SPOTLIGHT

CAN DNA BE USED TO BRING THE EXTINCT BACK TO LIFE?

— FELIX PATTON FILES THE LAST IN THIS SERIES ON DE-EXTINCTION

FELIX PATTON



is a rhino ecologist, who writes and broadcasts about the species from Africa and Europe. He has an MSc in Conservation Biology and a PhD based on research into individual rhino identification

and social behaviour. He is a frequent contributor to SWARA.

Perhaps the most exciting and controversial new conservation paradigm is that of De-extinction – principally the use of biotechnology and genomic science to both save near extinct species and bring back extinct species with the aim of restoring them to their former range.

Specimens of many extinct species are available in museum and university collections and some fossils. From these it may be possible to extract samples of the DNA, the carrier of the genetic blueprint of the organism,



Model of the Woolly Mammoth.

from which its full set of genes can be read and analysed. That data may, (or may not), be transferable as working genes into their closest living relatives, effectively bringing the extinct species



Mural of now extinct species previously found in Mexico from the National Museum of Anthropology, Mexico City.

back to life. What started mostly as a scientific challenge has led to molecular and conservation biologists rapidly developing techniques that will ultimately result in producing live individuals of now extinct species. HOTO BY: MAMMUT/FLYING PUFFIN

The World cannot expect to see the re-appearance of the dinosaur as it is too long gone for useful DNA to be extracted. But it could see the Wooly Mammoth as the Sooam Biotech Research Foundation in Korea has it in its long-term sights. This raises concerns over the ethics of bringing back a species into an entirely alien environment.

However, rather than be concerned with long gone species, it is perhaps better to consider those species that are today on the edge of extinction and how the technology can revive their populations. Species such as the Javan, Sumatran and Northern White Rhino





and even the cheetah are examples of such.

What of the progress so far? The first living birth of an extinct

animal, a Pyrenean ibex, was recorded in 2009. The foetus survived to term but died minutes after birth from lung abnormalities. The individual was created using cryo-preserved DNA implanted into eggs from domestic goats with the resulting embryos transplanted into the Spanish ibex or goat-ibex hybrids for gestation.

This technique is being tried to revive the Australian Gastric-brooding frog which went extinct in 1980. DNA is being implanted into eggs of the barred frog and live embryos produced.

A different technique involving genetic engineering is being progressed



Top Left: Pyrenian Ibex sketch 1898. Below: Southern Gastric-brooding Frog giving birth in a laboratory. Top Right: Stuffed passenger pigeon.

in order to revive the Passenger Pigeon. The basic concept is to "change" the genome of the band-tailed pigeon to match that of the Passenger Pigeon by converting viable band-tailed DNA into viable Passenger Pigeon DNA. The demise of this species, first recorded in 1534, was dramatic. In the 1800's there were billions of these birds but during the 1870s millions upon millions of birds were consumed for food with their feathers used in the making of bed mattresses and pillows. Live birds were caught and shipped by the thousands for trap-shooting tournaments. There were at least one billion Passenger Pigeons alive in 1878, but by 1890 only tens could be spotted anywhere. On September 1, 1914 Martha, the last surviving individual living at the Cincinnati Ohio Zoo, died and the Passenger Pigeon became extinct.

Seventy-seven Passenger Pigeon specimens including bones and tissue were sampled with the result that one specimen was considered of high enough quality to obtain a full genome and more than 20 specimens useful for population biology research with bits of DNA from all over the genome. However to actually assemble the genome is no easy task. For a start

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Top Left: Ben Novak preparing Passenger Pigeon DNA Libraries at the University of California Santa Cruz Paleogenomics Lab.

Top Right: DNA sequences of passenger pigeon DNA as viewed during the assembly process. Each line is a small sequence of nucleotides and comes from a different species/sample which are aligned together to reveal differences in their sequences.

Below Right: Life restoration of an Aurochs bull . Bottom: A Maremmaner bull.

it consists of 1.3 billion base pairs of genetic code. These have to be pieced together from the fragmented bits of DNA left in museum skins, almost all of the fragments less than 150 base pairs in length. More than 80 million fragments of DNA are needed to be able to put the whole genome together in a way that overcomes misinterpreting damage to the DNA as genuine mutations. Some DNA will be contaminants— from bacteria that were living on dust in the museum drawer, or from the human DNA of museum curators over the past century.

