, tissue plasminogen activator to dissolve blood clots during heart attacks, bacteria that eat oil spills, goats and cows that produce milk that contains lifesaving drugs, animals with built-in disease resistance, biodegradable plastics, and gene therapy. These examples are only the tip of the iceberg.

NUTS AND BOLTS OF GENETIC ENGINEERING

In 1953, James Watson and Francis Crick proposed the double helix structure of the DNA molecule, and they hinted about the potential importance and impact of this discovery. But not even the two Nobel Prize-winning scientists could have predicted the amazing pace at which molecular biology would advance over the next half century.

In the late 1960s, a great deal of interest surrounded the topic of gene cloning, and scientists speculated that it might be possible to clone DNA by cutting and pasting DNA from different sources (**recombinant DNA technology**). Although such terms as "gene cloning," "recombinant DNA technology," and "genetic engineering" may seem to describe the same process, these techniques are in fact different, interrelated

Recombinant DNA. Combination of DNA molecules from different biological sources.

methodologies. Recombinant DNA technology is commonly used to make gene cloning possible, whereas genetic engineering often relies on recombinant DNA technology and gene cloning to modify an organism's genome.

Because recombinant DNA is the core technology of biotechnology, details of basic enabling technologies of genetic engineering will be presented.

Restriction Enzymes

Restriction enzyme
Enzyme used to recognize a specific DNA sequence and cut it to produce DNA fragments.

The discovery of two essential components, **restriction enzymes** and plasmid DNA, made gene cloning and DNA recombinant techniques possible, and gene cloning became a reality in early 1970s. Restrictions enzymes are DNA-cutting enzymes, and plasmid DNA is a circular form of self-replicating DNA that scientists can manipulate to carry and clone other pieces of DNA.

In order to transfer a gene, or a piece of DNA, it must be excised from the chromosome. In 1970, working with the bacterium *Haemophilus influenza*, Hamilton Smith from Johns Hopkins University isolated *Hin*dIII, the first restriction enzyme to be well characterized and used for DNA cloning. Restriction enzymes are also called restriction endonucleases (*endo* = within, *nuclease* = nucleic acid cutting enzyme) because they cut within DNA sequences. Smith demonstrated that *Hin*dIII could be used to cut or digest DNA into small fragments, and in 1978 he shared a Nobel Prize with Werner Arber and Daniel Nathans for their discoveries on restrictions enzymes and their applications.

Restriction enzymes are found primarily in bacteria where they play a defensive role against invading bacteriophage (a virus that attacks bacteria) by cutting the viral DNA into small fragments. They are named according to a specific protocol based on the genus and species names of the bacteria from which they are isolated. For example, *Hpa* I and *Hpa* II represent the first and second restriction enzymes isolated from *Haemophilus parainfluenzae*.

Restriction enzymes cut DNA only at a specific site called a recognition sequence or restriction site (Figure 10–2). The nucleotide sequence at these sites is palindromic (read the same from either end) and consists of four, six, or eight nucleotide pairs. The four- and six-base cutters are most commonly used in molecular cloning research. A restriction enzyme may cut straight across the double helix, producing blunt ends, or produce a staggered cut when the ends of the cut

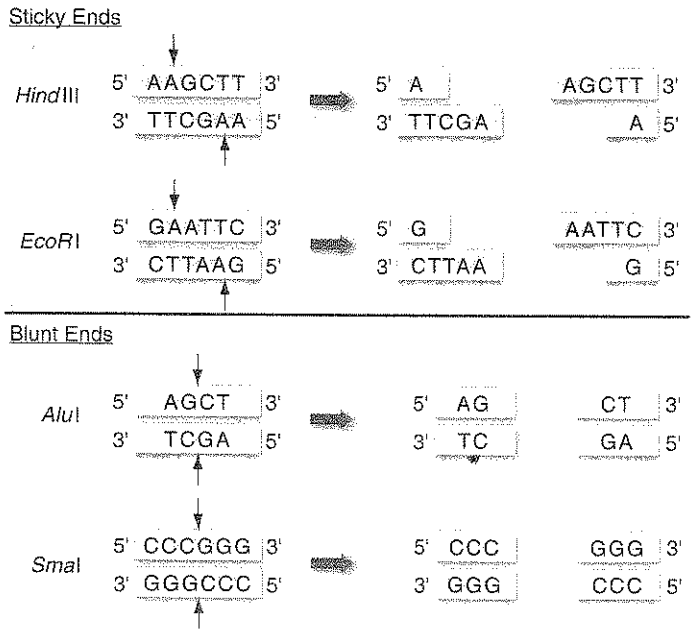

Figure 10–2
Selected restriction enzymes and their specific recognition sites. HindIII and EcoRI produce overhanging or sticky ends, AluI and SmaI produce blunt ends.

have an overhanging piece of single-stranded DNA. These are called sticky ends, because they are able to recombine with any DNA molecule that contains the complimentary sticky end. The union can be made permanent by another enzyme, DNA ligase, which forms covalent bonds along the backbone of each strand. The result is a molecule of recombinant DNA.

Gene Cloning

Cloning Process used to produce genetically identical copies of biological entities.

Cloning in biotechnology refers to the process of creating copies of (1) DNA fragments (molecular cloning), (2) cells (cell cloning), or (3) whole organisms. Cloning is frequently used to amplify DNA fragments containing whole genes, and it has practical application ranging from genetic fingerprinting to large scale protein production.

Cloning of any DNA fragment involves essentially four steps.

1. **Fragmentation**—cutting and separating a piece of DNA. The DNA of interest is obtained by first extracting the DNA from the organism and then cutting out the specific DNA sequence using restriction enzymes.
2. **Ligation**—gluing together pieces of DNA. When two different DNA samples are cut with the same sticky-end restriction enzyme, all the fragments will have identical overhangs or complementary ends. This allows DNA fragments from two sources to be linked together, or ligated, using DNA ligase, the same enzyme that ligates the Okazaki fragments during replication. Ligase is much more efficient with overhanging sticky ends, but can also link blunt ends. A ligation procedure is where the DNA fragment of interest is inserted into a **vector**, a small piece of DNA into which a foreign DNA fragment can be inserted. The vector (which is usually circular) is linearized using the same restriction enzyme, and it is incubated with the DNA fragment of interest under appropriate conditions with the ligase (Figure 10–3).

Vector A DNA molecule that carries foreign DNA into a host cell, replicates inside a bacterial (or yeast) cell and produces many copies of itself.

Several kinds of vectors are used in genetic engineering research to accomplish various purposes (Table 10–1). Some of them are suitable for cloning small pieces of DNA, whereas other are used for larger pieces of DNA. Regardless of type, all vectors contain the same essential features: (1) selectable marker,

