

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 seek to identify and test novel type 1 diabetes prevention and reversal strategies.

The *Special Statutory Funding Program for Type 1 Diabetes Research* has sparked a major expansion of research efforts aimed at preventing or reversing type 1 diabetes. 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. However, in susceptible individuals, these disease-causing T cells initiate an inflammatory process in the pancreas that eventually leads to the destruction of beta cells. The other arm of the immune system—the B cells—produces antibodies that also recognize beta cell proteins. 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 type 1 diabetes patients 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.

Attempts to turn advances in understanding the autoimmune basis for type 1 diabetes into a cure have been made for nearly three decades. Importantly, more progress has been achieved in the last 5 years than in the previous 25 years combined, due largely to support by the *Special Funding Program*. Until greater knowledge of genetic and environmental causes of type 1 diabetes is achieved, strategies to prevent or reverse the disease are currently focused on intervening in the immune system’s misguided assault. These strategies must be two-pronged: they must squelch autoimmunity in those who are at risk for or already have the disease, while maintaining or restoring the patient’s own insulin producing capacity. (Goal III addresses another approach for reversing the disease by transplanting insulin-producing cells obtained from donor pancreatic tissue or regeneration of beta cells.)

The immune system provides critical protection against infection, so it is vital that any treatment that modifies its activities is as selective as possible in damping down autoimmunity, while leaving the protective aspects of the immune defense system intact. This delicate balancing act will be achieved by leveraging knowledge about the immune system in general, combined with insights into disease causation, to the development of new treatments.

The *Special Funding Program* supports research consortia and clinical trials networks that promote: collaboration between basic and clinical investigators; delivery of therapeutic agents from the bench to the bedside; improved measurements of disease markers to facilitate the conduct of clinical trials; identification of novel therapeutic strategies; and testing of promising therapeutic agents in people. Notably, the *Special Funding 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 (e.g., immune system function, beta cell biology, genetic and environmental causes), more strategies for disease prevention and reversal will be identified, which will feed into this critically important pipeline made possible by the *Special Funds*.

HIGHLIGHTS OF SCIENTIFIC PROGRESS

While numerous significant advances have emerged since the beginning of the Special Funding Program, many of the research efforts to prevent and reverse type 1 diabetes are still in progress, and the full impact of these projects will not be realized for several years. The advances made possible by the Special Funding Program thus far are therefore only the beginning of the scientific gains that can be expected in the future.

Identification of Insulin as a Possible Disease-

Initiating Autoantigen in Type 1 Diabetes: For years, researchers have struggled to determine which beta cell proteins are key targets of autoimmune attack. Findings in a mouse model of type 1 diabetes, supported by the Autoimmune Disease Prevention Centers, as well as research in humans, now support the notion that the insulin molecule itself is an important, potentially disease-initiating autoantigen. Additionally, other studies have recently identified antigenic targets of the cellular immune response in non-obese diabetic (NOD) mice (islet specific glucose-6-phosphatase catalytic subunit related protein and dystrophin myotonia kinase). There is also continuing interest in the potential role that proteins of neuroendocrine origin may play in the disease in both human type 1 diabetes, as well as in animal models. To a large extent, many of these recent discoveries regarding autoantigen identification were dependent on the development of improved tools for characterizing the immune response associated with beta cell destruction, as well as on access to human tissues made available for research purposes.

Development of Outcome Measures for Clinical

Trials: In addition to immune markers, a variety of metabolic markers and their associated tests have also proven valuable to studies of human type 1 diabetes. Long-term studies of type 1 diabetes patients have shown that preservation of the ability to make even small amounts of insulin is strongly

associated with improved control of blood glucose, less hypoglycemia, and reduced risk of eye, kidney, and other diabetes complications. Endogenous insulin production can be determined in patients requiring insulin treatment by measuring C-peptide, a byproduct of insulin production which is co-secreted from the beta cell with insulin. Particularly notable are the recent improvements in the ability of researchers to determine the metabolic activity of individuals with or at-risk for type 1 diabetes. These improvements (e.g., standardization of the C-peptide assay, information on how best to stimulate and characterize residual insulin production in type 1 diabetes patients on insulin therapy) were made possible by the *Special Funding Program*. The FDA is considering use of these measures, which would make clinical trials shorter and less expensive, as a basis for approval of new therapies.

Completion of the Diabetes Prevention Trial Type-1 (DPT-1) Oral Insulin Arm in Type 1 Diabetes TrialNet:

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 insulin autoantibodies seemed to benefit from oral insulin treatment, though this result was not definitive. TrialNet is planning a trial to confirm the suggested benefit of oral insulin therapy in people with elevated insulin autoantibodies.

Tolerance and Regulation of the Immune System:

Recent studies of animal models have provided insights into type 1 diabetes, such as ascertaining the molecular and cellular defects that underlie the failure to maintain tolerance to beta cells; and identifying immune system cells that are key to regulating tolerance in type 1 diabetes. It is important to note that currently, many of these disease aspects can only be addressed through studies of animal models due to issues of both practicality and technical ability—providing but one of many examples of the importance of animals to type 1 diabetes research. Progress toward understanding tolerance and regulation of the immune response in human type 1 diabetes has also occurred, implicating defects in many cell types as potentially causative in autoimmune disorders such as type 1 diabetes. Similarly, several genes have been identified that contribute to susceptibility to autoimmune disorders because of their ability to modify immune reactivity.

