In people with type 1 diabetes, insulin-producing β (beta) cells in the pancreas are destroyed. Without insulin, the tissues of the body cannot absorb or use glucose (sugar), the major cellular fuel. Scientists are therefore pursuing strategies to replace the destroyed β cells and restore insulin production, including by transplantation of β cells produced in the laboratory. In research described in this chapter, scientists generated functional β cells in the laboratory from the skin cells of people with type 1 diabetes by first reprogramming the skin cells to become stem cells, and then coaxing the stem cells to become β cells.

This image depicts a mixed cell population composed of mature stem cell-derived, insulin-producing β cells (stained green or both green and red), cells that may be on their way to become insulin-producing β cells (stained red only), and cells that do not belong to the β cell lineage (stained blue). While many research questions remain before a stem cell-derived β cell transplant procedure will be ready for testing in humans, this advance is a significant step forward toward developing cell replacement therapy for type 1 diabetes.

*Image courtesy of Dr. Douglas Melton, Harvard University, and Dr. Jeffrey Millman, Washington University. Originally published in Millman JR, Xie C, Van Dervort A, Gürtler M, Pagliuca FW, Melton DA, Nat Commun 7: 11463, 2016, available at: [www.nature.com/articles/ncomms11463](http://www.nature.com/articles/ncomms11463)*. Image copyright 2016 from Millman, et al., and reprinted under a Creative Commons Attribution License ([https://creativecommons.org/licenses/by/4.0/](https://creativecommons.org/licenses/by/4.0/)).
Diabetes, Endocrinology, and Metabolic Diseases

NIDDK support of basic and clinical research in the areas of diabetes, endocrinology, and metabolic diseases spans a vast and diverse range of diseases and conditions, including diabetes, osteoporosis, cystic fibrosis, and obesity. Together, these diseases and conditions affect many millions of Americans and can profoundly decrease quality of life. Many of these diseases are complex—an interplay between genetic and environmental factors contributes to disease development.

Diabetes is a debilitating disease that affects an estimated 29.1 million people in the United States—or 9.3 percent of the total population—and is the seventh leading cause of death.¹ Compared with people of similar age without the disease, overall rates of death are about 1.5 times higher in people with diabetes, and rates of death from cardiovascular disease are 1.7 times higher.¹ Although rates of diabetes-related complications have declined substantially in the past two decades, and the rate of new cases of diagnosed diabetes has begun to fall, disease burden remains significant as the number of people with diabetes is still very high.²,³ Diabetes can affect many parts of the body and is associated with serious complications, such as heart disease and stroke, blindness, kidney failure, and lower-limb amputation. In addition to these human costs, the estimated total financial cost for diabetes in the United States in 2012—including costs of medical care, disability, and premature death—was $245 billion.⁴ Effective therapy can prevent or delay diabetic complications, but up to one-quarter of Americans with diabetes are undiagnosed and therefore not receiving therapy.³

Diabetes is characterized by the body’s inability to produce and/or respond appropriately to insulin, a hormone that is necessary for the body to absorb and use glucose (sugar) as a cellular fuel. These defects result in persistent elevation of blood glucose levels and other metabolic abnormalities, which in turn lead to the development of disease complications. The most common forms of diabetes are type 1 diabetes, in which the body loses its ability to produce insulin; and type 2 diabetes, in which the body becomes resistant to insulin signaling, with subsequent impaired insulin production. In addition, a significant proportion of pregnant women each year are diagnosed with gestational diabetes, a form of diabetes that is similar to type 2 diabetes but unique to pregnancy. Untreated, any form of diabetes during pregnancy increases the risk of serious complications for the mother and baby before, during, and after delivery.

Type 1 diabetes, formerly known as juvenile diabetes, affects approximately 5 percent of diagnosed diabetes cases in adults, and the majority of diagnosed cases in children and youth.¹ It most often develops during childhood but may appear at any age. Type 1 diabetes is an autoimmune disease in which the immune system launches a misguided attack and destroys the insulin-producing β (beta) cells of the pancreas. If left untreated, type 1 diabetes results in death from starvation: without insulin, glucose is not transported from the bloodstream into the body’s cells, where it is needed. Thus, people with type 1 diabetes require lifelong insulin administration—in the form of multiple daily injections or via an insulin pump—to regulate their blood glucose levels. The NIDDK's landmark Diabetes Control and Complications Trial (DCCT)/Epidemiology of Diabetes Interventions and Complications (EDIC) study demonstrated that keeping blood glucose levels as near to normal as safely possible reduced the risk of

eye, kidney, nerve, and heart complications associated with type 1 diabetes. However, despite vigilance in disease management, with current technologies to test blood glucose levels and administer insulin, it is still not possible for people with type 1 diabetes to control blood glucose levels as well as functional pancreatic β cells do. Thus, researchers are actively seeking new methods to improve blood glucose monitoring and insulin delivery. In this regard, a milestone was achieved this past year when the U.S. Food and Drug Administration approved the first commercial “hybrid artificial pancreas” device that automatically links glucose monitoring and insulin delivery. The NIDDK supported early research that contributed to the development of the approved device and continues to support research to test and improve artificial pancreas technologies. Researchers are also working to develop β cell replacement therapies, such as islet transplantation, to cure type 1 diabetes.

Type 2 diabetes is the most common form of the disease, accounting for about 90 to 95 percent of diagnosed diabetes cases in U.S. adults. The risk for developing type 2 diabetes is associated with older age, obesity, family history of diabetes, history of gestational diabetes, impaired glucose metabolism, physical inactivity, and race/ethnicity. Type 2 diabetes occurs at higher rates among racial and ethnic minority populations in the United States, including African Americans, Hispanic and Latino Americans, American Indians, some Asian Americans, and Native Hawaiians and Pacific Islanders. Gestational diabetes is also a risk factor: about half of women with gestational diabetes will develop type 2 diabetes within 5 to 10 years after giving birth.

In people with type 2 diabetes, cells in muscle, fat, and liver tissue do not respond to insulin properly. As a result, the pancreas initially produces more insulin to compensate. Gradually, however, the pancreatic β cells lose their ability to secrete enough insulin to restore balance, and the timing of insulin secretion becomes abnormal, causing blood glucose levels to rise. Treatment approaches for controlling glucose levels include diet, exercise, and oral and injected medications, with insulin often required as the disease progresses. There are also an estimated 86 million U.S. adults who have a condition called “prediabetes,” in which blood glucose levels are higher than normal but not as high as in diabetes. This population is at elevated risk of developing type 2 diabetes. Fortunately, the NIDDK-supported Diabetes Prevention Program (DPP) clinical trial has shown that people with prediabetes can dramatically reduce their risk of developing type 2 diabetes with diet and exercise changes designed to achieve a 7 percent reduction in body weight. To a more limited degree, the safe and well-tolerated drug metformin can also help prevent or delay type 2 diabetes. Moreover, follow-up research has shown that the benefits of reduced diabetes risk from weight loss or metformin can persist for at least 15 years.

Type 2 diabetes was previously called “adult-onset” diabetes because it is predominantly diagnosed in older individuals. However, this form of diabetes is increasingly being diagnosed in children and adolescents, and it disproportionately affects youth from racial and ethnic minority populations in the United States. Believed to be related to increasing rates of pediatric obesity, this is an alarming trend for many reasons. For example, the NIDDK-supported Treatment Options for type 2 Diabetes in Adolescents and Youth (TODAY) clinical trial showed that the disease may be more aggressive and difficult to treat in youth compared to adults. This is worrisome because the onset and severity of disease complications correlate with both the duration of diabetes and control of blood glucose levels; thus, those with early disease onset are at greater risk with respect to complications than those who develop the disease later in life. In addition, increasing rates of type 2 diabetes in girls may lead to more women who enter pregnancy with diabetes, and maternal diabetes during pregnancy—either onset of type 2 diabetes before pregnancy or the development of gestational diabetes during pregnancy—confers an increased risk of type 2 diabetes in offspring. Thus, the rising rates of diabetes and prediabetes in young women could lead to a cycle of ever-growing rates of diabetes. Therefore, the advent of type 2 diabetes in youth has the potential to worsen the enormous health burden that diabetes already places on the United States.

The NIDDK is supporting research to better understand metabolism and the mechanisms that lead to the development and progression

\[ ^{5} \text{Kim C, et al. Diabetes Care 25: 1862-1868, 2002.} \]
of diabetes and the many other endocrine and metabolic diseases within the NIDDK’s mission; such research will ultimately spur the design of potential new intervention strategies. In parallel, based on knowledge from past scientific research investments, the NIDDK is vigorously pursuing studies of prevention and treatment approaches for these diseases.

**TYPE 1 DIABETES—HEALTH BENEFITS OF GOOD GLUCOSE MANAGEMENT**

**Intensive Blood Glucose Management for Those with Type 1 Diabetes Preserves Heart Health and Reduces Risk of Early Mortality:** A long-term NIDDK study reports that keeping blood glucose (sugar) as close to normal as possible for an average of 6.5 years early in the course of type 1 diabetes reduces cardiovascular (heart and blood vessel) disease and can reduce mortality to rates close to those seen in people of similar age in the general population. The landmark Diabetes Control and Complications Trial (DCCT) began in 1983. The DCCT randomly assigned half its participants to an intensive blood glucose management regimen designed to keep blood glucose levels as close to normal as safely possible, and half to the less intensive conventional treatment at the time. When DCCT ended in 1993, it was clear that intensive management had significantly reduced eye, nerve, and kidney complications, but at that time the participants were too young to determine their rates of cardiovascular disease. All DCCT participants were taught the intensive management regimen and invited to join the Epidemiology of Diabetes Interventions and Complications (EDIC) study. EDIC continued to monitor participants’ health, and overall blood glucose management has since been similar in both DCCT treatment groups.

To study the long-term effects of the different treatments tested in the DCCT, researchers examined differences in cardiovascular problems, which can take many years to develop, between the former intensive and conventional treatment groups. After an impressive average 30-year follow-up, DCCT/EDIC researchers found that those who practiced intensive blood glucose management during the DCCT still had significantly reduced cardiovascular disease compared to those who did not, despite having similar blood glucose management for 20 years after the DCCT ended. Compared to the former conventional treatment group, the former intensive management group had a 30-percent reduced incidence of cardiovascular disease and 32 percent fewer major cardiovascular events (such as non-fatal heart attack, stroke, or death from cardiovascular disease) after 30 years of follow-up. These results were similar for both men and women who participated in the studies. However, the beneficial effects of intensively managing blood glucose during the DCCT appeared to be wearing off over time. For example, after 20 years of follow-up, DCCT/EDIC researchers reported that the former intensive treatment group had a 42-percent reduced risk of cardiovascular disease compared to the former conventional treatment group. After 30 years of follow-up, that number had fallen to 30 percent. Even with this reduction in protection, these new data show that a finite period of near-normal blood glucose management early in the course of type 1 diabetes can have beneficial effects on cardiovascular health for up to 30 years.

Historically, those with type 1 diabetes have had a higher mortality rate than the general population. Previous DCCT/EDIC analyses compared intensive versus conventional blood glucose management and showed that those in the former intensive treatment group had reduced mortality compared with that of the former conventional treatment group. Now, mortality in the DCCT/EDIC study from its inception through 2014 was compared to 2013 national mortality data. Researchers found that overall mortality when both DCCT/EDIC treatment groups were combined was no greater than what would be expected in the general U.S. population. However, they found that the mortality rate in the former conventional treatment group was 31 percent higher than that seen in the general population. While the former intensive treatment group’s mortality rate was below that in the general population, the difference was not statistically significant. Researchers also found participants’ long-term blood glucose control affected mortality rates, and those who had worse control had correspondingly worse mortality rates. This effect of blood glucose control on lifespan was more pronounced among women than among men. In general, these results suggest that the increased mortality historically seen in those with type 1 diabetes can be reduced or eliminated through careful management of blood glucose.
Overall, these findings add to DCCT/EDIC’s decades of evidence demonstrating how people with type 1 diabetes can dramatically increase their chances of living long, healthy lives by practicing early, intensive blood glucose management.