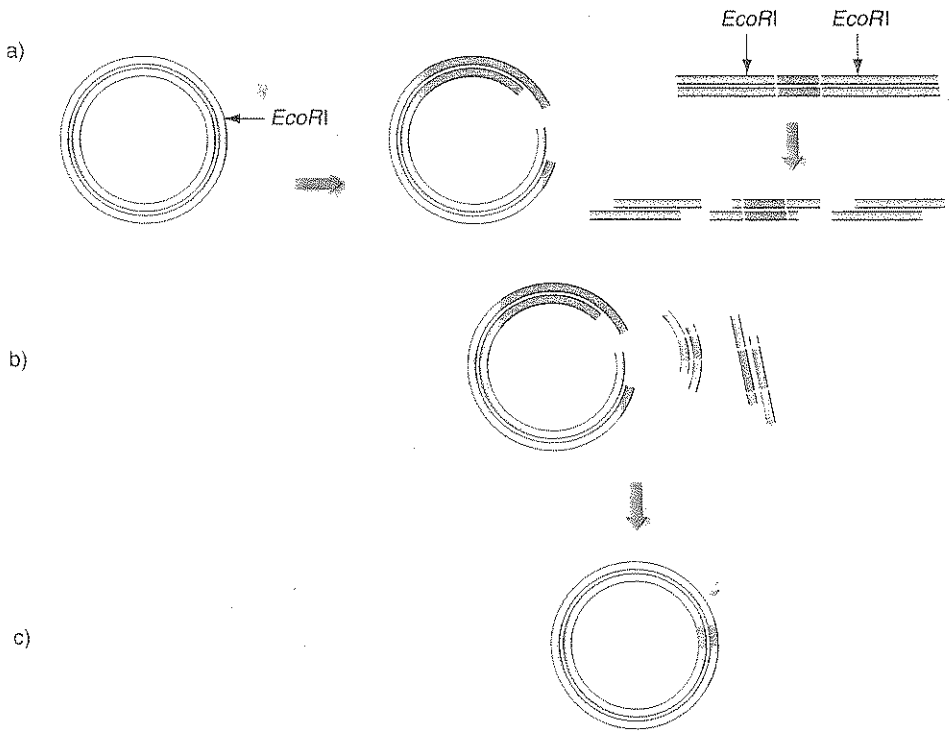

Figure 10–3
DNA cloning. a) Plasmid DNA and foreign DNA are cut with the same restriction enzyme (EcoRI) to produce complementary sticky ends. b) The digestion results in a linearized vector and a fragment of the foreign DNA containing the gene of interest. c) The two fragments of DNA can join by base pairing, forming a circular molecule. The enzyme DNA ligase will form covalent bonds to create the recombinant plasmid.

Table 10–1
DNA VECTORS AND THEIR APPLICATIONS

Vector type	Maximum insert size	Applications	Limitations
Bacterial plasmid vectors	6–12 kb	DNA cloning, protein expression, subcloning, direct sequencing of insert DNA	Restricted insert size; limited expression of proteins; copy number problems; replication restricted to bacteria
Bacteriophage vectors	25 kb	cDNA, genomic and expression libraries	Packaging limits DNA insert size; host replication problems
Cosmid	35 kb	cDNA and genomic libraries, cloning large DNA fragments	Phage packaging restrictions; not ideal for protein expression; cannot be replicated in mammalian cells
Bacterial artificial chromosomes (BACs)	~300 kb	Genomic libraries, cloning large DNA fragments	Replication restricted to bacteria; cannot be used for protein expression
Yeast artificial chromosomes (YACs)	200–1,000 kb	Genomic libraries, cloning large DNA fragments	Must be grown in yeast; cannot be used in bacteria
Ti vector	Varies	Gene transfer in plants	Limited to use in plant cells only; number of restriction sites randomly distributed; large size of vector not easily manipulated.

(2) origin of replication, and (3) restriction sites. Commonly used vectors include plasmids, bacteriophages, bacterial artificial chromosomes (BACs), yeast artificial chromosomes (YACs), fosmids, P1, and PACs.

3. **Transformation**—insertion of the newly formed DNA pieces into cells. The plasma membrane of cells is selectively permeable and does not normally admit large molecules such as long DNA fragments. However, cells can be treated in various ways (e.g., exposure to short electric shock, high ionic strength salts, etc.) so that the permeability of the plasma membrane is altered. These cells are now considered competent. The competent cells are now capable of taking up DNA from the extracellular environment, a process known as DNA transformation. A cell that has successfully taken up foreign DNA is referred to as a *transformed cell.*
4. **Screening/Selection**—selection of cells successfully transformed with the new DNA. Only a small percentage of cells will take up foreign DNA. Therefore, a screening process, or selection, is necessary to distinguish between the large number of untransformed bacteria and the transformed ones. Antibiotic selection has been the most widely used approach; however, newer vectors often incorporate other popular selection strategies, such as "blue-white" screening (Figure 10–4). For antibiotic selection, the most common strategy is to use a plasmid vector with genes encoding resistance to two different antibiotics (e.g., ampicillin and tetracycline). Cloned DNA fragments are then ligated into a restriction site within one of the antibiotic resistance genes (for example, ampicillin), resulting in disruption of the gene and prevention of antibiotic resistance protein synthesis. Transformed bacterial cells are then plated onto either an agar plate with no antibiotic or a plate with a second antibiotic (tetracycline). Nontransformed bacteria that do not contain a plasmid cannot grow in the presence of either antibiotic because they lack a selectable marker (one of the essential feature of a vector). Therefore, plating cells on antibiotic plates selects against the growth of nontransformed cells because the antibiotic will kill the bacteria. Bacteria containing plasmid with or without foreign DNA will grow in the presence of ampicillin, but only recombinant bacteria (containing a plasmid with the incorporated foreign DNA) will grow in the presence of tetracycline, because the foreign DNA ligated into the plasmid will have disrupted the tetracycline resistance gene.

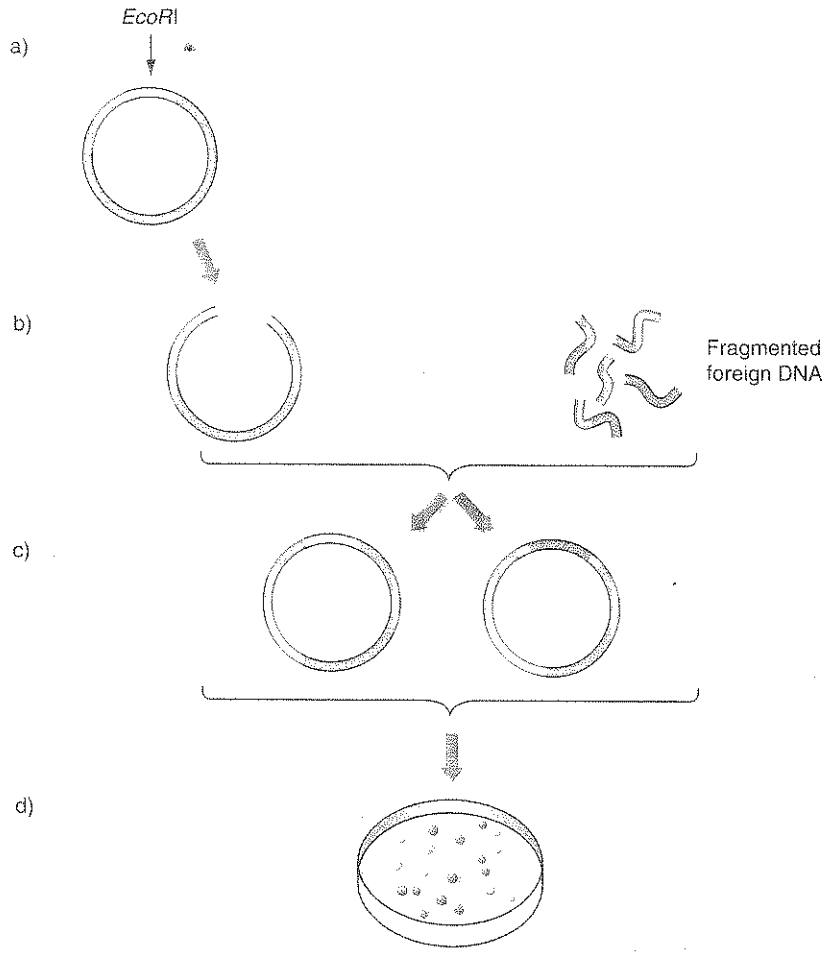

Figure 10–4
Cloning foreign DNA into a plasmid and blue-white screening. a) Plasmid vector is digested with EcoRI restriction enzyme that recognizes the restriction site within the LacZ gene. b) The linearized vector is incubated with fragmented foreign DNA. c) Following incubation, plasmid vectors will close with or without including foreign DNA. Successful insertion of foreign DNA disrupts the LacZ gene and production of functional β–galactosidase. d) Vectors are plated on agar plates containing X-gal, a modified galactose sugar that is metabolized by β-galactosidase to form a chemical which is spontaneously oxidized to the bright blue pigment. Cells in the blue colonies do not carry any cloned DNA fragments, whereas the white colonies contain vectors carrying foreign DNA fragments.

Polymerase Chain Reaction

Polymerase chain reaction Molecular biology technique used to amplify specific DNA.

