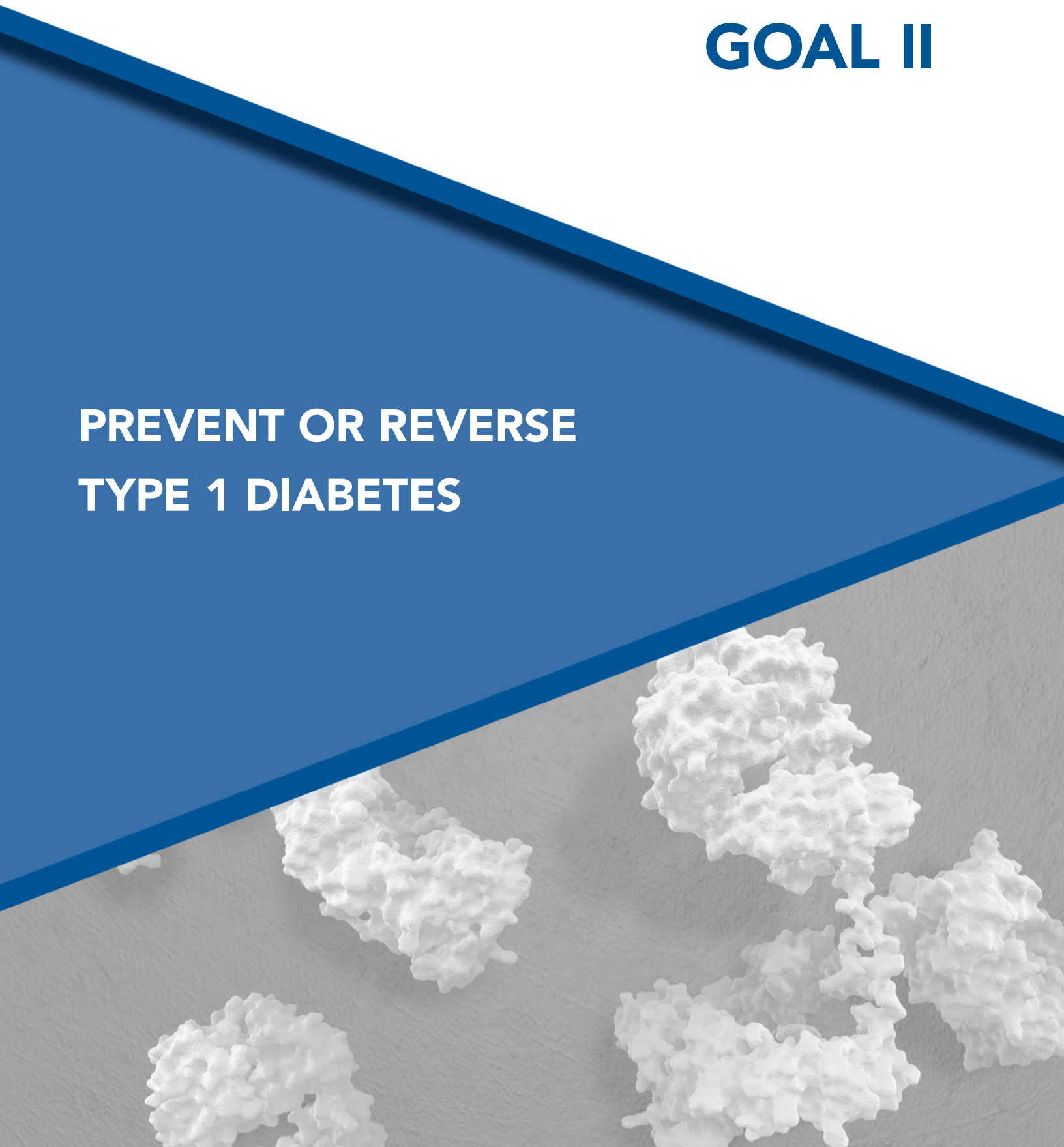
GOAL II

PREVENT OR REVERSE TYPE 1 DIABETES



The *Special Statutory Funding Program for Type 1 Diabetes Research* has enabled the establishment of large-scale, collaborative research groups and clinical trials networks that are identifying and testing novel type 1 diabetes prevention and reversal strategies. In addition to the significant research progress described in this chapter, information on the program evaluation related to Goal II can be found in Appendix A (Allocation of Funds), Appendix B (Assessment), and Appendix C (Evaluation of Major Research Consortia, Networks, and Resources).

Attempts have been made for nearly 3 decades to turn advances in understanding the autoimmune basis for type 1 diabetes into a cure. Progress in the last decade has been fueled by support from the *Special Statutory Funding Program for Type 1 Diabetes Research* (*Special Diabetes Program* or *Program*). In just the past several years, scientists have learned a great deal about the immune system and how its normally protective functions go awry in type 1 diabetes and other autoimmune diseases. New discoveries and technologies have led to better understanding of beta cell development and biology (Goal III), steps forward toward imaging beta cells and autoimmunity in living animals and potentially people (Goal VI), and more effective and safer ways to intervene in the autoimmune process (Goal II). These advances have accelerated other clinical efforts to develop therapeutic approaches to prevent or reverse type 1 diabetes, as discussed in this chapter.

Type 1 diabetes is an autoimmune disease that results when the body's own immune system launches a misguided attack on the insulin-producing beta cells in the pancreas. Harmful immune system cells, including some T cells, are normally eliminated during their maturation or regulated thereafter. However, in susceptible individuals, these disease-causing T cells evade elimination or regulation and initiate an inflammatory process in the pancreas that eventually leads to the destruction of beta cells. The initiation of autoimmunity is marked by the appearance of pancreas protein-specific antibodies made by autoreactive B cells. These "autoantibodies" are well-established markers that predict a person's risk of developing type 1 diabetes. Tests of these antibodies together with tests for genes affecting type 1 diabetes risk in the siblings or offspring of people with type 1 diabetes can predict with great reliability whether the unaffected relatives will develop the disease. This predictive tool, coupled with other new technologies, has given researchers the remarkable ability to design and conduct primary prevention clinical trials.

Graphic: Computer model image of antibodies—proteins that the body's immune system produces to protect itself from foreign substances. In people with type 1 diabetes, the immune system produces antibodies against insulin-producing cells in the pancreas. Image credit: Kenneth Eward/Photo Researchers, Inc.

