"Kepler contributed enormously, and now we're excited to go on to the next steps." —DAVID LATHAM, PAGE 10

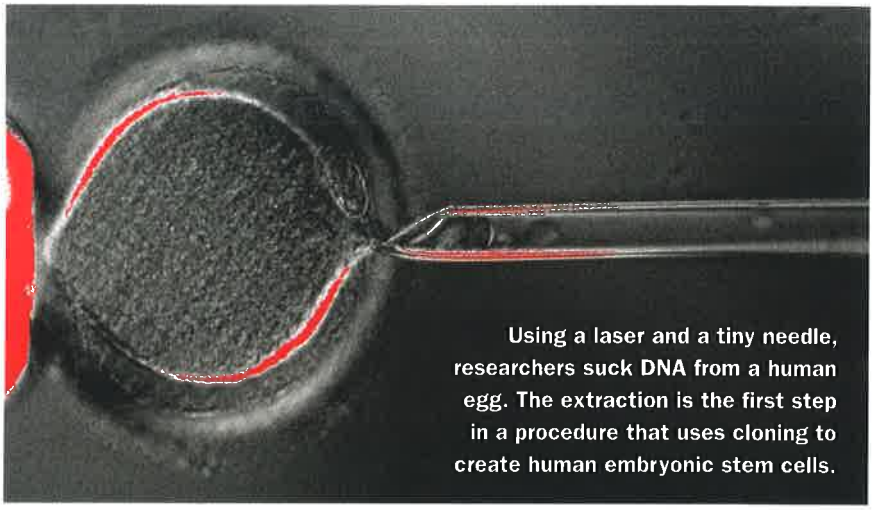
In the News

STORY ONE

Human cloning advance raises personalized medicine hopes

Embryonic stem cells made with nuclear transfer method

By Meghan Rosen



Using a laser and a tiny needle, researchers suck DNA from a human egg. The extraction is the first step in a procedure that uses cloning to create human embryonic stem cells.

COURTESY OF M. TACHIBANA

For the first time, scientists have created human embryonic stem cells by transferring the nucleus of a mature cell into an egg. The cloning technique could nudge the dream of personalized medicine closer to reality, researchers suggest May 15 in *Cell*.

"It's a huge, landmark achievement," says stem cell biologist George Daley of Children's Hospital Boston and Harvard University. Creating embryonic stem cells by nuclear transfer in humans, he says, is "the next major technological advance since Dolly."

The famous sheep was the first mammal cloned by the nuclear transfer technique, which inserts the nucleus of a cell from one adult animal into the egg of another. Since Dolly's birth in 1996, scientists around the world have tried to duplicate the technique in human cells.

If cloned human cells could be made to grow into normal embryos, the technique could supply fresh stocks of embryonic stem cells.

Unlike adult cells, which have already followed a path to become, say, heart cells or neurons, embryonic stem cells are uniquely poised to become any cell in the body. By making these stem cells from a patient's own tissues, once-untreatable conditions might be cured by replacing damaged cells with healthy ones.

Until now, the only way to get embryonic stem cells was from leftover embryos made through in vitro fertilization. These cells are useless for personalized medicine because they are not genetically matched to a patient, but they are extremely valuable for laboratory experiments. In 2001, however, President George W. Bush set new regulations that choked off federal funding for embryonic stem cell research. Scientists could use only discarded embryos created for reproductive purposes, and all embryos discarded after 9 p.m. August 9, 2001, were off limits. The rules sent researchers racing to find alternative ways to make embryonic stem cells.

In 2007, one new technique to create the cells dazzled scientists in the field. By dosing human cells with a small cocktail of molecules, researchers pushed a reset button that turned adult cells back into embryonic-like ones called induced pluripotent stem cells, or iPS cells.

"For the last six or seven years, virtually all of us have ended our nuclear transfer efforts and switched over to iPS cells," Daley says.

But a team led by Shoukhrat Mitalipov of the Oregon National Primate Research Center in Beaverton kept plugging away at nuclear transfer, first using rhesus macaques, and then human cells.

Using cloning to create embryonic stem cells in humans has proven tricky, says Kathrin Plath, a stem cell biologist at UCLA. No one knew why the technique worked in some other mammals but not humans.