Advances in Preventing or Reversing Type 1 Diabetes:

Recent years have seen much excitement about possible treatment strategies stemming from proof-of-principle

experiments in animal models. These include, but are not limited to: anti-CD3, which depletes and/or modifies the function of T cells; CTLA4-Ig, which antagonizes immune activation, e.g., “costimulatory blockade;” and anti-thymocyte globulin, which also depletes T cells. In addition, research on immunosuppression associated with islet transplantation efforts, as described in Goal III, also contributes to the identification of agents that could be used to control autoimmunity in the setting of disease prevention or reversal. Those agents that demonstrate adequate safety profiles have and will continue to move forward in human type 1 diabetes clinical trials, through such programs as Type 1 Diabetes TrialNet or the Immune Tolerance Network. Anti-CD3 is one example of an agent that has seen experimental translation from animal models to investigations in humans. Two research trials of anti-CD3 reported the ability of this agent to preserve metabolic function when administered to people with recent onset type 1 diabetes. With time, it is hoped that this or other agents will become proven components of a cure for type 1 diabetes by promoting disease reversal.

EVALUATION OF MAJOR RESEARCH CONSORTIA, NETWORKS, AND RESOURCES RELATED TO PREVENTING AND REVERSING TYPE 1 DIABETES

With the increase in Special Funds that became available in FY 2001, unique, innovative, and collaborative research consortia, clinical trials networks, and resources for the diabetes research community were launched. This section evaluates the progress of these ongoing efforts thus far and describes the impact that the efforts have already had—and have the potential to have—on type 1 diabetes patients.

Type 1 Diabetes TrialNet (TrialNet)

TrialNet is an international network of investigators, clinical centers, and core support facilities that recruits patients and conducts research to advance knowledge about type 1 diabetes and to test strategies for its prevention and early treatment. TrialNet supports the development and implementation of clinical trials of agents aimed at preventing the disease in at-risk patients and slowing the progression of type 1 diabetes in new onset patients. The network's "Natural History Study" will enhance understanding of how the disease develops in individuals at risk and will thus help in the formulation of future trials. Biological samples collected from study volunteers are being stored at the NIDDK Central Repository, and these valuable resources will be made available to the broader scientific community for further research on type 1 diabetes.

Highlights of Progress

The progress that TrialNet has made as of March 1, 2006, includes:

- *Completed the Diabetes Prevention Trial Type-1 (DPT-1) clinical trial of insulin for the prevention of type 1 diabetes in individuals at moderate and high risk for disease development:* The trial showed that oral or injected insulin administration did not delay or prevent the disease in relatives of type 1 diabetes patients. However, in a subset of the moderate risk patients studied (those with high titers of insulin-reactive autoantibodies), protection may have been observed. Because this result was not definitive, TrialNet is developing an additional study to further evaluate the role of oral insulin in delaying or preventing type 1 diabetes in this subset (scheduled for launch in late 2006).
- *Launched the Natural History Study:* This trial was begun to identify risk factors for type 1 diabetes and document disease characteristics and progression. The Natural History Study will also identify and maintain a pool of individuals who would be candidates for participation in clinical trials. The first phase of the Natural History Study involves identification of those at risk by using a blood test for the presence of diabetes-related autoantibodies to screen close relatives of people with the disease. Thus far, over 13,100 individuals have been screened. Of these, over 300 were found to have positive autoantibodies (indicating increased type 1 diabetes risk) and are undergoing evaluation at regular intervals to monitor for signs of progression to type 1 diabetes. The study expects to increase to a rate of screening of about 20,000 individuals per year. Participants will be offered enrollment in diabetes prevention and early intervention studies as they become available.
- *Launched the Mycophenolate Mofetil-Daclizumab (MMF/DZB) Clinical Trial:* This trial will test whether two immunosuppressive agents, MMF and DZB, will stop the ongoing destruction of any beta cells that are still functioning in new onset type 1 diabetes patients, and if the combination of the two drugs is superior to MMF alone in this regard. Type 1 diabetes results from progressive autoimmune destruction of beta cells. Preservation of remaining beta cells at disease onset is clinically important, because the ability to secrete even small amounts of insulin can make the disease easier to

control and help minimize complications associated with years of inadequate glycemic control. To date, about half of the required participants have been enrolled.

