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Research Article

THE GAME OF CLONE

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ABSTRACT

S0 you think you want to be a mom- at least some time in the near future. How exciting! Having a child is a wonderful experience that will enrich your life forever. But the decision to have a child shouldn't be taken lightly. Having trouble in getting pregnant? So are about 15 % of married women have this problem. Harward Medical school and Finnish researchers recently compare the data from 36 countries on rates of fertility, milk consumption and women's ability to digest milk. In Thailand 98 % of adults are" lactose tolererant "Women that are infertile can have their own babies with the help of implementing the cloned embryos in to their bodies. In cloning techniques defective genes could be eliminated, faster recovery from traumatic injury and infertility could be eliminated. The primary issues associated with the human cloning are possibility of faster aging, potential loss of individuality and may reduce the over all value of human life.(1)

INTRODUCTION

There are various aspects of cloning; animal cloning, human cloning and second therapeutic cloning. However, animal cloning and therapeutic cloning are in clinical practice (2). The animal cloning can be used for various commercial applications including agriculture, research, and medicine. The advancement of medical molecular biology and cutting-edge biomedical engineering technologies enabled us to the manipulate gene of interest for desired phenotypes without affecting animal physiology (3). There is the enormous potential of animal cloning in modern time which can fulfill shortage of food, agriculture, and domestic animals and therapeutic applications as well. (4)'Foreign' genes have been transplanted into zebrafish, which are widely used in laboratories, and embryos cloned from these fish express the foreign protein (5) Combining such genetic techniques with cloning of pigs (achieved for the first time in March 2000) would lead to a 133 reliable supply of suitable donor organs (6). Cloning could be used to create better animal models of diseases, which could, in turn, lead to further progress in understanding and treating those diseases (7). Cloning creates a genetically identical copy of an animal or plant. Many animals - including frogs, mice, sheep, and cows - had been cloned before Dolly. Human identical twins are also clones (8). For example, when two genetically identical cloned mice embryos are combined, the aggregate embryo is more likely to survive to birth (9). Commercially, genetically engineered animal clones are used to produce pharmaceuticals and other products in milk, immune tolerant pig cells and organs for xenotransplantation research and cloned animals for agricultural purposes (10). Hans Adolf Edward Dreisch showed that by shaking a two celled sea urchin embryo, you could separate the cells, which then developed into two separate sea urchins (11). Steen Willadsen separated one cell from an 8-cell lamb embryo, and then was able to use a small electric shock to fuse it into an egg cell with no nucleus. This turned into an embryo, and days later they placed the embryo into a surrogate mother sheep, who had three baby lambs (12). In 1996, Dolly was the first mammal created by somatic cell nuclear transfer. Ian Wilmut and Keith Campbell transferred the nucleus from an adult sheep udder cell into an enucleated egg to try and make a lamb. There were 277 attempts, but only one created an

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embryo, which was then placed into a surrogate mother. The result was a lamb named Dolly. (13)

The first in vitro fertilized (IVF) offspring was a rabbit born in 1959. Since that time, IVF offspring have been born to mice, rats, hamsters, cats, guinea pigs, squirrels, pigs, cows, monkeys (14).

The overall technique for IVF is similar among species and involves significant manipulations in vitro, or outside the body of animals (15).

In livestock species, oöcytes are collected from the ovaries of either living or deceased animal whose genetic potential is desirable. Transvaginal aspiration can obtain ovaries from live animals, or from a deceased 137 animal at the time of slaughter. Slaughterhouse ovaries are cross-sectioned, and the contents of all of the follicles are collected; mature oöcytes are collected, evaluated for quality, and used for fertilization (16). Embryo splitting may be considered the first true "cloning" procedure involving human intervention and was first described by Willadsen and Polge in 1981 when monozygotic twin calves were produced (17).

Transgenic sheep and goats have been produced that express foreign proteins in their milk. Transgenic chickens are now able to synthesize human proteins in the "white" of their eggs. These animals should eventually prove to be valuable sources of proteins for human therapy. Methods of creation of transgenic animals For practical reasons, i.e., their small size and low cost of housing in comparison to that for larger vertebrates, their short generation time, and their fairly well-defined genetics, mice have become the main species used in the field of transgenics. The three principal methods used for the creation of transgenic 139 animals are DNA microinjection, embryonic stem cell-mediated gene transfer, and retrovirus-mediated gene transfer (18).

1. DNA microinjection, involves the direct microinjection of a chosen gene construct from another member of the same species or a different species, into the pronucleus of a fertilized ovum. (19).
2. The insertion of DNA in to fertilized ovum is transferred into the oviduct of a recipient female, or foster mother that has been induced to act as a recipient by mating with a vasectomized male. (20).
3. This method involves prior insertion of the desired DNA sequence by homologous recombinetion into an in vitro culture of embryonic stem (ES) cells. Stem cells are undifferentiated cells that have the potential to differentiate into any type of cell and therefore to give rise to a complete organism (20).
4. These cells are then incorporated into an embryo at the blastocyst stage of development. The result is a chimeric animal. ES cellmediated gene transfer is the method of choice for gene inactivation, the so called knock-out method. This technique is of particular importance for the study of the genetic control of developmental processes.. Retrovirus-mediated gene transfer To increase the probability of expression, gene transfer is mediated using a carrier or vector, generally a virus or a plasmid. (21).
5. Transmission of the transgene is possible only if the retrovirus integrates into some of the germ cells. Depending on the technique used, the F1 generation may result in chimeras. (22),
6. The animal cloning relies on advanced molecularbiology technology and cutting edge bioengineering. The enzymes are used in animal cloning are broadly from the category of DNA manipulation enzymes. These enzymes are restriction digestion enzyme, ligation enzymes and DNA amplifying enzymes such as a polymerase. (23).
7. Animal cloning has provided a vast range of transgenic animals. The benefits can be grouped into Agriculture, Medicine, Industry. Breeding Farmers have always used selective breeding to produce animals that exhibit desired traits (e.g., increased milk production, high growth rate). When technology using molecular biology was developed, it became possible to develop traits in animals in a shorter time and with more precision. Also, it offers the farmer an easy way to increase yields. The quality Transgenic cows exist that produce more milk or milk with less lactose or cholesterol, pigs and cattle that have more meat on them, and sheep that grow more wool. In the past, farmers used growth hormones to spur the development of animals, but this technique was problematic, especially since the residue of the hormones remained in the animal product. Disease resistance Scientists are attempting to produce disease-resistant animals, such as influenza-resistant pigs, but a very limited number of genes are currently known to be responsible for resistance to diseases in farm animals.
8. Medical Applications a) xenotransplantation Patients die every year for lack of a replacement heart, liver, or kidney. Transgenic pigs may provide the transplant organs needed to alleviate the shortfall. (24).

