Human proteins in animal milk

Living Medicine Factories

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The beginnings of genetic engineering date back to the 1970's. Today this technology permits us to produce animals that manufacture human proteins – one of the most expensive medicines

Human proteins can even be made in bacteria, and this process is at present the simplest, cheapest and quickest means of doing so. However, not all proteins can be obtained in this way. This is why higher organisms - fungi, plants, and animals - are also used in such processes. In these systems, proteins are subjected to all the modifications needed for them to function properly. Complete proteins can be obtained quickest though the use of mammalian cell cultures. These, however, are extremely expensive, a factor that restricts their application.

Transgenic animals are produced by introducing alien genes into their cells. In order for a gene to be transmitted to subsequent generations, it must be introduced into the reproductive cells. The drawback of this solution may be the long time it takes for the transgenic organism to develop. This is especially important in the case of farm animals, e.g. pigs, sheep or cattle. If the process of genetic modification was fully successful, however, then we can expect to see the alien proteins in the milk, urine, blood or semen of animals, and the eggs of poultry.

Recombinant proteins, also called fusion proteins, are obtained in genetic engineering by attaching a sequence that encodes a "fused" part to the DNA sequence that encodes a specific protein. The additional piece increases the stability of the protein, facilitates and frequently speeds up the isolation process. It is later removed via enzymatic processing.

Milking the hormones

One of the proteins whose production has has gained a lot of attention recently is the growth hormone necessary

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A researcher from the Agricultural Academy in Poznań prepares the DNA samples of the transgenic animals for the PCR analyses

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Transgenic rabbits: female (left) and male – prospective factories of the human growth hormone

for the proper development of vertebrates. This hormone is involved in the metabolism of mammals, stimulating the synthesis of proteins and the degradation of fats. Attempts to introduce constructs containing the growth hormone gene into the genome of animals were already made back in 1982. In the case of farm animals, this approach was used to try to accelerate their growth and to reduce the cost of breeding. Today such animals are gradually being released onto the market.

The only human growth hormone preparation recognized by the World Health Organisation (WHO), used for example in treating dwarfism in children, girls with Turner's syndrome or AIDS patients, is obtained from *in vitro* bacteria cultures. It has very good biological properties, but is relatively expensive. Extraction from the human pituitary glands was previously the main source of human growth hormone, although major constraints on this are the availability of material, and especially the possibility of transmitting prions, for example. The alternative is to use transgenic animals as bioreactors, producing the human growth hormone in their milk or blood. The greatest attention has focused on the mammary glands, which can be used as bioreactors because during lactation milk may be obtained in large volumes (1 kg protein/day from a cow or 200 g/day from goats). By using "triggers" (promoters) of genes specific to the mammary glands, the production of human proteins can be restricted to these organs alone. The presence of foreign proteins in milk should not affect the animal's organism.

My research group decided to join forces with the team of Professor Z. Smorąg from the National Research Institute of Animal Production in Balice, and focus on obtaining animals that produce proteins with pharmaceutical significance in their milk. We designed gene constructs so that the foreign protein would not be active in the animals, with activation occurring only during laboratory purification. In our research we combined the Whey Acid Protein (WAP) gene promoter with the fusion part and gene of the human growth hormone (gene construct WAP: 6xHisHGH). The end of the human growth hormone gene was modified, including by adding a sequence recognized by the enzyme thrombine. Only after purification is the protein transformed by thrombine into its active form. Some animals produce so much foreign protein in their mammary glands that obtaining this protein from the milk becomes industrially feasible.

Cocktail against allergies

A second area of our research involves replacing allergens isolated from natural sources, which are used diagnostically, with preparations containing recombined allergens, selected carefully for composition and content – thus producing a so-called "cocktail" with clearly defined activity. The possibility of producing unlimited volumes of recombined allergens led us to contemplate using prophylactic vaccines in allergy therapy. If the allergen were to be administered with other vaccines within 1–2 years from birth, then it could lead to partial or complete inhibition of the development of the immunological response involving IgE-class antibodies. The use of recombined allergens is also foreseen in *in vitro* diagnostics, where their possible application in DNA microarray technology is particularly interesting.

The list of proteins that can be obtained in transgenic animals is not yet very long. The first protein - α-1-antitripsine – is slated to be launched on the market in 2004. At present we have a collection of a dozen-odd gene contructs that encode proteins of pharmacological significance, planned to be introduced into rabbits, pigs, goats and cattle. ■

Further reading:

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