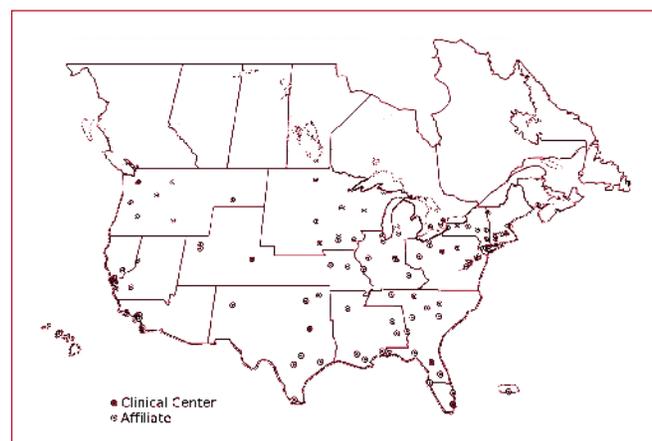
- *Completed recruitment for a clinical study to compare reliability of two tests for beta cell function—the Mixed Meal Tolerance Test (MMTT) and intravenous Glucagon Stimulation Test (GST):* The study has met its recruitment target; 138 patients completed the study. Residual beta cell function (insulin secretion) in patients with type 1 diabetes is known to result in improved glycemic control, reduced hypoglycemia, and reduced risk for complications. This beta cell function is currently best measured by determining levels of C-peptide. C-peptide is useful as an outcome measure in clinical trials: for example, those testing agents to preserve beta cell function in new onset diabetes. There are different ways to stimulate insulin production and, concomitantly, C-peptide production, but it has not been clear which of these conditions is optimal for enabling C-peptide measurement. The MMTT/GST clinical trial compared the reliability and burden on patients of two test conditions for stimulating insulin/C-peptide: one, MMTT, is a liquid meal; the other, GST, is an injection of the hormone glucagon. Results of this study, soon to be reported, will inform the design of future type 1 diabetes clinical trials to prevent or reverse type 1 diabetes in which C-peptide must be measured to determine if the intervention is successful.
- *Launched the T cell Validation Study:* The purpose of this study is to learn which T cell assays are most reliable and reproducible in identifying differences between people with and without type 1 diabetes. This study will facilitate further research toward understanding how type 1 diabetes occurs. Thus far, about half of the required participants have been enrolled toward a goal of 60-100 (30 with diabetes, 30-100 controls).
- *Designed a study to test the effects of the agent “rituximab” (anti-CD20) on progression of type 1 diabetes in new onset patients for launch later in 2006:* This study addresses the role of B cells in the autoimmune destruction of beta cells. In type 1 diabetes, B cells produce antibodies directed against components of the beta cell. Although B cells and autoantibodies do not directly attack insulin-producing cells, it is thought that they may exacerbate such an attack by certain T cells. Rituximab is approved by the FDA for use in other autoimmune diseases. This study will investigate the therapeutic potential of rituximab in type 1 diabetes by investigating whether it can help lower the number of immune B cells in newly diagnosed type 1 diabetes patients and thereby prevent destruction of insulin-producing beta cells that remain at diagnosis.
- *Designed a pilot study to test the role of omega-3-fatty acids in preventing type 1 diabetes:* The Nutritional Intervention to Prevent (NIP) Diabetes Study, for launch later in 2006, is based on observations from epidemiologic studies that children who have received more omega-3 fatty acid (such as from fish)—either in the womb or during the first year of life—have a lower risk of developing type 1 diabetes. This pilot feasibility study will enroll either infants or pregnant women and will randomly assign them to one of two groups: (1) daily omega-3 fatty acid supplements; or (2) placebo (no supplements). During the course of the study, the participants will undergo assessments for immunological markers of type 1 diabetes. Upon fulfillment of the study’s objectives, researchers plan to launch a full-scale trial to test if omega-3 fatty acid supplementation can prevent type 1 diabetes.
- *Designing a study to further evaluate the role of oral insulin in delaying or preventing type 1 diabetes in a subset of the population studied in DPT-1:* See first bullet; expected launch in late 2006. Extensive standardization of antibody measurement is under way to optimize the conduct of this study.

- **Designing Anti-CD3-Exenatide Clinical Trial:** The immunomodulatory agent anti-CD3 has been shown to slow the loss of metabolic function in newly diagnosed type 1 diabetes patients. However, this effect wanes over time. This trial will test whether anti-CD3, combined with exenatide, a newly approved drug for type 2 diabetes that increases insulin production and shows the added benefit of preserving or increasing beta cell mass in animal models, will slow or stop the ongoing destruction of residual beta cells in new onset type 1 diabetes patients. Recruitment will begin in late 2006. This combination is also being considered for a prevention study in relatives of type 1 diabetes patients who are at high risk for developing the disease.

Anticipated Outcomes

TrialNet is an international clinical research network focused on individuals at risk for or newly diagnosed with type 1 diabetes. Its efforts span the time period from birth in those at high genetic risk to the development of signs of increased risk (for example, autoantibodies), when prevention strategies are particularly urgent, and on through the time soon after diagnosis, when residual beta cell function may afford a unique opportunity for interventions to mitigate disease severity. TrialNet will test the ability of agents to slow or prevent type 1 diabetes, sparing those at risk from developing this devastating disease. TrialNet is also studying agents that can modulate the immune system of recently-diagnosed patients so as to preserve remaining beta cell function and thus make it easier for them to control glucose levels and reduce their burden of complications. In addition to the conduct of clinical trials, the network's extensive recruitment of individuals at risk or with new onset disease is facilitating research into disease progression. Other clinical studies conducted by TrialNet will improve the methodologies used in future type 1 diabetes clinical trials, for example, to assay T cells involved in autoimmune attacks and to stimulate C-peptide production as a measure of residual beta cell function. The infrastructure of TrialNet is also used to enhance other efforts supported by the *Special Funding Program*, such as aiding the Type 1 Diabetes Genetics Consortium with identification of families with two siblings affected with type 1 diabetes and collection of samples for genetic studies from these families.

There is a rigorous process for consideration of studies proposed for conduct through this coordinated clinical research infrastructure, which involves review by experts in diabetes and immunology. As new therapeutic agents are identified through additional studies supported by the *Special Funding Program*, TrialNet's standing infrastructure will be indispensable for the testing of these promising agents in patients. Furthermore, the knowledge gained from TrialNet's Natural History Study will help to spur the design of new prevention and treatment approaches. TrialNet's current position of strength is the result of years of effort in outreach to the diabetes care and research communities, intensive training in



Type 1 Diabetes TrialNet is a network of 18 Clinical Centers working in cooperation with affiliated sites throughout the United States, Canada, Finland, United Kingdom, Italy, Germany, Australia, and New Zealand. This map shows the broad distribution of TrialNet sites throughout the U.S. and Canada. (Image courtesy of Type 1 Diabetes TrialNet.)

research procedures, including sample collection and storage for mechanistic assays (in collaboration with the Immune Tolerance Network), and the establishment of close collaborative ties among clinical diabetes and immunology researchers. TrialNet investigators also take proactive roles in critically reviewing, identifying, and prioritizing promising candidates for trials, considering both clinical feasibility and scientific merit.