HIGHLIGHTS OF RECENT RESEARCH ADVANCES RELATED TO GOAL II

Rituximab Slows Progression of Type 1 Diabetes in Newly Diagnosed Patients: Researchers in Type 1 Diabetes TrialNet (TrialNet) reported that the drug rituximab slowed the decline of the function of insulin-producing beta cells in people newly diagnosed with type 1 diabetes. Rituximab destroys B cells of the immune system and has been approved by the U.S. Food and Drug Administration (FDA) for treatment of B cell non-Hodgkin's lymphoma and some autoimmune disorders, such as rheumatoid arthritis. Because B cells are thought to play a role in type 1 diabetes, scientists tested whether four separate infusions of rituximab shortly after diagnosis could slow disease progression. After 1 year, people who had received the drug produced more insulin, had better control of their diabetes, and did not have to take as much insulin to control their blood glucose levels, compared to people receiving placebo. The finding will propel research to find drugs targeting the specific B cells involved in type 1 diabetes because drugs such as rituximab that broadly deplete B cells can increase the risk of infection.

***Deaf1* Gene May Play a Role in Type 1 Diabetes:** Scientists in the Cooperative Study Group for Autoimmune Disease Prevention identified a gene that may play a role in the development of type 1 diabetes. In a mouse model of the disease, scientists found that cells in the animals' pancreatic lymph nodes make two forms of a gene called *Deaf1*. One form encodes full-length, functional Deaf1 protein, while the other encodes a shorter, nonfunctional variant form. Additional experiments in mice suggested that the functional form of Deaf1 may control the production of molecules needed to eliminate immune cells that can destroy insulin-producing cells in the pancreas, thus preventing type 1 diabetes. Researchers also found that levels of the variant form of Deaf1 were higher in people with type 1 diabetes compared to levels in people without the disease. The research suggests that the development of type 1 diabetes may in part be due to increased levels of the Deaf1 variant protein in pancreatic lymph nodes, which may, in turn, lead to reduced production of molecules that are required to "educate" the immune system not to attack the body's own cells, including the insulin-producing cells of the pancreas.

New Markers Discovered for Identifying Type 1 Diabetes-susceptible Individuals Prior to Disease Onset: Scientists in the Beta Cell Biology Consortium (BCBC) discovered a new autoantibody that is an excellent additional marker for identifying pre-clinical type 1 diabetes, and improves the ability to predict disease when combined with previously known autoantibodies. With the discovery of this fourth major autoantibody, called ZnT8, and analysis of large cohorts of children, autoantibody prediction of type 1 diabetes risk continues to gather strength, with increasing evidence for its feasibility both for relatives and, more importantly, for the general population. Approximately 1 million Americans express multiple autoantibodies targeting islet proteins and are at high risk of progression to type 1 diabetes. Prediction using autoantibodies, combined with increasing refinement of genetic and metabolic prediction, sets the stage for prevention trials at multiple stages of the disease.

Elucidating Mechanisms Underlying Tolerance: Type 1 diabetes is thought to arise from a defect in immune tolerance, the "normal" state in which the immune system is non-reactive to healthy cells and tissues. Scientists

have learned much about the cellular and molecular mechanisms controlling tolerance induction in recent years. In particular, some gene expression regulators (transcription factors, *e.g.*, Foxp3) are important for the proper function of regulatory T cells, which suppress misdirected immune responses, while other transcription factors (*e.g.*, Aire) function to allow the removal of autoreactive T cells during development. Other factors known to be important for tolerance induction or maintenance include those involved in immune cell signaling or modulating immune responses (co-stimulatory molecules and cytokines); the biology of regulatory T cells; and the function of dendritic cells, a type of antigen presenting cell, in the process. This knowledge has enabled the design of several successful strategies for imposing a state of tolerance for example, to transplantation antigens in normal rodents, but this has proven to be much more challenging for restoring self-tolerance in rodent models of diabetes. Numerous observations suggest that similar deficiencies in tolerance induction play an important role in human type 1 diabetes, including the association between alleles of the gene encoding insulin (Ins gene), their expression level in the thymic stroma (where removal of autoreactive T cells takes place), and diabetes incidence; the development of diabetes in patients with mutations in the *AIRE* and *FOXP3* genes; and the observations of defective regulation of activated T cells by regulatory T cells in type 1 diabetes patients. These findings are helping pave the way to future approaches that could restore a state of immune tolerance in people with type 1 diabetes.

Development of Sophisticated Mouse Models of Human Disease for Study of Type 1 Diabetes: Increasingly sophisticated stocks of mice modeling human disease have been developed that could provide the means to understand clinically relevant components of type 1 diabetes pathogenesis. These particular mouse models are mice that are either engrafted with functional human cells or tissues, genetically engineered to express human genes, or both, and can recapitulate aspects of the pathogenic process. These mice have been used to identify targets of the human immune response against transplanted islet cells, and have led to insights into the destructive cell populations that are important to this process. Mice engrafted with functional human immune systems may permit certain human immune responses, including autoimmune responses, to be manipulated in small animal models. As these types of studies cannot be done in people, such mouse models could facilitate the conduct of important translational research, providing insights into safety and efficacy before enrolling participants into clinical trials.

Preventing Type 1 Diabetes

A major goal of type 1 diabetes research is to identify strategies to prevent the disease. Ideally, effective interventions to prevent type 1 diabetes should selectively inhibit harmful immune processes, without the need for lifelong suppression of the patient's entire immune system. One major trial that tested a novel prevention strategy and laid the foundation for future clinical trials was the Diabetes Prevention Trial–Type 1 (DPT-1), which was conducted from 1994 to 2003. The trial tested whether insulin administered orally or by injection could prevent type 1 diabetes in relatives of people with the disease. Although the DPT-1 prevention strategies did not prove generally effective, a subset of trial participants who had higher levels of insulin autoantibodies seemed to benefit from oral insulin treatment, though this result was not definitive.

Importantly, the researchers' estimates of risk for disease based on genetic and antibody tests proved to be remarkably accurate. DPT-1 thus demonstrated that it is possible to identify people at risk for type 1 diabetes based on genetic evaluation and tests of antibodies in blood. It also showed that trials requiring massive screening efforts—tens of thousands of people—and involving intensive treatment regimens could be efficiently accomplished.