**BETA CELLS AND DIABETES**

**Making Beta Cells from People with Type 1 Diabetes:** Scientists generated functional β (beta) cells from skin cells of people with type 1 diabetes. In type 1 diabetes, a misguided attack by the immune system leads to destruction of insulin-producing β cells found in clusters called islets in the pancreas. Although administration of insulin via injections or a pump is life-saving, it does not mimic the exquisite blood glucose (sugar) control of the pancreas. Therefore, scientists are pursuing strategies to replace the destroyed β cells. One way to do that is through islet transplantation—an experimental procedure using islets from a cadaveric donor. The procedure has shown promise for people with difficult-to-control diabetes, but has significant challenges: donor islet tissue is limited, and immunosuppressive medications, which have toxic side effects, are required to prevent rejection of tissue transplanted from another individual. Toward overcoming the first barrier, scientists recently developed a new laboratory production method to make large quantities of β cells—called stem cell-derived β (SC-β) cells—from human stem cells. This method could, with further development, be used to make β cells from a sample of cells from a person with type 1 diabetes in the quantities needed for transplantation back into that same person. These cells would likely require protection from the autoimmune attack, but might not require toxic immunosuppressive medications to prevent rejection of the tissue.

To investigate this possibility, in new research, scientists used skin cells from three people with type 1 diabetes (T1D cells) and three people without diabetes (ND cells). By introducing specific factors into these cells and using the new large-scale production method they developed, they made the skin cells become stem cells—cells that could subsequently become any cell type. They then, by introducing other factors, coaxed these stem cells to become SC-β cells (T1D SC-β cells and ND SC-β cells). Cells from the two different origins showed no differences in the ability to become SC-β cells, indicating for the first time that cells from a person with type 1 diabetes could be used to make SC-β cells.

Next, the scientists demonstrated that the T1D SC-β cells functioned like healthy β cells. For example, in laboratory culture, T1D SC-β cells secreted insulin in response to glucose; they also released insulin in response to diabetes drugs that are known to stimulate insulin secretion, demonstrating their potential for use in screening for new diabetes drugs. The T1D SC-β cells also functioned in live animals: when T1D SC-β cells were transplanted into male mice, they produced insulin in response to glucose and controlled the animals’ blood glucose levels.

Many research questions remain before an SC-β cell transplant procedure will be ready for testing in humans. First, it remains possible that differences between T1D SC-β and ND SC-β cells could appear over a longer time period than in the study. Second, it is not known how the T1D SC-β cells will interact with the recipient’s immune system; for example, it is not yet clear whether these cells could still be rejected, even though they were derived from the recipient’s own cells; and it is likely that these cells would be subject to the same autoimmune attack that destroyed the person’s original β cells. Third, individual differences in type 1 diabetes may affect the production, function, or transplant success of T1D SC-β cells. Thus, further research will illuminate the potential of T1D SC-β cells as a therapy for type 1 diabetes. Nonetheless, these results mark another significant step forward toward a cell therapy for type 1 diabetes, and also provide a valuable resource for drug screening and studying the development of the disease.

New Biomaterial Protects Transplanted Insulin-producing Beta Cells from the Immune System: Scientists have developed a new biomaterial that can protect transplanted β (beta) cells and allow them to function for months in a mouse model of type 1 diabetes without the need for immunosuppression. Transplantation of organs or cells from one person (or animal) to another usually requires the recipients to take immunosuppressive drugs to prevent their immune systems from attacking and rejecting the transplant. Such an immune attack can lead to scar tissue formation (called fibrosis) around the transplant, and eventual death of the transplanted material. Immunosuppressive drugs, however, carry their own serious risks and side effects, and a method to protect transplanted tissues without immunosuppression would greatly benefit people with many diseases. One of these diseases is type 1 diabetes, in which the insulin-producing β cells are destroyed by a misguided immune attack. Transplanting lab-grown β cells into people whose own β cells are not functioning properly is a promising experimental treatment for this disease. Researchers are developing methods for large-scale, laboratory production of β cells that release insulin in response to elevated glucose (sugar) levels. However, the misguided immune response that destroyed the β cells of a person with type 1 diabetes may also attack transplanted β cells. Thus, to realize β cell transplantation’s potential fully, it is important to identify ways to protect the transplanted cells from the host’s immune system without immunosuppression.

One candidate biomaterial being tested for its transplant-protecting properties is a low-toxicity, inexpensive material called alginate. Previous alginate compounds were only able to protect transplanted β cells and enable them to regulate hosts' blood glucose levels for short periods, as the alginate elicited significant immune responses, fibrosis around the transplants, and eventual death of the transplanted cells. Scientists hypothesized that varying the chemistry of alginate might create an alginate variant that could protect transplanted tissues without provoking a strong immune response. To test this idea, the researchers generated a library of chemically altered alginate variants, and evaluation of the library revealed variants that produced substantially reduced immune reactions and fibrosis when tested in rodents and non-human primates. However, could the new biomaterials protect living, transplanted tissue?

To test this, researchers asked whether or not the most promising new alginate variant (called TMTD alginate) could protect lab-grown β cells against immune attack in a male mouse model of type 1 diabetes in which β cells had been destroyed with a chemical. They found that transplanted β cells encapsulated in TMTD alginate caused a weaker immune response and significantly less fibrosis than cells encapsulated in other alginate compounds. The encapsulated β cells were able to respond to and regulate the mice’s blood glucose levels in a normal range. This “cure” of the mice’s diabetes lasted until researchers removed the transplanted cells after 174 days, during which the alginate protected the human β cells from immune system attack without the need for immunosuppression. After 174 days, the TMTD alginate-coated transplant capsules still contained living β cells, still produced insulin, and had caused only minimal fibrosis at the implantation site.

Further research is needed to determine how well TMTD alginate can protect various types of transplanted materials in people. Additionally, more research is needed to determine how well TMTD alginate can protect β cells transplanted into mice or people with type 1 diabetes, where there is an ongoing misguided immune attack against the β cells. Given these hurdles, this new biomaterial’s ability to protect transplanted β cells in mice with chemically induced type 1 diabetes is a significant step forward in developing a long-term cellular therapy for this disease.


Newly Discovered Proteins in Beta Cells Are Targets for Autoimmune Attacks Implicated in Type 1 Diabetes: Researchers have found that immune cells can target naturally occurring fused protein fragments found in β (beta) cells, a discovery that may explain how the type 1 diabetes autoimmune attack is initiated and open up new disease treatment and prevention opportunities. Type 1 diabetes is caused by the immune system launching a misguided attack that destroys the insulin-producing β cells in...
the pancreas. Immune cells called T cells are thought to participate in this attack, and determining what proteins they target—and how the attack might be prevented—is a subject of keen interest. Studies of T cell lines known to attack β cells in mice have identified several relevant T cell targets, including fragments of β cell proteins. However, some of the identified protein fragments are small and only weakly stimulate T cells, suggesting that they are only a part of the T cell’s target.

Because fragments of the insulin protein also can trigger an immune response in type 1 diabetes, researchers hypothesized that “hybrid insulin peptides” (HIPs) made of insulin’s C-peptide fragment fused to another immune-triggering protein fragment might be the T cells’ true β cell target and elicit a stronger—and thus more damaging in people—response. To test this idea, researchers created a library of HIPs and tested their ability to stimulate an immune response in diabetes-causing mouse T cell lines. The researchers found that HIPs made of the C-peptide fragment and either of two naturally occurring fragments of other β cell proteins activated the diabetes-causing T cells over 10,000 times more strongly than the protein fragments alone. This observation supported the idea that T cells target the fusion proteins more strongly than the individual protein fragments. Further experiments determined that one of these HIPs was present in mouse β cell extracts and that HIP-reactive T cells were present in the pancreas and spleen in a female mouse model of type 1 diabetes. Thus, these HIPs are produced and recognized by T cells naturally in mice. The researchers also found T cells that react to similar HIPs (C-peptide fused to two different human β cell protein fragments) in the pancreata of two human males with type 1 diabetes. Whether these HIP-reactive T cells have a role in causing type 1 diabetes still needs to be determined.

Overall, this study has identified a novel class of insulin-fused targets for T cell attacks, which may be critical initiators of β cell destruction. Further research could determine whether these and other fusion proteins mediate the autoimmune attacks that cause type 1 diabetes and other autoimmune diseases, and whether this process can be slowed or halted to prevent disease.

Not All Beta Cells Are Alike—Discovery Helps Explain Altered Insulin Secretion in Type 2 Diabetes:
Researchers have discovered that human pancreatic islets have four separate subtypes of β (beta) cells, and that islets from people with type 2 diabetes have abnormal percentages of the different subtypes. Human islets have long been known to have several distinct cell types, including β cells that release insulin in response to glucose. Until now, all β cells were thought to be alike. However, because previous research hinted at the possibility of functional differences among β cells, scientists sought to determine whether there are distinct types of β cells. To examine this question, the scientists developed novel antibodies—immune proteins that each bind and recognize only very specific structures—that can distinguish between different proteins present on the surface of β cells. This allowed them to sort β cells from islets from male and female donors. In this way, they were able to distinguish four separate β cell subtypes, which they designated β1 through β4, all of which proved able to produce insulin. The percentages of each subtype were similar in islets from 17 donors without diabetes; β1 was generally the most abundant and β4 was typically rarest. It was a much different picture, however, in islets from men and women with type 2 diabetes. For example, in islets from most people with type 2 diabetes, β3 and β4 cells were more abundant, and β1 less abundant, than in people without the disease.

These observations led the researchers to wonder whether an altered distribution of β cell subtypes may contribute to the glucose control problems associated with type 2 diabetes. To address this, they examined whether the newly discovered subtypes functioned differently. Although they found that overall gene expression (how genes are “turned on” or “turned off”) was similar, some genes were indeed expressed at different levels in the subtypes, including genes known to play a role in type 2 diabetes and insulin secretion, suggesting functional differences among the subtypes. Importantly, the scientists also found that insulin secretion in response to glucose differed among the subtypes, with β1 cells being the most glucose responsive. In contrast, β4 cells had the
highest basal insulin secretion rate: that is, they secrete more insulin (although still not a lot) when glucose levels are low and insulin is not needed. These observations suggest that differences in the percentages of \( \beta \) cell subtypes might contribute to the altered timing of insulin secretion and poor glucose control seen in people with type 2 diabetes.

This research has shed important new light on \( \beta \) cell biology, showing that not all \( \beta \) cells are alike and that the distribution of the four newly discovered \( \beta \) cell subtypes is altered in people with type 2 diabetes. Further research is needed to understand the origin of the subtype differences, as well as to determine whether these differences could be capitalized upon for type 2 diabetes treatment.

Type 1 diabetes is characterized by the loss of insulin-producing β (beta) cells in the pancreas. Replacing those lost β cells, which are part of clusters of cells called islets, could improve the health of people with the disease and reduce the significant burden associated with managing the disease. This, therefore, is an important goal of type 1 diabetes research. Decades of research, including by the now-concluded NIDDK-supported Beta Cell Biology Consortium, revealed key insights into how β cells develop and function, laying the path for cell-based therapies. To build on this success, in 2014 the NIDDK launched the Human Islet Research Network (HIRN), a new team-science program to pursue innovative strategies to protect and replace β cells in people with diabetes.

More than 80 scientists with diverse expertise belong to HIRN’s four independent consortia working on different, but complementary, research goals using human cells and tissues. One consortium is focused on discovery of biomarkers of β cell injury that will be important for testing strategies to stop β cell destruction early in the disease process. Another is combining advances in generation of functional human pancreatic β cells with tissue engineering technologies to develop micro-devices that will support functional human islet growth for transplantation. A third is developing approaches to model the interaction of the immune system and β cells in type 1 diabetes, and a fourth is investigating methods to increase or maintain functional β cell mass. A Coordinating Center and Bioinformatics Center aid HIRN investigators in sharing data and resources within the Network as well as with the broad scientific community to facilitate scientific interactions and accelerate research.

HIRN investigators have gotten off to a quick start, and several exciting research advances are described in this publication, including the discovery of four distinct subtypes of β cells and the generation of β cells using stem cells from people with type 1 diabetes. Additionally, HIRN has developed a website (www.hirnetwork.org) to provide detailed information about the Network, its investigators, projects, and progress.

