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AN OVERVIEW ON ADVANCES IN TRANSGENIC ANIMALS

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ABSTRACT The dependence of man on animals such as cattle, sheep, poultry, pig and fish for various purposes(milk, meat, eggs, wool etc) is well known. Improvement in genetic characteristics of livestock and other domestic animals(example, high milk yield, weight gain etc), in early days was carried out by selective breeding methods. This technique primarily involves a combination of mating and selection of animals with improved genetic traits. Although selective breeding is very time consuming and costly, it was the only method available till some years ago, to enhance genetic characteristics of animals. For larger animals with a long gestation period, it might take several decades to create a desired character by conventional breeding. With the advent of modern biotechnology , it is now possible to carry out manipulation at the genetic level to get the desired characteristics in animals. Transgenesis refers to the phenomenon of introduction of exogenous DNA into the genome to create and maintain a stable heritable character. The foriegn DNA that is introduced is called transgene. And the animal whose genome is altered by adding one or more transgenes is said to be transgenic. The transgenes behave like other genes present in the animal genome and are passed onto the offspring. Thus, transgenic animals are genetically engineered or genetically modified organisms with a new heritable character. It was in the 1980s, the genetic manipulation of animals by introducing genes into fertilized eggs became a reality.

KEYWORDS : Transgenic Animals, Transgenesis, Transgene, Genetically Modified Organisms

INTRODUCTION

The technology has already produced transgenic animals such as mice, rats, rabbits, pigs, sheeps and cows. Although there are many issues surrounding transgenesis, this research focuses on the basics of technology and its application in agriculture, medicine and industry. Foriegn DNA is introduced into the animal, using recombinant DNA technology, and then must be transmitted through germ lines so that every cell, including germ cells, of the anime contains the same modified genetic material.

Genes can be altered artificially, so that some characteristics of an animal are changed. For example, an embryo can have an extra, functioning gene from another source artificially introduced into it which can knock out of the functioning of another particular gene in the embryo. Animal that have their DNA manipulated in this way are known as transgenic animals

In genetically modified organisms (GMO) or genetically engineered organisms(GEO) genetic material has been altered using genetic engineering techniques. These techniques are genetically known as recombinant DNA technology. With this technology, DNA molecules from different sources are combined into one molecule to create a new set of genes. This DNA is then transferred into an organism, giving it modified or novel genes. Transgenic animals, which are a subset of GMOs, are organisms that have inserted DNA that originated in different species.

A transgenic animal develops from genetically altered gamete or fertilized ovum. Transgenic plants can derive from those sources as well as from somatic cells. Different vectors and gene transfer techniques are sometimes used in plants because their cell walls, which are not present in animals, are difficult to penetrate. Transgenic technology permits rapid introduction of new traits. For example, a gene that confers an agriculturally useful characteristic - such as the ability to withstand a particular pesticide - is isolated from one species and inserted into a vector; then the recombinant vector is placed into single plant cells whose cell walls have been removed. A whole plant generated from a genetically engineered cell has the gene for the transferred trait in all of its cells.

The complex interactive processes of living mammals are not reproducible in vitro. However, transgenic animals provide a means of evaluating genetic modifications in terms of anatomical and physiological changes in a complex system. Transgenic models are more precise in comparison to traditional animal models, for example the Onco-mouse with its increased susceptibility to tumour development enables results for carcinogenicity studies to be obtained within a shorter time-frame, thus reducing the course of tumour development in experimentally affected animals. The majority of transgenic animals produced so far are mice, the animal that pioneered the technology. The first successful transgenic animal was a mouse. A few years later, it was followed by rabbits, pigs, sheeps and cattle.