Testing Prevention Strategies in Type 1 Diabetes TrialNet: The accomplishments of the DPT-1 laid the foundation for current research efforts to prevent the disease. DPT-1 served as the prototype for the present-day Type 1 Diabetes TrialNet, which completed the oral insulin arm of the DPT-1. TrialNet is a large, multidisciplinary, international network established to screen relatives of people with type 1 diabetes for their risk of developing disease and to support the creation and implementation of clinical trials of agents to slow progression of and/or prevent the disease. TrialNet receives support from the *Special Diabetes Program* and is led by NIDDK in collaboration with NIAID, NICHD, NCRR, NCCAM, JDRF, and ADA.

TrialNet is building on the results of the DPT-1 that suggested that oral insulin may prevent or delay type 1 diabetes in people with high levels of insulin autoantibodies. TrialNet developed a new clinical trial to test oral insulin administration in this subset of people. The trial, which requires screening large numbers of people, was started in 2007. Because of the huge effort and expense involved in finding high-risk individuals for prevention studies, a promising strategy is to study the prevention potential of drugs that slow disease progression in individuals newly diagnosed with type 1 diabetes. Thus, TrialNet is developing a prevention trial with an agent called anti-CD3, which targets the immune system and previously has been shown to slow disease progression in people newly diagnosed with type 1 diabetes. Another prevention trial plans to test whether injections of a bioengineered form of a protein made by the insulin-producing beta cells, called glutamic acid decarboxylase (GAD), could prevent the disease in at-risk people. This trial is under development and anticipated to start after more safety data are available in children newly diagnosed with type 1 diabetes. TrialNet also completed a pilot trial testing whether omega-3 fatty acid supplements could affect an immune marker in babies who have a relative with type 1 diabetes. While measurable differences in omega-3 fatty acids were achieved, there was no difference in the immune marker studied, so a full trial will not be launched.

Testing Dietary Intervention To Prevent Type 1 Diabetes: Another important prevention trial supported by the *Special Diabetes Program* is the Trial to Reduce IDDM in the Genetically At Risk, or TRIGR, which began in 2001 and is expected to be completed in 2017 (also see information about TRIGR in Goal I). TRIGR is led by NICHD in collaboration with NIDDK, as well as the JDRF and several other non-federal sources. TRIGR has completed recruitment of newborns into this study, which is testing whether weaning infants to an extensively hydrolyzed formula, as compared to standard cow's milk formula, will reduce the risk of developing type 1 diabetes-predictive antibodies and, ultimately, type 1 diabetes. The design of the TRIGR study was based on both animal studies and a pilot study in humans. The pilot study included 242 children at high risk for the development of type 1 diabetes; data from the study showed that levels of autoantibodies predictive of type 1 diabetes were lower in children who received

the intervention formula—the extensively hydrolyzed formula—compared to children fed conventional formula. Based on those results, and on similar observations made in animal studies, the large-scale TRIGR trial was launched. If TRIGR shows that a dietary modification in infancy could reduce type 1 diabetes, it would have a significant positive impact on patient care.

Preserving Function of Insulin-producing Beta Cells and Reversing Type 1 Diabetes

For people who have already developed type 1 diabetes, reversing or slowing beta cell loss is a key goal because prevention is no longer possible. It was historically thought that people diagnosed with type 1 diabetes did not have any functional beta cells, but research has demonstrated that, at disease onset, 10-20 percent of people's beta cells remain.[22] The basis for immune intervention in type 1 diabetes is to suppress autoimmunity in order to "rescue" the remaining beta cells from immune destruction. The goal of preserving beta cell function is very important because data from NIDDK's landmark Diabetes Control and Complications Trial (DCCT) showed that people whose pancreas continued to produce some insulin had better diabetes control, less hypoglycemia, and reduced rates of disease complications. Preserving remaining beta cells may also allow for the possibility for patients to regrow pancreatic tissue. Research supported by the *Special Diabetes Program* is also pursuing strategies to coax new beta cell formation in the pancreas (see Goal III).

Testing Therapies in Newly Diagnosed Patients in TrialNet: Because of the importance of preserving beta cell function and possibly reversing disease, the *Special Diabetes Program* vigorously supports research toward these goals. For example, TrialNet has supported five trials testing therapies in newly diagnosed patients. In 2009, TrialNet reported the result that the drug rituximab preserved the function of insulin-producing beta cells in people newly diagnosed with type 1 diabetes. Improved insulin production was maintained 1 year after the drug was administered. This drug had been approved by FDA for treatment of certain cancers and autoimmune disorders. It was historically believed that therapies to combat type 1 diabetes should focus on the T cells of the immune system, which attack and destroy the insulin-producing beta cells. However, rituximab targets other cells of the immune system—B cells. Therapies targeting B cells had been tested in mouse models of type 1 diabetes, but they had not been tested in people. Thus, rituximab was the first drug targeting B cells to be tested in people. While the drug was only given transiently after onset of type 1 diabetes and the effect of rituximab had dissipated at 2 years, the observation that a therapy that targets B cells could preserve beta cell function in people is therefore a novel finding and suggests that other therapies to target B cells may be effective for type 1 diabetes prevention or early treatment.

In addition to testing rituximab, TrialNet is also testing the ability of other therapies to halt beta cell destruction in new-onset type 1 diabetes. Three clinical trials are ongoing testing a vaccine using a recombinant form of the beta cell protein GAD administered with the immune adjuvant alum; a drug called abatacept (which inhibits T cell activation); and early and intensive blood glucose

[22] Skyler JS and Marks JB: Immune Intervention. In *Diabetes Mellitus: A Fundamental and Clinical Text, 3rd Edition*, edited by LeRoith D, Taylor SI, and Olefsky JM (pp. 701-709). Philadelphia, PA: Lippincott Williams & Wilkins, 2004.

control using a closed-loop system shortly after disease onset. TrialNet also completed a trial testing the combination of two drugs (MMF/DZB) for treating newly diagnosed patients and found no effect on slowing beta cell loss.