The **polymerase chain reaction** (PCR) was developed in the mid-1980s by Kary Mullis, who won a Nobel Prize in chemistry for his invention. PCR is a rapid and versatile method for amplifying target DNA sequences that turned out to be a revolutionary technique that had an impact on numerous areas of molecular biology. The technique allows scientists to clone genes or gene fragments for identification and analysis, and it also allows for manipulation of an already identified gene.

PCR amplifies small samples of DNA into large amounts, much as a copier makes many duplicates of one document. The concept behind a PCR reaction is remarkably simple. A few specific reagents are required: template DNA, primers, DNA precursors, and heat-stable DNA polymerase. The template DNA is the double stranded DNA sample of interest, and extremely small amounts (even a single cell) are sufficient. The primers are a pair of short nucleic acid sequences (oligonucleotides) that bind specifically to a matching or complementary sequence on the template DNA. One primer anneals (binds) to the 5' end of the **sense strand**, and the other anneals to the 3' end of the **antisense strand** of the template sequence. The third reagent is a supply of the four DNA bases (A, T, C, G) in the form of deoxynucleotide triphosphates (dATP, dCTP, dGTP and dTTP). These serve as the DNA precursors or the building blocks of DNA. The fourth is *Taq* DNA polymerase, which is the enzyme necessary to actually makes the copies. *Taq* polymerase is the most widely used polymerase for PCR because it is very stable at high temperatures. *Taq* polymerase was isolated from *Thermus aquaticus*, a bacterium that can tolerate high temperatures and that was first isolated from the hot springs of Yellowstone National Park.

Sense strand is the coding strand of DNA which as the same sequence as the mRNA (except for possessing T instead of U).

Antisense strand is the DNA that is complementary to mRNA.

The basic mechanism of PCR relies on a 3-step process: (1) heat denaturation of the template, (2) annealing of the primers, and (3) making a **complementary copy** using DNA polymerase. The process requires changing the temperature in a cyclic manner to provide the necessary temperature for each step to occur. PCR is carried out in a thermocycler, a machine designed to change the temperature of its heat block rapidly so each cycle can be completed in minutes. Each cycle consists of three steps:

1. **Denaturation**, typically at about 94–96°C, causes separation of the target DNA into single strands.
2. **Annealing**, from about 50°C to 70°C, depending on the length and sequence of the primer, allows the primers to bind (anneal) to the complementary bases at opposite ends of the target sequence.
3. **Elongation**, typically at about 70°C–75°C, allows the DNA polymerase to copy the target DNA by binding to the 3' ends of each primer and adding nucleotides to synthesize a complementary strand of DNA.

At the end of each cycle, the amount of template DNA has been doubled. The PCR is a chain reaction because newly synthesized DNA strands will act as templates for further DNA synthesis in subsequent cycles. After about 25 cycles of DNA synthesis, the products of the PCR will include, in addition to the starting DNA, about 10^5 copies of the specific target sequence, an amount that makes it easily visualized as a discrete band of a specific size when subjected to gel electrophoresis.

Gene Mapping

Gene mapping refers to the creation of a genetic map by identifying genes at specific locations on chromosomes. It is a critical step in the understanding of any genetic trait, whether a genetic disease or an economically important trait in livestock. Searching for a specific gene somewhere within the vast genome without a gene map is similar to trying to find a friend's house in an unknown city without a street map. There are two different ways of mapping: *genetic mapping* and *physical mapping*. Genetic mapping uses crossbreeding experiments—or, in the case of humans, the examination of family histories to determine the relative position of genetic markers or genes within a genome. Physical mapping uses molecular techniques to determine the absolute position of these markers or genes within the genome. Both genetic and physical maps provide a similar order of markers and genes, however, the scale does not need to be identical.

Gene mapping Mapping of genes to specific locations on chromosomes.

Genetic mapping uses linkage analysis to determine the relative position of genes on chromosomes. The genetic distance is measured by the frequency of **cross-overs** between genes on the same chromosomes. One map unit, or one centimorgan (cM) is equivalent to 1% recombination. The underlying principle in linkage analysis is that two genes that are close together on a chromosome will be separated less frequently than two genes that are more distant. Pedigree analysis in experimental populations created specifically for genetic mapping is used to determine recombination frequency between genes and genetic markers and to order them along a chromosome. However, crossovers are not uniformly distributed across the genome or any chromosome, and recombination hotspots exist along all chromosomes where crossovers are more likely to occur than in any other regions of the chromosome. As a consequence, the genetic map distance estimated based on frequency of crossovers is not always a precise measurement of the actual distance between markers.

Cross-over Exchange of genetic material between homologous chromosomes

Physical mapping uses molecular biology techniques for a direct physical examination of DNA molecules. A physical map gives the physical location of genes on chromosomes and provides precise information about the distance between markers

BOX 10–1

NATIONAL CENTER FOR BIOTECHNOLOGY INFORMATION'S (NCBI) MAP VIEWER: A TOOL FOR INTEGRATING GENETIC AND PHYSICAL MAPS

The ultimate goal of a genome project is to have the complete DNA sequence of the organism being studied integrated with the genetic and physical maps in order for the all the genes and other genetic markers to be easily located within the DNA sequence. The NCBI Map Viewer, accessible at http://www.ncbi.nlm.nih.gov/mapview/, provides a graphical display of the available genome sequence data for a subset of organisms. You can view and search the complete genome, simultaneously display up to seven maps, and access detailed information for a selected map region.

and genes. There are numerous physical mapping techniques, but the most important fall into three categories:

1. **Restriction mapping** uses restriction endonucleases to locate the relative position on a DNA molecule.
2. **Fluorescent *in situ* hybridization** (FISH) maps the location of markers by hybridizing the marker to intact chromosomes.
3. **Sequence tagged site** (STS) mapping, in which short sequences are mapped by PCR and/or hybridization analysis of genome fragments.

The different types of maps vary in their degree of resolution. The physical map that provides the most detail is the *sequence map*. The sequence map shows the order of genes and genetic markers on a chromosomes and the distance between them measured in base pairs.

BIOTECHNOLOGY AND/OR GENETICALLY ENGINEERED ORGANISMS

The potential number of genetically engineered plants, animals, and microorganisms is limitless. Genes are isolated daily from several species in research laboratories around the world. One day, the entire genome of any animal or plant of interest will be known. The human genome has been successfully mapped and sequenced. Maps of other species are well on their way to elucidation. The number of possibilities that will then be possible are effectively infinite. Many applications are and will be used in agriculture and include novel foods, pesticides, feed additives, and animal drugs. Areas of interest and potential in agriculture include animals that produce leaner meat, plants with insect and herbicide tolerance, and bacteria that produce drugs for livestock. Some of the most exciting applications for genetically engineered products are in the realm of human medicine. These are tied to agriculture in some cases because transgenic animals or field crops have the potential to produce the products. Some of the products and procedures developed for humans will eventually be available for animals. Others have important uses in food processing. Transgenic microorganisms are being developed to minimize pollution from livestock excretion, and for use in the pulpwood, ethanol, textile, detergent, compost, waste treatment, and pharmaceutical industries. Some of the applications of biotechnology are discussed in the following section.

Applications of Biotechnology

Developments in biotechnological techniques have provided new mechanisms that improve crop yields and enable genetic advances in animal agriculture. Indeed, genetic engineering has greatly enhanced the process of introducing desirable gene

into an organism by directly transferring the gene responsible for the beneficial trait. However, the end results of genetic engineering are not all that different from crossbreeding or genetic selection. Selection of desirable traits in plants has been used for over 10,000 years to produce plants with increased yields and resistance to disease. Instead of crossbreeding for a number of years to acquire a desired trait, advanced molecular technology allows scientists to identify and insert a single gene responsible for a specific trait. Genetic alterations in plants have been used to introduce *input traits*; these are traits that improve the plant's health by making it resistant to pests, disease, or herbicides. Such alterations are beneficial to the grower because the grower uses fewer inputs to grow their crops. Other alterations that affect the property of oils or starch in the seeds and thereby lead to improved nutritional content, texture, uniformity, and appearance of the plant are called *output traits*. Although this terminology is most frequently used in relation to agronomic production, the principles are the same for animal products. The introduction of specific genes into the genome of farm animals to produce transgenic animals has been available for more than 20 years, and recent lines of transgenic livestock enable major genetic advances that benefit producers and consumers.