More information about HIRN and other programs supported by the Special Statutory Funding Program for Type 1 Diabetes Research is described in a June 2016 report: http://bit.ly/t1dreport.
COMBATING TYPE 2 DIABETES IN YOUTH

TODAY Study Helps Predict Whose Blood Glucose Will Rise Tomorrow: A new study suggests an approach to help clinicians with adolescent patients who have type 2 diabetes distinguish whose diabetes is likely to remain adequately controlled with standard metformin therapy, and whose will need more careful monitoring and potentially more aggressively stepped-up treatment to stave off rapid disease progression. The Treatment Options for type 2 Diabetes in Adolescents and Youth (TODAY) clinical trial was the first major trial to test approaches to managing type 2 diabetes in the small but rapidly growing number of adolescents and young adults with the disease. The study showed that type 2 diabetes often progresses more rapidly in young people than in middle-aged and older people with the disease. By the end of the 4-year trial, the standard first-line drug metformin was insufficient to control blood glucose (sugar) adequately in about half of the participants. (Adequate blood glucose control was defined by the study as keeping participants’ HbA1c—a measure of long-term blood glucose control—below 8 percent.) Treatment with both metformin and another medication, rosiglitazone, worked somewhat better than metformin alone. However, even this combination of medications failed to maintain adequate glucose control in a high proportion of the adolescents in the study. But interestingly, most of those whose diabetes was not well controlled by metformin (or metformin with rosiglitazone) saw their blood glucose rise outside the prescribed range very early. In fact, metformin had failed to maintain good blood glucose control in about one-fourth of the participants within less than 1 year. After that, fewer and fewer additional participants saw their blood glucose become too high. This suggests that while it is critical to find better means of controlling blood glucose in young people with type 2 diabetes, in many cases (like those whose blood glucose stayed in control for the entire study) the safe, inexpensive drug metformin will actually be sufficient. An important question then, is how can we tell which young people with diabetes are likely to need more aggressive treatment to control their blood glucose?

To address this, the authors of the current study compared characteristics of the group whose blood glucose remained in control during the TODAY trial with characteristics of those in whom the drugs failed. Characteristics like age, race/ethnicity, socioeconomic status, and measures of obesity did not correlate with success or failure of treatment; but one measure that offered quite good predictive power was how well the participants responded to initial treatment with metformin. When TODAY began, study scientists checked to ensure that participants’ diabetes had not already progressed to the point that metformin would be unable to provide adequate glucose control. To find out, they gave metformin for 2 to 6 months before the intervention formally began to all of those interested in joining the trial who otherwise met study requirements. Only those whose HbA1c was below 8 percent during this initial treatment with just metformin were able to participate in the full trial. The researchers of the current study found that for those accepted to participate, HbA1c values during this pre-study metformin-treatment test ranged from near normal (non-diabetic) for some participants to just under the 8 percent cut-off for others. In the new findings, researchers showed that these initial metformin treatment results were very good predictors of whether metformin or metformin plus rosiglitazone would succeed in the longer term: those participants whose HbA1c remained below 6.3 percent during the pre-study metformin test were much more likely to have well-controlled diabetes after 4 years than those whose HbA1c was higher during this initial period. An HbA1c of 6.3 percent is actually considered to be within the non-diabetic range (usually considered to be under 6.5 percent), and is below suggested treatment targets for adults (typically 7 percent), so health care providers may think their adolescent patients are doing very well even if their HbA1c is somewhat higher. However, this study shows that young people for whom metformin alone cannot keep HbA1c below 6.3 percent are at substantially increased risk for rapid type 2 diabetes progression, and should be carefully monitored in case more aggressive treatment is needed to keep blood glucose under control. Further research will be needed to determine whether additional treatment aimed at lowering HbA1c below 6.3 percent would help stave off disease progression in young people for whom metformin alone does not achieve that level of blood glucose control.

SCREENING FOR PREDIABETES

Improving on Methods for Diagnosing Prediabetes in Africans: Research by NIDDK Intramural scientists has shown that it may be possible to improve on a screen for diabetes risk in Africa, a part of the world which is expected to see explosive growth of the disease in the coming decades. Although it was long associated with more prosperous countries, type 2 diabetes is increasingly becoming a problem in low- and middle-income countries, according to the International Diabetes Federation, which also estimates that in sub-Saharan Africa over two-thirds of people with diabetes are unaware they have the disease, the lowest rate of diagnosis anywhere in the world. Therefore, improving diagnosis is vitally important to bring life-saving diabetes treatment and prevention approaches to Africans who urgently need them. The hemoglobin A1c test (often referred to as “HbA1c”) was originally validated as a method for monitoring the effectiveness of diabetes treatment through the NIDDK’s landmark Diabetes Control and Complications Trial. In recent years, it has become accepted also as a means of diagnosing diabetes and prediabetes. Unlike other widely used tests for diagnosing type 2 diabetes and prediabetes, like the fasting plasma glucose and oral glucose tolerance test (OGTT), HbA1c has the advantage that it does not require previous fasting. However, HbA1c is not a perfect diagnostic test. For example, it does not correlate perfectly with average glucose levels: individual genetics have been shown to matter, particularly at near-normal glucose levels. Further, the test has not been validated for diagnosis of prediabetes or diabetes in people of completely African descent. NIDDK scientists reasoned that other tests that also do not require prior fasting—tests of total glycated albumin (GA) or of fructosamine—might potentially perform better than HbA1c for African patients. As a reference standard, they used OGTT, a test that is particularly time and labor intensive because it requires following a person’s response to an orally administered quantity of glucose over a period of hours, but also is most likely to be reliable, because it directly measures the effectiveness of the body’s response to glucose. In a study of 217 male and female African immigrants living in the United States, HbA1c tests correctly detected only half of the prediabetes cases identified by OGTT, while GA and fructosamine identified slightly fewer. However, the study showed that combining GA with HbA1c led to detection of significantly more of the prediabetes cases: 78 percent of those found through OGTT. On the minus side, combining HbA1c and GA tests also increased the number of false positives (i.e., reduced the specificity of the HbA1c test alone). However, it may be more practical to conduct OGTT tests only on individuals who turned up positive via a combined HbA1c/GA test than on a larger fraction of the at-risk population. It is important to remember that no test can determine exactly who will, in the fullness of time, eventually go on to develop type 2 diabetes. But quick and easy blood tests may prove to be a valuable tool for improving prevention, diagnosis, and treatment of type 2 diabetes, so that Africa may avoid some of the ravages of this disease.


GENETICS OF TYPE 2 DIABETES

Genetic Trait in Pima Indians Linked to Increased Birth Weight and Elevated Risk for Type 2 Diabetes: A genetic analysis in Pima Indians of the American Southwest has identified a rare mutation linked to elevated birth weight, that is later associated with higher risk of type 2 diabetes. Pima Indians have among the highest rates of diabetes in the world. To understand their unique genetic risk factors, and find ways to help alleviate this health disparity, NIDDK Intramural researchers examined the DNA sequences in and around a pair of genes thought to be involved in type 2 diabetes pathogenesis in 7,710 Pima study volunteers. They found that 3.3 percent of the participants had a previously uncharacterized variation in the gene ABCC8, which encodes a protein with a key role in regulating insulin secretion. The resulting genetic change—designated R1420H—was similar to known mutations that inactivate ABCC8 and initially cause the pancreas to release more insulin than is needed but, for unknown reasons, later lead to a decline in insulin production, typically followed by type 2 diabetes.

A review of medical records showed that Pima babies who inherited a copy of the R1420H variant from one parent (and a normal copy of ABCC8 from the other parent) tended to have a higher birth weight than siblings born with two normal copies of the gene. This is consistent with the expectation that these babies would be born with an excess of insulin, because insulin is a major growth factor during gestation. Further, the researchers found that as people with a copy of the R1420H version of ABCC8 grow up, they had double
the risk of type 2 diabetes relative to Pima peers with two normal copies of \textit{ABCC8}. This is despite having a lower body mass index (a measure of weight relative to height), on average, than other Pima adults at any given age; and Pima with a copy of R1420H develop the disease, on average, 7 years earlier than Pima with two normal copies of the gene. Why do genetic variants in \textit{ABCC8} initially result in too much insulin, and eventually in too little? As with other mutations in \textit{ABCC8} and its molecular partners, the reason is unclear and may be determined through further research. While rare, R1420H is common enough among the Pima that 1 in 3,600 babies born in the community would be expected to be born with two copies, and have no properly functioning \textit{ABCC8}. One of the participants in the study had this genetic condition, and a review of medical records showed that he was born with severe hypoglycemia (low blood glucose), yet developed diabetes before the age of 4. These new findings could lead to tests to identify newborn Pima babies with two copies of the mutation, allowing early and effective intervention to improve the babies’ health, and reduce their chances of death in infancy. Such a test would also help identify babies born with a single copy of R1420H, individuals who may benefit from keeping a careful eye on their blood glucose as they grow up, to allow timely intervention to prevent or treat type 2 diabetes. In a larger sense, also, the findings build on our knowledge of the genetics of type 2 diabetes. However, since R1420H is found in just 3.3 percent of the Pima population, this study does not resolve the question of what other genetic factors put this group at such a high risk for diabetes: those answers remain to be discovered.


**Variation in a Glucose Transporter Affects Response to the Type 2 Diabetes Drug, Metformin:** New research indicates that a common variation in the gene encoding a protein that allows glucose (sugar) to move in and out of cells has a surprising impact on the effectiveness of the first-line anti-diabetes medication metformin.

Metformin is a very widely used, safe, and helpful treatment for type 2 diabetes, but it is more effective in some people than in others, and scientists are trying to understand why. An international consortium of investigators looked at genomic variation in over 13,000 volunteers of varying ancestry who were taking metformin. They found that a common variation in the gene for a glucose transporter protein, GLUT2, had a significant impact on metformin effectiveness. (The gene encoding GLUT2 is known as \textit{SLC2A2}.) Before treatment, people with two copies of a version of the gene (designated “C”) typically had somewhat worse blood glucose control, as detected by higher levels of HbA1c, a marker for glucose levels. Yet, these individuals had slightly better (lower) HbA1c when taking a standard dose of metformin than did people with two copies of the other version (“T”) of the GLUT2-encoding gene. This effect was most pronounced in people who were obese, but was also seen in those who were not. People with one copy of each version had an intermediate response to metformin. GLUT2 allows glucose to move passively in and out of cells in the liver, an organ with a critical role in regulating blood glucose levels. The GLUT2 that is produced by the C and T versions of the gene is the same, equally capable of allowing glucose movement. However, the researchers found that liver cells with the C version make less GLUT2 than liver cells with the T version. This suggests that in the absence of metformin, individuals with type 2 diabetes and the C version are at a disadvantage compared to those with the T version when it comes to regulating blood glucose levels, but that metformin treatment overcomes and even slightly reverses this effect. Metformin still works in people with two copies of the T version of the gene, but more of the drug—or an additional medication—would be needed to achieve the same degree of HbA1c reduction.

This discovery has broad applicability, because the C and T versions of the gene are both common in a wide variety of racial/ethnic groups, albeit to differing degrees. For example, about 70 percent of African Americans have at least one copy of C, while 24 percent of Latinos do. With further research, tests to reveal a patient’s GLUT2 gene version could one day help further precision medicine by allowing health care providers to tailor metformin dosage for that individual, so that he or she takes neither more nor less of the medication than needed.

The AMP Type 2 Diabetes Knowledge Portal (www.type2diabetesgenetics.org/) online library and discovery engine has greatly expanded data and search capabilities to accelerate the pace of scientific advancement. Simplified, customizable navigation of aggregated data from more than 100,000 DNA samples from research supported by the NIH and other institutions facilitates new understanding of diabetes by increasing users’ ability to share and evaluate content.

A product of the NIH’s Accelerating Medicines Partnership for type 2 diabetes (AMP T2D), the portal—which opened in 2015—enables user-friendly exploration of international networks of human genetic information linked to type 2 diabetes. At present, researchers and the public can search for information by gene, genetic variant, and region; access summaries of genetic variants; and run customized genetic analyses using versatile tools. Personal identifying information will remain confidential.

Anyone can now query detailed data from the portal. Previously, only approved researchers could access that content, while others could view aggregate results. A Google account is all that is needed to use the portal, which is also available in Spanish. Because the power of the portal depends on community participation, people are encouraged to submit data, comments, and other materials. Administrators also continue to expand the network to include more national and international content, such as the recent addition of data from European collaborators.