Testing Therapies To Induce Tolerance in the Immune Tolerance Network: The *Special Diabetes Program* also supports the Immune Tolerance Network (ITN), which is an international group of researchers dedicated to evaluating therapies to reprogram harmful immune responses to reduce autoimmunity, allergy, and asthma; and to improve islet, kidney, and liver transplantation (see Goal III for ITN research related to islet transplantation). The ITN is led by NIAID in collaboration with JDRF and NIDDK through its oversight of the *Special Diabetes Program*; the Network also works closely with TrialNet. The ITN also carries out laboratory tests to understand how the body responds to treatments studied in clinical trials and to find better markers of immune function. The ITN is trying to move away from current approaches of suppressing the immune system, in which people take drugs for the rest of their lives and may encounter significant side effects. Rather, the ITN is looking to induce "tolerance," in which a short-term therapy re-educates the immune system so that it does not destroy the body's own cells. The focus of the ITN's current type 1 diabetes research portfolio is to preserve beta cell function in newly diagnosed patients. For example, the Network completed a Phase I clinical trial testing a novel vaccine (insulin-B chain peptide) and found that it elicited a detectable immune response in the patients. In addition, the ITN is building on the results of research showing that the anti-CD3 monoclonal antibody could preserve patients' beta cell function. The Network is testing whether multiple doses of anti-CD3 have additional benefits. Other therapies being tested in newly-diagnosed patients include thymoglobulin, and IL-2 plus sirolimus.

Creating a Pipeline of New Therapies for Type 1 Diabetes Prevention and Reversal

The *Special Diabetes Program* has enabled the creation of a pipeline of therapeutic agents for testing in clinical trials and has also created the infrastructure to test them. As new knowledge is gained about the underpinnings of disease development, more strategies for disease prevention and reversal will be identified, which will feed into this critically important pipeline made possible by the *Program*. For example, the TEDDY study (see Goal I) is examining environmental triggers of type 1 diabetes. If scientists identify a possible environmental trigger, such as a dietary factor, then researchers in clinical trials networks, such as TrialNet, could test the ability of that factor to prevent disease in at-risk people. The *Special Diabetes Program* also supports pre-clinical research resources that foster translational research from the bench to the bedside. For example, the Type 1 Diabetes-Rapid Access to Intervention Development (T1D-RAID) program helps scientists ready agents for testing in clinical trials to bridge basic research discoveries with clinical trials in people.

This research pipeline has already resulted in the movement of therapies from discovery in the laboratory to clinical trials. For example, researchers in the Non-human Primate Transplantation Tolerance Cooperative Study Group (see Goal III), which is supported by the *Special Diabetes Program* and led by NIAID, demonstrated long-term survival of islets after transplantation when the animals were given a novel

mixture of medicines that target the immune system. Based on those findings, the ITN approved a clinical trial to test this therapy in newly diagnosed type 1 diabetes patients, to determine if the medicines can slow progression of disease. The T1D-RAID program is generating the medicines for use in the trial. This example demonstrates how the *Special Diabetes Program* supports the discovery, manufacture, and testing of promising therapeutic agents—creating a robust pipeline of agents that can improve the health of people with type 1 diabetes.

Improving Predictive Abilities and the Ability To Conduct Trials on Type 1 Diabetes Prevention and Reversal

Clinical trials to test type 1 diabetes prevention strategies require screening large numbers of people to identify at-risk individuals. For example, scientists in the DPT-1 screened about 100,000 relatives of people with type 1 diabetes to enroll 372 into the study—an undertaking that was both time- and resource-intensive. In addition, TrialNet plans to screen 20,000 people annually to identify those who are eligible to enroll in prevention trials.

The *Special Diabetes Program* supports a broad range of research to streamline this process by improving methods to assess risk and identify people who may benefit from prevention therapies. Researchers have made progress in identifying novel markers of the disease process in order to improve the ability to identify people at risk. For example, scientists in the BCBC (see Goal III), which is supported by the *Special Diabetes Program*, discovered that antibodies to a protein called ZnT8 are an excellent marker for pre-clinical type 1 diabetes and greatly improve predictive abilities when combined with previously identified disease markers. Some research studies supported by the *Special Diabetes Program* are now screening for the presence of these antibodies in people in their studies. Identification of new type 1 diabetes susceptibility genes by the Type 1 Diabetes Genetics Consortium (see Goal I) can also pave the way toward using those new genes to improve predictive abilities. The efforts to improve predictive abilities are not only important for enhancing the ability to conduct clinical trials with fewer people, but are also critical for identifying at-risk individuals in the general population, so that as many people as possible can benefit when new prevention strategies are proven effective.

Because clinical trials to prevent or reverse type 1 diabetes occur at various sites throughout the United States and the world, it is critically important to have standardized tests so that data can be combined and compared. C-peptide is a byproduct of insulin production and thus useful as a marker of beta cell function. Indeed, C-peptide is used as an outcome measure to indicate insulin production in clinical trials focused on type 1 diabetes prevention and reversal, including trials supported by industry to gain regulatory approval of new drugs. To standardize C-peptide measurement in clinical trials, the *Special Diabetes Program* supports the C-peptide Standardization Program, which is led by CDC in collaboration with NIDDK. The scientists have made progress toward optimizing measurement techniques and standardizing results of C-peptide tests conducted at laboratories throughout the world by developing a highly precise reference method.

Understanding the Regulation of the Immune System

While clinical trials are ongoing, parallel research efforts are continuing to investigate the underlying causes of type 1 diabetes. Significant research progress on understanding the underlying mechanisms of type 1 diabetes have laid the foundation for conducting the clinical trials already described in this chapter. Researchers are building on the success to date and are now vigorously trying to understand the interactions between the environment and the immune system, as well as the means by which genes influence immune responses resulting in autoimmunity. Scientists are also studying the roles that the less specific arms of the innate immune system may play as contributors to the complex underpinnings of type 1 diabetes. Recent research has also suggested that inflammatory cells, such as mast cells and neutrophils, and cells of the innate immune system, such as natural killer cells, may play a role in type 1 diabetes. At the same time, scientists are studying the role of other important immune system cells, including dendritic cells, that control immune response and tolerance. Research to define the key components and the molecular defects that provoke the immune system to attack and destroy the beta cells is key to predicting, diagnosing, treating, and ultimately preventing this autoimmune process.