Crops Thus far, genetically modified crops outnumber all other **genetically manipulated organisms (GMO)**. In the United States and the Western world, genetically altered foods are very prevalent. Estimates suggest that from 60% to as much as 80% of the processed foods purchased from the supermarket today contain ingredients derived from genetically modified crops.

Genetically Modified Organism (GMO) Any organism that has been modified by altering one or more genes using recombinant deoxyribonucleic acid (rDNA) technology.

GMOs provide a promising strategy for increasing global food production by reducing crop losses and increasing yields, while at the same time conserving the little unused farmland remaining. Already, several million acres of these crops are planted each year in the United States and around the world. The use of recombinant technology to produce GMOs has already proven to reduce the need for chemical pesticides and tillage, which can enhance the nutritive value of crops. Several thousand have been field tested, with a much smaller number approved for commercial use. The number of transgenic plants that are field tested in the United States alone roughly doubles annually. Corn, squash, tomatoes, cotton, canola, rice, and soybeans have gained the most attention to date. However, many other species and a multitude of traits are under study. Areas of interest include manipulating protein quality, vitamin content, and energy content to help prevent diseases and deficiencies by delivering optimal levels of key nutrients. The most well known of these is Golden Rice, a genetically altered variety of rice with higher levels of beta-carotene, an organic compound that gets converted in the body to vitamin A. Genetically modified foods that use recombinant technology to introduce or concentrate nutrients in plants provide a means to overcome deficiencies of these nutrients where rice is a staple food. Transgenic plants are being created that have resistance to various pests and diseases, thereby reducing the need for chemical pesticides. Plants capable of withstanding adverse growing conditions, such as extremes in temperatures and poor soil conditions, are being developed. Plants resistant to herbicides have been developed and are widely cultivated around the world. Horticulturists are creating transgenic ornamentals. The possibilities seem endless, and several of these crops are being developed and tested for commercial use, including: transgenic decaffeinated coffee beans, potatoes engineered to have higher starch content that will absorb less oil when deep fried, tropical plants growing in the snow, fruit that ripens on demand, crops such as peanuts and wheat with decreased allergenic compounds, and so on.

A very controversial area being explored is the possibility of creating edible vaccines with transgenic plants. Transgenic plants have already been successfully created to

produce several plantbodies (antibodies produced in plants), and vaccine production in a food that is eaten raw is necessary to prevent denaturation of the vaccine. Research is underway to use bananas and potatoes as carriers of vaccines for diseases such as cholera, diarrhea, and hepatitis B. Edible vaccines are directed at human disease with an emphasis in developing countries; however, it also seems quite likely that transgenic plants will be developed to produce human insulin and other hormones at a fraction of the current cost.

Micropropagation

Micropropagation is a biotechnology that takes cells of a desired plant and uses them to generate another plant. This process is also called *plant tissue culture*. Because just a few cells can generate a whole plant, many identical plants can be made in a very short time from just a single leaf of a desirable plant. Micropropagation can be combined with genetic engineering to produce large numbers of a plant that has been engineered to do something considered of value.

Safety of Food and Feed Derived from Genetically Modified Crops Currently, in the United States and abroad, the safety of food and feed derived from genetically modified (GM) crops is assessed by internationally accepted procedures for evaluating any associated risks. Comparative assessments of GM crops with the appropriate non-GM counterparts are performed in laboratory research and field research trials. Those GM crops demonstrating unacceptable agronomic characteristics as a result of the inserted transgene are not further developed or commercialized. Scientific evidence continues to indicate that the risks associated with GM crops are not any greater than the risk posed by plants produced by traditional breeding methods. GM foods currently available have passed risk assessment evaluations that include agronomic and morphological characterizations, compositional and nutritional analysis, as well as potential to provoke allergic reactions (allergenicity) and specific toxicity tests. Additionally, no effects on human health have been shown to result from consumption of GM foods.

Transgenic Animals

Whole Organism Cloning Similar to transgenic plants, transgenic animals carry a segment of foreign DNA (the transgene) in their genome that can be inherited by their offspring. To date, transgenic animals are not nearly as plentiful as are transgenic microorganisms and plants. The techniques for producing transgenic animals are more complicated and expensive and are not yet as successful. However, techniques are improving rapidly. As genome sequencing projects are completed for various farm animal species, the ability to target the modification of individual genes will become even more practical. Several hundred scientific papers are published each year describing techniques for producing transgenic animals. The transgenic animal that is subsequently used to establish the transgenic line is referred to as the **transgenic founder** animal.

Transgenic founder A transgenic animal that is subsequently used to establish a transgenic line of animals.

The first successful generation of a transgenic animal used pronuclear microinjection of foreign DNA into one-cell mouse zygotes. This was the same technique used in production of the first transgenic livestock in 1985. Microinjection of cloned DNA into the pronucleus of a fertilized ovum continues to be a widely used method for producing transgenic animals. However, this technique is inherently inefficient. More recently, nuclear transfer techniques have been adapted to allow for more exact modification of the genome. There are many potential practical applications for the use of transgenic livestock, including enhancing growth rates, improved muscle-to-fat ratio of carcasses, improved resistance to disease, enhanced feed nutrient use, improved milk production and composition, and enhanced reproductive performance and increased prolificacy. Additionally, transgenic livestock species are being used in biomedicine with the use of "**gene pharming**," presently at the level of commercial marketing.

Gene pharming Derived from the combination of farming and pharmaceuticals. Gene pharming uses transgenic livestock to produce biologically active pharmaceuticals for human medicine.

BOX 10–2

AGRICULTURAL BIOTECHNOLOGY TIMELINE, IN CHRONOLOGICAL ORDER

Year	Description
1908	First U.S. hybrid corn produced by G. H. Shull of Carnegie Institution through self-pollination.
1919	Word biotechnology coined by Hungarian immigrant Karl Ereky.
1926	Hybrid corn becomes commercially available in the United States, causing corn yields to triple over the next 50 years.
1963	New wheat varieties (later known as Green Revolution grains), developed by Norman Borlaug, increase yields by 70% and later save millions of lives.
1983	First biotech plant is produced—scientists create a tobacco plant resistant to an antibiotic.
1985	First field trials for GM crops resistant to insects, viruses, and bacteria are held in the United States.
1986	EPA approves the release of the first crop produced through biotechnology—tobacco plants.
1993	U.S. Food and Drug Administration allows companies to market GM seed.
1994	The first GM food product, the Flavr Savr tomato, is approved in the United States.
1996	Ingard® insect resistant (Bt) cotton is grown commercially in Australia.
1996	Herbicide-tolerant GM soybean available in the United States.
2000	German and Swiss scientists develop Golden Rice, a GM variety with genes added that produce a vitamin A precursor capable of preventing some forms of blindness.
2000	The first entire plant genome, for *Arabadopsis thaliana*, is sequenced.
2003	Indonesia allows consumption of imported biotech foods, and China and Uganda accept biotech crop imports.
2006	The American Dietetic Association (ADA) reaffirms its support of agricultural biotechnology as a way to enhance the quality, value. and variety of food for human consumption.
2008	The USDA's crop production report predicts that the United States will produce the second largest corn crop and fourth largest soybean crop in U.S. history, partially as a result of the biotech varieties that are better able to tolerate weed and insect threats.

