Animal Pharming: The Industrialization of Transgenic Animals December 1999

Animal pharming, the process of using transgenic animals to produce human drugs, is staking its claim in a lucrative world market. Transgenic animals are animals which have been genetically transformed by splicing and inserting foreign animal or human genes into their chromosomes. The inserted gene, when successful, enables an animal to make a certain pharmaceutical protein in its milk, urine, blood, sperm, or eggs, or to grow rejection-resistant organs for transplant.

Global demand continues to grow for human proteins and vaccines. These proteins serve numerous therapeutic purposes such as treatments for cystic fibrosis, hemophilia, osteoporosis, arthritis, malaria, and HIV (see Table 1). Transgenic animals can also produce monoclonal antibodies (antibodies specifically targeted towards disease proteins) which are used in vaccine development.

Transgenic animals are costly to produce and they have high value. The cost of making one transgenic animal ranges from $20,000 to $300,000, and only a small portion of the attempts succeed in producing a transgenic animal. A Wisconsin firm that clones transgenic calves for human pharmaceutical production estimated that one transgenic animal can produce, in its lifetime, $200 to $300 million worth of pharmaceuticals.

In 1998, less than one percent of the world supply of human therapeutic proteins came from production of recombinant proteins (proteins which are formed by laboratory manipulation of genes in plants, bacteria, or animals). That tiny percentage of overall production, however, was valued at almost $12 billion, or 50 percent, of a $24 billion global market for human proteins[1]. A recent Financial Times article reported that a herd of 600 transgenic cows could supply the worldwide demand of some pharmaceuticals, for example, human serum albumin used in the treatment of burns and traumatic injuries.

With greater integration of computers into laboratory functions, molecular biologists have drastically reduced the time needed to identify and isolate genes. As gene sequencing has become increasingly automated, each known sequence is recorded and stored in a data base. Animal producers are using the body of genetic information to produce transgenic animals.

Automated gene sequencing and the biological advantages of animals, when compared to more

traditional methods of recombinant protein production, have combined to make pharming a preferred alternative. Traditional methods of recombinant protein production use laboratory cell cultures of transgenic bacteria, yeast or animal cells to produce proteins. Inherent disadvantages in traditional methods, when compared to using animals as bioreactors, include (1) cell and bacterial cultures require constant monitoring and sampling, (2) expansion is more costly, because substantial plant machinery must be purchased and maintained, and (3) isolating and purifying proteins is more difficult than purifying proteins from an animal's milk or bodily fluid.

Overall, animals as bioreactors are more cost effective because the product is efficiently passed through the milk, with an average yield of 53 percent and with 99 percent purity. The purifying process may become more simple if harvesting proteins from poultry eggs and urine becomes viable.

Using animals as bioreactors is also cost-effective and advantageous because animals naturally carry the cellular mechanisms needed to produce complex proteins. Genes require certain cellular mechanisms to help them produce proteins. These mechanisms are present in a living animal, but they may be difficult or impossible to replicate in a cell culture.

Unit cost per protein should be significantly less when animals are used as bioreactors to produce human proteins. Only a fraction of the raw material, capital equipment, and maintenance costs needed for traditional cell culturing are required with transgenic animals. Table 2 highlights one private firm's estimates regarding the superiority of transgenic human protein production through eggs or goat milk.

The technology used to develop transgenic animals is somewhat mature, however, the industrialization of bio-pharming is new. The first transgenic animal, a mouse, was produced in 1981. In an effort to determine which genes were involved with cancer, a gene was inserted into the mouse that made it susceptible to cancer. In 1985, the first transgenic farm mammal was produced, a sheep called "Tracy". Tracy had a human gene that expressed high levels of the human protein alpha-1-antitrypsin. The protein, when missing in humans, can lead to a rare form of emphysema.

Transgenic animals with alterations to the germ line are commonly produced through microinjection (see Figure 1). Changes to the germ line are heritable from generation to generation within the herd, and this heritability has potential to facilitate long-term productivity gains. For example, fish can be bred for increased expression of a growth hormone, although the industries are currently wary of consumer preferences for gene-modified product. Another example is the recently developed "enviro pig", a pig which had a phytase gene placed in its salivary glands to allow better utilization of phosphorus in feedstuffs. The genetically modified "enviro pig" may prove to be part of the solution to animal waste issues.