Cell-based Immune Modulation Therapy: Researchers supported by the *Special Diabetes Program* have made strides toward using dendritic cell therapy for preventing and treating type 1 diabetes in animal models. Some types of dendritic cells are involved in activating T cells, which are the cells that are central to the attack on the beta cells in type 1 diabetes, while others can induce tolerance to beta cells. Recent studies have tested approaches to modify dendritic cells to prompt the elimination of errant T cells to prevent the destructive immune attack. For example, scientists have modified dendritic cells in culture and then used them to successfully prevent or delay type 1 diabetes in a mouse model of disease. Other researchers have modified dendritic cells directly in the mouse. For instance, researchers used "microspheres" to deliver certain molecules to the animals that modify the dendritic cells in a way to make them suppress, rather than incite, T cell attacks on beta cells. A single injection of microspheres containing these suppressive molecules significantly delayed onset of diabetes in a mouse model of type 1 diabetes; several consecutive injections prevented the disease altogether. In mice that already had diabetes, the microsphere therapy reversed the disease. These studies demonstrate that modifying dendritic cells is a possible therapeutic approach for preventing, delaying, or reversing type 1 diabetes. Future research will help to determine if dendritic cell therapy might have the same dramatic benefits in people.

Identifying Targets of Autoimmune Attack in Type 1 Diabetes: For years, scientists have struggled to determine which beta cell proteins are key targets of autoimmune attack. A major advance in this area was made by the *Special Diabetes Program*-supported Cooperative Study Group for Autoimmune Disease Prevention (Prevention Centers), which is led by NIAID in collaboration with NIDDK and JDRF. The research group showed that they could prevent the disease in the non-obese diabetic (NOD) mouse model of type 1 diabetes by eliminating a primary sequence of insulin previously known to be a target of autoimmunity. Insulin is also known to be a key target in humans. These findings suggest that autoimmune reaction against insulin may

be a critical initiator of the pathway toward beta cell destruction in humans.

Understanding How Risk Genes Cause Disease: The Prevention Centers have made other significant contributions toward understanding the underpinnings of type 1 diabetes. For example, the Centers have spearheaded the "NOD Roadmap" project, which is generating a comprehensive time course of disease in the NOD mouse model and providing other key data. One important finding from this project was the identification of a gene that may play a role in the development of type 1 diabetes. Scientists found that cells in the animals' pancreatic lymph nodes make two forms of a gene called *Deaf1*. One form encodes full-length, functional Deaf1 protein, while the other encodes a shorter, nonfunctional variant form. Research suggested that the functional form of Deaf1 may control the production of molecules needed to eliminate immune cells that can destroy insulin-producing cells in the pancreas, thus preventing type 1 diabetes. Researchers also found that levels of the variant form of Deaf1 were higher in people with type 1 diabetes compared to levels in people without the disease. The research suggests that the development of type 1 diabetes may in part be due to increased levels of the Deaf1 variant protein in pancreatic lymph nodes, which may, in turn, lead to reduced production of molecules that are required to "educate" the immune system not to attack the body's own cells, including the insulin-producing cells of the pancreas.

Uncovering the Relationship Between Gut Bacteria and Type 1 Diabetes: Insights from research supported by the *Special Diabetes Program* has also found that the trillions of bacteria and other microbes that live in the gut can blunt the immune system attack that causes type 1 diabetes. During the past decades, researchers observed increased incidence (number of new cases) of type 1 diabetes in developed countries, which was thought to be due to changes in the environment, including the microbes that live in our bodies. Supporting this idea, previous studies found that the incidence of type 1 diabetes in mice susceptible to this disease can be affected by microbes in their environment. Thus, researchers supported by the *Special Diabetes Program* set out to further explore the possible connection between type 1 diabetes and microbes. They discovered that a complex interaction between the immune system and bacteria in the gut may help to lower the risk of developing type 1 diabetes in the mouse model of the disease. The widespread use of antibiotics and more aggressive cleanliness of modern society can alter the mix of microbes living in our body. This research suggests that an unexpected consequence of this environmental change may be an increased risk of autoimmune diseases like type 1 diabetes.

Promoting Translational Research: Researchers supported by the *Special Diabetes Program* have made progress in generating increasingly sophisticated stocks of mice with the expectation that they will show greater fidelity to human type 1 diabetes, and which may provide the means to understand clinically relevant components of type 1 diabetes pathogenesis. These mice are either engrafted with functional human cells or tissues, genetically engineered to express human genes, or both, and can recapitulate aspects of the pathogenic process. These mice have been used to identify targets of the human immune response against transplanted islets and have led to insights into the destructive immune cell populations that are important to this process. Mice that are engrafted with functional human immune systems may permit certain human immune responses to be

manipulated in small animal models. As these types of studies cannot be done in people, such mouse models could facilitate the conduct of important translational research, providing insights as to safety and efficacy before enrolling people into clinical trials.

Summary

This chapter highlights some of the significant research progress that has been made possible by the *Special Diabetes Program* toward the goal of preventing and reversing type 1 diabetes. Without support from the *Special Diabetes Program*, it would not have been possible to establish large, collaborative networks, such as TrialNet, at an unprecedented scale. As basic research supported by the *Program* continues to identify possible new therapeutic targets, networks such as TrialNet and the ITN are poised to test these new therapies in people. Progress has already been achieved, and additional progress is expected in the future as new therapies are identified and tested in the people who could benefit from them.

RESEARCH CONSORTIA AND NETWORKS RELATED TO TYPE 1 DIABETES PREVENTION AND REVERSAL

Evaluation of research consortia and networks supported by the *Special Diabetes Program* and related to Goal II is found in Appendix C. Highlights of research progress are summarized below.

Type 1 Diabetes TrialNet: TrialNet is an international network that screens large numbers of people and conducts clinical trials of agents to prevent type 1 diabetes in at risk people and to slow progression of the disease in people who are newly diagnosed. TrialNet has screened over 74,000 people for type 1 diabetes risk to identify those eligible for participation in three ongoing or planned disease prevention trials. TrialNet also supports trials in people newly diagnosed with type 1 diabetes that hope to delay disease progression. TrialNet reported a novel finding that therapies targeting B lymphocytes of the immune system may be a strategy for preventing or reversing type 1 diabetes.

Immune Tolerance Network (ITN): The ITN is an international group of researchers dedicated to evaluating therapies to reprogram harmful immune responses to reduce autoimmunity, allergy, and asthma; and to improve islet, kidney, and liver transplantation (see Goal III for ITN research related to islet transplantation). ITN has developed eight clinical trials in people newly diagnosed with type 1 diabetes to test novel therapies to slow disease progression. In addition, the ITN completed the first multicenter study of islet transplantation in sites across North America and Europe, laying the groundwork for the Clinical Islet Transplantation Consortium (see Goal III).