External Evaluation by Expert Panel

To supplement evaluation and guidance by TrialNet's Data Safety and Monitoring Board, leading scientific and lay experts were asked to evaluate the progress of TrialNet at an *ad hoc* planning and evaluation meeting convened by the NIH in January 2005 (see Appendix 3). Comments from the panel review included:

- ▶ The concept of a standing infrastructure to test promising therapeutic agents to prevent or slow the onset of type 1 diabetes is of critical importance.
- ▶ TrialNet's completion of the DPT-1 was a significant achievement.
- ▶ It is important to conduct studies that will add scientific knowledge to the field of type 1 diabetes research, even if results are negative.
- ▶ The panel discussed several factors that have limited the rapidity with which TrialNet implements new protocols, including: (1) expansion from the U.S. to international sites; (2) effects of decentralized leadership on management complexity; (3) revised screening procedures; (4) large number of required patients; and (5) current limited availability of therapeutic agents to test.
- ▶ Panel members commended TrialNet for critically analyzing protocols before they are approved and implemented; it is not appropriate to test proposed agents solely because the TrialNet infrastructure exists.

- ▶ Caution in selecting proposed agents for testing should be balanced with the importance of promoting the testing of agents that potentially may be a "breakthrough" in the prevention or treatment of type 1 diabetes.

Actions Taken in Response to Expert Panel Recommendations

TrialNet took the following actions in response to recommendations of the expert panel at the *ad hoc* planning and evaluation meeting convened by the NIH in January 2005:

Recommendation: Increase the Number of Protocols

- ▶ TrialNet currently has one completed protocol, three active protocols, and four more scheduled for launch later in 2006. Two additional studies are nearing launch with the Immune Tolerance Network (ITN) as the lead, but with full partnership and recruitment participation from TrialNet. These studies will bring TrialNet near its goal of providing a therapeutic opportunity for nearly every category of diabetes risk.

Recommendation: Identify Highly Innovative Projects

- ▶ TrialNet has implemented a new process for strategizing and prioritizing the best ideas for implementation. The "Strategies and Prioritization" Committee has been established, which includes top scientists in the diabetes field both within and outside of TrialNet. This Committee considers the universe of ideas, whether formally submitted to TrialNet or gleaned from the field.

Recommendation: Decrease Time Required for Protocol Implementation

- ▶ TrialNet has restructured and streamlined the protocol review and implementation process. A subgroup of representatives from the Executive Committee, including the TrialNet Chairman's office, the coordinating center, and the NIDDK, met numerous times to evaluate the procedural roadblocks and to develop a better process. The new process has shortened the review of proposals considerably, as subcommittees now review proposals based on scientific merit, clinical feasibility, ethics, strategy, and prioritization.

Recommendation: Create a More Rigorous Mechanism To Critically Assess the Scientific Rationale for and Aid in Prioritization Among Agents Proposed for Study

- ▶ TrialNet has established a rigorous Scientific Review Committee, including members outside of TrialNet. This Committee also considers strategies to improve the innovation and impact of the proposals.

Recommendation: Institute an Advisory Group Consisting of External Scientists with Expertise in Both Basic and Clinical Research

- ▶ TrialNet is establishing an External Advisory Committee (EAC) as recommended, which will meet in 2006. In addition, the NIDDK has undertaken to critically review the operations of TrialNet, starting with a review of the Data Coordinating Center on March 2-3, 2006, by a panel of experts in the operation of multicenter clinical trials. The recommendations of this panel regarding the TrialNet Data Coordinating Center will be shared with the EAC. The NIDDK has also appointed a panel of advisors to review TrialNet's procedures and policies for the implementation of mechanistic studies and mechanistic sample collection and distribution. This panel is scheduled to

meet in April 2006, and its recommendations will also be shared with the EAC.

Recommendation: Enhance Collaborations Between TrialNet and the Immune Tolerance Network (ITN)

- ▶ TrialNet is committed to working together with ITN at every level. Leadership from both networks have had numerous meetings to discuss joint recruiting, comparable reimbursement of recruiting and clinical costs, prioritization of studies, and sharing data and mechanistic assay policies and procedures. ITN and TrialNet will directly partner on two studies in which ITN takes the lead role. In addition, ITN and TrialNet will work together to implement immunophenotyping using common reagents and a core facility, and for consistency of quality control and data analysis (see section on "Actions Taken in Response to Expert Panel Recommendations" under the ITN later in this chapter for additional examples of collaboration).

Ongoing Evaluation

Ensuring continued and ongoing evaluation of TrialNet's study design and progress are a Data and Safety Monitoring Board (DSMB), Steering Committee, and other TrialNet committees, as well as external scientific experts. TrialNet protocol (study) proposals are evaluated by the TrialNet Steering Committee, and relevant protocols are also evaluated by the ITN Steering Committee. The DSMB reviews all TrialNet protocols and informed consent materials and must approve them before implementation. Multiple subcommittees within TrialNet have also been established to address relevant issues. To critically assess the scientific rationale and aid in prioritization among agents proposed for study, TrialNet has established a rigorous Scientific Review Committee, including members external to the TrialNet network. This Committee also considered strategies to improve the innovation and

impact of the proposals. TrialNet is also establishing an External Advisory Committee that will have its first meeting later in 2006, following earlier external advisory committee assessments of specific components of TrialNet, such as the function of the Data Coordinating Center.

Coordination with Other Research Efforts

TrialNet coordinates its efforts with multiple other type 1 diabetes research consortia and networks supported by the *Special Funding Program*. Collaboration, coordination, and resource sharing serve to synergize research efforts and accelerate research progress. Examples of coordination with other consortia are given below. For a full description of ongoing collaborative efforts, please see Appendix 2.