Due to the expense and difficulty in producing a transgenic animal, cloning is the method of choice to produce multiple transgenic animals without altering the animal's genes as traditional breeding would do. Cloning is the process of replicating an identical gene, cell, or organism from a single ancestor. Since Dolly the sheep was cloned by nuclear transfer in 1997, advances in cloning technology have

made it easier to build a genetically tailored livestock herd. Several farms of cloned transgenic animals have emerged throughout the world, including a Wisconsin operation with 37 cloned cows in 1999, 17 of them transgenic. Cloning an animal is still costly, ranging from $100,000 to $200,000 in 1999, but cloning costs are projected to decrease to $5000 in the next few years as the practice becomes more common[2].

Concerns regarding transgenic animals have developed along with the new technologies. These concerns include safety of the food supply, safety of the pharmaceutical product, and keeping the transgenes out of non-transgenic animals.

As of December 1999, no specific legislation had been enacted to direct federal oversight of transgenic animals. Therefore, the U.S. Food and Drug Administration (FDA) has considered transgenic animals according to animal drug provisions of the Federal Food, Drug and Cosmetic Act. In 1994, the FDA issued a report titled "Points to consider in the manufacture and testing of therapeutic products for human use derived from transgenic animals." This report set guidelines for the development, maintenance and disposal of transgenic animals.

Because some transgenic animals are produced for food rather than for pharmaceuticals, the U.S. Department of Agriculture, Food Safety and Inspection Service (USDA-FSIS) has issued guidance on food safety issues related to transgenics. In 1994, the FSIS published "Points to consider in the food safety evaluation of transgenic animals from transgenic animal research." This FSIS guidance, revised in 1997, highlighted animal regulations that must be met before animals may be submitted for slaughter. Regulations require the producer to supply information about the animal including any drugs, biologics or chemicals that were administered, or any genetic alterations. The FSIS also offers continuing education courses for USDA employees on food safety issues in transgenic animals.

The Animal and Plant Health Inspection Service (APHIS) of the USDA has also begun to explore implications of increased production of transgenic animals. As commercial use of transgenic food animals increases, APHIS will encounter new questions concerning animal health and disease control. Animal vaccines produced by transgenics will fall under APHIS regulation, just as transgenic plants and arthropods are already overseen by APHIS.

Summary

As transgenic animals, pharming, and cloning become more mainstream, a small yet growing portion of the animal production industry will shift its operations from farming livestock for meat production, to pharming transgenic animals for pharmaceutical production. The world market is growing for human pharmaceutical products. Producing transgenic animals is still relatively expensive, however, costs are trending down and transgenic animals have certain advantages over traditional laboratory methods for producing human proteins. More commercial use of transgenic animals in food production is also likely.

Regulators will need to review existing policies and guidelines regarding transgenic animals. New policies regarding transgenic and cloned animals may be necessary to ensure the safety and health of

humans and animals. Ongoing public debate regarding transgenic technologies will ensure that further research and analyses will be demanded by animal producers, regulators, environmentalists, and the general public.

References

1) The Biotechnology Revolution Keeps Evolving.", Research Studies-Business Communications Inc. Page:1 06/25/99
2) Netherton, Martha "Barnyard biotech.", Georgia Trend, Page 84, July 01, 1999

For more information or references contact:

Margaret Rittner or David Cummings
Centers for Epidemiology and Animal Health
Telephone: (970) 490-8000
David.j.cummings@aphis.usda.gov
http://www.aphis.usda.gov/vs/ceah/cei/

Table 1: Pharming products currently in development.

Animal	Drug/protein	Use
sheep	alpha1 anti trypsin	deficiency leads to emphysema
sheep	CFTR	treatment of cystic fibrosis
sheep	tissue plasminogen activator	treatment of thrombosis
sheep	factor VIII, IX	treatment of hemophilia
sheep	fibrinogen	treatment of wound healing
pig,	tissue plasminogen activator	treatment of thrombosis
pig	factor VIII, IX	treatment of hemophilia
goat	human protein C	treatment of thrombosis
goat	antithrombin 3	treatment of thrombosis
goat	glutamic acid decarboxylase	treatment of type 1 diabetes
goat	Pro542	treatment of HIV
cow	alpha-lactalbumin	anti-infection
cow	factor VIII	treatment of hemophilia
cow	fibrinogen	wound healing
cow	collagen I, collagen II	tissue repair, treatment of rheumatoid arthritis