Trial To Reduce IDDM in the Genetically At-Risk (TRIGR): TRIGR completed recruitment of 2,160 newborns for a trial examining whether hydrolyzed infant formula compared to standard cow's milk-based formula decreases the risk of developing type 1 diabetes in at-risk children. Researchers are now following the enrolled children.

Cooperative Study Group for Autoimmune Disease Prevention: The Prevention Centers engage in scientific discovery to advance knowledge toward the prevention and regulation of autoimmune diseases, such as type 1 diabetes, and create improved animal models of disease to better understand immune mechanisms. The Study

Group identified insulin as a primary target of the immune attack in a mouse model of type 1 diabetes, and also identified a gene (*Deaf1*) that may contribute to the development of type 1 diabetes and be a target for therapy.

Standardization Programs: Three different standardization programs are improving reliability in measurement of autoantibodies, C-peptide, and hemoglobin A1c (HbA1c) (also see Goal V). The ADA has built on the tremendous success of the HbA1c standardization program to set treatment goals for glucose control in all forms of diabetes based on the test and more recently recommended HbA1c as a more convenient approach to diagnose type 2 diabetes.

Type 1 Diabetes–Rapid Access to Intervention Development (T1D-RAID): This program provides services to scientists to help them ready agents for testing in clinical trials, thus bringing novel prevention and treatment approaches to people who could benefit from them. T1D-RAID has manufactured several agents that are being tested in ongoing clinical trials supported by the *Special Diabetes Program*. The Pre-clinical Testing Program associated with T1D-RAID has developed better methods for using rodent models for pre-clinical testing and has initiated testing of several new possible therapeutics. In addition to agents related to prevention and reversal of type 1 diabetes, the T1D-RAID and the Pre-clinical Testing Program are providing resources for agents related to preventing or treating diabetes complications (Goal V).

Investigator Profile

Mark D. Pescovitz, M.D.*

Leading a Clinical Trial Testing Rituximab in People with Newly Diagnosed Type 1 Diabetes

Mark D. Pescovitz, M.D.

Mark D. Pescovitz, M.D., is Professor of Surgery and Professor of Microbiology and Immunology at Indiana University School of Medicine, in Indianapolis, Indiana. Through his participation in Type 1 Diabetes TrialNet, which is supported by the Special Statutory Funding Program for Type 1 Diabetes Research, *he leads a clinical trial testing the ability of a drug, called rituximab, to preserve the function of insulin-producing beta cells in people with newly diagnosed type 1 diabetes. This profile describes how he came to be involved in type 1 diabetes research, the origin of the rituximab trial, and how Type 1 Diabetes TrialNet facilitated the trial.*

"I've always had a strong interest in immunology," says Dr. Pescovitz. "Because my father was a surgeon, I also had an interest in surgery. My combined interests in immunology and surgery led me to focus my career on being a transplant surgeon, but with a strong interest in immunology."

Upon his arrival as a faculty member at Indiana University in 1988, Dr. Pescovitz started a pancreas transplant program, beginning his long-standing interest in the management of type 1 diabetes. Because of the need to suppress people's immune systems after they undergo an organ transplant, Dr. Pescovitz was involved in developing several new immunosuppressive agents. He subsequently became interested in applying those agents to the treatment of type 1 diabetes, which results from the misguided attack of the immune system on the insulin-producing beta cells in the pancreas. Thus, therapies targeting the immune system could be a way to prevent or reverse the disease.

At about the same time that Dr. Pescovitz and a colleague were planning for a trial to test an immunosuppressive agent in new-onset type 1 diabetes, NIDDK was soliciting applications to establish a new collaborative clinical trials network, called Type 1 Diabetes TrialNet. Because the goals of the network dovetailed with his research interests, he and his colleagues at Indiana University applied and successfully competed to become a TrialNet clinical center, which began his involvement in the network. TrialNet is an international network of investigators, clinical centers, and core support facilities that recruits patients and develops and conducts clinical trials testing strategies for type 1 diabetes prevention and early treatment.

** This profile is printed in honor and memory of Dr. Pescovitz whose death in 2010 was a major loss to the diabetes community.*

A New Clinical Trial Concept

Rituximab destroys B lymphocytes (also called B cells). These are the part of the immune system that produce antibodies. The connection between rituximab and treating type 1 diabetes came when Dr. Pescovitz attended an annual B lymphocyte summit sponsored by Genentech, Inc., which markets Rituxan® (rituximab). At that time, most of the research discussed at the summit was focused on using rituximab to treat lymphoma; the drug had been approved by the U.S. Food and Drug Administration (FDA) in 1997 for the treatment of B cell non-Hodgkin's lymphoma. However, Dr. Pescovitz recalls, "At one of the meetings, I heard a presentation about rituximab being used successfully in early studies for the treatment for rheumatoid arthritis. Rheumatoid arthritis is similar to type 1 diabetes in that they are both autoimmune diseases thought to be T cell mediated and associated with autoantibodies that are not necessarily pathogenic. It was an easy leap for me to think that, if rituximab works for rheumatoid arthritis, maybe it will work for type 1 diabetes." With that thought, a new clinical trial concept was born.

Launching a Clinical Trial Through Type 1 Diabetes TrialNet

While there was research being done testing B lymphocyte therapies in mouse models of type 1 diabetes, "there was nothing being done in humans," says Dr. Pescovitz. In fact, immune therapies being tested in people targeted other cells in the immune system, called T cells, which attack and destroy insulin-producing beta cells. Thus, rituximab, which targets B lymphocytes, was a novel approach for treating people with type 1 diabetes.

