

Top Research Highlights

Help for Beta Cells: Regeneration, Survival, and Reprogramming

Although the field of beta cell regeneration hardly existed five years ago, it has quickly become one of the more promising areas of type 1 diabetes (T1D) research, due to JDRF support and leadership. During this time, JDRF has funded a number of scientists, each of whom is continuously refining our knowledge of how beta cells grow, divide, and function in the body. While this is still an emerging area of research, through the continued support of JDRF, these findings are opening up entirely new directions that may lead to the discovery of new therapies for T1D. And JDRF's support is advancing the translation of basic research by encouraging or helping form collaborations between academic and industry partners.

Within the field of beta cell regeneration, there are a number of potential therapeutic strategies. One area of research focuses on stimulating the production of new beta cells. A second area focuses on promoting beta cell survival via: keeping existing beta cells healthy; slowing or preventing the progression of T1D in people recently diagnosed with the disease; or protecting newly regenerated or transplanted beta cells in people with established disease. A third area focuses on converting other types of cells in the pancreas (where beta cells reside), or other cell types located elsewhere in the body into new,

functioning beta cells—an approach called “reprogramming.”

These therapeutic strategies for beta cell regeneration survival, and reprogramming could—in conjunction with other therapies that halt the immune attack that destroys beta cells—help to provide a cure for a person with T1D.

In this issue, we focus on four recent advances in research aimed at discovering new approaches for beta cell regeneration, survival, and reprogramming. These include new insights on how to make aging beta cells act young again, drug targets to promote beta cell regeneration and survival, and promising results for converting other cell types into beta cells. These developments highlight our commitment to funding the brightest scientists to address some of the most challenging problems in T1D research.

The following is a “Pocket Guide” to help familiarize you with terms that appear repeatedly in this issue:

- **Biomarker (biological marker):** an indicator of a biological state—a substance or characteristic that is objectively measured and evaluated and can serve as an indicator of a normal biological process, a disease-causing process, or a response to a drug
 - **Gene:** the part of DNA that contains blueprints for building proteins. Each human cell contains the same set of approximately 30,000 genes on a DNA molecule.
 - **Nucleus:** the command center of the cell, which stores DNA and coordinates the cell's activities, including cell division
 - **Receptor:** a protein that sits on a cell's surface or within a cell and receives information that the cell needs in order to function
 - **Reprogramming:** a process by which one type of cell in the body is converted into a different type of cell that takes on the function of the new cell, for example, converting an alpha cell in the pancreas into a beta cell
- **Beta Cell Proliferation:** a process by which beta cells multiply to form new, functional beta cells
 - **Beta Cell Regeneration:** a process by which the body restores beta cells lost to disease or injury, or increases beta cell number in response to increased metabolic demands such as pregnancy, obesity, or growth

We hope you enjoy this special issue.

Researchers Make Old Beta Cells Act Young Again

As children age, they get better at doing a lot of things. But growing new beta cells isn't one of them. In fact, by the time children reach the age of 10 to 12 years, their beta cells have largely lost their ability to divide and make new cells. Now, JDRF-funded researchers at Stanford University have identified a pathway responsible for this age-related decline in beta cell production and have shown that they can coax older beta cells into dividing as frequently as they did when they were younger.

The work, which was published last month as an advance online publication in *Nature*, provides the most complete picture to date of the molecular gears and levers that bring beta cell regeneration to a near halt. The finding not only paves a path forward for developing strategies to treat type 1 and type 2 diabetes, it also has broader implications for treating aging in general.

"The ability to regenerate damaged or lost tissue declines with age in many tissues in the body, not only in beta cells," says Seung Kim, M.D., Ph.D., who led the study. "So this is not just a diabetes-specific result—it is part of a greater story of how to treat aging and make older tissues act young again."