Researchers had to figure out the best way to ease out an egg's DNA, slip in a new nucleus and then cue the egg to divide and grow. In 2011, scientists came close, but the egg stalled out after three divisions, producing just eight cells.

A key change to the protocol was adding caffeine to the eggs before DNA transfer, says stem cell biologist James Byrne of UCLA, who was not involved in the new work. Caffeine acts like a set

For the latest news, visit **www.sciencenews.org**

of chemical reins, holding back the egg's development until researchers inject a new nucleus. The new protocol also features other tweaks such as examining the eggs under polarized instead of ultraviolet light, which can be more damaging to the egg.

Using the new method, researchers made embryonic stem cells from a donor egg and the nucleus of a young boy's skin cell. The new cells can grow and divide to form a mass of embryonic stem cells just like those derived from fertilized embryos, Mitalipov said in a press briefing May 14.

Since then, questions have been raised about image duplications and manipulations in the Mitalipov group's *Cell* paper. In the past, such discoveries have led to charges of scientific misconduct and even fraud, but so far there has been no evidence that the research is invalid and the journal stands by the paper.

When the researchers ground the cells up and compared the genetic bits with those in embryonic stem cells, they didn't see much of a difference. Virtually all of the new cells' genes were reset to their embryonic states.

What's more, Byrne says, the approach is much more efficient than traditional methods of producing embryonic stem cells. Instead of burning through thousands of eggs to make a single embryonic stem cell line, Mitalipov's group succeeded with as few as two eggs.

The new cells may have advantages over iPS cells in treating some genetic flaws that lurk in mitochondria, little cellular power plants that carry their own DNA. By putting the nucleus of a patient's skin cell into a fresh egg with healthy mitochondria, scientists could conceivably make a customized therapy that erases the defects, Mitalipov said.

"Most scientists who practice nuclear transfer think it's unethical and unsafe to try human reproductive cloning."
GEORGE DALEY

The work "is certainly impressive," says developmental biologist John Gurdon, who shared the 2012 Nobel Prize in physiology or medicine for pioneering the nuclear transfer technique to clone a frog.

Next, Gurdon says, researchers ought to compare the new embryonic stem cells with iPS cells. A side-by-side look might provide clues to how resetting adult cells actually works. If they can figure out why Mitalipov's nuclear transfer method is so successful, researchers might be able to improve the technique to make iPS cells and avoid having to retrieve eggs from volunteer donors.

Improving iPS cells could also help scientists skirt the ethical issues of human cloning. One of the biggest issues with nuclear transfer is the possibility that it could be misused for human fertility treatments, says Daley.

"The animals produced by cloning are abnormal," he says. "Most scientists who practice nuclear transfer think it's unethical and unsafe to try human reproductive cloning."

But, he says, used for research, the technique could address many important scientific questions.

Embryonic stem cells made using this method also have the potential to treat spinal cord injuries and diseases such as diabetes or Parkinson's, says Dietrich Egli, a stem cell biologist at the New York Stem Cell Foundation. "I'm very confident that such cells will be used for therapies in humans in the future." ■

Back Story | HISTORY OF CLONING RESEARCH

Advancement in animal cloning has occurred against a backdrop of ethical debates and scientific fraud.

1962 John Gurdon clones a frog using nuclear transfer.

1996 Ian Wilmut, Keith Campbell and colleagues create the first cloned mammal, a sheep they named Dolly (shown below).

2001 President George W. Bush bans federal funding for research that uses stem cells, with the exception of a small number of existing cell lines. The decision severely restricts stem cell research in the United States.

2004, 2005 South Korean researcher Woo Suk Hwang claims to have cloned human cells.

2006 Seoul National University reports that Hwang's work was fabricated.

2007 Shinya Yamanaka creates induced pluripotent stem cells from humans.

2009 President Barack Obama reverses Bush's 2001 decision.

2011 Researchers try to create human embryonic stem cells using nuclear transfer, but the eggs either stall out after only a few divisions or need an extra set of chromosomes to grow.

2013 For the first time, Shoukhrat Mitalipov and colleagues use cloning to create human embryonic stem cells.

GETTY IMAGES