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—April 2012

Cells Converted Directly to Muscle In Mice Reverse Heart Damage

CIRM-funded research in Deepak Srivastava's lab at the Gladstone Institutes has shown that the fibroblast connective tissue in the heart that would normally form scar tissue after a heart attack can be directly reprogrammed to become beating heart muscle instead. The work was published online April 18 in *Nature.*

Srivastava's team had previously shown that heart fibroblasts could be converted to heart myocytes, or muscle tissue, in the petri dish. They now have shown that by injecting the three genetic factors that are key to embryonic heart development directly into the adult heart, fibroblasts can be reprogrammed to become muscle cells. The team had induced a heart attack-like lesion in the mice earlier and found that heart function improved after three months and that individual cells had properties of heart myocytes.

Developing ways to induce the heart to repair itself rather than having to transplant new tissue, or a whole new heart as is often the only option for many patients today, would be a great boon to the nearly one million Americans who survive a heart attack each year. When the team used the three factors that go by the acronym GMT along with a protein that promotes the growth of new blood vessels, thymosin Beta4, they saw even more dramatic improvement in heart function.

See DeWitt and Trounson: Direct conversion in the heart: a simple twist of fate. The EMBO J, April 2012

Chemical Directs Own Stem Cells to Become Cartilage, Repair Joints

CIRM funded research lead by Peter Schultz at Scripps has shown that a single chemical can induce the body's endogenous, or own, stem cells to become functional cartilage in two different mouse models of osteoarthritis. The April 5 online publication in *Science* included collaborators from Massachusetts General Hospital and the Novartis Research Foundation.

Several clinical studies are underway using mesenchymal stem cell transplants to try to repair joints damaged in osteoarthritis, a condition that impacts 27 million Americans. But like the heart study above, getting the body's existing cells to do the job could have significant advantages. To find their one compound that worked, the team screened 22,000 chemicals using a high throughput lab system that could image changes in mesenchymal stem cells that indicated they were becoming cartilage. When the compound, kartogenin, was injected into the animal's joints once a week for four weeks the team saw cartilage formation and noted improvement in

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the ability of the mice to walk. On the molecular level, they were able to show that kartogenin was able to turn on or turn off the various factors that are known to encourage stem cells to become cartilage instead of other types of tissue.

Method Seems to Solve Scale-up Issue for Most Endodermal Tissues

A team led by Paul Gadue at Children's Hospital of Philadelphia has developed a method to mature pluripotent stem cells into a type of endodermal cell that is able to renew itself on an unlimited basis. The paper published in the April 6 *Cell Stem Cell* Vol. 10 (371-384) cited collaborators from Boston University and the McEwen Centre in Toronto.

It has generally been difficult to mature pluripotent stem cells into adult endodermal tissues—the various tissues of the gut, such as liver, pancreas or intestine—and have the final tissue be fully functional. Most attempts have also produced mixed types of cells and have not been usable for large-scale production. The current team, instead of using a steady sequence of steps to get to the final adult tissue, decided to first make cells that closely resemble the definitive endoderm, the sheet of cells in the early embryo that folds to form the primitive gut tube that all the gut tissues arise from during development. They called these cells Endodermal Progenitors (EP) and developed a cell culture method that would let them keep the cells in this progenitor state indefinitely and multiply their numbers exponentially. They grew their cells through 20 passages and expanded them 16 fold.

However, the cells could readily be instructed to form specific adult tissue. In both cell culture and in immune compromised mice, the human cells were able to mature into liver, pancreas and intestinal tissue that appeared to have appropriate function. They saw pancreatic cells that were responsive to glucose levels, for example. Also important, they found no sign of tumor formation.

Lung Is an Endodermal Tissue that Requires a Different Method

A team led by Darrell Kotton at Boston University has developed a system to derive and grow progenitor cells for lung and thyroid, two tissues that have generally been difficult to study. The team, with collaborators from New York University, the University of Vermont and Mount Sinai School of Medicine published their results in the same issue of *Cell Stem Cell* Vol. 10 (398-411) as the paper above.

Lung and thyroid tissue come from the same definitive endoderm that forms the primitive gut tube in the early embryo that was the focus of the study above. However, lung and thyroid come from the opposite end of that primitive tube from the pancreas, liver and intestine created by the earlier team, of which Kotton was a member. The progenitors for lung and thyroid exist so fleetingly in the embryo that they have been little studied and the growth factors that lead to them have been difficult to detect. Kotton's team used a complex process of elimination to determine which factors were essential, and more important, which factors needed to be turned off to avoid the cells maturing into another type of tissue.

Starting with mouse embryonic stem cells, they were able to derive progenitor cells for both lung and thyroid and were able to seed the lung cells on a scaffold and generate the two types of cells that normally line the air sacs of lungs. This system should help open a door onto the development of this poorly understood vital organ as well as help to model lung diseases and potentially provide replacement cells for therapy.

Lung Model for Cystic Fibrosis also Created

A Harvard team developed a protocol similar to Kotton's team above to derive lung progenitor cells from mouse embryonic stem cells and then adapted that method to develop human lung progenitors from induced Pluripotent Stem Cells (iPSCs) made from cells of a patient with cystic fibrosis. Led by Jayaraj Rajagopal the team's work was featured on the cover of the same April 6 issue of *Cell Stem Cell* Vol. 10 (385-397) as the two studies above.

The team used cells from a patient with the delta-508 mutation, which is responsible for 90 percent of cystic fibrosis (CF) in the U.S. But the cells also contain the G551D mutation, which causes just two percent of CF, but is the only cause of CF for which there is an available drug. This means that as scientists move forward to use these cells to test potential CF drugs, they have a control to determine if the disease-in-a-dish cells are truly behaving like cells would in a human. If the cells respond positively to the existing drug for the rare mutation, there is a better chance that a positive response to a drug for the common mutation will be affirmed in a human clinical trial.

All three of these papers on endodermal tissue have benefited from a trend in the field to go back and look really hard at normal embryonic development and then try to mimic those steps in the lab. In this case the team first created definitive endoderm from pluripotent cells, then converted that into foregut endoderm, then lung endoderm, and finally into lung progenitors.