

# Researchers achieve major breakthrough in cell reprogramming

September 30 2010

---

(PhysOrg.com) -- A group of Harvard Stem Cell Institute (HSCI) researchers has made so significant a leap forward in reprogramming human adult cells that HSCI co-director Doug Melton, who did not participate in the work, said the Institute will immediately begin using the new method to make patient and disease-specific induced pluripotent stem cells, know as iPS cells.

“This work by Derrick Rossi and his colleagues solves one of the major challenges we face in trying to use a patients own cells to treat their disease,” said Melton, who is also co-chair of Harvard’s Department of Stem Cell and Regenerative Biology. “I predict that this will immediately become the preferred method to make iPS cells from patients and, indeed, at the HSCI we are turning our entire iPS core to using this method,” Melton said.

The findings today were given advance on-line publication by *Cell Stem Cell*.

Rossi’s group at the Immune Disease Institute at Children's Hospital Boston has used synthetic [mRNA](#) to reprogram adult human skin cells, fibroblasts, turning them into cells that are apparently identical to human embryonic stem cells, the initial building blocks of all the organs of the body. They have then used other mRNA to program the new cells, which they are calling RiPS (RNA-iPS), cells to develop into specific cells types - in the current study they created muscle cells.

Because the mRNA carries [genetic instructions](#), but does not enter the DNA of the target cells, the resulting tailored cells should be safe to use in treating patients, Rossi said, unlike the iPS cells now being created around the world.

“Our findings address three major impediments to clinical translational use of iPS cells,” Rossi said in an interview. “The method: does not in any way breach genomic integrity as it does not necessitate integrating genes or viruses into the target cells’ DNA; it is orders of magnitude more efficient at producing iPS cells than conventional iPS methods, which were notoriously inefficient; and it gives us a way to directly program and direct the fate (development) of the iPS cells towards clinically useful cell types.”

The Rossi group indeed appears to have in one fell swoop solved not one, but all three of the major problems with which researchers have been grappling ever since Japanese researcher Shinya Yamanaka announced in 2006 that he had used four genes to convert fully developed adult skin cells - or fibroblasts - into cells with all the properties of embryonic stem cells. The cells, which Yamanaka dubbed induced pluripotent stem (iPS) cells could, like embryonic stem cells, be induced to become any human cell type.

But Yamanaka used virus to insert the genes into genome of the target cells, and that created at least two major impediments to using the iPS cells to treat human disease - the main goal of stem cell researchers:

First, the use of the integrating viruses raised the very real possibility that cancers might inadvertently be triggered; and second, inserting the genes into the genome could lead to changes that would alter the properties of the resulting iPS cells so that they would not be identical to human [embryonic stem cells](#).

Ever since Yamanaka's discovery scientists have been looking for other ways to turn adult cells into iPS cells, both to study diseases by creating cells lines carrying the genes of diseased patients, and to create patient-specific cell lines to use to treat individual patients. Now, with Rossi's RiPS cells, they may have what they've been seeking.

“Most approaches for generating iPS cells involve some sort of integration into the genome, usually viral,” Rossi explains, “so clearly the development of a technology that does not breach genomic integrity is very important. Gene therapy trials unfortunately taught us the danger in leaving viruses in the genome as some patients developed cancers that were driven by the integrated viruses. So when one thinks about strategies for regenerative medicine, you need to envisage utilizing cells whose genome has not been breached. We believe that that utilizing RNA to generate transplantable cells and tissues is a ideal solution because ,to the best of our knowledge, RNA is completely non-integrative,” Rossi says.

What Rossi and his team have done is create artificial messenger RNA - mRNA - that carries the instruction sets from the four genes used by Yamanaka. So the mRNA tells the adult cells to reprogram, just as the Yamanaka genes do, but it does so without disturbing the integrity of the adult cell's genome. Thus the resulting RiPS cells may be more identical to [human embryonic stem cells](#) since they do not contain viral transgenes. Indeed, when Rossi and colleagues compared the RiPS cells they made to human ES cells, they found a much closer match than when iPS cells were generated with viruses.

Next comes the question of how to turn the RiPS cells, cellular blank slates, into the kind of cells the researchers need to treat patients, for example, the insulin-producing beta cells destroyed in diabetics, or the motor neurons that degenerate in the brains of patients with Parkinson's.

