CALIFORNIA INSTITUTE FOR REGENERATIVE MEDICINE

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PRESIDENT'S UPDATE ON ADVANCES IN STEM CELL SCIENCE

Highlights of recently published papers from CIRM grantees and other leading research teams around the world—October 2012

Eggs Created from Stem Cells Produced Viable Offspring

A team led by Mitinori Saitou at Kyoto University used mouse embryonic stem cells and mouse iPS cells to create viable eggs that produced healthy offspring that were also fertile and able to reproduce. The work was published online in *Science xpress* October 4.

The same Japanese team created sperm from stem cells last year. Creating functionally mature eggs required a much more complex process. The procedure that succeeded may not be reproducible in human cells. Like with sperm, the team first coaxed the stem cells into becoming early precursors of the desired germ cells using transgenes for the germ line lineage, in this case, precursors of oocytes, or eggs. But to get them to mature they had to culture them with already differentiated (somatic) ovary non-germ cells from embryonic ovarian tissue, which could limit the procedure's application in humans. But getting viable eggs required two more steps. After the cells had formed ovary-like structures in the dish, they were implanted in mice to further mature for a month. Then they were removed and fertilized in the lab before being implanted in female mice where they gave rise to normal-appearing mouse pups that themselves were fertile.

While there are many very high hurdles before a similar procedure could be used to assist infertile couples, it immediately opens up numerous research opportunities to start to unlock the many mysteries that surround normal egg development and maturation. This window into basic understanding could provide opportunities to aid infertile couples long before the stem-cell-to-egg procedure is perfected.

Swapping DNA Corrects Genetic Defect in Eggs

Researchers at the Oregon National Primate Research Center have taken human eggs that had defects in their mitochondrial DNA and removed the DNA from their nucleus and placed it in eggs with healthy mitochondrial DNA. They then fertilized the eggs and created viable stem cells lines from the resulting embryos in the work published in *Nature* online October 24.

In the current study, the team led by Shoukhrat Mitalipov used human eggs, which could not be ethically implanted in a human, before conducting further research on their safety. But three years ago, the team had conducted similar work with monkeys and when the modified eggs were implanted they produced healthy monkeys. In the current paper, they note that those young monkeys seem to be developing normally.

Mitochondria are the powerhouse mini organelles that provide energy for our cells. Because they reside in the inner cell outside of the nucleus where our chromosomes reside, we inherit mitochondrial DNA only from our mothers. We get copies of what she has in her egg and if she has one of the rare diseases linked to defects in the DNA in mitochondria, we get it, too.

One of the various mitochondria-linked diseases occurs in about one in 5,000 births. This new work theoretically gives women who know they carry one of the diseases the opportunity to avoid passing it down to the next generation. Mitalipov has been quoted saying that while the procedure needs to be tweaked to improve its efficiency, he expects the first child to be born using the method within three years.

For the research in *Nature* the team collected 97 eggs from volunteers. They made the DNA swap in 64 and left 33 for controls. After injecting sperm, 19 of the 64 manipulated eggs developed into early stage embryos, or blastocysts. By contrast, half of the control eggs developed normally. The team did see some problems with achieving fertilization in the manipulated eggs, but they said the success rate was high enough to result in a pregnancy in most egg harvest cycles.

Another Functional Tissue Formed from Stem Cells, This Time Thyroid

Researchers at Universite' Libre de Bruxelles led by Sabine Costagliola have coaxed mouse embryonic stem cells to become thyroid cells that were able to self-organize into the three-dimensional shape required to produce thyroid hormones. The work was published on line October 11 in *Nature.*

Costagliola's team genetically manipulated the embryonic stem cells so that they would express two genes that are never turned on at the same time except in the thyroid. This method efficiently drove the cells to become thyroid cells that the team expected to study in the dish. But to their surprise, those cells then formed the hollow spheres capable of trapping iodine that is needed for making thyroid hormones. When those spherical follicles were implanted in mice, eight out of nine mice that had previously had their thyroids destroyed, were fully rescued and showed normal levels of thyroid hormones in their blood.

One out of 3,000 humans is born with improperly functioning thyroid glands and face a lifetime of hormone replacement therapy that has to be constantly monitored and tweaked. This work suggests that stem cells, particularly genetically matched iPS cells made from the patients own cells, could provide a one-time permanent replacement for the hormones that are critical for regulating growth and metabolism.

Transplanted Neurons Survive in Sufficient Numbers for Therapy

A CIRM-funded team at UCSF led by Arturo Alvarez-Buylla has found that the key neurons that balance intercellular cross talk in the brain can be grown from stem cells and survive in the brain in sufficient numbers to be therapeutic. Their results from working with mouse stem cells were published online October 12 in *Nature*.

Brain cells called GABA-secreting interneurons balance the impact of the brain's excitatory neurons. The cellular changes seen in a number of diseases including epilepsy, Alzheimer's, Huntington's and Parkinson's are marked by disruptions in this balance. Brains of patients with those same diseases have been shown to have problems in the proper functioning of their GABA neurons. So, although it makes sense that transplanting healthy BAGA neurons could help these patients, the prevailing theory was that not enough of the new neurons would survive to be beneficial.

Most in the field had thought that the brain only had room for so many interneurons and transplanted healthy neurons would have to compete with the defective native neurons for space. But the UCSF team found that no matter how many new neurons they implanted, the same proportion always survived, suggesting it will be easier than folks thought to get enough nerves to survive to potentially impact and correct patients' disease.

Progeny of Brain Stem Cells Help Manage Actions of Other Brain Cells

A CIRM-funded team at Stanford led by Tony Wyss-Coray has found that the daughters of neural stem cells, neural progenitor cells, secrete proteins that help to orchestrate the actions of other cells in the brain. The paper was published in the November issue of *Nature Neuroscience* Vol 15(11).

The proteins identified by the team seem to have a particular influence on the actions of brain cells called microglia. Those cells serve as the brain's immune system, clearing pathogens, but also cleaning up cells damaged from disease or injury. Microglia seem to have a role in many neurodegenerative diseases either through over activity damaging good neurons, or failure to clean up and remove damaged cells. The neural progenitor cells (NPCs) seems to be able to issue orders that set microglia on a proper course of action.

This finding may seem like just a brick in the basic biology of understanding how our brain cells work together. But the findings actually provide evidence for why neural stem cells or NPCs seem to speed recovery from stroke in animal models even though the cells don't tend to engraft and make connections to native tissue. Knowing this method of action can be critical in convincing the Food and Drug Administration to allowing clinical testing of these cells in patients.