Coordinating Patient Recruitment Efforts:

- ▶ TrialNet and the ITN jointly introduced and advertised the TrialNet Natural History Study and the ITN Insulin Vaccine Study.
- ▶ The SEARCH for Diabetes in Youth study is helping TrialNet recruit eligible participants.
- ▶ All 14 North American TrialNet centers are participating as recruitment centers for the Type 1 Diabetes Genetics Consortium (T1DGC) North American Network.
- ▶ T1DGC assisted TrialNet in establishing international recruitment sites.
- ▶ TrialNet, The Environmental Determinants of Diabetes in the Young (TEDDY), and the Trial To Reduce IDDM in the Genetically At-Risk (TRIGR) have coordinated recruitment efforts to ensure that they are not adversely competing for patient participants in their studies.

Enhancing Data Comparison Among Studies:

- ▶ TrialNet, TEDDY, and T1DGC share the same North American laboratory for measurement of autoantibodies. This coordination will permit direct comparison between

results obtained in each study.

- ▶ TrialNet uses laboratories certified through the HbA1c standardization program.
- ▶ The C-peptide Standardization Program included two laboratories from TrialNet in an international comparison effort, the results of which illustrated the need to identify and minimize the major sources of variation in C-peptide measurements in multicenter, multi-laboratory clinical studies.
- ▶ The Diabetes Autoantibody Standardization Program (DASP) provides tools that TrialNet laboratories use to standardize autoantibody data.
- ▶ T1DGC, TrialNet, SEARCH, and TEDDY are all sharing either the same laboratories or laboratory reagents to analyze genetics data. This coordination will permit comparisons of genetics data across all four studies, effectively increasing the power of each in learning which genes play a role in disease onset.

Sharing of Other Resources and Information:

- ▶ TrialNet uses services of the ITN, such as biological sample preparation and processing and certification of laboratory coordinators, for the MMF/DZB clinical trial, the anti-CD20 clinical trial, the T Cell Validation Study, and the Natural History Study. These ITN services will be used for most TrialNet protocols currently in development.
- ▶ Protocols potentially of interest to ITN and TrialNet are considered by both consortia to assess the possibility for joint sponsorship.
- ▶ TrialNet and ITN use a common DSMB.

Coordinating Research Studies Involving Newborns:

- ▶ TrialNet investigators meet with investigators participating in other type 1 diabetes research studies involving newborns (TEDDY and TRIGR) to discuss opportunities for enhancing coordination and collaboration.

- ▶ TEDDY has shared the following materials with TrialNet investigators who are studying newborns in the NIP Diabetes Study: genetics-screening procedures, data forms, and parts of the Manual of Operation concerning follow-up of high-risk children. Through concerted action to define exclusive study geographic areas, investigators in the two studies have also avoided direct competition for eligible study participants.

TrialNet Administrative History

Date Initiative Started	2001
Date <i>Special Program</i> Funding Started	2001
Participating Components	NIDDK, NIAID, NICHD, NCRR, JDRF, ADA
Website	www.diabetestrialnet.org

TrialNet consists of 18 centers in North America, Europe, Australia, and New Zealand working in cooperation with 66 additional patient recruitment sites.

Immune Tolerance Network (ITN)

The ITN is an international consortium of over 70 scientists and physicians dedicated to evaluating therapies to reduce autoimmunity and other adverse immune responses by inducing, maintaining, and monitoring “tolerance” in humans for islet, kidney, and liver transplantation; autoimmune diseases; and allergy and asthma. The goal of immune tolerance research is to identify strategies to reprogram the immune system in a highly specific way to prevent or inhibit disease-causing or aberrant immune responses. Such damaging autoimmune processes include those that destroy insulin-producing beta cells in type 1 diabetes, or the immune response that destroys transplanted islets. It is important, however, that these strategies not dampen the body’s normal disease-fighting immune mechanisms. Investigators within and outside of the ITN are invited to submit clinical trial proposals for review. The ITN then assists investigators with study development, monitoring, and analysis; access to cutting-edge technologies; and a wide range of other expert scientific and technical support. Clinical trials are augmented by mechanistic studies designed to uncover basic biological features of clinical tolerance which will, in turn, help guide the design of future clinical trials.

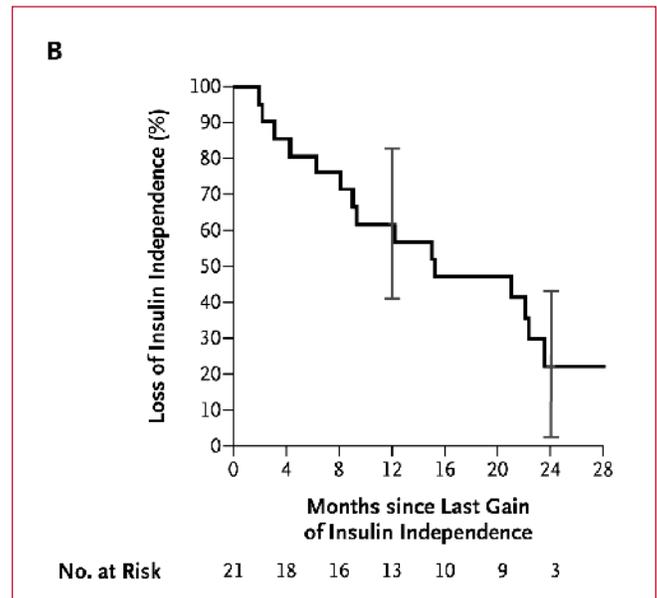
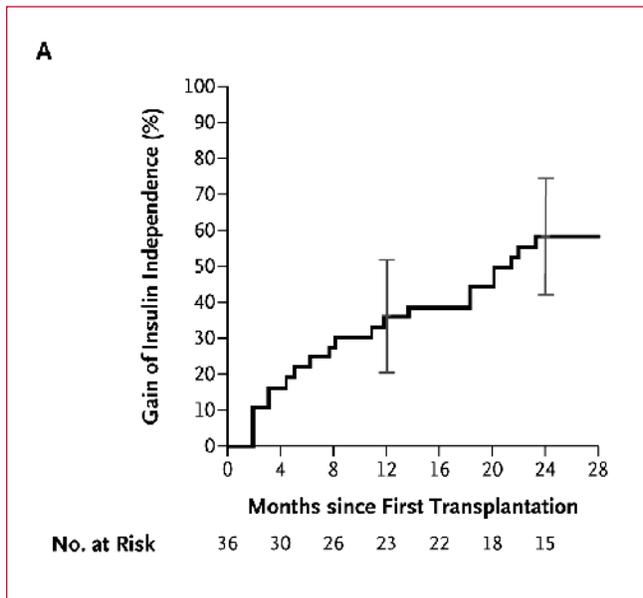
Highlights of Progress