Funding for the portal is provided through grants from the NIDDK and the Foundation for the National Institutes of Health. Additional support is provided by the Carlos Slim Foundation, Mexico City. The awards are part of a larger partnership of academic investigators, the NIH, and five pharmaceutical companies. AMP T2D was conceived to translate findings on genetic risk factors in type 2 diabetes into valid targets for new therapies or treatments and provide insights into the pathogenesis and heterogeneity of diabetes.
A Molecular Signal That May Lead to Insulin Resistance and Type 2 Diabetes: New research in mice has identified a compound that triggers increased uptake of “fatty acids”—a category of fat molecules—by blood vessels and muscle cells, and that may thereby contribute to insulin resistance. Experimental evidence suggests that in type 2 diabetes, accumulation of fatty acids in muscle cells may play an important role in reducing the cells’ capacity to respond to insulin by absorbing glucose from the blood; but the mechanism by which fatty acids become concentrated in muscle cells to have this effect has remained unclear. Researchers considered the possibility that a protein called PGC-1α may be involved: among its many roles in the body, it is responsible for increasing the ability of muscle cells to utilize fatty acids by signaling the cells to make more of the enzymes that break them down for fuel. The scientists reasoned that PGC-1α may also trigger secretion of a chemical signal to adjacent blood vessels that acts as a request to supply the muscle cells with more fatty acids. To test this idea, the scientists grew muscle cells that make extra PGC-1α, and then collected the liquid they had grown in—which would contain anything they secreted. When the scientists then transferred this liquid to cultures of blood vessel cells, they found that it caused a marked increase in uptake of fatty acids.

By comparing substances secreted by normal muscle cells with those from cells with elevated levels of PGC-1α, they were able to single out a small molecule designated “3-HIB” as the likely signaling molecule. PGC-1α causes muscle cells to increase the breakdown of valine, an essential amino-acid building block of protein, into 3-HIB, among other compounds; and some of the resulting 3-HIB is then secreted. The researchers found that simply adding 3-HIB to cultured blood vessel cells induced them to increase their uptake of fatty acids. Further, they found that mice given 3-HIB in their drinking water accumulated more fatty acids in their muscle cells than mice not consuming the compound, and also became insulin resistant. (The experiments reported were performed with male mice; female mice may or may not have responded similarly.) Interestingly, previous research has shown that elevated blood levels of valine (as well as certain other amino acids) are associated with a higher risk of developing type 2 diabetes in humans, but the reasons for the correlation remained mysterious. This new study suggests one possible explanation: that excess signaling by 3-HIB, a breakdown product of valine, may contribute to the development of type 2 diabetes. If confirmed, a therapeutic agent that interferes with valine breakdown or 3-HIB signaling may one day help treat or prevent type 2 diabetes.


Newly Identified Molecule Modulates Glucose Release by the Liver: Scientists have discovered a molecule that shows potential as a therapeutic target in people with type 2 diabetes and metabolic syndrome. The molecule, a protein called asprosin, was identified by studying people with a congenital condition that, among other things, causes them to have partial lipodystrophy, or lack of fat tissue in certain areas of the body. Lipodystrophy is often accompanied by insulin resistance and increased insulin levels (a response to insulin resistance), and thus a high risk of developing type 2 diabetes, as well. However, the researchers found two patients who, surprisingly, were not insulin resistant, had lower than normal insulin levels, and normal blood glucose (sugar) levels. By conducting genetic analyses in these two people and examining published scientific reports describing this condition in several other people, the researchers discovered that they all had mutations in a gene coding for a protein called fibrillin. These mutations caused cells to make and secrete much less asprosin, which is derived from fibrillin.

But what does asprosin do? To find out, the researchers conducted experiments in healthy male mice and found that asprosin levels in the bloodstream dropped with the onset of eating. In other experiments, analyzing both mice and humans, they showed that asprosin levels rose in response to fasting. Further experiments in mice and mouse cells revealed that asprosin targets cells in the liver that store and release glucose to help regulate blood glucose levels—a critical metabolic function. When asprosin interacted with these cells, glucose was released into the bloodstream. At the same time, insulin levels rose quickly to counteract the rise in glucose levels, and appeared to help suppress asprosin’s stimulation of glucose release from the liver.
cells. These results suggest that asprosin may play a key role in regulating blood glucose in response to food intake.

Excess release of glucose from an insulin-resistant liver is a problem in type 2 diabetes. The researchers observed that asprosin levels were much higher than normal in both male humans and male mice with insulin resistance and elevated insulin levels, suggesting that asprosin may be involved in this metabolic dysfunction. To see if targeting asprosin might be of therapeutic use, the scientists conducted several experiments, including “blocking” its activity in insulin-resistant, obese male mice by using an antibody directed against the molecule. Encouragingly, injection of a single dose of the antibody caused both asprosin and insulin levels to drop for several hours in the mice, while their blood glucose levels remained stable. In combination with other experimental results, these findings suggest that blocking asprosin leads to a reduced glucose burden in the blood that can be regulated with less insulin. While additional research needs to be pursued in both animal models and people, including investigating whether asprosin activity is the same in females and males, these and other experimental results suggest that artificially lowering asprosin levels may be a new approach to help improve conditions and diseases rooted in insulin resistance, such as metabolic syndrome and type 2 diabetes.


**METABOLIC REGULATORS OF HEALTH AND DISEASE**

**Brain Cells That Control the Body’s Response to Heat:** New research in mice identifies a subset of cells in a specific region of the brain that controls how mammals likely respond to heat. It is critical that our bodies are able to maintain a stable temperature, because sustained periods when core body temperature is too hot or too cold can be dangerous. Mammals have developed various ways to combat temperature fluctuations, such as sweating to cool down or shivering to warm up. In addition to these involuntary responses, mammals also change their behavior, like burrowing for warmth or seeking cool locations. Temperature is sensed by the skin, and this information is relayed to the brain where it is translated into physiological and behavioral responses. Past studies have pointed to a specific region of the brain called the preoptic area, or POA, as being strongly associated with temperature regulation. Previous experiments have shown that heat stimulation of the POA generated substantial regulatory responses to cool the body, while damaging this region eliminated this response. However, the specific cells in the POA that receive temperature information and how they coordinate the appropriate response are poorly understood.

To uncover these details, researchers first sought to identify genes that are “turned on” when both male and female mice were exposed to heat. The levels of gene activity were then used as markers to reveal the brain cells that were activated when the mice were exposed to heat (“warm-sensitive neurons”). Through a series of experiments, the scientists discovered the identified brain cells are specifically and rapidly activated by warm temperatures that trigger temperature-regulating reactions in the mice, indicating that these cells are sufficient to regulate the complex response to heat. The team used a genetic technique to insert a light-responsive protein into the specialized cells to control their activity. This allowed the researchers to turn on the warm-sensitive cells with light. Activation of the cells triggered a rapid decline in core body temperature of the mice. The scientists observed an increase in tail temperature to dissipate heat, and a decrease in the temperature of brown adipose (fat) tissue, which generates heat. In addition to the involuntary physiological responses, experimental activation of the cells also induced behavioral changes. The mice sought out cooler temperatures and decreased nesting activity. Using imaging techniques, the researchers were also able to visualize the heat-sensitive cells making connections with areas of the brain that regulate physiological and behavioral responses to heat. No differences were observed between male and female mice.

These results provide new insights into our understanding of how body temperature is regulated by the brain. The identification of neurons that act as mission control centers to receive environmental information and disseminate messages to alter body temperature in response is a breakthrough and a potential target for therapeutic manipulation.
research is needed to determine if analogous cold-sensing cells exist.


(Information adapted from original article by Dr. Tianna Hicklin, published on October 4, 2016 in NIH Research Matters).

**Identification of Exercise Molecule That Promotes Physical Endurance:** Researchers discovered that musclin, a protein released by skeletal muscle in response to physical activity, enhances exercise capacity in mice. The benefits of exercise are numerous and varied, but how these benefits are achieved at a molecular level remains poorly understood. Skeletal muscle releases proteins, called myokines, and understanding the activity of myokines could lead to new therapies that provide the benefits of exercise. In this study, scientists focused on the relationship between physical activity and a previously discovered myokine named musclin. To determine whether or not musclin was associated with exercise, they looked at the levels of musclin in a group of mice that exercised on a treadmill daily in comparison to a sedentary group. In addition to finding increased levels of musclin in the muscle of the active mice, musclin was increased in the mice’s blood, suggesting that it could have both local and systemic effects.

To identify the effects of exercise-induced musclin, the scientists genetically engineered mice to lack musclin. Compared to mice with musclin, the mice without the protein showed less physical endurance; they tolerated less time, distance, and overall workload on the treadmill. When these mice were given musclin though an infusion, they increased their exercise to normal levels. This demonstrated that musclin was responsible for the differences in physical endurance, and that musclin has potential as a therapeutic to increase exercise tolerance, making it easier for people to exercise. Disruption of musclin also altered the oxygen consumption of the mice during exercise and the size of mitochondria—the energy powerhouses of the cell—in the mice’s muscles, revealing how musclin affects the production of energy in the muscles.

This study showed that levels of musclin are increased in mouse skeletal muscle in response to exercise, and that loss of musclin decreases exercise endurance and oxygen consumption. Importantly, the scientists showed that human muscle cells in the laboratory have musclin, but additional research will be needed to establish whether human musclin acts in the same way as the mouse version. By finding a link between musclin and physical activity, this study identifies musclin as a potential therapeutic to help people receive the benefits of exercise.

Subbotina E, Sierra A, Zhu Z, ... Zingman LV. Musclin is an activity-stimulated myokine that enhances physical endurance. *Proc Natl Acad Sci USA* 112: 16042-16047, 2015.

**CYSTIC FIBROSIS RESEARCH**

**Understanding Differences Between Mouse and Human Cystic Fibrosis Suggests a Potential Way To Reduce Infections:** New research has shown why mice that have the mutation that causes cystic fibrosis (CF) avoid the repeated, serious bacterial lung infections that gradually cause severe lung damage in people with the disease; if a medicinal approach can be found that safely makes the human CF lung environment more like that of mice, this may help people with CF avoid some of the disease’s most dangerous and debilitating effects.

Soon after the discovery that a mutation inactivating the CFTR protein is the cause of cystic fibrosis, scientists created mice with the same mutation in hopes of better understanding the human disease, and to more easily test promising new approaches to CF treatment. To their surprise, however, mice lacking functional CFTR were not nearly as sick as people without it are. Among other differences, the mice are much more resistant to bacterial infections of their lungs, and have mucous in the lungs that is not quite as thick as in human CF. Since then, researchers developed other animal models that more closely resemble the human form of the disease—such as CF pigs and ferrets. But a nagging question remained: what makes CF mice uniquely resistant to infection? A variety of explanations have been suggested. One recently offered hypothesis has to do with acidity—the fluid lining the lungs (“airway surface liquid”) is more acidic in people with CF than in people without the disease. Recent findings suggest this acidification makes it harder for the lungs to fend off bacteria, while simultaneously contributing to the viscosity and
stickiness of airway surface liquid in CF. Bolstering this hypothesis, airway surface liquid also becomes acidic in pigs with CF—but not in CF mice.

New research indicates that acidity does indeed account for much of the difference in infection susceptibility between humans and mice with CF, and identifies the specific protein responsible for acidification of airway surface liquid in people with the disease. The researchers noted that acidity could result either from an excessive transport of acid-causing protons into the airway surface liquid, or from a failure of CF lungs to pump out enough acid-neutralizing ions to counteract protons that are normally there. Indeed, fully functioning CFTR is needed to move bicarbonate, an acid-neutralizing molecule, out of airway cells and into the airway surface liquid. Because CF mice have as much trouble transporting bicarbonate as do humans with the disease, the researchers reasoned that the mouse advantage likely stems from having fewer acidifying protons to neutralize in their airway surface liquid. A careful comparison of mouse and human airway cells suggested that this was, in fact, the case, and implicated a protein called ATP12A that moves protons out of airway cells into the surface liquid, in exchange for potassium ions that it moves into the cells. Mouse airway cells have almost no ATP12A on the outer surface of their airway cells, while those of people and pigs have quite a bit of the protein. In an experiment with cultured human and pig CF airway cells, applying ouabain, a chemical that inhibits proton transport by ATP12A, prevented the cells from acidifying the liquid they were growing in, and made the cultures significantly more resistant to infection, while also reducing the viscosity of the liquid. They achieved similar results by removing potassium ions from the culture liquid, which also prevents ATP12A from transporting protons. In contrast, when they experimentally introduced ATP12A into the airways of CF mice, they found that these mice were much more susceptible to lung infection.