Transgenic techniques are also being used in commercial aquaculture. Microinjecting cloned gene sequences into fish eggs to produce a transgenic fish is considerably easier than microinjecting DNA into the pronuclei of mammalian zygotes. This has resulted in at least 35 species of fish and shellfish that have been genetically engineered. Most transgenic fish have been have been altered for traits such as growth rate, disease resistance, temperature tolerance, and flesh quality. For example, a fast-growing Atlantic salmon has been engineered to contain a gene for either bovine or Chinook salmon growth hormone (Figure 10–5). The salmon reportedly has a much as a 2-6 fold increase in growth rate compared to the

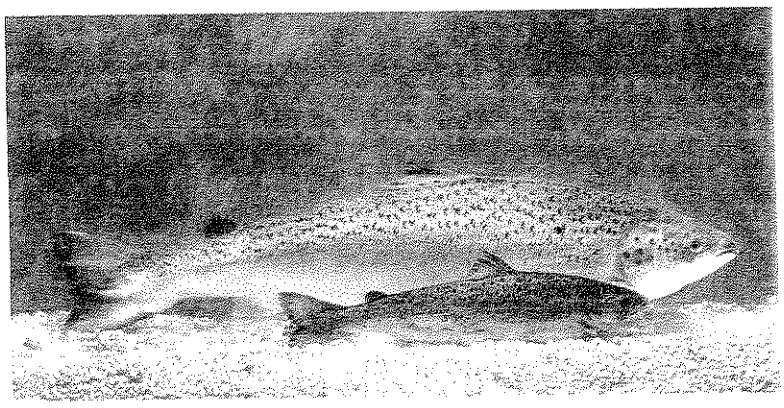

Figure 10–5
Size comparison of an AquAdvantage® Salmon (background) versus a non-transgenic Atlantic salmon sibling (foreground) of the same age. (Photo courtesy of Barrett & MacKay. Photo-barrettmackay.com)

Figure 10–6
Dolly, the first cloned sheep produced through nuclear transfer from differentiated adult sheep cells, created at the Roslin Institute in Edinburgh, ignited international controversy about reproduction through cloning. Dolly died on February 14, 2003, euthanized because of an incurable lung infection. (Photo courtesy of, The Roslin Institute, The University of Edinburgh.)

non-transgenic salmon. Similarly, a fast-growing transgenic tilapia, with genetic material from rainbow trout, striped bass, and carp, has also been developed. The prospect of improving nutritional value of fish has also been investigated. Consumption of fish containing omega-3 fatty acids has many human health benefits, and transgenic technology could provide a means to transfer the beneficial fats to other foods.

Somatic cell All cells in the body other than gametes.

Another technique is cloning by *nuclear transfer* from somatic cells. This is the process of producing a "twin" of an animal by transplanting the nucleus from a **somatic cell** of the animal into an egg with the nucleus (and thus its original genetic material) removed. The egg with the new genetic material is allowed to develop in a female as would a naturally fertilized egg. The young animal bears no resemblance to the egg donor. All of its genetic material came from the nucleus donor. Thus, the newborn is genetically identical to the donor of the nucleus. The genetic material can come from an embryo (accomplished in the early 1980s), or, as the creators of the sheep Dolly have shown us, an adult animal. Dolly was introduced to the world in 1997 as the first clone of an adult mammal (Figure 10–6). She was produced from a normal somatic cell taken from the udder of her "twin," Tracy. Of course, a genetically engineered animal could be cloned, but the genetic manipulation and the cloning are separate. Figure 10–7 demonstrates how animal cloning is accomplished. Table 10–2 presents a chronology of landmark accomplishments in cloning.

Figure 10–7
How to clone an animal.

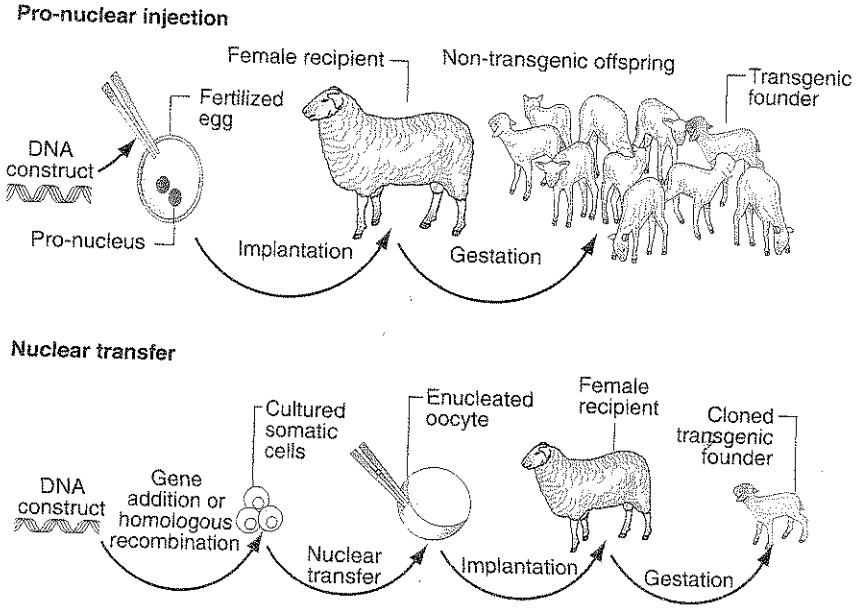

Table 10–2
LANDMARK CLONING EXPERIMENTS CLONING, IN CHRONOLOGICAL ORDER

Species	Year	Description
Carp	1963	The world's first cloned fish produced in China by inserting the DNA from a male carp cell into a female carp egg.
Mice	1986	The mouse Masha, cloned by Russian scientists, was the first successfully cloned mammal.
Sheep	1996	Dolly, a Finn-Dorset ewe, was the first mammal to have been successfully cloned from an adult cell. She was cloned at the Roslin Institute in Scotland and lived there from 1996 to 2003.
Rhesus monkey	2000	Tetra was cloned using a method that splits the original cells in an embryo to make multiple identical animals.
Gaur	2001	Noah, a baby Asian ox and the first endangered species cloned, died two days after birth.
Cattle	2001	Alpha and Beta, two cloned calves at the University of Pennsylvania College of Veterinary Medicine.
Cat	2001	CopyCat, a brown tabby and white domestic shorthair, was the first cloned pet.
Dog	2005	Snuppy, a male Afghan hound, was the first cloned dog.
Rat	2003	Ralph, the first cloned rat.
Mule	2003	Idaho Gem was the first clone born in the horse family and the world's first cloned mule. On June 4, 2006, Idaho Gem finished third in the Winnemucca Mule Race. This was the first showdown between cloned and natural-born mules.
Horse	2003	Prometea, a Haflinger female, was the first horse cloned and the first to be born from, and carried by, its cloning mother.
Water buffalo	2009	Samrupa, the first cloned water buffalo, died five days later due to lung infection.
Camel	2009	Injaz, the first cloned camel.

The potential advantages of cloned animals are many. Cloning should prove useful in propagating transgenic animals with genes that cause them to produce important pharmaceuticals in their blood or milk. In addition, large numbers of genetically identical animals could be available for research purposes. For agriculture, outstanding animals could be cloned for production purposes, thereby making leaps in genetic progress in herds and reducing variability in production characteristics. Combining cloning with rDNA technology is considered one of the great potential benefits of cloning.

Achieving a useful transgenic animal is complicated and expensive, and only a very small number of attempts succeed. Once a transgenic animal with a desired characteristic is achieved, cloning would allow the rapid development of a whole herd of these animals.

The technique that produced Dolly was extremely inefficient. This hurdle needs to be overcome before practical uses can be made of cloning. Progress is being made for several species on this front, but efficiencies are still low. Several species have now been cloned. However, cloning research is still in a very basic stage and a long way from routine applications.

BOX 10–3
WHY MAKE TRANSGENIC ANIMALS?

The answer is not so simple. However, some of the reasons are: (1) to gain new knowledge; (2) to decipher the genetic code; (3) to study the genetic control of physiological systems; (4) to build genetic disease models; (5) to improve animal production traits; and (6) to produce new animal products. This question is certain to be debated, refined, and considered well into the future.

From Trends in Biotechnology 2007; 25(5): 204–210.