Nutritional supplements and pharmaceuticals Products such as insulin, growth hormone, and blood anti-clotting factors may soon be or have already been obtained from the milk of transgenic cows, sheep, 142 or goats. Research is also underway to manufacture milk through transgenesis for treatment of debilitating diseases such as phenylketonuria (PKU), hereditary emphysema, and cystic fibrosis (25).

Human gene therapy Human gene therapy involves adding a normal copy of a gene (transgene) to the genome of a person carrying defective copies of the gene. (26).

Major Advances and Discoveries

Industrial Applications In 2001, two scientists at Nexia Biotechnologies in Canada spliced spider genes into the cells of lactating goats. The goats began to manufacture silk along with their milk and secrete tiny silk strands from their body by the bucketful. By extracting polymer strands from the milk and weaving them into thread, the scientists can create a light, tough, flexible material that could be used in such applications as military uniforms, medical microsutures, and tennis racket strings (27). Challenges and issues Most of the ethical concerns about cloning relate to the possibility that it might be used to clone humans. There would be enormous technical difficulties. As the technology stands at present, it would have to involve women willing to donate perhaps hundreds of eggs, surrogate pregnancies with high rates of miscarriage and stillbirth, and the possibility of premature aging and high cancer rates for any

children so produced. However, in 2004 South Korean scientists announced that they had cloned 30 human embryos, grown them in the laboratory until they were a hollow ball of cells, and produced a line of stem cells from them. The further ethical discussion was raised in 2008 when scientists succeeded in cloning mice from tissue that had been frozen for 16 years. In the USA, President Clinton asked the National Bioethics Commission and Congress to examine the issues, and in the UK the House of Commons Science and 143 Technology Committee, the Human Embryology and Fertilisation Authority and the Human Genetics Advisory Commission all consulted widely and advised that human cloning should be banned (28).

Significant Gap in Research

The Council of Europe has banned human cloning: in fact, most countries have banned the use of cloning to produce human babies (human reproductive cloning). However, there is one important medical aspect of cloning technology that could be applied to humans, which people may find less objectionable. This is therapeutic cloning (or cell nucleus replacement) for tissue engineering, in which tissues, rather than a baby, are created. In therapeutic cloning, single cells would be taken from a person and 'reprogrammed' to create stem cells, which have the potential to develop into any cell in the body. When needed, the stem cells could be thawed and then induced to grow into particular types of cell such as heart, liver or brain cells that could be used in medical treatment. Reprogramming cells are likely to prove technically difficult. Therapeutic cloning research is already being conducted in animals, and stem cells have been grown by this method and transplanted back into the original donor animal. In humans, this technique would revolutionize cell and tissue transplantation as a method of treating diseases. However, it is a very new science and has raised ethical concerns. In the UK a group headed by the Chief Medical Officer, Professor Liam Donaldson, has recommended that research on early human embryos should be allowed. The Human Fertilisation and Embryology Act was amended in 2001 to allow the use of embryos for stem cell research, and consequently, the HFEA has the responsibility for regulating all embryonic stem cell research in the UK (29).There is a potential supply of early embryos as patients undergoing in-vitro fertilization usually produce a surplus of fertilized eggs.

Where the Research Go Next

As far as animal cloning is concerned, all cloning for research or medical purposes in the UK must be approved by the Home Office under the strict controls of the Animals (Scientific Procedures) Act 1986. This safeguards animal welfare while allowing important scientific and medical research to go ahead. Summary The animal cloning is a growing arena of molecular biology and have a great scope in medical and biomedical applications. Interestingly, the creation of transgenic animals has resulted in a shift in the use of laboratory animals — from the use 144 of higher-order species such as dogs to lower-order species such as mice — and has decreased the number of animals used in such experimentation,26 especially in the development of disease models. This is certainly a good turn of events since transgenic technology holds great potential in many fields, including agriculture, medicine, and industry. The therapeutic application of animal cloning is gaining new momentum as conventional approaches largely fail to cure several life-threatening diseases and disorders. The use of animals for the production of recombinant proteins, antibodies, growth hormone and enzymes is important in current prospect. The chimeras research is an applied aspect of animal cloning give rise a diverse animal model not only to study disease biology but also finding a cure.

Current Debate

It is a hot debate to argue the pro and cons of cloning. It is controversy. At a certain point of time, it was considered as profound achievement in medicine. But now it is becoming a heated topic in debate from all over the world. The main advantages with cloning are it eliminates the defective genes. It is extremely powerful tool to bring about the huge changes for the entire world. It gives a new meaning of genetic modification and could eliminate infertility. It can cure some disorders by replacing damaged tissues and organs within the human body. However, it has several adverse effects like goes against religious ethics and decreases overall value of human life.

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