This study suggests that preventing airway acidification could be a new approach to CF therapy. The experimental strategies used in the study, eliminating potassium, or administering ouabain, are not likely to be feasible therapeutically, because potassium is essential for normal function of critical tissues, and ouabain can be quite toxic. However, less toxic ATP12A inhibitors may be identified; and another potential treatment approach might be to neutralize acid in the airway surface liquid using a solution of bicarbonate or other non-toxic buffer. Future research is needed to determine whether these sorts of approaches can safely and effectively improve infection resistance and/or thin the viscous airway surface liquid of CF patients.


**SCREENING FOR LYSOSONAL STORAGE DISEASES**

**Improving Newborn Screening for Lysosomal Storage Diseases:** Scientists developed a new method to screen newborns for six lysosomal storage diseases simultaneously. Lysosomal storage diseases are a group of inherited metabolic conditions in which certain molecules accumulate in harmful amounts in the body's cells and tissues. Each of these diseases is caused by the lack or deficiency of a different protein (enzyme) responsible for degrading waste molecules in lysosomes, which are structures found in cells throughout the body where critical cellular recycling processes occur. The symptoms of the various lysosomal storage diseases vary significantly, and often are not immediately apparent at birth, but each involves the toxic buildup of metabolites that can damage multiple organs in the body. These diseases can cause significant pain; neurological, heart, liver, or kidney problems; and premature death. Because several lysosomal storage diseases are treatable, and early treatment can lead to better outcomes, there is interest in increasing current newborn screening to include treatable lysosomal storage diseases. In many states, newborns are screened for specific disorders using dried blood spots on screening cards. Previous research demonstrated that lysosomal enzymes (such as those missing in this cluster of diseases) retain their activities when the dried blood spots are rehydrated, indicating that screening assays based on direct enzymatic activity could be possible.
In this study, researchers developed and evaluated a new test designed to efficiently and simultaneously screen newborns for Pompe, Mucopolysaccharidosis-I, Fabry, Gaucher, Niemann Pick-A/B, and Krabbe lysosomal storage disorders. In this pilot study, they performed the test on around 43,000 de-identified newborn dried blood spots. Because the samples were de-identified, the researchers could not determine whether their test had successfully pinpointed all cases of these diseases from among the samples. However, by sequencing the key gene from each apparent case of a lysosomal storage disorder the test did identify, the researchers were able to infer what appeared to be a reasonable, low rate of false-positives, that would presumably be corrected by further testing. This technical achievement is a proof of principle, showing that this type of approach may one day soon lead to improved and expanded newborn screening for lysosomal storage disorders.

Islet Transplantation—A Promising Treatment for Difficult-to-treat Type 1 Diabetes

Decades of research, funded by NIDDK and others, is bringing a potential life-changing treatment—islet transplantation—closer to a reality for people with type 1 diabetes. People with type 1 diabetes are unable to take in glucose (sugar) from the blood and use it to fuel their bodies. Without glucose, the cells of the body starve. In 1921, scientists identified the factor that directed glucose from the blood into cells—the hormone insulin—and changed the course of treatment for the disease. Insulin is produced by β (beta) cells, which inhabit the pancreas in clusters known as islets. It is now known that, in people with type 1 diabetes, the body mounts a misguided immune attack against its own β cells. Some or all of the β cells are destroyed, leaving the body without the ability to produce insulin.

A person with type 1 diabetes can take insulin, by injections or with an insulin pump, which is a life-saving treatment. To estimate how much insulin their bodies may need, people with the disease must closely monitor their diet, exercise, and daily routine. Despite careful management of diabetes, it is difficult to mimic the exquisite blood glucose control of the pancreas. While taking insulin treats excess glucose in the blood (hyperglycemia), too much insulin can lead to a lack of glucose (hypoglycemia) in the brain and dangerous situations including coma and death. This absence of normal glucose control leads to diabetic complications.

Even with today’s improved, long-lasting formulations of insulin, this treatment is burdensome and does not work for everyone. Despite vigilant insulin administration, people with difficult-to-control diabetes (also called “brittle diabetes”) may have episodes of severe hypoglycemia with memory loss, confusion, altered or irrational behavior, difficulty in awakening, seizures, or loss of consciousness. Such episodes may make driving or caring for young children unsafe. Repeated episodes can lead to “hypoglycemia unawareness,” in which a person does not realize that he or she has dangerously low blood glucose levels, and thus does not recognize the situation and/or is unable to self-administer treatment. It is critical, therefore, to work toward improved treatments, prevention strategies, and possible cures for type 1 diabetes.

Researchers believe that islet transplantation could be a potential alternate treatment, especially for people with brittle diabetes. In current islet transplantation procedures, the islets are removed from the other cells of a deceased organ donor’s pancreas using specialized enzymes. The islets are purified, processed to maintain their viability and improve engraftment, and counted in a laboratory to ensure that there is a sufficient number for the transplant. After X-rays and ultrasound guide placement of a thin, flexible tube called a catheter through a small incision in the upper abdomen, the surgeon infuses the islets slowly through the catheter into the portal vein of the liver. Once implanted, the islets engraft into the liver tissue and begin to make and release insulin. Full islet function and new blood vessel growth from the new islets take time. Transplant recipients usually take insulin injections until the islets are fully functional. Medications suppressing the immune system are needed for islet transplantation, and must be continued as long as the transplanted islets function to prevent rejection of the transplant. Because of the side effects of these medications, islet transplantation is only considered appropriate for people with incapacitating hypoglycemia despite therapy from physicians skilled in treating type 1 diabetes or for...
those with kidney transplants who already require immunosuppression to preserve the function of the transplanted kidney. Currently, islet transplantation is an experimental therapy, one that can only occur within a clinical trial. For this procedure to transition to a therapy that can be conducted outside of clinical trials, human islets would need to be approved as a cellular biological product by the U.S. Food and Drug Administration (FDA). A Biologics License Application (BLA) will soon be submitted to the FDA, representing decades of research progress to develop innovative solutions to significant challenges.

Transplantation To Restore Insulin Production—From Whole Pancreas to Islets

The idea of transplanting insulin-producing tissue into a person with type 1 diabetes is not a new one. In the late 19th century, scientists discovered that removal of the pancreas caused a dog to develop diabetes, and that transplantation of healthy pancreatic fragments into a diabetic dog could prevent the mortality associated with removing the pancreas. This seminal finding launched the field of pancreas transplantation, and the following year an English surgeon transplanted pancreatic fragments from sheep into a 15-year old boy with severe diabetes. The boy demonstrated temporary improvement of his diabetes, but died several days after the transplant. Doctors have since improved whole pancreas transplantation from human donors, but the procedure involves invasive surgery and, therefore, is best when it can be performed at the same time as another organ transplant, such as a simultaneous kidney and pancreas transplant.

In 1972, scientists achieved the first successful islet transplantation in laboratory rats, reversing their chemically induced diabetes. This success invigorated the field and described several techniques for harvesting, purifying, and transplanting the islets that laid the path toward human clinical trials. However, scientists had difficulty repeating the procedure in larger animal models due to differences in the structure, size, and shape of rodent islets compared to large mammalian islets. In addition, researchers had difficulty in extracting and purifying sufficient numbers of human islets for transplantation. A major advance toward improving approaches to isolate human islets occurred in 1988. Researchers developed an automated method that involved minimal traumatic action on the islets, continuous digestion of the pancreas to release islets, removal of islets to avoid over-digestion, and minimal human intervention in the digestion process. With this new method, islets could be purified more easily, in larger numbers, and in better quality. This advance renewed interest in the possibility of human islet transplantation as a treatment for type 1 diabetes.

Challenges to Islet Transplantation

Another challenge to islet transplantation and to any organ transplantation is the potential rejection of the islets as “foreign” tissue. Scientists think it is likely that many early attempts at islet transplantation—and transplantation of other organs—failed because the recipient’s immune system attacked the transplanted tissue. In addition to the immune system attack on the transplanted tissue as foreign, the continued autoimmune attack on β cells, the same misguided attack that destroyed a person with type 1 diabetes’s own β cells, can reduce the success of islet transplantation. The field of transplantation was propelled forward with the development of immunosuppressive drugs, which dampen the transplant recipient’s immune system.

In 1989, NIDDK-supported researchers combined the new method for isolating human islets with advances in immunosuppression to report the first human islet transplantation that resulted in insulin independence—being free from the need for insulin injections or a pump, because the body was making its own insulin. Ten days after the transplant, the recipient stopped taking insulin
and maintained acceptable blood glucose levels for 12 days. After this period, her glucose levels rose, and she needed to take insulin again. Continued improvements in immunosuppressive medications and procedures to harvest, isolate, purify, and preserve islets led to additional reports of successful islet transplantations with insulin independence for longer periods, even over a year post-transplant, although results varied from patient to patient and study to study.

Immunosuppressive drugs became standard therapy for transplants, but with serious associated risks. Their use can lead to significant side effects, including increased susceptibility to bacterial and viral infections, fatigue, decreased kidney function, mouth sores, and gastrointestinal problems. The long-term side effects are not fully known, and taking immunosuppressive medications also increases the risk of developing certain cancers. These immunosuppressants are thought to affect also the long-term viability of the transplanted islets, as studies suggest that they are toxic to the islets over time. Thus, researchers were searching for ways to enhance the standard therapy using immunosuppressives to improve islet viability and transplantation outcomes.

Significant progress toward that goal was made in 2000, when researchers in Edmonton, Canada, utilized a new protocol that tested a novel combination of immunosuppressive drugs. In a small study, seven people with type 1 diabetes achieved normal blood glucose levels following islet transplantation using this new “Edmonton protocol.” Each patient received a large number of islets in two or three transplants. Progress in methods for isolating and storing islets from donor pancreata prior to transplantation also added to the success of the trial. However, while patients maintained normal blood glucose levels for a period after the transplant, the islets tended to lose their insulin-producing function over time. In addition, it remained to be demonstrated whether Edmonton’s success could be replicated at other sites around the world in a standardized way.

**Standardizing Islet Transplantation**

The Immune Tolerance Network, an international consortium led by the National Institute of Allergy and Infectious Diseases (NIAID) in collaboration with the NIDDK and JDRF (formerly Juvenile Diabetes Research Foundation International), took on the challenge to replicate the Edmonton protocol in a multi-center trial. In the first multi-center trial of islet cell transplantation, from 2001 to 2006, nine sites in North America and Europe successfully replicated the Edmonton protocol in 36 people with type 1 diabetes. One year after transplant, 44 percent of the participants achieved insulin independence with good glycemic control, and another 28 percent, although still requiring some insulin, had partial graft function that completely protected them from severe hypoglycemic episodes. Protection from severe hypoglycemic episodes is an important outcome, as hypoglycemic episodes endanger the lives of people with type 1 diabetes, and fear of hypoglycemia is one reason people with the disease do not achieve the recommended blood glucose levels. At the 5-year evaluation after their final transplant, 17 percent were still insulin-independent.

Although insulin independence declined over time in the study participants, this important study demonstrated that the success of the Edmonton trial could be replicated in a standardized way at other locations. Refinements in techniques for preserving the pancreas and preparing the islets, as well as initiating immunotherapy prior to transplantation to improve islet engraftment, led to high rates of insulin independence in another study of eight single-donor islet transplants. These studies laid the groundwork for a large, international, multi-center network to study and refine islet transplantation technology and provide data toward submission to FDA of an application for islets as a biological product.
The Clinical Islet Transplantation Consortium: Working Towards FDA Approval of Islet Transplantation

As knowledge of islet cell biology and the processes associated with transplantation and immune rejection increased, and pre-clinical studies evaluating new approaches to immunomodulation in conjunction with islet transplantation in animal models progressed, a means was needed by which to study these new approaches rigorously. In 2004, NIDDK and NIAID established the Clinical Islet Transplantation (CIT) Consortium to provide a well-coordinated, collaborative approach to find islet transplantation methods that have higher success rates and fewer risks. To find participants that would benefit the most from this treatment, CIT researchers screened over 8,000, enrolled about 450, and transplanted over 125 participants. CIT has conducted eight clinical trials, with associated immunologic, metabolic, and mechanistic studies, of islet transplantation in individuals with difficult-to-control type 1 diabetes despite intensive medical management.