LIVESTOCK

Agricultural Applications

The production of transgenic livestock is a rapidly developing field that has the potential to impart nearly limitless benefits on the efficiency of animal agriculture. In production species, modification of milk by genetic engineering provides the opportunity to change milk's composition or to produce novel milk proteins. For example, of the production species, nutrient availability in the milk of sows can be especially limiting to the survival and growth of their offspring. Transgenic pigs expressing bovine alpha-lactalbumin in the mammary gland have higher lactose content and increased milk production, which in turn leads to improved survival and development of the piglets. Genetic engineering has also been pivotal in gaining understanding of the mechanisms of the genes involved in the control of growth and of carcass composition. Results from studies in mammals and fish illustrate that transgenic methods can be used to increase growth rates and protein production. Another aspect of manipulating carcass composition is introducing beneficial fats from fish into livestock. The University of Missouri has genetically engineered a line of pigs that has enhanced levels of omega-3 fatty acids, potentially leading to bacon and pork chops that might be considered heart healthy. Environmentally friendly pigs known as Enviropigs were developed at the University of Guelph to address the problem of manure-related pollution. These pigs produce phytase in their saliva in order to use phosphorous from feed more efficiently and thus reduce the amount of phosphorous as a pollutant in manure. Numerous instances show how transgenic technology is used in livestock production to modify disease resistance, reproductive performance, and even hair and fiber characteristics. The benefits associated with the development of useful transgenic animal agriculture are tremendous. However, it should be mentioned that at the time of publication, no transgenic animal has been approved for human consumption.

Disease Resistance

Genetic engineering is also being investigated as a tool to increase an animal's ability to resist disease. Transgenic dairy cows that are resistant to mastitis, an infectious disease of the mammary gland, are being produced. These cattle secrete an antimicrobial peptide that protects the mammary gland against the bacteria known to cause the infection. Transgenic chickens and turkeys have been developed that resist avian diseases. And, in the future, the production of prion-free, scrapie-free and BSE-free livestock may be available.

Biomedical Applications

The production of pharmaceutical proteins in the blood or milk of transgenic animals is referred to as "gene pharming." Targeting expression to the mammary gland is desirable primarily due to the large quantities of protein that can be produced in this organ. Several products secreted in the milk of transgenic goats and sheep have advanced to clinical trials. An anticoagulant produced in the mammary gland of transgenic goats was approved by the FDA in February 2009 (Figure 10–8). ATryn, a human anti-thrombin-α, is indicated for use in people with antithrombin deficiencies who are predisposed to blood clots during surgery or childbirth. ATryn became the first transgenic animal drug approved for use in the United States. A novel development in biologically active molecules derived from the mammary gland of transgenic animals is the production of an antidote against organophosphorus compounds used in agriculture and chemical warfare. Transgenic goats that produce the antidote, butyrylcholinesterase, can produce sufficient quantities to protect all humans at risk of organophosphorus poisoning.

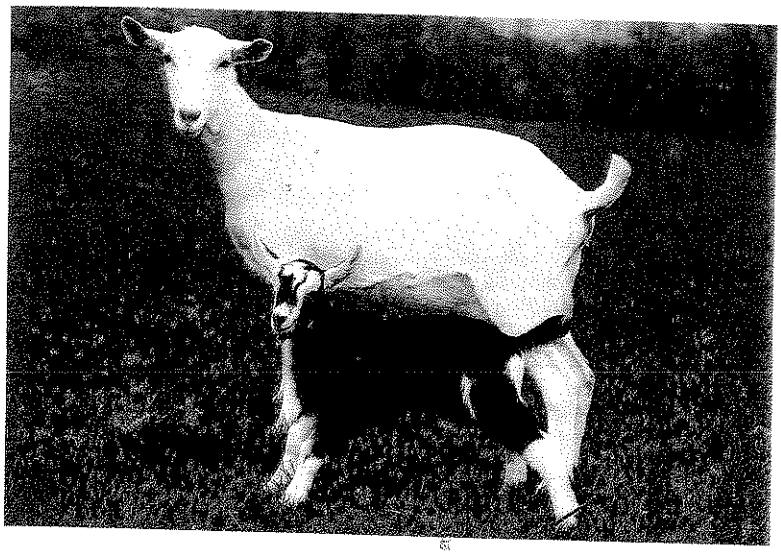

Figure 10–8
On February 6, 2009, ATryn, an anticoagulant protein derived from the milk of transgenic goats, became the first animal drug to be approved in the United States. The protein could prove to save lives for those who are at high risk of blood clots during surgery and childbirth. (Photo courtesy of GTC Biotherapeutics. Used with permission.)

Animals Engineered as Sources of Transplant Organs Companies are actively pursuing the development of transgenic animals that could donate organs to humans that would not be rejected by the human recipient. In the United States, there are far more people on organ transplant waiting lists than there are organs available. Researchers have been working toward narrowing the gap between the demand and availability for appropriate organs through **xenotransplantation**. The pig as a donor may be a suitable choice for several reasons: (1) the organ size of both species is similar, (2) there is not a large difference between the anatomy and physiology of pigs and humans, (3) pigs grow rapidly, and (4) pigs have short reproductive cycles with relatively large litters. The single largest hurdle is engineering a pig whose organs will not be rejected by a human recipient.

Xenotransplantation The use of organs from genetically engineered animals to transplant into humans.

Animals Engineered to Help Researchers Study and Treat Human Diseases Thus far, most transgenic animals have been designed for human-disease research. Several hundred transgenic rodent lines have been developed in order to study cardiovascular disease, cancer, several autoimmune diseases, sickle-cell anemia, and neurological diseases. These transgenic laboratory animals develop diseases in a manner very similar to that of humans because they have been engineered to do so, allowing researchers a model to study the human disease. Although rodents offer numerous benefits as models for human diseases, researchers realize that their physiology, anatomy, and lifespan are vastly different from humans. Some diseases—such as neurodegenerative disorders, atherosclerosis, certain types of cancer, and cystic fibrosis—require longer study periods than are possible with mouse models. In these cases, farm animals that exhibit similar pathologies may be more appropriate models for developing effective treatments.

TRANSGENIC PETS

The first transgenic pet marketed in the United States, the GloFish®, is a zebra fish (*Danio rerio*) that carries the fluorescent protein gene from a sea anemone. Originally engineered to detect pollutants in waterways, they have been on the pet market in the United States since 2003. "Starfire Red®" "Electric Green®" and "Sunburst Orange®" are now available and more varieties are likely to follow. GloFish® are also useful in understanding cellular disease and development, cancer, and gene therapy (Figure 10–9). Yorktown Technologies, a Texas-based company, initiated commercial sales in the

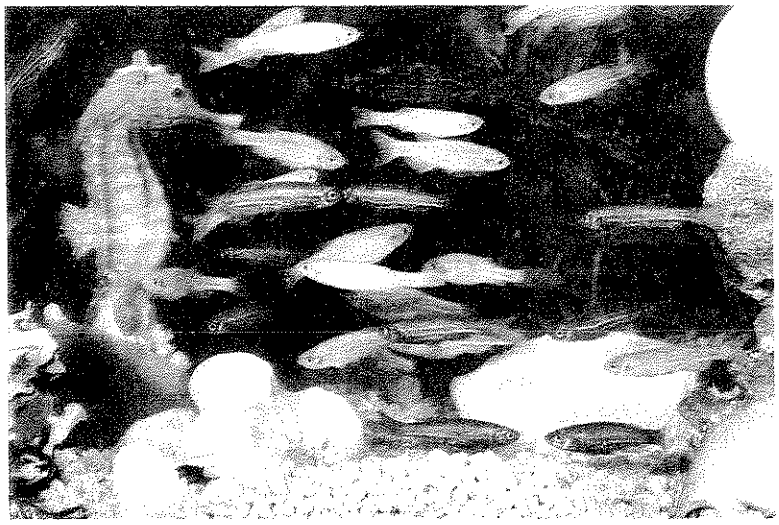

Figure 10–9
The GloFish® fluorescent fish is a zebra fish (*Danio rerio*) that carries fluorescent protein gene from other marine organisma. Originally engineered to detect pollutants in waterways, they have been on the pet market in the United States since 2003. "Starfire Red®" "Electric Green®," and "Sunburst Orange®" are now available and more varieties are likely to follow. These fish are also useful in understanding cellular disease and development, cancer, and gene therapy. (Photo courtesy of www.glofish.com. Used with permission.)