In their work, the researchers systematically tracked the activity of a protein called platelet-derived growth factor (PDGF) across different ages in mice. In young mice, they found that PDGF normally docks on its receptor on the surface of the beta cell and sends a series of signals via the so-called "PDGF pathway" to the cell's nucleus. These signals instruct beta cells to divide, or form new beta cells. However, the researchers found that, as the mice grew older, the number of PDGF receptors on their beta cells

declined or reduced with age. It turns out that without its receptor, PDGF can't dock and activate the PDGF pathway, and beta cell proliferation is not set in motion.

To further test whether the reduced number of PDGF receptors is behind beta cells' waning ability to proliferate as they age, the researchers artificially increased the number of PDGF receptors on older beta cells to see if they could restore the cells' ability to divide and generate new cells. Not only did older beta cells begin to divide in living mice in response to PDGF, but the researchers also observed similar results in human beta cells treated in a Petri dish, suggesting that a drug developed for mice may also work for humans.

"What is exciting about our work is that with the advent of new genetic techniques and the completion of the Human Genome Project, we were able to identify a protein within the beta cell that we can manipulate to get older beta cells to grow and proliferate in a desirable, controlled manner," says Dr. Kim, who is also a Howard Hughes Medical Institute investigator.

However, it wasn't always so easy. In the past, researchers have used other techniques to trigger older beta cells to start dividing, but they have been met with consistently frustrating results. "If you tweak other proteins in the cell, a different picture emerges," says Dr. Kim. "You can get these cells to grow, but they will literally lose their identity. They will either stop making insulin, or they'll grow just fine but they will grow uncontrollably or into other cell types."

By better understanding the mechanisms that control and govern how beta cells divide and proliferate, researchers could potentially transform treatments for diabetes, says Patricia Kilian, Ph.D., JDRF's scientific program

director of regeneration research. The cascade of events leading from the beta cell's surface to its nucleus could inspire scientists with new ideas on how to develop drugs that safely promote the regeneration of beta cells to replace those lost in T1D.

Key Point: *Scientists have identified a pathway that allows beta cells to divide when they are young, but which turns off as the cells age. Finding drugs that could activate this pathway may provide a new way to make older beta cells divide again. This may be part of a strategy to maintain a quantity of beta cells that is sufficient to restore insulin production in T1D.*

A Well-Known Hormone Turns On Beta Cell Survival Genes

The key job of a beta cell is to respond to blood glucose levels to trigger insulin secretion. However, when things go awry as they do in T1D, and glucose levels are too high or elevated, the remaining beta cells can become "overworked." What results is stress on the beta cell and the potential to form harmful particles known as free radicals—events that can result in beta cell death. A team of researchers at the Salk Institute for Biological Studies in La Jolla, CA, has now figured out how a well-known hormone can protect beta cells from the harmful conditions associated with T1D and prolong their survival. The work holds promise for several areas of T1D research, from improving drugs that stimulate beta cell survival to devising new antirejection drugs for transplants.

Researchers and pharmaceutical companies have long been interested in a hormone known as glucagon-like peptide-1 (GLP-1), because of its ability to make beta cells function better overall. In animal studies, not

only can it enhance glucose-stimulated insulin secretion by beta cells, it can also enhance beta cell survival and proliferation.

However, until now, it wasn't known in detail how GLP-1 actually works, mechanistically, to help regenerate beta cells in animals. The point is important to understand, since in people with type 2 diabetes, drugs that are forms of GLP-1 are used because of their ability to enhance insulin secretion, which results in improved blood glucose levels. However, an effect on beta cell regeneration in humans has not been demonstrated conclusively.

The new research findings provide a path for improving GLP-1 drugs to have greater effect on human beta cell survival and regeneration for both type 1 and type 2 diabetes. The study reveals the series of events that is set in motion inside the beta cell when GLP-1 latches onto its receptor on the cell's surface.

In their work, lead researcher Marc Montminy, M.D., Ph.D., and his team treated beta cells with GLP-1 and then tracked the activity of the cells' genes. To their surprise, they found that GLP-1 activates not one set of genes, as they previously thought, but two sets. The first set of genes becomes active shortly after GLP-1 docks onto its receptor. The second set of genes is activated hours after the first set turns off. This second set of genes is very interesting, because it is activated by hypoxia inducible factor 1 α (HIF-1 α), a genetic switch that is linked to cell survival.