With sufficient specimens and enough recorded history to establish a great understanding of the species' past, it will be possible to plan its future in a knowledgeable and responsible manner. This makes it a model species for thoroughly testing the process of deextinction.

A third technique, known as back breeding, is being used to bring back the extinct species of cattle, the Aurochs which was once widespread

> throughout Europe, North America and Asia on open savannah-like landscapes to marshes, forests and lower mountains. Hundreds of plant and animal species developed in co-evolution with the vast herds of these Europe's heaviest land mammals and other large grazers. The last Aurochs individual died in 1627.

Two projects working on Aurochs are focusing







on cross-breeding the most suitable primitive breeds of cattle from throughout Europe to produce a new fully self-sufficient cattle breed which will be a very close, though not exact, copy of the Aurochs. Several old cattle breeds have been proven to still be very close genetically to the Aurochs.

One project is developing the 'Tauros' which will have all the right characteristics of the Aurochs – the colour, size, behaviour and the way of grazing. It will not be one single breed but a number of lineages bred in and suited to different regions of Europe. Seven breeds have been selected for a cross breeding programme including the Maremmaner primitive.

In November 2012 the Taurus Foundation and Rewilding Europe signed a strategic partnership agreement with the goal to re-populate the new European rewilded areas (see SWARA April-June 2014) with wild bovines again. In turn this should result in the preservation and restoration of many species on decline in the relevant ecosystems especially where large herbivores are missing while bringing the potential for economic recovery from eco-tourism.

PHOTO BY: PETER VON WESENDONK



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PHOTO BY: KATHY



As the DNA of the Aurochs has been completely reconstructed, it should be possible to use genome editing to remove or reduce unwanted traits and eliminate wrong genes from the new genomes produced by the back breeding.

It may be abhorrent to some to work on species that are long gone but the application of these technologies can be used to revive todays near extinct wildlife populations. Species such as the Javan, Sumatran and Northern White Rhino and even the cheetah could benefit although there are already people who are against the use of any form of genetic engineering even for saving species. Take, for example, the Northern White Rhino which today is represented by only seven live but nonbreeding individuals in the world and extinct in its historic range. Once, the species was widespread through many

African countries to the west of the Albert Nile in Uganda with the most recent population to die out in Garamba National Park, Democratic Republic of Congo. This large herbivore would have made a major contribution to shaping the ecosystems in these countries and without it, biological diversity would have declined and will continue to do so.

The nearest living relative to the Northern White rhino is the Southern White rhino of which there are some 20,000 today, all of whom have been derived through the generations from less than 50 that remained in 1900 in South Africa. Despite this small remnant population, there has been no indication of inbreeding or genetic abnormalities. It is likely that quality DNA samples from Northern White rhino individuals around this number can be obtained from the live animals, cryo-preserved material from animals that died in captivity, samples taken during notching or treatment for injuries and archived material from zoos and museums. Genome sequencing can compare the DNA from all these sources and only the best chosen for the production of Northern White rhino embryos in the laboratory. These could then be implanted into Southern White rhino females to act as surrogate mothers. This all sounds quite easy and, of course, it would not be as more research would be needed to perfect the material and methods needed to obtain long lasting, live embryos.



Top Left clockwise: Barrosa bull, Chianina cow, Northern white rhino in Garamba National Park, Northern white rhino and Javan rhino.

Heading closely towards extinction are the Javan rhino with only some 35 to 45 individuals in the wild and none in captivity, and the Sumatran rhino with numbers below 200. It has recently been announced that there are more tigers in captivity than in the wild; that lions are extinct in 25 African countries and virtually extinct in a further 10 and that over a thousand rhinos were poached in 2013. The list of species doomed by human action is endless. The de-extinction projects currently under way will create roadmaps for others to follow in the future and especially will highlight the pitfalls to be avoided. It is a potential human answer to a human problem. The question being asked is, is the technology an intervention too far?

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