United States at a price of $5.00 each. The FDA, which has jurisdiction over genetically engineered animals, found no evidence that these transgenic fish posed any greater threat than their non-genetically modified counterparts, either to the environment or to human health.

MEDICINE

Genetic Testing

Genetic testing Process using a variety of laboratory techniques to determine if a person has, or is likely to get, a genetic condition or disease.

Genetic tests involve direct examination of DNA molecules from blood or other tissues to find genetic disorders. About 900 such tests are available, and they are used by doctors for several reasons:

- Prenatal diagnosis screening: finding possible genetic disorders in unborn babies
- Carrier screening: finding out if unaffected individuals are carriers of disease genes, which might be passed on to their children
- Sex determination
- Identity/forensic testing
- Embryo or newborn screening: screening embryos or newborns for disease
- Presymptomatic testing: testing for genetic diseases in adults before they cause symptoms and testing for estimating the risk of developing late-onset cancers
- Confirming a diagnosis in people with disease symptoms

Although there are no physical risks associated with most genetic testing, many of the risks are emotional, social, or financial in nature. Also, genetic testing can provide only limited information about some conditions. This is especially the case when not all mutations associated with a disease are known or when known mutations present different risks to different people. A genetic counselor can provide information about the pros and cons of genetic testing and can explain in detail the risks, benefits, and limitations associated with a particular test.

Gene Therapy

Gene Therapy Transferring specific genes into mammalian cells with the goal of treating genetic disorders.

Gene therapy is a technique for correcting defective genes responsible for disease development, by insertion, alteration, or removal of genes within an individual's cells. The most straightforward use of gene therapy is to treat hereditary defects due to single genes. Introducing a functional copy of the gene can then cure the defect. Other

forms of gene therapy involve directly correcting the causative mutation or modifying the regulation of a particular gene.

In most gene therapy studies, a normal gene is inserted into the genome to replace an abnormal, disease-causing gene. A carrier molecule called a vector must be used to deliver the therapeutic gene to the patient's target cells. Currently, the most common vector is a virus that has been genetically altered to carry normal human DNA. Viruses have evolved a way of encapsulating and delivering their genes to human cells in a pathogenic manner. Scientists have tried to take advantage of this capability and manipulate the virus genome to remove disease-causing genes and insert therapeutic genes.

Target cells, such as the patient's liver or lung cells, are infected with the viral vector. The vector then unloads its genetic material, containing the therapeutic human gene, into the target cell. The generation of a functional protein product from the therapeutic gene restores the target cell to a normal state.

About 75% of gene-therapy trials have used viral vectors because of their efficiency. However, nonviral vectors are inherently safer, and a variety of alternative approaches have been investigated. The simplest method is the use of naked DNA, which introduces the therapeutic DNA directly into target cells. This approach is limited in its application because it can be used only with certain tissues and requires large amounts of DNA.

About 10% of gene therapy trials have used liposomes, artificial lipid spheres with an aqueous core that can be filled with DNA. The liposomes are capable of passing the DNA through the membrane of the target cell.

Pharmacogenomics

Pharmacogenomics Branch of pharmacology that looks at how variation in human genetics leads to variation in response to drugs.

Pharmacogenomics is the study of how an individual's inherited variation in different genes affects drug response. Pharmacogenomics hold the promise of personalized medicine, when drugs and drug combinations will be tailor-made for individuals and adapted to an individual's own genetic makeup. Right now, most drugs are one size fits all—a drug is formulated and tested in a population. If there are more benefits than risks, generally the drug gets approved. However, although some individuals will respond well to a certain drug, others may suffer adverse effects. Genetic testing may predict whether a patient is a good candidate for treatment with a particular medication. In the near future, pharmacogenomics might be used to make consistently accurate diagnoses, which are essential for treatment to be effective.

BOX 10–6

FARM GENOMICS

It took more than 6 years, 300 international researchers from 25 countries, and $53M US to fully sequence the genome of L1 Dominette: an inbred 8-year-old Hereford cow. The bovine was the first livestock species to be sequenced. By the end of the bovine genome project, researchers had discovered approximately 2.44 million single nucleotide polymorphisms (SNPs) on the bovine genome. In 2007, the Illumina Bovine50SNP Beadchip was developed through a collaboration of several research institutions. The high density SNP chip contains 58,000 SNPs, selected to be equally spaced along the genome. It is now widely used to genotype dairy bulls for breeding. The DNA information from the SNP chips is used to enhance the genetic evaluation in a new approach called genomic selection. The genetic value associated with each specific nucleotide at these 58,000 genomic positions is combined with pedigree and performance information to generate predicted transmitting abilities or PTAs. The PTAs that include the genomic information are much more reliable, especially for young animals without any performance data, and can be used to test bulls before they produce any sperm and calves before they produce any milk.

MICROORGANISMS AND RECOMBINANT DNA TECHNOLOGY

Drug and Vaccine Production

Biopharmaceuticals are pharmaceuticals produced using biotechnology. Bacteria have long been used to manufacture drugs for humans. In 1982, human insulin of recombinant DNA origin was the first biopharmaceutical produced this way for therapeutic use. Recombinant human insulin provides a reliable and continual supply of insulin for diabetics. Prior to development of the recombinant human insulin, the primary source of insulin were almost exclusively derived from pork and beef pancreas glands collected from packing plants. However, with the increasing number of insulin-dependent diabetics it was feared that demand may exceed the supply available from porcine and bovine pancreatic glands. Additionally, many individuals are allergic to nonhuman insulin. Recombinant human insulin was the first commercial health care product derived from recombinant DNA technology. This product alone generated $13.3 billion in sales in 2009.

To date, the number of biopharmaceuticals on the market is just over 200. The floodgates have opened on this category of drugs: In 2010, the Food & Drug Administration approved 21 biotech drugs with several hundred new drugs tested each year. Certainly, they don't all reach market. However, dozens of new drugs could be added each year. This research will escalate even more with time. Among the approved biopharmaceuticals are hormones and growth factors, antibodies such as TNF-α used to treat rheumatoid arthritis, and blood-related proteins to stimulate red blood cell growth to help patients on dialysis. Some of these products replace previous treatments. However, a significant number are completely new therapeutic agents with no predecessors. Two biotechnology-produced vaccines have been approved for the prevention of cancer. Gardasil and Cervarix protect against the types of human papilloma virus (HPV) that cause most cervical cancers.

Additionally, vaccines for animals have been approved and used, including rabies vaccines for wild animal populations. Vaccines for baby pig scours, foot rot, sheep measles, and infectious bursal disease have also been developed.

Recombinant bovine somatotropin (rBST) has been among the most successful products of genetic engineering made available to agriculture. It is produced commercially by transgenic bacteria. When administered to dairy cows, BST causes greater milk production. Because of the high fixed costs and labor demands of a dairy, it is generally more profitable to increase average production of the cows in the herd. BST is discussed at length in a later chapter.

Recombinant Bovine somatotropin (rBST) Synthetic bovine growth hormone produced by recombinant DNA technology and given to dairy cows to increase milk production.

Biotechnology and Pest Management

The use of biotechnology to control pests dates back to 100 A.D., when the Chinese used powdered chrysanthemums as an insecticide. More recently, several bacteria have been genetically altered to improve their ability to kill or repel pests. Some have gained approval for commercial use and are being used as pesticides.