TrialNet requires researchers both inside and outside of the project to submit clinical trial proposals for consideration by the network. Thus, Dr. Pescovitz submitted his clinical trial proposal to TrialNet, which would provide the funding and infrastructure for the trial, as well as to Genentech, which would provide the drug. Both groups approved the concept to test rituximab in people newly diagnosed with type 1 diabetes. The trial enrolled 87 patients who received four separate infusions of either rituximab or a placebo. In November 2009, Dr. Pescovitz and his colleagues published the results of the trial in the *New England Journal of Medicine*. The trial showed that, after 1 year, the people receiving rituximab produced more insulin, had better control of their diabetes, and did not have to take as much insulin to control their blood glucose levels, compared to people receiving placebo. The patients are now being followed to assess longer-term outcomes. The exciting results suggest that rituximab delays progression of type 1 diabetes; additional research is needed before the drug would be approved for treating patients. The success of the trial also suggests that other therapies to target B lymphocytes may be useful for treating or preventing the disease—knowledge that could inform future clinical trials.

Dr. Pescovitz stressed that the trial did not happen overnight. "It took 5 years from concept proposal to looking at the results, and 6 years from concept proposal to publication," he explains. "It takes a long time to do these trials, and you cannot expect instantaneous answers. Clinical research, in particular, takes long time horizons to pursue and to see results." Because of ethical considerations to the dedicated patients involved in trials, and the long timeframes necessary to conduct trials and adequately follow participants, Dr. Pescovitz says, "These trials also require a long time horizon in terms of funding." If there had been uncertainty as to whether

there would be sufficient funds to complete the trial, "I couldn't have started the study," he says.

The Benefits of Type 1 Diabetes TrialNet

Rituximab was an FDA-approved drug for the treatment of B cell non-Hodgkin's lymphoma and subsequently for rheumatoid arthritis and chronic lymphocytic leukemia. However, Dr. Pescovitz notes that, "Industry did not have an interest in developing the drug for the treatment of type 1 diabetes. So if it hadn't been for someone like me, industry would not have moved forward on their own. They were happy to provide the drug to be used in the trial, but I needed to have support and patients."

The support and patients came in the form of TrialNet. "TrialNet was the perfect fit because it had the large infrastructure that provided access to patients and it had the clinical support structure in place. It was a perfect combination," says Dr. Pescovitz. Dr. Pescovitz could have applied for NIH funding for the trial through an investigator-initiated R01 grant mechanism. However, in this case, he explains that, "Using an R01 mechanism would have been more cumbersome, difficult, and taken a longer period of time. TrialNet was the perfect fit for this type of trial."

Dr. Pescovitz also says that his research has benefited from interactions and collaborations with other scientists in TrialNet. "I am a transplant surgeon and an expert in immunology, but not a diabetes guy," he says, "and I think there is great synergism in having people like me interacting with people who are experts in diabetes." He also stressed the importance of collaborations on mechanistic studies. "There are a group of top diabetes immunologists from around the world who sit at a table and provide ideas as to how to look at mechanism—not just whether a therapy works or not, but how or why it works. Those are collaborations facilitated by TrialNet." Understanding the mechanism by which a drug works could shed light on the molecular underpinnings of disease and open up new avenues for therapy.

The rituximab trial tested the drug in newly diagnosed patients, but TrialNet also conducts clinical trials to prevent the disease in at-risk individuals. Dr. Pescovitz explains, "Prevention trials are an entirely different type of trial that only an organization like TrialNet can do because you have to screen large numbers of patients to find those who are eligible." Indeed, TrialNet has screened over 70,000 people to date to identify those eligible for prevention trials and screening is ongoing. Dr. Pescovitz also notes that industry is unlikely to undertake such a large-scale effort to study type 1 diabetes prevention. Thus, TrialNet provides a unique infrastructure—made possible by the *Special Diabetes Program*—to conduct these important studies.

Through TrialNet, Dr. Pescovitz was able to move forward a new idea for type 1 diabetes treatment to testing it in people. TrialNet enabled the conduct of this trial in a more streamlined timeframe than would otherwise have been possible. The trial not only identified a potential new therapy to slow progression of type 1 diabetes, but also suggested that other therapies targeting B lymphocytes may be effective. TrialNet remains critically important for testing these and other new and emerging therapies to improve the health of people with type 1 diabetes, and the creativity and innovation of scientists, such as Dr. Pescovitz, remains a cornerstone of realizing progress in type 1 diabetes research.

Patient Profile

The Gould Family

Dedicated To Participating in Research To Be Part of a Cure for Type 1 Diabetes

Dave and Ellen Gould of Nashville, Tennessee have eight children ranging in age from 2 to 17. Within the last 5 years, four of their children have been diagnosed with type 1 diabetes. Even though their lives are busier than most people can imagine, the Goulds make time not only to participate in clinical research studies, but also to tell others about the importance of research toward combating type 1 diabetes and finding a cure.

Their passion and dedication was evident when Ellen testified in Congress at a hearing held in conjunction with the Juvenile Diabetes Research Foundation's 2009 Children's Congress. In her testimony, Ellen related how, on a Saturday morning several months earlier, the family was awakened by then 12-year-old son, Sam, who collapsed in his room, incoherent, because of a dangerously low blood sugar level. "It took us 20 minutes to get him back to normal," Ellen said. "But what happens the next time if we don't hear him? As their mother, I just want to reach out and make it better–but I can't. I can't cure this disease; I can't make it better for my kids. I need help. Finding a cure means everything to my family, and we are willing to be part of the solution."

To that end, the Goulds are participating in NIDDK's Type 1 Diabetes TrialNet, an international network of researchers exploring new strategies to prevent, delay, and reverse type 1 diabetes. TrialNet is also supported by the *Special Statutory Funding Program for Type 1 Diabetes Research*.

The Gould family.
Back row, left to right: Sam, Patrick, Ellen, Dave, Andrew, and Nicholas.
Front row, left to right: Maggie, Annie, Sarah, and Oliver.
Photo credit: Amy McIntyre

"Finding a cure means everything to my family, and we are willing to be part of the solution," said Ellen.

"There are a lot of smart people working on a cure for this disease," Dave said in a later interview. "I'm an optimist. I believe a cure is coming, and if my family can help speed it up a bit by being part of an important study, all the better."

"Diabetes Is Part of Our Family"

Type 1 diabetes is a chronic disease in which the body's immune system launches a misguided attack and destroys the insulin-producing cells of the pancreas. People with the disease need daily administration of insulin, either by injection or with a pump, and must monitor their blood sugar levels vigilantly.

"Diabetes is part of our family," Ellen said. "We're constantly filling prescriptions, scheduling doctors'

appointments, filling out forms for school and various activities, educating others—and making sure our kids are safe," she added.

