Reprogrammed adult cells treat sickle-cell anemia in mice

December 6 2007

Mice with a human sickle-cell anemia disease trait have been treated successfully in a process that begins by directly reprogramming their own cells to an embryonic-stem-cell-like state, without the use of eggs. This is the first proof-of-principle of therapeutic application in mice of directly reprogrammed “induced pluripotent stem” (IPS) cells, which recently have been derived in mice as well as humans.

The research, reported in *Science Express* online on December 6, was carried out in the laboratory of Whitehead Member Rudolf Jaenisch. The IPS cells were derived using modifications of the approach originally discovered in 2006 by the Shinya Yamanaka laboratory at Kyoto University.

The scientists studied a therapeutic application of IPS cells with the sickle-cell anemia model mouse developed by the laboratory of Tim Townes of the University of Alabama at Birmingham. Sickle-cell anemia is a disease of the blood marrow caused by a defect in a single gene. The mouse model had been designed to include relevant human genes involved in blood production, including the defective version of that gene.

To create the IPS cells, the scientists started with cells from the skin of the diseased mice, explains lead author Jacob Hanna, a postdoctoral researcher in the Jaenisch lab. These cells were modified by a standard lab technique employing retroviruses customized to insert genes into the cell’s DNA. The inserted genes were Oct4, Sox2, Lif4 and c-Myc,

known to act together as master regulators to keep cells in an embryonic-stem-cell-like state. IPS cells were selected based on their morphology and then verified to express gene markers specific to embryonic stem cells. To decrease or eliminate possible cancer in the treated mice, the c-Myc gene was removed by genetic manipulation from the IPS cells.

Next, the researchers followed a well-established protocol for differentiating embryonic stem cells into precursors of bone marrow adult stem cells, which can be transplanted into mice to generate normal blood cells. The scientists created such precursor cells from the IPS cells, replaced the defective blood-production gene in the precursor cells with a normal gene, and injected the resulting cells back into the diseased mice.

The blood of treated mice was tested with standard analyses employed for human patients. The analyses showed that the disease was corrected, with measurements of blood and kidney functions similar to those of normal mice.

“This demonstrates that IPS cells have the same potential for therapy as embryonic stem cells, without the ethical and practical issues raised in creating embryonic stem cells,” says Jaenisch.

While IPS cells offer tremendous promise for regenerative medicine, scientists caution that major challenges must be overcome before medical applications can be considered. First among these is to find a better delivery system, since retroviruses bring other changes to the genome that are far too random to let loose in humans. “We need a delivery system that doesn’t integrate itself into the genome,” says Hanna. “Retroviruses can disrupt genes that should not be disrupted or activate genes that should not be activated.”

Potential alternatives include other forms of viruses, synthesized

versions of the proteins created by the four master regulator genes that are modified to enter the cell nucleus, and small molecules, Hanna says.

Despite the rapid progress being made with IPS cells, Jaenisch emphasizes that this field is very young, and that it's critical to continue full research on embryonic stem cells as well. "We wouldn't have known anything about IPS cells if we hadn't worked with embryonic stem cells," says Jaenisch. "For the foreseeable future, there will remain a continued need for embryonic stem cells as the crucial assessment tool for measuring the therapeutic potential of IPS cells."

Source: Whitehead Institute for Biomedical Research