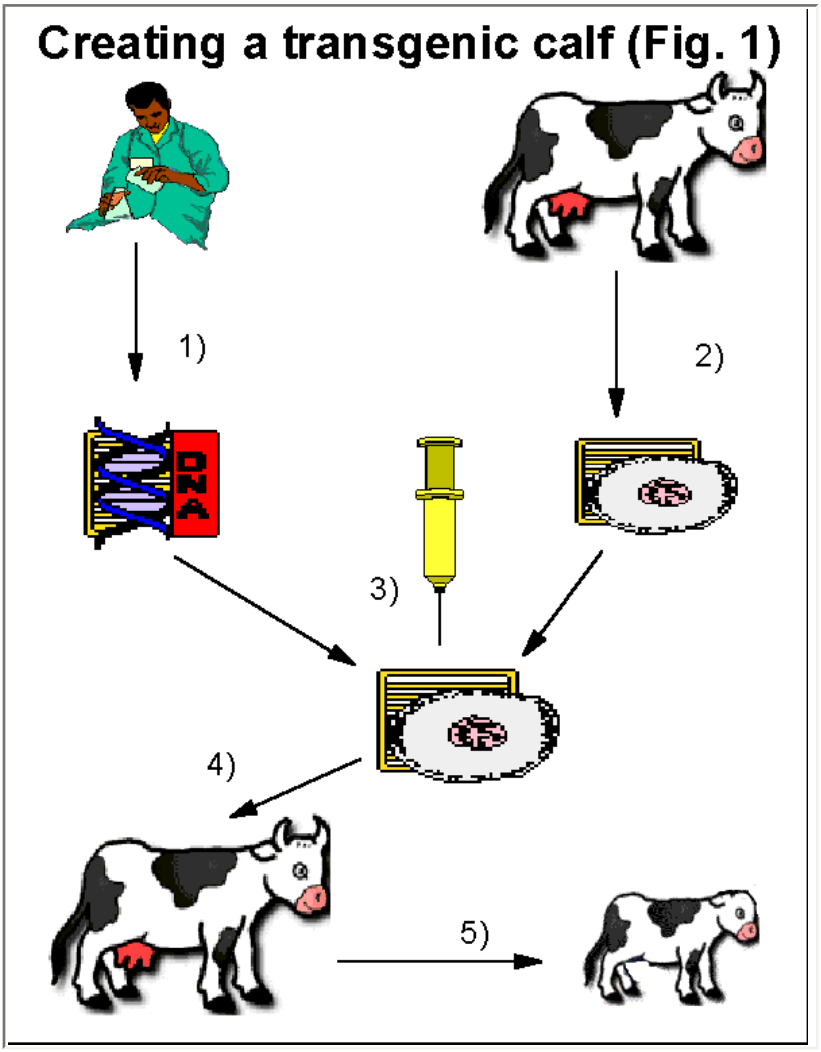
cow	lactoferrin	treatment of GI tract infection, treatment of infectious arthritis
cow	human serum albumin	maintains blood volume
chicken, cow, goat	monoclonal antibodies	other vaccine production

Table 2: AviGenics comparison of production inputs and costs for monoclonal antibodies* using traditional cell culture versus using transgenic poultry or goats

	Traditional Cell Culture	Poultry Eggs	Goat Milk
Raw Material Volume (kg)	170,000	250	21,000
Capital Equipment Cost, or Cost per Animal (dollars)	100 Million	1,000	10,000 to 50,000
Equipment Maintenance Costs, or Keeping Cost per Animal (dollars)	100,000	10	2,500
Unit Cost per Protein (dollars per gram)	100	0.10 to 0.25	2 to 20

*100 kg of raw material per year
Source: AviGenics www.avigenics.com

Figure 1: Creating a transgenic calf



(1) A human gene responsible for producing a desired protein is isolated in a laboratory.

(2) An animal is given hormonal treatment to produce a large number of embryos, and the embryos are collected from the oviduct.

(3) The human gene is inserted into the fertilized egg via micro injection. DNA of the pronucleus is injected into the fertilized embryo.

(4) The transgenic embryo is placed in a surrogate host which gives birth to the transgenic animal.

(5) The offspring is tested for the new gene.