History

By 'transgenic' one means an organism in which a gene has been changed or added from another organism. In 1946, Max delbruck showed that genes from 2 different viruses could be combined to form a new kind of virus. This is called recombination. We had to wait until 1972 for paul berg to knowingly join two DNA strands from different sources into one plasmoid. This was the first so-called recombinant DNA, which is the basis of transgenics we are seeking. The first chimeric mice were produced in 1974. The cells of two different embryos of different strains were combined together at an early stage of development to form a single embryo that subsequently developed into a chimeric adult, exhibiting characteristics of each strain. In 1973, Herbert Boyer combined a gene from a bacterium with a gene from a virus and got a recombinant DNA that was then introduced into bacteria E.coli. The first transgenic organism came into existence. The recombinant E.coli expressing a salmonella gene, led to concerns in the scientific community about potential risks from genetic engineering.

The first transgenic animals were mice created by Rudolf Jaenisch in 1974. He successfully managed to insert foreign DNA into the early stage of mouse embryos. The resulting mice carried the modified gene in all their tissues. Subsequent experiments, injecting leukemia genes to early mouse embryos using a retrovirus vector, proved the genes integrated not only to mice themselves but also to their progeny.

Methodology

The production of transgenic mice was the cumulation of previous advances in the areas of recombinant DNA and the micromanipulation of mammalian cells. Although the production of synthesized mice could be the first step towards the introduction of genetic materials into intact mammalian organisms, this approach only allowed the introduction of the whole genome into chimeric mice by aggregation of genetically different embryos at a cleavage stage. Another approach to the transfer of genes into the germ line resulted in the insertion of discrete sequences of DNA. This technique is based on the introduction of teratocarcinoma cells into synthesized cavities. A more efficient method is based on the use of embryonic stem cells. These cells can be transformed or selected for a particular phenotype and they also contribute frequently to the germ cell population of chimeric animals. Besides it, the retroviruses have been successfully used to transform embryos by infection.

There are three methods for introducing a foreign gene into mice, and in fact the same methods are applicable to other animals as well

1. Retroviral vector method
2. Micro-injection method
3. Embryonic stem cell method
4. Transfection of fertilized/unfertilized egg or embryo
5. Transfer of whole nucleus
6. Transfer of whole individual chromosomes or fragments
7. Transfection of cultured mammalian cells

8. Targeted gene transfer

Transfer Of Whole Nucleus

Transfer of whole nucleus from a somatic cell of a superior donor to the enucleated egg can be achieved using the following steps

1. Enucleation of unfertilized eggs is achieved by centrifuging Cytochalasin-B - treated cells, such that the nuclei detach from the eggs and pellet at the bottom of the tube, leaving enucleated eggs in the supernatant.
2. Karyoplasts are similarly obtained from the blastula stage of the developing embryos of the donor.
3. Karyoplasts derived from the donor are incubated with enucleated eggs in the presence of PEG, and fusion is achieved.
4. The manipulated eggs are transferred to the uterus of the surrogate mother for development.

Technique has also been developed, where a superior developing embryo may be bisected into 2 parts using a surgical blade. Each bisected part of the embryo may be separately transferred to an enucleated unfertilized egg. Such a manipulated egg may then be transferred to the uterus of a surrogate mother for further development.

Transfer Of Whole Individual Chromosomes Or Fragments

Chromosomes may be isolated from the metaphase cells by hypotonic lysis. Incubation of these isolated chromosomes with whole cells after co-precipitation with calcium phosphate results in their incorporation in the nuclei. Chromosomes once isolated may also be subjected into fractions using density centrifugation or flow cytometry and individual specific chromosome pairs may be inserted into recipient cells.

CONCLUSION

Innovative technologies to enhance experimental gene transfer efficiency in different species are desperately needed. Such enabling techniques would not only bring the cost of individual projects into a reasonable realm but would also increase the likelihood of breakthrough studies in many disciplines. Apart from mouse modeling, multigene targeted blastomere aggregation and nuclear transplantation studies, DNA microinjection into pre-implantation embryos has been the only reproducible means for heritable gene transfer in the preponderance of species.

There is no doubt about the benefit of biotechnology especially genetic engineering to man and the amount of transformations they bring to society. The production of genetically modified or altered animals and plants has many applications and advantages in many areas, the alteration of natural gne is however against nature. When it is applied to animals and plants it is beneficial to man, even though it has some harmful effects in their original genes. But man will not stop here and continue his exploration and experimentation to invent and innovate.

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