For the Goulds, the beginning of a life dominated by type 1 diabetes started 5 years ago when their oldest son, Patrick, then 12 years old, was diagnosed with the disease.

"I was watching him lose weight, and as a mother, I knew something was wrong," Ellen said. "I even asked Dave, 'do you think it could be diabetes?'"

As fate would have it, the family was on vacation when Ellen came upon a 1974 edition of Life Magazine at a flea market. The cover story just happened to be "Does Your Child Have Diabetes?" It was all the impetus she needed. As soon as the vacation ended, Ellen brought Patrick to their pediatrician where a blood test revealed that he had the disease. "Patrick's diagnosis came as a complete shock," Ellen said. "There's no history of diabetes in Dave's or my families."

Eighteen months later, their daughter Sarah, then 6 years old, began losing weight and urinating frequently at night. She too was diagnosed with type 1 diabetes. "Sarah took it very cavalierly, just like a trooper," says Ellen. But when Ellen and Dave told Patrick of his sister's diagnosis, "he just broke down and cried. Since having been diagnosed, Patrick had always dealt with his diabetes well and never really complained. But at that moment we knew how bad it was for him," said Ellen.

Shortly after Sarah was diagnosed, the family's endocrinologist told them about TrialNet, in which researchers were looking for children whose siblings had type 1 diabetes to see if other children in the family were at risk for developing the disease.

Dave said that at first he and Ellen didn't want to have their other children screened for the disease. "We just didn't want that cloud hanging over our heads," Ellen added. However, the more they thought about it, the more they began to realize that "maybe we can learn something from this. We also felt strongly that we needed to be part of this search for a cure, and the more we thought about it the more enthusiastic we became," said Dave. It was through a TrialNet screening that the Goulds learned that then 10-year-old Sam also had type 1 diabetes.

Participating in a Clinical Trial To Prevent Type 1 Diabetes

A clinical trial being conducted by TrialNet is building on the results of a previous NIDDK-supported clinical trial, called the Diabetes Prevention Trial-Type 1 (DPT-1). The DPT-1 studied whether injected or oral insulin administration could prevent or delay type 1 diabetes in persons at high- or moderate-risk for the disease. While the DPT-1 did not find an overall protective effect of injected or oral insulin, a subset of trial participants who had higher levels of a certain predictive marker of the disease (insulin autoantibodies) seemed to benefit from oral insulin treatment, though this result was not definitive. TrialNet is now building on these observations, and has launched a clinical trial to determine if oral insulin therapy could prevent the disease in people with elevated insulin autoantibodies.

Through a TrialNet screening, the Goulds learned that 4-year-old Oliver had elevated levels of insulin autoantibodies, which made him eligible to enroll in the TrialNet oral insulin prevention study. The Goulds enrolled Oliver into the study, which randomly assigns participants to receive either an insulin pill or a placebo (inactive pill without insulin). Those participating in the

trial do not know whether they are getting the insulin or the placebo. This randomization allows researchers to compare the two groups to determine if oral insulin could prevent or delay the development of type 1 diabetes.

Oliver has since developed type 1 diabetes—the fourth of the Goulds' children to be diagnosed with the disease—and, until the study is over, the family will not know whether he received insulin or placebo. "When we decided to enroll Oliver in the study, friends would ask, 'if you don't know whether he's receiving insulin or placebo, why did you enroll him?'" said Dave. "Ellen's and my response to them is: that's what research is. You have to be willing to accept that when you get into a study like this." He quickly added that, "We would be ready, willing, and able to do it all again. The best thing about TrialNet is that it's helping all of us move closer to preventing or delaying type 1 diabetes."

Likewise, significant knowledge will be gained no matter the outcome of the trial—it is only through a rigorous clinical trial that researchers definitively learn which therapies work and which ones don't. When effective therapies or preventative approaches are found, other patients and people at risk can benefit from them. If a potential intervention turns out to be ineffective, then scientists know to explore other avenues to find therapies that work. It is thanks to the dedication of the Goulds and other families that this important new knowledge can be gained.

"When we decided to enroll Oliver in the TrialNet study, friends would ask, 'if you don't know whether he's receiving insulin or placebo, why did you enroll him?'" said Dave. "Ellen's and my response to that is: that's what research is. You have to be willing to accept that when you get into a study like this." He quickly added that, "We would be ready, willing, and able to do it all again."

Fortunately, the Goulds' other four children—Maggie, Annie, Nicholas, and Andrew—so far have tested negative for early markers of type 1 diabetes. But that doesn't mean that they are not affected by the disease. According to Ellen, 3-year-old Annie asks "When I am I going to get diabetes?" and 2-year-old Maggie tries putting Patrick's glucose meter on her finger to test herself. Describing his perspective on this disease, 13-year-old Nicholas said, "I'm really glad I don't have it. I see what my brothers and sister have to go through every day. I try to help as best I can, but I'm worried about them."

But Dave lays claim to being the family's ultimate worry-wart. "Ever since Sam's low blood sugar episode, I'm up with every bump I hear during the night, checking their bedrooms."

Through another study being conducted by TrialNet—the Natural History Study—the Goulds' four children who don't have type 1 diabetes will continue to be screened annually. For the four who do have the disease, the best news to date is that they are all doing well and show no signs of complications from the disease.

"When I was first diagnosed," Patrick said, "I got a note from someone in the Juvenile Diabetes Research Foundation, and it said 'Hang in there. There's a cure coming. Take as good care of yourself as you can; you're not going to have to do this much longer.' My message to others with type 1 diabetes is the same: There's a cure coming. Hang in there."

As for his mother's testifying in front of Congress with her urgent message for finding a cure for her children and all the others who must deal with type 1 diabetes every minute of every day, Patrick said: "She was awesome!"

For information about participating in Type 1 Diabetes TrialNet, please call 1-800-HALT-DM1 or visit www.diabetestrialnet.org

EMERGING RESEARCH OPPORTUNITIES RESULTING FROM THE *SPECIAL DIABETES PROGRAM*

The *Special Statutory Funding Program for Type 1 Diabetes Research* has fueled the emergence of a wide range of research opportunities. These opportunities were identified in a strategic planning process as being critically important for overcoming current barriers and achieving progress in diabetes research. Key questions and research opportunities relevant to type 1 diabetes, including those related to type 1 diabetes prevention and reversal, are outlined in Appendix F.