

Cloning of animals and human beings

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Abstract: Every plant propagated by cutting is a clone of the original. Thus cloning of plants has been done since antiquity. Cloning of animals is something new. This paper reviews recent experiments with cloning of animals.

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1. Introduction

The cloning of plants dates to antiquity. Any plant propagated by cutting is a clone of the original plant. The Bartlett Pear, for instance, originated from a single seedling around 1770, and has been propagated largely through cuttings. Many houseplants are propagated through cuttings.

Cloning of animals can occur in nature. Earthworms and starfish, if cut in two, will grow to replace the “missing” part, and the two resulting creatures are clones of the original. In some cases an egg cell divides into twins, producing two animals that have the same genetic heritage. However, intentional cloning of higher animals is much more difficult than it is with plants. Only within recent years have techniques been developed that permit cloning of animals.

2. Dolly, the first cloned animal

Dolly, a sheep, was born on July 5, 1996. She was the first successful animal born of attempts to clone higher animals (Wilmut et. al). She was cloned from a dead adult sheep using frozen cells. It took 277 tries to get one successful birth.

Dolly suffered from many health problems during her lifetime. She apparently had arthritis. She also may have been “born old.” Olknikov (1973) observed that the telomeres, at the ends of chromosomes, shortened as cells divided throughout the life of an individual. Since Dolly came from the chromosomes of an adult sheep, her telomeres were already shorter than those of a newborn sheep. This may have contributed to her health problems.

Dolly was euthanized in February 2003, after suffering from lung cancer caused by a virus. (Whitfield 2003). In addition, she suffered from arthritis.

2. Reproductive Cloning

Dolly was an example of reproductive cloning, which is intended to produce a duplicate of the original animal. The process, called “somatic cell nuclear transfer” (SCNT), involves removing the genetic material from an egg cell and replacing it with the genetic material taken from a cell from the original animal. Division of the egg cell is then stimulated electrically or chemically. When cell division has proceeded far enough, the cell mass is transplanted to the uterus of the animal that will serve as the mother for the clone.

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The result is an animal that is genetically identical with the animal from which the genetic material was taken. In effect this can be considered delayed twinning. However, the clone may not be an exact duplicate of the donor of the DNA. Only chromosomal or nuclear DNA is transferred in SCNT. The mitochondria of the egg cell remain in the egg. Thus the clone inherits some genetic material from the egg donor.

3. Subsequent cloning successes

3.1 Mice

Wayakama et. al. (1998) succeeded in cloning mice by transferring the nuclei of cumulus cells into enucleated oocytes (egg cells). They were able to create two lines of cloned mice. Line A started with 2 pups born out of 131 egg cells (0.6% successful); Line B resulted in 4 pups born out of 96 egg cells (4.2% successful).

3.2 Mules

A mule is produced by breeding a male donkey (62 chromosomes) to a female horse (64 chromosomes), producing an offspring with 63 chromosomes. Mules are sterile. In this experiment (Woods et. al. 2003), a donor fibroblast cell line was established from a 45-day mule fetus. The nuclei from these were implanted in egg cells collected from mares (horses). The resulting mule oocytes were implanted in mares. A total of 334 oocytes were prepared by this method. Of these, 305 were transferred to recipient mares. Of these, 21 resulted in 14-day pregnancies. One live mule was born of this procedure.

This was the first successful cloning of an animal that cannot normally reproduce. However, it should be noted that the mule fetus would have been able to develop into a normal mule. This experiment is different from those in which the genetic material is taken from a skin cell or other similar source, rather than from a fetus.

3.3 Horses

A cloned horse was born on May 28, 2003, weight 79 pounds (Galli et. al. 2003). A team headed by Cesare Galli, at the Laboratory of Reproductive Technologies, Cremona, Italy, carried out the project. Galli's team took a single skin cell from a 6-year-old mare and fused it with an egg cell whose own genetic material had been removed. That newly combined cell grew in a lab dish into an embryo, which was transferred into the mare's uterus for a standard 11-month gestation.

The team created 841 embryos by this process, growing them in the laboratory. All but about two dozen embryos died during their first week in laboratory dishes. Seventeen embryos were transferred to surrogate mother horses. This resulted in four pregnancies. Two of these ended spontaneously within two weeks. A third aborted after six months. Only one was born normally. The successful birth came from the same mare that provided the genetic material for insertion into the egg cell. Thus this mare gave birth to her own twin sister.

At birth, the foal appeared to be normal, and was suckling within an hour after being born. The clone's markings differ from those of her genetic twin, because there is some chance influence in markings as well as genetic influence.

4. Endangered or Extinct Species

The idea of recreating an extinct species through cloning was popularized by the novel and the movie *Jurassic Park*. While it may never be possible to recreate dinosaurs, the idea is not completely far-fetched. Recent attempts have succeeded in cloning the last survivor of an endangered species, and in cloning an extinct species. These techniques may have significant value in restoring endangered species.