The progress that the ITN has made as of March 1, 2006, includes:

- *Conducted first multicenter trial of islet cell transplantation:* Nine sites in North America and Europe have successfully replicated the “Edmonton protocol” for islet transplantation in the ITN’s multicenter study from 2001-2006. Islet transplantation is still an experimental treatment for type 1 diabetes. (For extensive information on islet transplantation, see Goal III.) The Edmonton protocol was a revolutionary new procedure developed in Canada that greatly improved the outcomes for islet transplantation. As of 1 year post final transplant, 16 of the 36 (44 percent) enrolled participants in the ITN trial have achieved insulin independence with good glycemic control; 5 of 36 patients achieved insulin independence with a single donor islet infusion. Twenty-four of 36 patients remain C-peptide positive (a measure reflecting beta cell function) but continue to require small doses of insulin. Insulin independence declined over time in study participants. Importantly, even among patients who still required insulin injections, the presence of functioning transplanted islets led to an absence of severe hypoglycemic events due to hypoglycemia unawareness. The results of this study confirm and extend the demonstration that islet transplantation may become an alternative to whole pancreas transplantation. They also highlight the continued need for safer, more tolerable anti-rejection therapies. This effort has also established a network of qualified investigators and centers for future islet transplantation studies and will serve as a baseline for future tolerance studies.
- *Determined that autoantibody titers may predict islet transplant success:* Among the aberrant immune processes that occur in type 1 diabetes is the production of “autoantibodies” that recognize beta cell components. Autoantibody levels were measured pre-transplant in patients enrolled in the ITN multicenter study of the Edmonton Protocol. Investigators found that pre-transplantation levels of autoantibodies to two beta cell proteins correlate indirectly with long-term graft survival and insulin-free status following the transplant. If confirmed, this result may lead to the development of biomarkers of graft survival. It also underscores the need to abrogate both the immune system’s reaction to transplanted donor cells and the ongoing autoimmune response.
- *Supported research on a potential therapeutic agent for new onset type 1 diabetes, called the “hOKT3gamma1(Ala-Ala) monoclonal antibody,” or “anti-CD3” monoclonal antibody:* Residual beta cell function in type 1 diabetes patients is

associated with improved glycemic control and reduced hypoglycemia and risk for complications. Thus, it would be greatly beneficial to be able to blunt autoimmunity before all beta cells are destroyed. The anti-CD3 monoclonal antibody being studied is a genetically-engineered antibody that recognizes T cells, a type of immune cell involved in the autoimmune attack. Results from an ITN pilot study confirmed previous work showing that a single course of treatment with the anti-CD3 monoclonal antibody reduced insulin requirements for glycemic control during 18 months of follow up. In addition, this treatment attenuated the decline in patients' endogenous insulin production (as assessed by C-peptide levels in response to the MMTT). The ITN has also launched a larger, open-label, phase II study. As of February 2006, the study had recruited 7 of a planned 81 patients to investigate whether a second course of this anti-CD3 monoclonal antibody administered 1 year after the first treatment, with standard diabetes management, is able to have prolonged or improved effects in people with recently diagnosed type 1 diabetes compared to standard diabetes management alone.

- *Demonstrated that a combination of assays detects type 1 diabetes with high sensitivity and specificity:* ITN investigators showed that no single assay (such as an autoantibody test or any of several other types of assays) distinguishes normal individuals from those with type 1 diabetes. However, the combination of an autoantibody test and two types of assays for T cells identified a high proportion of patients with type 1 diabetes with no false positives. Additional patients are being recruited for this study for further optimization of these techniques. With refinements, assays such as these will play an important role in large-scale screening efforts to identify individuals at risk for development of type 1 diabetes, but who lack first-degree relatives with this disease.
- *Began a study to determine the safety of a potential diabetes antigen-specific immunotherapy, incorporating the insulin B chain, in patients with new onset type 1 diabetes:* Insulin is one of several beta cell proteins recognized by the immune system in type 1 diabetes. It has been hypothesized that treatment with insulin may arrest or slow ongoing autoimmunity in type 1 diabetes so as to preserve beta cell function. ITN-supported scientists have completed enrollment of 12 participants in a study, with follow-up through March 2007. This double-blinded, phase I/II pilot study was designed to examine the safety of a potential insulin B-chain peptide. The study was unblinded in December 2005 to evaluate the interim data in consideration of conducting a larger phase II study in the near future. To date, three participants have completed follow-up on the study. Results of the unblinded data are being reviewed by both the ITN and TrialNet.
- *Plan to launch trial testing thymoglobulin therapy in newly diagnosed type 1 diabetes patients:* Researchers plan to conduct an early phase II study of the safety and efficacy of treating new onset type 1 diabetes patients with an antibody to T cells (thymoglobulin) to determine if it can induce tolerance and thereby preserve beta cell function in these patients. The FDA placed this protocol on clinical hold in November 2005, pending modifications to the study design. The study team will revise the protocol, respond to the FDA's letter, and launch the trial for enrollment in Summer 2006.