"The HIF switch was previously known to help beta cells survive, but this is the first time that it has ever been linked to GLP-1," says Dr. Montminy. "That's the real accomplishment—it's a connection that wasn't evident before."

The work may help improve the effects of a currently available drug called exendin-4, which is prescribed to people with type 2 diabetes to help their beta cells secrete insulin. The drug is a form of GLP-1.

Since activating HIF-1 α may help beta cells survive, this work also has implications for islet transplantation. When islets are isolated outside of the body, they become less robust, so when they are transplanted back into the body, they need time to become healthy and strong again. Activating HIF-1 α before islets are transplanted could help ensure that they flourish in their new home.

"These new insights could help research improve drugs that are used to treat type 1 and type 2 diabetes," says Andrew Rakeman, Ph.D., senior program manager of beta cell therapies at JDRF. "By better understanding the mechanisms by which drugs like exendin-4 work, we have new strategies for prolonging the benefits that these drugs have on beta cell survival, and potentially for preventing beta cell loss."

Key Point: *A hormone called GLP-1 has long been thought to have a positive impact on beta cell survival, and now scientists have started to uncover how it works. The new findings provide scientists with insights to improve the effects of currently available drugs that may prevent beta cell loss and promote beta cell survival in people with T1D or in those in the transplant setting.*

New Drug Target and Potential Biomarker for Beta Cell Regeneration

One of the most important goals of T1D research is to discover molecules that stimulate the growth of beta cells.

Another goal: to find drugs that target those molecules. Now, JDRF-funded researchers, in collaboration with the pharmaceutical company Hoffmann-La Roche, have done both, discovering not only a protein that regulates beta cell growth, but also a drug that stimulates it.

The discovery, led by Markus Stoffel, M.D., Ph.D., a professor at the Swiss Federal Institute of Technology Zurich, represents a significant advance in identifying a new drug target for beta cell regeneration and potentially new biomarkers that can track the effectiveness of such a drug.

The work builds on a discovery made five years ago, when Dr. Stoffel and his team first showed that a once-obscure protein called transmembrane protein 27 (Tmem27) sits on the outer surface of beta cells. At the time, they found that the more Tmem27 was present on beta cells, the more beta cells were present in mice. They also found that when Tmem27 was snipped in two, the protein was completely inactivated, and fewer beta cells were present in the mice.

In other words, when Tmem27 is whole or intact, beta cells proliferate. When Tmem27 is cut in two, they do not.

"We hypothesized that if we could prevent Tmem27 from being snipped, we could get more beta cells to grow and proliferate," says Dr. Stoffel, who is also a 2010 recipient of JDRF's Gerold & Kayla Grodsky Basic Research Scientist Award. "This observation gave us the rationale to look for molecules that snip, and thus inactivate, Tmem27."

After screening thousands of possible culprits, Dr. Stoffel and his team found the molecular scissors they were looking for: beta-site APP-cleaving enzyme 2 (Bace2), which cuts Tmem27 at the exact same

spot on the protein every time. To confirm the role of Tmem27, mice were genetically engineered such that they lacked the molecular scissors. These mice were found to have larger islets and higher numbers of beta cells within those islets. Not only were there improvements in beta cell size and function, but the genetically engineered mice lacking the Bace2 gene were able to metabolize and clear glucose from the blood more efficiently than control mice with Bace2. These findings suggest that an inhibitor of Bace2 might be useful for promoting beta cell regeneration.

Having found the molecule responsible for inactivating Tmem27, Dr. Stoffel and his team wanted to find compounds to inhibit the enzyme to prevent Tmem27 cleavage. They worked with scientists at Hoffmann-La Roche to investigate chemical compounds that would inhibit Bace2. They found inhibitory compounds and showed that, when given to mice, those compounds inhibited Bace2 as expected and stimulated the proliferation of new beta cells.