One of these trials, a phase III study of islet transplantation, enrolled 48 people with type 1 diabetes, impaired awareness of hypoglycemia, and frequent, severe hypoglycemic events despite expert care. At 2 years after transplantation, more than 70 percent of participants demonstrated excellent blood glucose control (with an overall average HbA1c level less than 6 percent), and freedom from severe hypoglycemic events, with restored hypoglycemia awareness. These findings indicated that islet transplantation is an effective treatment for people who have severe hypoglycemic events even with the best medical care. Though these results are impressive, the procedure had significant side effects. Although less toxic, the immunosuppressive drugs still had substantial risks and were demonstrated to decrease kidney function as seen in other transplantation trials. Therefore, this procedure should only be undertaken by people whose type 1 diabetes cannot be controlled by other means and for whom hypoglycemic episodes are life-threatening.

Manufactured islets are not currently approved by the FDA, meaning that islet transplantations cannot be conducted outside of clinical trials. Much like a new drug, islets produced for islet transplantation need to be produced at a high quality consistently. This CIT trial utilized a standardized protocol for generation of islets for transplantation across multiple sites, so that the results could be used as the basis to apply for FDA licensure of islets as a biologic product. Licensure would ensure the purity, potency, and safety of a standard islet product. Once the islet product is FDA-approved, it could transition islet transplantation from an experimental treatment to one that could be performed in regular practice, outside of clinical trials. At that point, the procedure could be covered by third-party insurers. To continue to advance this field as rapidly as possible, scientists need access to information on every islet transplant that takes place, not just those in their local facilities. Thus, the NIDDK created the Clinical Islet Transplantation Registry in 2000 to collect data on islet transplantations for use by the scientific community and the public. By collecting and analyzing these data, the Registry is helping to define the overall risks and benefits of islet transplantation as a treatment option for people with difficult-to-control type 1 diabetes, which is informing future research efforts.

Looking to the Future

Additional efforts are under way to tackle several of the ongoing challenges to long-term success in islet transplantation. A major challenge is the lack of islets available for transplant. Currently, only donor pancreata are used for islet transplantation; a limited number of donor organs are available in general, and they need to match the recipient on several criteria. Additionally, these organs are only available for islet transplantation if the organ has been deemed...
unsuitable for whole pancreas transplantation. Compounding this problem is that studies have demonstrated that large numbers of islets, often from multiple donors, are required to improve the likelihood of success. Thus, it is imperative to identify alternate islet sources. Toward this goal, researchers are exploring ways to generate human islets in the laboratory, and recent advances in this field, including the creation of stem cell-derived β cells from people with type 1 diabetes reported in this chapter, are accelerating progress. The NIDDK’s newly established Human Islet Research Network is conducting basic research studies to pursue innovative strategies to protect and replace β cells in people with diabetes, including developing approaches to grow human islets in the laboratory (see Feature in this chapter). Scientists are also developing ways to protect transplanted islets to improve their longevity and function and to reduce the need for the toxic immunosuppressive medications. Researchers are evaluating new immunosuppressive medications, new combinations of immunosuppressive medications, and non-immunosuppressive medications, as well as ways to induce tolerance in the immune system to protect the transplanted islets. Another strategy being studied to protect newly transplanted islets from immune system attacks is to encapsulate them in a special material; promising results in this area are also described in this chapter. Advances in these fields will be critical for developing a durable cellular therapy for more people with type 1 diabetes.

Researchers have shown that individuals with difficult-to-control type 1 diabetes who receive transplanted islets can have greatly reduced episodes of hypoglycemia, and some remain free of insulin injections for extended periods of time. Islet transplantation, at its current state of development, has been shown to be an effective therapy for this population, a small subset of people with type 1 diabetes. In addition to efforts to improve islet transplantation and other methods to replace β cells, improved technologies for managing diabetes are also being developed, and it is hoped that the population with difficult-to-control diabetes will decrease as these technologies emerge. To develop all possible options to treat type 1 diabetes, researchers continue to pursue islet transplantation. A continued, multi-pronged research approach with pivotal trials of state-of-the-art methods of islet transplantation in humans, as well as basic and pre-clinical efforts, will advance the procedure. As techniques improve and islet transplantation becomes safer and with fewer side effects, the benefits of this treatment may outweigh the risks, making it an option for a larger number of people with type 1 diabetes.
Dr. Alan R. Saltiel—Out of Balance: Obesity, Inflammation, and Diabetes

Dr. Alan R. Saltiel is a professor at the University of California, San Diego School of Medicine, where he is the Director of the school’s new Institute for Diabetes and Metabolic Health. Dr. Saltiel is an internationally recognized expert on the hormone insulin and its role in obesity, diabetes, and cellular signaling. He has published more than 280 original papers, has developed drugs for diabetes and cancer, and holds 18 patents. Dr. Saltiel has received numerous awards, including the Rosalyn Yalow Research and Development Award from the American Diabetes Association, the Hirschl Award, and both the John J. Abel Award in Pharmacology and the Louis S. Goodman and Alfred Gilman Award in Receptor Pharmacology from the American Society for Pharmacology and Experimental Therapeutics. He is a member of the National Academy of Medicine (formerly the Institute of Medicine) and a fellow of the American Association for the Advancement of Science. Dr. Saltiel was appointed to the National Diabetes and Digestive and Kidney Diseases Advisory Council in January 2015.

The seriousness of the obesity epidemic has been well-established: approximately one-third of U.S. adults live with obesity, and obesity increases the risk for type 2 diabetes, kidney disease, and many other devastating diseases and conditions. As researchers seek new ways to treat and prevent obesity, one question is: how, on the cellular level, does obesity harm the body such that excess fat increases risk of health problems? Dr. Saltiel’s research on the links between obesity, inflammation, and diabetes strives to answer this question.

At the National Diabetes and Digestive and Kidney Diseases Advisory Council meeting in May 2016, Dr. Saltiel shared some of his research group’s findings about how obesity causes an inflammatory response that has widespread effects on metabolic health. This research supports a model of metabolic health and disease in which the body views obesity as a stress that it attempts to remedy by triggering inflammation. That inflammation can lead to a shift in the metabolic “set point” that maintains weight and blood glucose (sugar) at heightened levels, but ultimately, the continued stress of obesity and inflammation can lead to insulin resistance and type 2 diabetes.

Warning Signal: Inflammation and Fat (Adipose) Tissue

Inflammation has been linked to obesity for decades, since inflammatory signaling proteins and inflammation-associated immune cells were found in fat (adipose) tissue. In the mid-2000s, these immune cells were still being characterized, and their role in obesity was unclear. Dr. Saltiel’s research team discovered that in lean mice, adipose tissue was rich in a type of immune cell called an M2 macrophage. These M2 cells were known to function in tissue repair and to perform anti-inflammatory functions. If mice were fed a high-fat diet, however, the fat cells became stressed, releasing signaling proteins that recruited pro-inflammatory M1 macrophages as well. The M1 cells eventually overcame the anti-inflammatory activity of the M2 cells, an inflammatory state set in, and the fat cells became resistant to the actions of the hormone insulin. This insight was an important advance in understanding the link between inflammation and
diabetes, because insulin resistance is a precursor to, and also a feature of, type 2 diabetes.

One observation that intrigued Dr. Saltiel was that even though fat cells would become insulin resistant when mice were given a high-fat diet, the cells would still continue to store energy—building up body fat from calories. Insulin promotes fat storage in adipose tissue (in addition to its role in blood glucose control). But were signals other than insulin contributing to fat storage? The answer, Dr. Saltiel’s group found, was yes. Mice fed a high-fat diet not only showed hallmarks of low-grade inflammation but also increased activation of a signaling protein called IKKε. To elucidate IKKε’s role in inflammation and obesity, Dr. Saltiel’s group genetically engineered mice to lack the IKKε gene. When those mice were put on a high-fat diet, they gained less weight, had much less inflammation, and had much better insulin responses than did normal mice put on the same diet. They also increased their calorie burning (energy expenditure). The researchers concluded that IKKε was, indeed, responsible for inflammation in response to a high-fat diet and also for downstream effects on glucose and fat storage. This effect also seemed to be related to a change in calories burned: the mice lacking IKKε had increased body temperature and an increase in oxygen consumption, both indicators of increased energy expenditure.

**Breaking the Cycle: Treating Obesity by Preventing Inflammation**

To shed more light on these findings and investigate IKKε signaling as a potential therapeutic target, the researchers sought an inhibitor of IKKε that could be tested in mice. Dr. Saltiel and his colleagues performed an extensive test of over 150,000 chemical compounds for their ability to inhibit IKKε, and one “hit” was a drug called amlexanox. Further investigation into the scientific literature found that amlexanox had been developed in 1985 and had been used in Japan to treat asthma, though how it worked was not fully determined. The drug also had very few reported adverse side effects.

To determine if inhibiting IKKε would have an effect on obesity, Dr. Saltiel’s group gave amlexanox to mice on a high-fat diet. The drug protected the mice from the associated weight gain. Amlexanox also caused already obese mice on a high-fat diet to lose weight, and if the amlexanox was withdrawn, the mice regained the weight. Thus, amlexanox could both protect against and reverse the effects of a high-fat diet, promoting weight loss. Additionally, amlexanox treatment helped restore other signs of metabolic health: compared to mice not receiving the drug, mice receiving amlexanox had improved glucose tolerance, better insulin sensitivity, and reduced adipose tissue inflammation on a high-fat diet.

But how did amlexanox cause the mice to lose weight? Amlexanox was not altering their food intake, but Dr. Saltiel’s group found that mice given amlexanox and a high-fat diet had a consistent increase in energy expenditure compared to mice not given the drug. Mice given amlexanox also had higher body temperatures, just like the genetically modified mice lacking IKKε. These results supported the idea that amlexanox was increasing the amount of calories the mice burned, which could explain the weight loss and protection against weight gain while on a high-fat diet.

**Releasing Energy: Restoring Fat Burning**

One possible way to burn more energy involves activation of brown fat, a special type of fat that has been increasingly studied for its ability to burn calories, in part to help keep the body warm. (The body can also generate fat with similar characteristics, called beige fat, from the more abundant type of body fat.) Could amlexanox be affecting the mice’s brown/beige fat? Dr. Saltiel and his colleagues studied the fat of mice receiving amlexanox and
found that their fat tissue had characteristics of brown/beige fat, including increased amounts of proteins involved in fat metabolism and smaller stored fat droplets compared to mice not receiving the drug. This confirmed that amlexanox was directly affecting the activation of brown or beige fat to burn more energy.

Dr. Saltiel was also interested in how IKKε factored into another metabolic problem commonly observed in obesity: a fat-burning defect called “catecholamine resistance.” Catecholamines (including adrenaline) are stress hormones that help mobilize the “fight or flight” response to danger. Catecholamine resistance is a physiological state in which cells become resistant to these hormones, resulting in reduced breakdown of fats to provide energy in stressful situations. Although researchers had noted the presence of catecholamine resistance in obese adipose tissue for decades, the underlying mechanism of this condition was unknown. Dr. Saltiel’s group discovered that elevated levels of IKKε reduced the ability of certain receptors (called β-adrenergic receptors) in the fat cells of obese mice to respond to catecholamines. This reduction in β-adrenergic receptor activity resulted in reduced fat breakdown. However, treating the cultured adipose cells with amlexanox blocked this effect and restored sensitivity to catecholamines. Amlexanox also restored catecholamine sensitivity in obese mice.

This research demonstrated that IKKε plays a role in glucose handling and in fat burning, and it explained further how blocking the activity of IKKε could lead to weight loss in mice. These results may also have important implications to human health. Other research groups had showed that the β-adrenergic pathway was not as active in the fat cells of people with type 2 diabetes as in fat cells of people without diabetes.