Here again Rossi and his team turned to mRNA. “Up to this point it’s been extremely difficult to direct cells to differentiate towards particularly fates, or cell types,” Rossi explains. Current methods for directing cellular differentiation usually rely on tightly controlling the environment in which the cells are developing, tailoring the growth media and other factors to coax the iPS cells to develop into a particular cell type. “We thought to use mRNA encoding cell type specific factors in order to drive the fate of iPS cells to the desired cell fate. We are beginning to know more about what factors are need to create certain types of cells - a great example was the demonstration by Doug Melton’s group that they could use just 3 specific factors to turn adult pancreatic exocrine cells into insulin-producing beta cells.”

But those experiments again required the insertion of gene-carrying viruses into the target cells, says Rossi, although the Melton group substituted chemicals for some of the viruses. To demonstrate that mRNA might be used to direct iPS cell fate,, Rossi and colleagues synthesized an mRNA with the instruction set for making muscle cells, and showed that they could use this to efficiently drive the fate of RiPS to [muscle cells](#), again without compromising the genome of these cells. “These results provide us with a new experimental paradigm that might safely be used in regenerative medicine,” he says.

At the same time they appear to have found a way to produce and program iPS cells in an experimental and medically useful way, Rossi’s group reports it has also found a method that is far more efficient than previous methods for producing iPS cells.

“Up until now, iPS cell generation has been an extremely inefficient process,” Rossi continues. “Our technique allows for iPS generation that’s significantly more efficient than conventional approaches.” It used to be that only 0.001 to 0.01 of starting cells could be reprogrammed to iPS cells. The study by Rossi and colleagues however reports iPS

conversion efficiencies in the range of 1 to 4% of starting cells. What that practically means is that iPS cells can be generated even if only very few starting cells is used. This may prove important for patients in which only very few starting cells can be obtained.

The icing on this particular cellular cake is that it includes yet a fourth finding with implications far beyond stem cell science - the Rossi group reports that it is found a way to overcome the natural cellular immunity to the insertion of foreign RNA.

“I am sure were not the only lab to have the idea of using RNA for cellular reprogramming,” says Rossi. “The problem is that when you introduce RNA into a cell, the cell thinks it is being infected by an RNA virus and retaliates by producing a massive interferon response that effectively shuts down cellular function and can prompt the cell to altruistic suicide as it tries to stop the ‘virus’ from replicating. In order to use RNA for cellular reprogramming we clearly needed to overcome this problem,” he explains. “Our approach was to modify the RNA so that it no longer set off anti-viral responses when introduced into cells. The modified-mRNA enabled us to efficiently express proteins in cells for days and weeks without causing any adverse reaction in the [cells](#). This in turn allowed us to reprogram cell to pluripotency, which is a process that requires several weeks of Yamanaka factor expression.

“Although we developed this technology for cellular reprogramming, it actually has utility far beyond that,” says Rossi, who has patented the technology, and is forming a company to develop it. “Basically our technology provides a means of transiently expressing any protein in a cell without eliciting the cell’s anti-viral response pathways. This could have potential therapeutic benefit in patients suffering from a protein deficiencies.”

This work was supported with funding from Derrick Rossi’s “startup

package,” the money newly hired faculty receive to establish their labs, and with a Seed Grant from the Harvard Stem Cell Institute.

Said Doug Melton, “It’s wonderful to see that HSCI seed grant funds given to outstanding, innovative, and imaginative young scientists like Rossi that can so quickly and dramatically change a field.”

**More information:** Paper: [www.cell.com/cell-stem-cell/fulltext/S1934-5909%2810%2900434-0](http://www.cell.com/cell-stem-cell/fulltext/S1934-5909%2810%2900434-0)

Provided by Harvard University

Citation: Researchers achieve major breakthrough in cell reprogramming (2010, September 30) retrieved 30 April 2024 from <https://phys.org/news/2010-09-major-breakthrough-cell-reprogramming.html>

This document is subject to copyright. Apart from any fair dealing for the purpose of private study or research, no part may be reproduced without the written permission. The content is provided for information purposes only.
---