The Immune Tolerance Network (ITN) conducted the first multicenter clinical study of islet transplantation using the Edmonton protocol. Panel A shows the interval between the first islet transplant and insulin independence (attained in 21 of 36 patients), and Panel B shows the subsequent loss of insulin independence among 16 of these 21 patients during the next 28 months. However, despite the loss of insulin independence, all patients with an islet transplant who continued immunosuppressive therapy maintained some insulin production. The partially functioning islet transplants protected patients from hypoglycemia. (Images courtesy of Dr. A.M. James Shapiro and reprinted from Shapiro, A.M. et al. *N Eng J Med.* 355: 1318-1330, 2006. Copyright © 2006 Massachusetts Medical Society. All rights reserved.)

Anticipated Outcomes

The efforts of the ITN are strengthening knowledge of the autoimmune response in type 1 diabetes, testing strategies for blocking destruction of patients' beta cells, and investigating approaches to improve success of transplantation of donor islets. ITN research on assays of the immune system to detect those at risk may help in mitigating the severity of disease onset. Once beta cell-preserving therapies are further developed and tested, improved identification of those at risk will permit more patients to begin early therapy. Research on tolerance-inducing agents brings hope of arresting the autoimmune destruction of beta cells; preservation of some insulin producing function would facilitate glucose control with less risk of hypoglycemia. For those who may undergo islet transplantation, modulation of the immune system is necessary, not only to block the diabetes-specific autoimmune

reactions that destroy beta cells, but also to prevent the general immune rejection that can occur with any transplanted tissue. When cells or organs are transplanted from a donor into a patient, the patient's immune system appropriately sees these as foreign (unless the donor is an identical twin). Consequently, immunosuppressive drugs are necessary to prevent transplant rejection. As another potential treatment strategy, scientists are exploring whether beta cells can be coaxed to regenerate to sufficient levels to restore greater insulin production. Such a treatment would also require blocking of the autoimmune response. However, long-term immunosuppression may carry increased risk of infections and certain types of cancer. Furthermore, many drugs that are effective in suppressing the immune system are actually harmful to beta cells. The ITN's research may lead to more specific drugs that blunt unwanted immune responses without impairing essential immune

functions or damaging beta cells. Thus, these efforts hold promise for improving the lives of patients with type 1 diabetes and for those at risk.

External Evaluation by Expert Panel

Leading scientific and lay experts were asked to evaluate the progress of the ITN at an *ad hoc* planning and evaluation meeting convened by the NIH in January 2005 (see Appendix 3). Comments from the panel review included:

- ▶ The goal of creating immune tolerance is critical to combating type 1 diabetes. Significant accomplishments and progress have been made.
- ▶ The ITN conducts studies that have the potential for long-term benefit to type 1 diabetes patients.
- ▶ Major strengths include the ITN's productive interactions with the transplant community and the emphasis on investigator-initiated studies.

Actions Taken in Response to Expert Panel

Recommendations

The ITN took the following actions in response to recommendations of the expert panel at the *ad hoc* planning and evaluation meeting convened by the NIH in January 2005:

Recommendation: Enhance Collaborations with TrialNet

- ▶ The ITN collaborates with TrialNet on the development and implementation of protocols in type 1 diabetes where both parties agree it is beneficial. The studies in which TrialNet and the ITN collaborate include: (1) Natural History Study; (2) MMF/DZB; (3) T Cell Validation Study; and (4) the Effects of Rituximab on the Progression of Type 1 Diabetes in New Onset Patients.
- ▶ The ITN supplies collection kits and training to laboratories for isolating peripheral blood mononuclear cells (PBMCs).
- ▶ The ITN provides RNA isolation on batched specimens.

- ▶ The ITN coordinates the transfer of frozen PBMCs, RNA, and plasma specimens to the NIDDK Repository from laboratories for studies, as applicable.
- ▶ The ITN coordinates the collection of blood and the transfer of samples from the clinical sites to the Flow Core Laboratory for analysis.
- ▶ Research staff from the ITN and TrialNet collaborate on joint studies, communicating daily and convening for 1 hour weekly to discuss critical site/study/technical issues. They also use this time to update each other regarding each Center's status in ongoing studies.
- ▶ The ITN and the TrialNet Coordinating Center participate in monthly meetings, which key study members attend to discuss the status of and any pending problems or issues with ongoing studies.

Recommendation: Share Resources with Researchers in Other Consortia

- ▶ The ITN's comprehensive website has publicly available information, such as protocols and descriptions of ongoing studies (www.immunetolerance.org).

Ongoing Evaluation

Several mechanisms exist to ensure continued and ongoing evaluation of the study design and the progress of the ITN. The ITN Scientific Review Committee is the major decision-making body that evaluates proposals for clinical and mechanistic studies. Recommendations are subject to final approval by the Network Executive Committee. Studies selected for implementation are developed by the Principal Investigator in collaboration with the ITN Clinical Trials Group, the Tolerance Assay Group, and industry partners. During protocol development and implementation, the NIAID provides regulatory, medical affairs, and project management support. Additionally, several external organizations, under contract with the NIAID, provide clinical monitoring, data management, and drug distribution services.

Coordination with Other Research Efforts

The ITN coordinates its efforts with multiple other type 1 diabetes research consortia and networks supported by the *Special Funding Program*. Collaboration, coordination, and resource sharing serve to synergize research efforts and accelerate research progress. Examples of coordination with other consortia are given below. For a full description of ongoing collaborative efforts, please see Appendix 2.