4.1 Cloning the last survivor

The idea of perpetuating an endangered species by cloning the last survivor is an attractive one. In one recent experiment, Princess, the last female of the Gloucestershire Old Spot pig bloodline in the US, was successfully cloned (Westphal 2002). The cloning was attempted only after several attempts at breeding the last remaining sow failed.

While in this case the donor was not exactly the last survivor, since there were three other bloodlines of the same breed in North America, the experiment does demonstrate the potential for perpetuating a species when there is one survivor left.

4.2 Cloning an extinct species

Cloning an extinct species requires that there be some usable genetic material available to serve as the nucleus of an egg. An attempt was made to clone Asian Banteng cattle from a specimen that died over 20 years ago. The Banteng is not extinct, but is endangered in its native environment. The cloning was done in an attempt to increase the genetic diversity of Bantengs in captivity. Cellular material was taken from a specimen preserved in the “frozen zoo” at the San Diego Zoo. Two Banteng calves were born. One appeared to be healthy. The other suffered from what has come to be called “large calf syndrome” and was euthanized shortly after birth. Cloned animals are sometimes much larger than they should be. This leads to heart failure and failure of other organs.

Another current attempt to clone an extinct species is that to clone the Tasmanian Tiger. The last specimen died in 1936. However, stuffed specimens exist in Australian zoos. Polymerase Chain Reaction was used to amplify the genetic material extracted from cells in these specimens. These DNA fragments must then be assembled into functioning chromosomes. Once this is done, the DNA can be inserted into an egg cell that may then be grown into a Tasmanian Tiger. This process has not yet been completed successfully for the Tasmanian Tiger, but is still in progress.

5. Cloning pets

Cats have been cloned successfully (Shin). This has led to the first commercial cloning enterprise. The firm Genetic Savings and Clone now offers to clone pet cats. Pet owners supply a sample of skin from the cat to be cloned. For a fee, this is stored until needed. For another fee (quoted at \$50,000), an attempt will be made to clone the cat from the stored skin sample. The skin is first cultured, to verify that it is viable. DNA from a skin cell is then inserted into an enucleated egg, which is implanted in a female cat.

While humans may become attached to their pets, it must be remembered that environment as well as heredity influences the development of an animal. Even a genetic copy of a pet cat, raised in a different environment, will not behave exactly like the cat from which the DNA was extracted. This people paying for a clone of their pet may be sorely

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disappointed. Moreover, considering the number of cats that are euthanized every year by animal control facilities, it seems pointless to spend large sums to replicate a particular cat.

6. Cloning Humans

6.1 Reproductive cloning

While there have been reports in the press from time to time of the cloning of human beings, none have been confirmed. There is strong political objection to reproductive cloning of humans.

Moreover, the idea of cloning human beings seems to make little sense. As has been noted, the normal reproduction of human beings requires no capital equipment, is undertaken voluntarily, and can be performed by unskilled labor.

6.2 Therapeutic cloning

The term therapeutic cloning refers to a process that is identical with that which would be used in reproductive cloning, but in which the embryo is destroyed and the stem cells harvested. The hope is that the stem cells could be used to treat various diseases or conditions, such as Alzheimer's or spinal cord damage.

The fact of identity with the start of reproductive is disguised by using the technical term "somatic cell nuclear transplant." Despite the verbal camouflage, however, the idea is to create an embryo human being with the deliberate intent of destroying him or her.

7. The Downside of Cloning

These successful attempts at cloning animals have received considerable attention. They do represent remarkable achievements that could not have been possible without the technology available today for manipulating individual cells. However, the story is not one of unalloyed success.

7.1 Low success rate

The success rate for having what amount to fertilized eggs carried to term is extremely low. Typical success rates were fractions of a percent to one or two percent. In the case of cloned mice, over six generations, there were 35 pups born from 3920 "fertilized" eggs, or a success rate of about 0.9%.

7.2 The health problems of clones

One of the issues regarding cloning is whether the cloned animals are inherently less healthy than animals born normally. As noted above, Dolly, the first cloned sheep, suffered from arthritis and lung cancer. "Large calf syndrome" is common enough that it has a name. It is not clear whether clones must inherently have health defects, but a high percentage of them turn out to. Whether this comes from deficiencies in the cloning procedure, or some other cause, is not clear yet.

8. Summary

Reproductive cloning of animals has been demonstrated in certain species. It appears to have some utility for replicating prize animals, for enhancing the preservation of endangered species, and for recreating extinct species.

Human therapeutic cloning requires the creation and deliberate destruction of an embryo. The potential for treatment of various diseases and conditions has been claimed, but not yet demonstrated. Moreover, what experiments have been undertaken have been complete failures.

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