**Back in Balance: A Model for Restoring Metabolic Health**

In conclusion, Dr. Saltiel described the current model of obesity as an unbalanced state. In a lean body, there is a balance between the β-adrenergic pathway, which encourages fat burning to mobilize energy resources, and insulin, which encourages fat storage. On a high fat diet, fat cells store energy and expand, creating stress. The stressed cells send out a chemical “cry for help,” recruiting pro-inflammatory M1 macrophages. These immune cells release more signaling proteins, which activate IKKε. IKKε then mediates responses associated with obesity: both insulin resistance and catecholamine resistance. This combination results in a state of metabolic inflexibility. Dr. Saltiel suggested that this metabolic inflexibility might also be part of the reason that those attempting to lose weight can find shedding pounds more difficult over time, as the body responds to weight loss with a decrease in the breakdown of fats.

Dr. Saltiel’s presentation clearly demonstrated how understanding the response to a high fat diet in mice can provide insight into human obesity and point to potential new treatments. If this research in mice can be translated into humans, the drug amlexanox—or other compounds that target IKKε—might be a promising avenue for restoring balance to this system by inhibiting IKKε and thus reducing inflammation, restoring energy balance, and decreasing weight. Dr. Saltiel and his research team have begun to test the effects of amlexanox on body weight and other metabolic measures in a preliminary study of a small number of people. His research group has recently found evidence that amlexanox may work through the same pathway in both mice and people. Dr. Saltiel hopes to perform larger clinical studies to determine if amlexanox might be a useful therapy for people with obesity.
Sisters Participate in Life-changing Clinical Trials—Testing Artificial Pancreas Technology for Managing Type 1 Diabetes

Michelle (left) and Paula

To keep blood sugar (glucose) levels within a healthy range, people with the disease (or parents of young children) must measure blood sugar levels with finger sticks or a continuous glucose monitor (CGM), calculate how much insulin to administer, and deliver that insulin via injection or pump. Paula and Michelle said that they typically measure their blood sugar levels four times a day, but it can be more often if their blood sugar is running too high or too low—then they need to monitor more often to make sure it comes back to a healthy range.

Additionally, while insulin therapy helps keep blood sugar from climbing too high, it brings with it the risk of potentially life-threatening episodes of low blood sugar (hypoglycemia). The risk of hypoglycemia greatly limits people’s ability to achieve recommended levels of blood sugar control—levels that have been shown by NIDDK-supported research to reduce the risk of long-term disease complications. Everyday experiences like eating, exercising, and illness can also affect blood sugar levels in unexpected ways, complicating people’s ability to predict changes in their blood sugar levels and determine how much insulin to take.

For these reasons, research is under way to develop new and improved tools to help people with type 1 diabetes manage their disease. Paula and Michelle are on the forefront of testing new technology—called an artificial pancreas—that could potentially help reduce the burden of managing the disease, as well as help people improve their blood sugar control.

Sisters Paula and Michelle were diagnosed with type 1 diabetes 6 months apart 41 years ago when Paula was 12 years old and Michelle was 6. Taking cues from their parents, the sisters made the best of their situation. When they were children and had to take insulin shots, the sisters would save empty syringes (without needles), and “pass them out to our cousins as water guns,” they recall with a laugh.

People’s ability to manage type 1 diabetes has greatly improved since the sisters were children. However, type 1 diabetes is still an extremely burdensome and difficult disease to manage.
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Signing Up for Their First Artificial Pancreas Clinical Trial

An artificial pancreas, or “closed-loop system,” currently being tested in research studies, is technology that would replace the function of the pancreas that does not work in type 1 diabetes: delivering insulin in response to blood sugar levels. It links three technologies: (1) a sensor, such as a CGM, that measures blood sugar levels and sends information to a computer; (2) an insulin delivery device, such as an insulin pump; and (3) a computer that calculates the amount of insulin needed and instructs the pump to deliver insulin.

Artificial pancreas technology would require minimal human input and mimic the exquisite control of blood sugar maintained by a healthy pancreas. Thus, the technology could help people with type 1 diabetes achieve recommended levels of blood sugar control while preventing hypoglycemia, as well as alleviate the enormous burden associated with current management strategies—improving the health and quality of life of people with the disease.

Paula and Michelle have participated in two artificial pancreas trials at the University of Virginia (UVA), located in Charlottesville. It was Michelle who initially heard about their first trial because she liked to keep tabs on artificial pancreas research. Through www.clinicaltrials.gov, which is a service of the National Institutes of Health (NIH) with information on trials funded by NIH and other sponsors, “I found they had quite a few trials in Charlottesville for the artificial pancreas,” she says. She talked to one of the UVA researchers and found she was eligible to participate in an upcoming trial. She told Paula about it, with the hope that she could also participate, and both sisters enrolled in the trial. The trial was funded by JDRF—an organization that has been a key research partner with the NIDDK to advance artificial pancreas research.

The technology they tested as part of that trial is called DiAs, short for “diabetes assistant,” and was developed by researchers at UVA. DiAs is an Android-based smartphone medical platform that serves as the “brains” of the operation; it is paired with a commercial CGM and insulin pump via Bluetooth. For safety reasons, during a clinical trial, the system allows the researchers to monitor real-time data from participants remotely so that they could intervene if the system doesn’t work properly. In “closed-loop” mode, DiAs runs computer algorithms that, based on CGM data, predict when blood sugar levels will fall or rise; in response, the system sends a signal to the insulin pump telling it to adjust insulin levels accordingly. The sisters had to input the number of carbohydrates (carbs) they were eating into DiAs, and to tell it when they were going to exercise, but otherwise the system took over control of managing their type 1 diabetes.

The first phase of the trial started in July 2014. “It was a 3-month trial that was to test the feasibility and safety of using an artificial pancreas at home,” explains Michelle. They first had to learn how to use the individual components of the artificial pancreas. Then, “we moved on to using the DiAs as an artificial pancreas at night. The last step was to use the system for 2½ weeks in 24-hour closed-loop mode,” she recalls. It didn’t take the sisters much time to adjust to using the new technology. “[It was] not hard to get used to,” remembers Paula. “We like technology; we like new gadgets” Michelle adds. “If you use a smartphone, you could definitely use an artificial pancreas.” In November 2014, they turned in their devices but then got them back in March 2015 when they started the
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second phase of the trial, in which they used DIAs at home for 6 months in 24-hour closed-loop mode.

Artificial Pancreas Technology—A Life Changer

“We found that using the DIAs gradually changed our lives,” Michelle says. “It’s been a taste of freedom so to speak,” Paula adds. As Michelle explains, the artificial pancreas “was in complete control. You didn’t have to think about anything. You just put your carbs in there, and it gave you insulin, and you didn’t really have to worry…. It was great!”

Use of the DIAs also helped the sisters sleep better, since low—as well as high—blood sugar levels could often interrupt their sleep. As Paula explains, “Before I got the DIAs, my sugar was dropping and then spiking in the night, and I would have to get up at least once during the night.” After starting on the device, though, she says that, “I was able to sleep through the night and my blood sugar levels were steady through the night and morning.” Michelle adds that, “I could get a full night’s sleep and would wake up … refreshed and ready to take on the day.”

The ability of the artificial pancreas to control blood sugar levels has positively affected other aspects of their lives. One of the things that the sisters liked most was having the ability to exercise and eat healthier, which, for them, resulted in losing some excess weight by the end of the trial. As Michelle explains, before using the device, “Every time I’d exercise I’d have to eat something,” because her blood sugar would drop so low. Even outside of exercise, the sisters had to eat extra calories during the day when their blood sugar dropped too low. However, the artificial pancreas protected against the very low blood sugar drops during exercise, so they didn’t have to eat a lot of food after exercising. And their blood sugar didn’t drop as often or as low when not exercising, so they didn’t need to consume as many extra calories during the day to raise it. “The fact that I wasn’t having to eat all the time … gave me the ability to exercise and eat healthier,” Paula reports. Michelle states, “It feels good to only eat when you are hungry and not because you have to keep your blood sugar up.”

At the end of the trial, the sisters achieved impressive personal results. Paula’s hemoglobin A1c (HbA1c) level—a measure of average blood sugar control—improved from 7.7 percent to 6.6 percent.

One major benefit of using the system was that the sisters experienced many fewer episodes of hypoglycemia. Michelle explains that if blood sugar levels dropped extremely low during the day, “It pretty much wipes you out. Especially if it does it quickly, it completely wipes you out in terms of energy levels.” Paula adds: “Your brain function slows down, too.” The sisters further explain that blood sugar drops could happen quickly and unexpectedly. According to the sisters, the artificial pancreas helped protect against such episodes by keeping their blood sugar levels in a healthier range, so they didn’t experience the extremely low blood sugar levels they once did. If their blood sugar did drop, it dropped at a much slower rate, making it easier to manage.

Relief from the frightening and debilitating symptoms of hypoglycemia made driving less of a worry. “Driving has always been a concern—I never wanted to have a low [blood sugar level] while driving, as the guilt would have eaten me alive if there had been an accident,” says Michelle. However, because her blood sugar was being well-controlled by the artificial pancreas, “During the study, driving became just an everyday task,” she reports happily.

“The [UVA] researchers are fantastic. They are striving to make our lives better… I cannot sing their praises enough,” emphasizes Michelle. “They are a great bunch of people,” states Paula.
(The American Diabetes Association recommends that adults with type 1 diabetes aim for HbA1c levels less than 7.0 percent unless there is a reason to set a higher target.) Paula exclaims, “That is my all-time lowest HbA1c ever!” Michelle started with an HbA1c level of 6.9 percent, which was already at recommended levels, so she didn’t think she’d see much change. However, her HbA1c level improved to 6.1 percent. Thus, both sisters achieved greatly improved blood sugar control with the artificial pancreas, while experiencing fewer episodes of hypoglycemia and improved quality of life.

The sisters recognized that the technology being tested in the trial was still a prototype device, and there are areas that need to be improved. For example, the device would often lose Bluetooth connectivity, requiring them to reconnect the components manually. (There were safety mechanisms in place that alerted them to connectivity problems.) For safety reasons, there were also loud and frequent alarms, although they reported that the alarms lessened overtime. “While this version was not the nirvana I had hoped for, it did allow me to relax a great deal,” states Michelle. They also stress that the research team has been extremely willing to hear their feedback about how to make the system better and more user-friendly. “They are definitely wide open to any improvements that we suggested,” the sisters report. Improving the “usability” of artificial pancreas technology is another important aspect of this research area.

Paula jokes that at the end of the trial, she and her sister weren’t thrilled about turning in their artificial pancreas devices: “We had plans to run away with them,” she laughs. More seriously, she adds: “While it was a sad day when I had to turn off the system and hand it over to the research team, I know that it brings us a step closer to having it available to the public.” She knows that today’s clinical trials are paving the way to what scientists hope will be U.S. Food and Drug Administration (FDA) approval of the technology, which could lead to it being available to people with type 1 diabetes outside of a research setting. Of the trial, Michelle reports: “I could not have had a better experience.”

Another Artificial Pancreas Trial—Project Nightlight

The sisters had such a great experience in their first trial that they eagerly signed up for a new NIDDK-supported trial, called Project Nightlight, which began in spring 2016. In that trial, they are using “inControl,” which is the commercial version of DiAs licensed by a start-up company called TypeZero Technologies; it uses the same algorithms as DiAs. Although the sisters only recently started the trial at the time they were interviewed for this profile, they already see two major improvements in the technology. First, the inControl system is much more user-friendly. Second, they aren’t having the connectivity problems that they experienced in the first trial. Thus, the technology has advanced in the short amount of time between the two trials.

