

# Cloning of endangered and extinct wild animals

Lena Hollund Surdal / 1<sup>st</sup> year veterinary / May 2009

Today's scientists are at all times looking for answers and everyday they get a little closer to a lot of them. Some think that only time can show what we actually have the possibility to discover. The world we live in has a lot of secrets, and not least about the secrets of life.

The secrets of life have started to get some answers, and people think they have found some answers in cloning. In the last decades scientists have discovered a lot of new things and we have had some big breakthroughs. One of the breakthroughs was the sheep Dolly that was born in 1997, she was the first cloned mammal. This has created a lot of questions, discussions, and reactions. Here I'm going to discuss some of the issues about cloning endangered and even extinct wild animals.



*Young Spanish ibex (Capra pyrenaica), Sierra de Gredos, Spain Photo: Jose Luis GOMEZ de FRANCISCO/naturepl.com*

## Cloning

Cloning has occurred naturally in nature at all times, but now humans have learned how to clone animals artificially. The nucleus (DNA) is moved from the donor cell to an enucleated recipient cell, and you then create an exact genetic match of the donor. This is called Somatic cell nuclear transfer and you can abbreviate it to SCNT. The offspring will have the same genetic complement as the original donor which can be an extinct or endangered animal, but the mitochondrial DNA will be from the recipient.

When you find clones in nature the offspring only have one parent. The parent reproduces asexually, and the offspring is called a clone. Then you get two genetically identical individuals. In plants you have a lot of examples of cloning, just think about plants like the potato and strawberry. In humans you have one example, and that's when you have two identical twins. Researchers have also found some animal species that reproduce asexually, for instance anemones and *Mycocepurus smithii*. According to a new study these ants have evolved to reproduce only when queens clone themselves.

## Endangered species

The first endangered clone of an endangered species happened in 2001. They had managed to clone an Asian ox the year before, but it died shortly after its birth. In 2001 they managed to successfully clone the European mouflon, a small wild sheep. This was a big step for those that, after Dolly was born

believed that cloning was the way for people to rescue populations of endangered and threatened species. After 2001 more endangered species have been tried to be cloned, on example is cloning of the endangered gray wolves (*Canis lupis*).

The gray wolf was nearly exterminated from south Korea by the 1960s, and has become increasingly rare in northern Korea. They wanted to know if nuclear transfer of postmortem wolf somatic cells into enucleated dog oocyte was a feasible method to produce a cloned wolf. They took tissue obtained from a male gray wolf 6 hour after death, transferred 372 embryos to 17 recipient dogs, and four got pregnant. One of them spontaneously delivered two dead pups, but the 3 other delivered 4 cloned wolf pups. One of them was much smaller than the other and died shortly after birth. The 3 live wolf had a normal wolf karyotype and amount of telomeric DNA. Then they concluded that this demonstrated the successful cloning of an endangered male gray wolf via interspecies transfer of somatic cells, isolated postmortem from a wolf, and transferred into enucleated dog oocytes. They also concluded that SCNT has potential for preservation of canine species in extreme situations, including sudden death, like they did with the dead wolf.

SCNT has been used in cloning several kinds of somatic cell-cloned mammals, so it has promise for maintaining endangered or extinct species.

For project like this interspecies can have some potential when there are limitation to obtaining con-specific oocyte. These 3 pups that was successfully born demonstrated the use of iSCNT using somatic cells collected postmortem from an adult gray wolf.

The clone result was that the pups and nuclear donor wolf had identical microsatellite patterns, and the recipient females had different. Their mitochondrial DNA was as expected identical to those of the domestic dog oocyte donor. So these wolf was a kind of hybrid, with genomic and mitochondrial DNA from two different species. (Mitochondria are organelles outside the nucleus that have their own genomes.)

If we want to use cloning to preserve endangered species, intraspecies and interspecies SCNT technique is something that have great potential as a tool. A lot of species have limited availability of oocyte and recipients, so this can be a good solution.

These researchers got better result on cloning this time compared to last project just because some small factors. Important factors was for example to know precisely time of ovulation, have good SCNT technique, and to know embryos required for successful pregnancy. This also show how fast scientists can learn by experience, and get new information.



### **Extinct species**

In year 2000 the last individual of the Pyrenean ibex (*Capra pyrenaica pyrenaica*), also called a bucardo, died. The scientists had capture this female a year before, and cells from a skin biopsy were obtained, multiplied and kept frozen in liquid nitrogen. Using DNA taken from these samples, the scientists were able to replace the genetic material in eggs from domestic goats, and transferred

to pure Spanish ibex or hybrids between Spanish ibex and domestic goats. Bucardo was one of the four subspecies of the Spanish ibex, but because of money and practical reason they also used hybrids. Divided by two experiment they transplanted the DNA into 439 eggs, then 57 was implanted in the recipients, 8 got pregnant, and 1 goat was born at the end.

The newborn ibex kid died shortly after birth (after 7 minutes) because physical defects in its lungs, but still this was a breakthrough for many people. This show that it can be possible to save endangered and newly extinct species by resurrecting them from frozen tissue. It also make it more possible to belief that they can reproduce such as the woolly mammoths, who scientists now are trying to get the fully genome from.

### **Research speed**

Cloning by nuclear transfer is nothing new. The technique was first reported in frogs in 1952, and another breakthrough was 5 July 1996, when Dolly was born. She was the genetic copy of a six-year-old Finn Dorset ewe. Since then a lot of animals has been duplicated, including pigs, goats, mules, horses, and rats. The researchers find new information about cloning and within stem cell research every year. US have approved animal clones as food in , and the world are using a lot of money on these kind of experiment. The first cat was cloned in 2002, and the first dog in 2005 (canine eggs are more difficult to work with). Some of the last information from researchers is by a South Korean research team. In April 2009 the research team told that they had cloned beagles with an implanted fluorescent gene. These dogs glow red under ultraviolet light, and some parts looks red even under normal light. The puppy was born in December 2007 through cloning with a gene that produces a red fluorescent protein. These show that the scientist are going forward and we are getting more information about it, and perhaps we can used it to save endangered and extinct species.

### **People have issues**

The largest issue about cloning is when it comes to the idea to clone human beings. And a lot of people are against cloning of animals because it looks like it eventually will end with clones of human beings, and you then get a big ethical dilemma. Cloning can be the last chance for some species, and a lot of the them are extincted or endangered because of human influence on them and the nature.

SCNT may preserve and propagate endangered species that reproduce poorly in captivity until natural habitats can be restored and populations reintroduced to their ecological units. People have already started to make so called frozen zoo. That is a cryonic facility for the long term storage of animal and plant genetic material, such as DNA, sperm, eggs, and embryos. The preserved material can then be used for artificial insemination, in vitro fertilization, embryo transfer, and cloning.

### **What to do?**

At this stage, cloning of animals is not possible for any but a few species that are closely related to domestic animals and it is very expensive. Further, the technology requires surrogate mothers and an ample supply of these is unlikely to be available for threatened species. But the researchers are discovering new things every year, so we should have the possibility open. Cloning may contribute to the conservation of endangered animal in the future. For example, biopsies can be collected from endangered animals, and used to expand numbers rapidly in captivity, whilst retaining the wild population. The founders could be used to generate the target population size with essentially no loss of genetic diversity. Then they have to collecting and storing the DNA of threatened species when population numbers are high, then they might be able to restore the original level of genetic variability through cloning.