In addition to identifying a new drug target for promoting beta cell regeneration, Dr. Stoffel's research may also provide the basis for a new biomarker for monitoring the effectiveness of T1D treatments. Because Tmem27 floats into the bloodstream when it is clipped, scientists could develop a test to measure the number of Tmem27 fragments floating around the blood, and use this test to gauge the number of beta cells in the body.

"This is an exciting and potentially impactful finding," says Patricia Kilian, Ph.D., scientific program director of regeneration research at JDRF. "It's an example of how researchers make an early observation and follow up on

it, and then take it to the next level, where it has translational potential—the potential to be developed into a drug that promotes the growth of beta cells for diabetes."

Key Point: *An important protein involved in beta cell growth is snipped in T1D. Scientists have now identified the culprit—a molecular scissors called Bace2—and identified a chemical compound that inactivates it in mice. This inhibitor renews beta cell growth and could potentially lead to new strategies to promote beta cell regeneration to treat T1D. This is a key example of how science can advance when academic and industry scientists work together.*

New Findings about the Transformers: Alpha Cells

Alpha cells and beta cells are the yin and yang of the pancreas. Whereas beta cells secrete the hormone insulin and lower blood glucose levels, alpha cells secrete the hormone glucagon, which raises them. Normally, the two work in tandem to regulate blood glucose levels and keep them in check. So what happens when nearly all of the alpha cells are destroyed? Surprisingly little, according to new research from the University of Geneva in Switzerland.

Led by Pedro Herrera, Ph.D., the researchers found that when they destroyed nearly all the alpha cells in the pancreas of a mouse, the body was able to maintain tight glucose control nearly as well as in a mouse with a full set of alpha cells. The results show that alpha cells are largely dispensable for maintaining tight glucose control and are, for the most part, not needed for beta cells to function.

"And that's really good news," says Dr. Herrera, the 2011 recipient of JDRF's

Gerold & Kayla Grodsky Basic Research Scientist Award. "Because now we know that if we can come up with some treatment that could convert almost all alpha cells to beta cells, it wouldn't disrupt the body's ability to maintain tight glucose control."

In 2010, Dr. Herrera and his team made a breakthrough in the field of beta cell regeneration. They showed that when they destroyed all of the beta cells in the pancreas, approximately 5 to 10 percent of the glucagon-producing alpha cells began spontaneously converting into insulin-producing beta cells, in an apparent attempt to restore balance between these two important cell types.

Even though only a small number of alpha cells converted into beta cells, Dr. Herrera and his team saw it as an opportunity. If the body is able to spontaneously convert alpha cells into beta cells, then perhaps there are opportunities to design drugs that mimic this effect and "reprogram" alpha cells to convert them to beta cells. If so, scientists could try to exploit those mechanisms to reprogram a large portion of alpha cells into beta cells—enough to restore glucose control in people with T1D.

But before they could take on that feat, the researchers first had to make sure that in fixing one problem, they did not introduce another. That is, they had to first see how the body responded when nearly all its alpha cells in the pancreas were depleted.

Amazingly, with 98 percent of the alpha cells destroyed, the few remaining alpha cells were able to keep up with the entire workload of secreting glucagon when glucose levels were low. Furthermore, the body's response to high levels of blood glucose remained unaffected, showing that a loss of alpha cells didn't change



the way that beta cells perform, either.

“This work suggests that if you switch most of the alpha cells in your body into beta cells, you will still be able to make enough of the hormone glucagon,” says Andrew Rakeman, Ph.D., senior program manager of beta cell therapies at JDRF. “It’s an important advance because it provides further support that a therapeutic strategy for converting alpha cells to beta cells is feasible.”

Key Point: *Alpha-to-beta cell conversion or “reprogramming” is an early, but promising, therapeutic strategy for T1D. Scientists have now shown that the body can function normally without most of its alpha cells, addressing a potential safety concern for alpha-to-beta cell conversion.*