For the 11-month trial, Project Nightlight is examining different functionalities of the inControl platform. For example, for the first part of the trial, Paula and Michelle can only use the closed-loop mode starting at dinnertime and continuing overnight. The hardest part for them is that they want to use it all the time! However, they know that some people with type 1 diabetes may prefer to use closed-loop control during the nighttime only, so it is important to test the device at night only, as well as in 24-hour use.
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Helping Others Through Sharing Research Experiences

Paula and Michelle have active and busy lives. Paula is married to David and has a 28-year-old son, Nathan; she is a licensed practical nurse (LPN) and works for a public school system. Michelle is married to husband Gray and has two daughters, 18-year-old Abby and 16-year-old Maddie; she works from home as a web content manager for a government contractor. Even with their busy schedules, Paula and Michelle have made helping others one of their priorities. In addition to their family, work, and participating in artificial pancreas clinical trials, they have made time to share their experiences in their trials through a blog: www.diabeticsisters.net. One of the reasons they enrolled in the trials and started the blog was because they wanted to help others—including family members—who may not be as comfortable with new technology as they are; five other family members on both sides of their family have type 1 diabetes. “I really wanted to not only help them, but anybody else who might be scared about this [new artificial pancreas technology],” says Michelle.

Hope Through Research

Paula and Michelle couldn’t say enough good things about the research team at UVA. “The researchers are fantastic. They are striving to make our lives better.... I cannot sing their praises enough,” emphasizes Michelle. “They are a great bunch of people,” states Paula. In addition to the group at UVA, there are several other research groups developing different artificial pancreas technologies. To propel research progress in this area, the NIDDK recently funded new advanced clinical trials testing different artificial pancreas systems. The goal is that these trials will pave the way toward generating data to satisfy safety and efficacy requirements for FDA approval of these systems. One of the recently funded trials is led by the UVA scientists.

With continued research, artificial pancreas technology represents a near-term approach that could transform the ability of people with type 1 diabetes to manage their disease with less burden while maintaining healthy blood sugar levels. As Michelle says and Paula echoes: “I personally cannot wait until this [artificial pancreas] equipment is released into the market... so that I can use it 24/7 until a cure is found.... It’s truly life-changing.”
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A Life in Public Health and Service to the Community Continues with Participation in the GRADE Study for Type 2 Diabetes

Earl with his wife, JoAnn

Earl, 71, is a self-described “news junkie” who enjoys travel and believes he was meant to go into public health. As a result, he spent over 34 years in a multi-faceted career helping people in Alabama and neighboring states to improve or manage their health—whether it was visiting communities during disease outbreaks, counseling cancer patients and their families, or encouraging people to join clinical trials. Thus, when he was diagnosed with type 2 diabetes, Earl was already keenly aware of the importance of clinical trials to improving health, with the upshot that he himself volunteered to be a participant in an NIDDK-supported clinical trial that is testing treatments for the disease—and, as he puts it, “Not a day have I regretted that. Not one day.”

Helping Others with Their Health

Earl grew up in a small, rural agricultural community near Birmingham, Alabama. He comes from a large family of 11 siblings—five boys and six girls—all but one of whom are still living. While some have moved to other parts of the country, five of them, including Earl, still live in Birmingham, and they and their families get together regularly. “Those are the benefits that come from having a large family,” Earl notes happily. Earl himself has a son from his previous marriage and two stepsons from his current marriage.

After graduating from college, Earl joined the Air Force for 4 years, serving through basic training alongside one of his brothers, after which he was posted to Japan. It was during his time there, serving as an Air Force medic, that he feels his future career began. “I took care of patients coming back from Vietnam. Everything from bedside care, assisting the nurses and physicians, to direct patient care, even emergency work,” Earl says, adding, “so I guess I was kind of destined, after that, to pursue some career here [in the United States] in ... public health after I left.”

Once he’d completed his active service in 1971, Earl immediately joined the Alabama State Health Department in communicable disease control. For 3½ years, he saw a lot of Alabama—“you know, ‘have car, will travel,’” he says jokingly. He could spend up to 2 to 3 weeks at a time in various, often rural, communities to locate affected individuals in disease outbreaks and get them treated by local physicians. Says Earl, “I think about those days, up and down the highway, sometimes 3,500 miles a month....” But, he adds firmly, “I wouldn’t trade it for anything.”

In 1974, Earl joined the University of Alabama at Birmingham (UAB) Comprehensive Cancer Center as a counselor for patients and their families while in
the hospital or in the outpatient clinic. He notes how being a part of the health care team was especially important for new patients and for people from small communities in Alabama—“patients often looked totally overwhelmed” after a visit from a large medical team, Earl remembers—and he became a “familiar face” for them, sometimes working with patients and their families for 2 to 3 years at a time. Earl admits that there were both high points and low points over the 18 years he was in this position, but emphasizes that, “probably, besides taking care of ... soldiers who were coming back from Vietnam, [this was] the most rewarding part of my career.”

During this time, Earl was also steadily working toward his Masters in Public Health degree in epidemiology. Between work and family, it was “one class per semester” for 11 years, says Earl. Then, while still working at UAB, he took on a position with a National Cancer Institute program called the Cancer Information Service (CIS). According to Earl, the CIS provided both a telephone-based service, which allowed people to call and get answers about their illness or about studies or programs nearby that might be able to help them, and an outreach program. As an outreach coordinator, Earl spent a lot of time encouraging people across Alabama and two other states to volunteer for clinical studies, often visiting their communities in person.

Health Challenges and Putting Knowledge into Action

At the same time as he was working with such dedication to help others with their health, Earl was hit by some health problems of his own. Once an avid tennis player, he had to exchange it for racquetball when he was diagnosed with psoriatic arthritis, a painful and potentially debilitating condition, in his thirties—“jarring news,” Earl says, though he feels he’s been fortunate with his outcomes and has not had to take any medication for the arthritic pain for a couple of decades. More problematically, he developed high blood pressure (hypertension) in his mid-thirties, which worsened as he got older and was a challenge for him and his physician to get under control. Earl has a strong family history of high blood pressure, which he believes contributed to the early deaths of his father and the brother he served with in the Air Force.

In the years following those diagnoses, Earl’s activity levels decreased, in large part due to arthritic pain, such that he even had to give up racquetball. Overtime, his weight crept up, with his highest weight hitting 262 pounds. About 12 to 14 years ago, when he was in his fifties, Earl’s doctor noticed that his numbers from a blood test that indicates risk of type 2 diabetes were creeping up, too. In 2007, Earl was diagnosed with the disease. Seven years later, with his diabetes still not under optimal control despite medication and some weight loss, Earl decided to do what he’d encouraged so many others to do: search to see if there was a clinical study at UAB that could help with his type 2 diabetes—and that is how he found the GRADE Study.

Type 2 Diabetes and the GRADE Study

In type 2 diabetes, the body becomes resistant to the action of insulin—the master hormone in the body controlling blood sugar (glucose) levels—and the pancreatic cells that produce insulin don’t function normally, causing blood sugar levels to rise. This, in turn, can cause damage to blood vessels, organs, and nerves throughout the body. Risk factors for type 2 diabetes include older age, obesity, and a family history of the disease; certain racial and
At the initial screening, a potential volunteer’s degree of blood sugar level control is measured by a test called the HbA1c test, to see if it meets the threshold for the study. That threshold is an HbA1c measurement of 6.8 percent or greater. (The normal range for someone without diabetes is an HbA1c of 5.7 percent or less.) A person who meets this screening criterion then receives additional medical tests over several weeks, and is given information about the study and about diabetes. After this period, eligible volunteers whose HbA1c levels are between 6.8 and 8.5 percent are randomly assigned to one of the four drug treatment combinations (a treatment “arm” of the study) being tested in GRADE.

The four medication classes and the specific drugs being tested in combination with metformin are:

- Sulfonylurea (glimepiride, brand name Amaryl®), which increases insulin levels directly;
- DPP-4 inhibitor (sitagliptin, brand name Januvia®), which indirectly increases insulin levels by increasing the effect of a naturally occurring intestinal hormone;
- GLP-1 receptor agonist (liraglutide, brand name Victoza®), which increases the amount of insulin in response to nutrients; and
- a long-acting insulin (glargine, brand name Lantus®).

Two of the medications, the sulfonylurea and the DPP-4 inhibitor, are taken orally, while the other two are taken by injection under the skin. Metformin (Glucophage®) is also taken orally. All medications are taken every day. GRADE volunteers receive the study medications for free, and attend at least four medical visits per year with their GRADE clinical study site staff so that health outcomes, including blood sugar levels, blood pressure, effects on risk factors for cardiovascular disease, any
When Earl talks about the GRADE Study, he has no end of praise. “I cannot say enough about the people in the clinic and how fantastic they’ve been every time—they have really and truly made me feel like a family member every time I’ve been there,” says Earl.

Feeling the Health Benefits at Home and Beyond

When Earl first entered the GRADE Study in September 2014, his HbA1c was 7.3 percent, and he weighed 254 pounds. Two years later, his HbA1c had dropped to 5.5 percent and he was 196 pounds. “I can’t remember the last time I was under 200 pounds,” says Earl.

Earl says he thinks his weight loss was gradual. “I didn’t notice it as much at first, until I started noticing my belt did not fit in the same place,” he says. His weight loss didn’t go unnoticed by family and friends, however. At a big family reunion in Illinois over the summer, “my sisters and brother in this area (Birmingham) who see me on a fairly regular basis were raving about how much weight I had lost, so I guess that was positive reinforcement. Especially my two youngest sisters ... they just couldn’t quit talking about how much weight [I’d lost], and how good I looked,” says Earl, adding “I guess even if you don’t think of yourself as being very vain, it certainly doesn’t hurt to hear that.”

He also has a lot more energy. Says Earl, “I do feel better, there’s no question about it. In fact, there was a time I would get out to wash the car and I would be tired after washing the car. Now, I can wash two cars and still have some energy left to do whatever it is...
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something else. And I don’t get tired climbing the stairs. It’s been remarkable.”

When asked about how he feels on the study medication in general, Earl says he’s noticed a decrease in appetite and that he doesn’t crave the same kinds of foods he once did—for example, fried chicken wings.

“Just one day, it just seemed my appetite for it was gone.” He adds, “Even in the morning, I don’t care that much for bacon anymore.... I just don’t have much of an appetite beyond one egg or grapefruit.”

Now, he says, he enjoys soups, and can “eat [beans] for days.”

The positive health changes Earl has experienced so far during his participation in the GRADE Study have also had a positive impact on other parts of his life. Although he retired from UAB in 2005, Earl has continued to work in another area that he’s been involved in for over 20 years, first with one of his brothers and now with his son—real estate. He says the business is “in an area [of Birmingham] that is low income, and so we feel like every time we buy a house and fix it up, we’re making a contribution to the community.” Moreover, Earl adds, “We’ve established a relationship with an organization here ... over the last 6 months [that helps] women who are homeless to find a place to live ... just another way of giving back.”

As he goes out daily during the week, “driving out to look at one of [the] properties, or just driving around to meet a tenant who wants to look at [one],” his weight loss and improved energy contribute to a better day.

While no one else in his family has diabetes, [Earl] knows of several friends who do, and says he would encourage people to join the GRADE Study “in a heartbeat.”

Spreading the Word About the GRADE Study

When Earl talks about the GRADE Study, he has no end of praise. “I cannot say enough about the people in the clinic and how fantastic they’ve been every time—they have really and truly made me feel like a family member every time I’ve been there,” says Earl. He recalls one example, a luncheon hosted by the clinic for GRADE volunteers in 2015, and how the UAB GRADE Study coordinator, Dana Golson, R.N., CDE, was there and welcomed every single person who attended as they came in the room. “If there’s an ambassador for GRADE, she’s it,” says Earl.

Because of his own past work in public health and the challenges he encountered in encouraging people to join clinical trials, Earl is also happy to see the diversity of GRADE participants in his own clinic group, including many African Americans, observing that, in clinical trials, “you need people who [represent] diverse populations in the study to come up with a conclusion that is valid.”

Earl also continues to be a health ambassador himself. While no one else in his family has diabetes, he knows of several friends who do, and says he would encourage people to join the GRADE Study “in a heartbeat.” He talks about the GRADE Study at church, with friends and family, and, Earl adds, “I have a group of my long-time [college] fraternity brothers, we get together on a monthly basis ... and I tell them about the study.”

When asked what is the strongest personal impression he’d use to encourage people to volunteer for the GRADE Study, Earl puts it simply and succinctly: “Feeling better, I think is what I’d focus on—on feeling better.”

As of January 2017, the GRADE Study is still actively recruiting participants at clinical sites across the country. For more information about the study and how to participate, please see http://gradestudy.com/