Bacteria Engineered for the Foods Industry

Microorganisms have long been a part of food processing. In fact, in 6000 B.C., the Babylonians made alcoholic beverages by using microorganisms to ferment fruits, and the Egyptians used yeast to bake bread around 4000 B.C. Now products of transgenic bacteria are also in use. Genetically altered bacteria produce a rDNA-engineered enzyme called chymosin or rennin, which is used in cheese making. Before microbial production of chymosin was available, its natural equivalent, rennet, was obtained from calf stomachs. The use of recombinant chymosin is desirable due

to greater purity and consistency than calf rennet. Rennet was approved by the FDA in 1990 and became the first protein produced from transgenic means to be used in food. Several chymosin products are on the market, including Chy-Max, Chymogen, and ChymoStar. Fast-acting yeasts have been developed that reduce the amount of time bread must rise before baking and also have better brewing characteristics. Among the valuable new tools of biotechnology are fast, easy tests that can be used on foods to detect disease causing microorganisms or their toxins. These important tools will help provide a safer food supply. Both DNA probes and monoclonal antibodies are being used in this way. An emerging area of food biotechnology will include **nutraceuticals**, which are products that have both nutrient and pharmaceutical properties. Genetically manipulated soybeans may produce beneficial fish oils. Microorganisms used in a fermentation process may add increased levels of antioxidant vitamins. Transgenic microorganisms have also been developed to produce a safe, consistent supply of flavors for the food processing industry. Many other uses are also being investigated.

Nutraceuticals Products perceived to have both nutrient and pharmaceutical properties.

SOCIETAL CONCERNS

Biotechnology and genetic engineering have moral, ethical, and religious issues associated with them that society must reach conclusions about. It is outside the scope of this chapter to deal with these issues in any meaningful way. However, some questions that society must answer include these:

- How far is it acceptable to alter the genetic code? For instance, are featherless chickens acceptable?
- Is altering the genetic code of a human acceptable?
- Is cloning an animal acceptable for any and all purposes?
- Is cloning a human acceptable? Is cloning perhaps acceptable for some situations but not others?
- Is it acceptable to generate human embryos specifically for the treatment of disease with no intention of allowing them to develop to term?
- Is it acceptable to change the genes of a human if those genes can then be passed on to offspring? If it is determined that this is acceptable, should changes be restricted to correcting for diseases or expanded to such things as height, pattern baldness, and eye color?
- How far are we willing to go in mixing genetic information from species to species? Would you knowingly eat lettuce with dog genes inserted into its genetic code? How about genes from fish? What about genes from bacteria? What about genes from carrots?
- What will become of bioreactor animals when their productive life is over? Should they be used as food?
- Should genetically modified food be labeled?

GM foods are also under scrutiny. In the view of many, significant risks are posed by GM foods. Because no confirmed cases of illness or disease are associated with human consumption of GM foods, the risks are potential rather than actual. The companies that produce and market GM foods detect and eliminate plants with undesirable traits during development and before they are commercially available. However, critics argue that detection techniques are not foolproof and need improving. The risks generating the most concern about GM foods include direct risks to human health such as the production of allergenic or new toxic proteins or enhanced production of existing toxic proteins; indirect risks to human health such as reduced levels of nutrients, decreased efficacy of antibiotics, more rapid development of

antibiotic-resistant bacteria, and greater human exposure to herbicides; direct risks to the environment and public welfare including turning crops into superweeds and the reduction in populations of beneficial nontarget species; and indirect risks to the environment and public welfare, including increased development of pesticide-resistant pests, loss of valuable biological pesticides, turning related species into superweeds, greater environmental exposure to herbicides, and threats to biodiversity.

Policy debates over these and other related issues are sure to rage for the foreseeable future.

SUMMARY AND CONCLUSION

Biotechnology is the development of products by a biological process using intact organisms or natural substances (e.g., enzymes) from organisms, or intact cells, plants, or animals to produce products that could not previously be produced. The use of rDNA through genetic engineering figures prominently in many of the modern or new applications of biotechnology. However, the term *agricultural biotechnology* is a broader term that describes the use of biological processes other than genetic engineering, such as micropropagation, plant and animal health diagnostics, vaccines, and biopesticides. Many different disciplines contribute to biotechnology including biology, genetics, molecular biology, biophysics, biochemistry, chemical engineering, and computer science. Consumers, agriculturists, and the agrifoods industry all have a stake in biotechnology. Safer foods, novel foods, cheaper foods, pharmaceuticals, diagnostics, and treatments all await the promise of biotechnology. Some of the more important applications include the following:

- Existing crops can be engineered for disease and insect resistance, drought tolerance, increased climatic tolerance, and the ability to produce their own fertilizers or be productive with less fertilizer. They can also be changed so they produce a different chemical makeup in their seed, such as more, less, or chemically different fats, proteins, and vitamins. Digestibilities can be improved, value enhanced, and new sources of products developed. Field crops with improved industrial applications are being developed. Food crops can be engineered to have greater nutrient content, improved taste, and better storage and processing properties.
- Microorganisms can be used to help increase crop yields, produce vaccines and other pharmaceutical agents, provide products for food processing, and so on.
- Livestock species can be improved for the useful traits, such as leanness, better growth, improved milk production, altered nutritional properties, and so on.
- Livestock can be altered to produce biologically active compounds in body fluids, including milk, for a variety of uses. These substances will be inexpensive compared to other methods of production.
- Diseases can be diagnosed and researched with the use of monoclonal antibodies and transgenic animals.
- Microorganisms can be used to help in the fight to reduce pollution and keep the environment clean.

The possibilities are as large as the size of the combined genome of all the world's species, and that is very big indeed.

STUDY QUESTIONS

1. Define *biotechnology*. What are traditional biotechnologies and what are new biotechnologies? What are products of each?
2. Define *recombinant DNA technology*. Describe how DNA can be fragmented and analyzed, how a gene is cloned and amplified.
3. Why is gene mapping important? What are the uses of a gene map? What is the difference between a genetic and a physical map?
4. What is a genetically modified organism? Is there any risk associated with genetically modified crops?
5. Describe ways in which genetically engineered plants are being put to use today. What are the major ways they have been engineered so far? What other traits do you think it would be useful to engineer into plants?
6. What is a clone? What was unique about Dolly the clone? Has that feat been duplicated in any other species? What are the uses of clones?

7. Why aren't there as many transgenic animals as there are transgenic microorganisms and plants?
8. What are the traits that would be useful to genetically engineer in food fish? Can you think of any traits of ornamental fish that would be beneficial to engineer into food fish?
9. What does it mean to refer to animals as bioreactors? How does this relate to pharming? What are the benefits of pharming as compared to microbial production of pharmaceuticals?
10. For what use have most transgenic animals have been developed to date? Why is this use so valuable?
11. How do genetically engineered organisms fit into the food-processing industry? What is a nutraceutical? Would it bother you to eat potatoes that produced fish oils?
12. What is genetic testing? What is gene therapy? What is pharmacogenomics?
13. What are some of the uses that microorganisms have been genetically harnessed to perform in terms of producing drugs and vaccines?
14. How is BST produced commercially? What is its value?
15. What are some of the concerns you have seen expressed in the media with reference to the ethics of genetic engineering? Is the list of social concerns in the text complete or do you think there are other issues to consider? Are there some in the text that you don't think are issues? Can you defend your position? In the end, what can one country do to stop genetic engineering of a particular type from happening?

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Chapter 10: Biotechnology & Genetic Engineering

3. Critical step in understanding of genetic traits, whether a genetic disease an an economically important trait in livestock b. Searching for specific genes c. Genetic analyses linkage, physical-association & physical characteristics.

4. An organism that has been modified by altering one or more genes using recombinant DNA technology.
B. NO passed Risk assessments

9.

10. Enhancing growth Rates, improved muscle-to-fat Ratio of carcass, improve resistance to disease, enhance feed nutrient use, improved milk production and composition, and enhanced reproductive performance and increased prolificacy

12. A. Process using a variety of laboratory techniques to determine if a person has, or is likely to get, a genetic condition or disease.
B. A technique for correcting defective genes responsible for disease development, by insertion, alteration, or removal of genes with an individual's cells.
C. Study of how an individual's inherited variation in different genes affects drug response.

13. Insulin, HPV- cervical cancer, vaccines for baby pig scours, foot rot, sheep measles & infectious bursal disease, BST- milk production

14. Produced by transgenic bacteria, causes great milk ~~production~~