Coordinating Patient Recruitment Efforts:

- ▶ The ITN and TrialNet jointly introduced and advertised the ITN Insulin Study and the TrialNet Natural History Study.

Collaborating To Enhance Islet Transplantation Efforts:

- ▶ Islet Cell Resource Centers (ICRs) isolate and supply human islets for multicentered clinical study sites using the Edmonton Protocol.
- ▶ The Collaborative Islet Transplant Registry (CITR) archives trial results.
- ▶ The Clinical Islet Transplantation Consortium (CIT) and ITN are sharing expertise and coordinating efforts in the planning of immunologic assays in CIT trials. ITN core labs will be used for selected assays in CIT trials.
- ▶ The CIT, the ITN, and the Non-Human Primate Transplantation Tolerance Cooperative Study Group (NHPC-SG) are interested in using similar reagents for islet transplantation or as immune modulators for the treatment of type 1 diabetes.
- ▶ The NHPCSG and ITN share information about scientific priorities and interests for research planning.
- ▶ ITN priorities for pre-clinical testing of new therapeutics are considered in evaluating NHPCSG Opportunities Pool applications. Several ITN high-priority strategies are currently funded as pilot projects.

Sharing of Other Resources and Information:

- ▶ TrialNet uses services of the ITN, such as biological sample preparation and processing and certification of laboratory coordinators, for the MMF/DZB clinical trial and the Natural History Study. These ITN services will be used for most TrialNet protocols currently in development.
- ▶ Protocols potentially of interest to ITN and TrialNet are considered by both consortia with the consideration of joint sponsorship.
- ▶ TrialNet and the ITN use a common DSMB (more information on TrialNet and ITN collaboration can be found in the section titled: “Actions Taken in Response to Expert Panel Recommendations”).
- ▶ ITN-supported investigators have used the Type 1 Diabetes-Rapid Access to Intervention Development (T1D-RAID) program for provision and pre-clinical testing of novel reagents.
- ▶ TRIGR and the ITN are coordinating their efforts in the area of T cell assays.
- ▶ ITN researchers receive islets for basic research studies through the ICRs.

ITN Administrative History

Date Initiative Started	1999
Date <i>Special Program</i> Funding Started	2001
Participating Components	NIAID, NIDDK, JDRF
Website	www.immunetolerance.org

The ITN consists of over 70 world leaders in the clinical and basic science of immune tolerance from academic research institutions around the world.

Cooperative Study Group for Autoimmune Disease Prevention (Prevention Centers)

The Cooperative Study Group for Autoimmune Disease Prevention (Prevention Centers) engages in scientific discovery to advance knowledge toward the prevention and regulation of autoimmune diseases such as type 1 diabetes. The Prevention Centers aim to create improved models of disease pathogenesis and therapy to better understand immune mechanisms. These models will provide opportunities for prevention strategies and be used as validation platforms with which to test new tools applicable to human studies. They will also encourage core expertise and collaborative projects designed for rapid translation from animal to human studies—emphasizing the development of surrogate markers for disease progression and/or regulation, which could be used in the context of clinical trials. This research will help uncover new approaches for halting the development of autoimmune diseases prior to clinical onset by mechanisms other than global immunosuppression.

Highlights of Progress

The progress that the Prevention Centers have made as of March 1, 2006, includes the following advances. Researchers are building understanding of the cells, molecules, and pathways involved in the autoimmune destruction of beta cells and are revealing opportunities for the design of new therapeutic approaches.

- Identified insulin as a primary target for the autoimmune response in the NOD mouse model of diabetes: Mice have two insulin genes, and generation of a NOD mouse lacking the insulin 1 gene revealed that it is required for development of insulinitis and diabetes. Subsequent experiments showed that diabetes did not develop in NOD mice engineered to produce a slightly altered insulin molecule not recognized by the mouse's immune system. This research suggests that autoimmune reaction against insulin may be a critical initiator of the pathway toward beta cell destruction.
- Developed biological tools to identify certain types of T cells that can attack beta cells based on recognition of the beta cell protein GAD65: These tools are "MHC class II tetramers," which are constructed to contain a segment of the GAD65 protein. Researchers can use these tools to retrieve, quantify, and characterize GAD65-reactive T cells from patients and individuals at-risk for the disease. Such T cells are a potential marker of early disease, and this research will increase understanding about the destructive autoimmune response that underlies type 1 diabetes.
- Characterized functional properties of cells called "CD4+CD25+ regulatory T cells," which can help protect against autoimmune disease by suppressing the activities of the autoimmune-reactive T cells, as well as functional defects in this T cell subset in humans with autoimmune disease.
- Initiated the "NOD Roadmap" project to build understanding of type 1 diabetes through intensified research on the NOD mouse model of the disease: This research will involve creating a comprehensive description of genes that are turned on, proteins that are made, and functioning of the immune system in the NOD mouse during development of insulinitis (a condition preceding type 1 diabetes) and diabetes.
- Developed tools for using proteomics technology (which enables analysis of large numbers of proteins) to facilitate detection of autoantibodies and other markers of autoimmune disease.
- Determined mechanisms by which blockade of a particular molecular interaction between immune cells can prevent or modulate the course of diabetes and other autoimmune diseases: In these studies, scientists administered to mice an agent that blocked the interaction between two important molecules. One molecule, called CD154, exists on the surface of many T cells, and another molecule, called CD40, is present on other types of immune cells. One of their findings

was that blocking the CD154-CD40 interaction resulted in induction of a novel type of cell that is able to prevent the onset of type 1 diabetes in mice.

- Funded 48 pilot projects, including investigations to test hypotheses about the biology of type 1 diabetes, and projects to develop reagents and resources for further research.

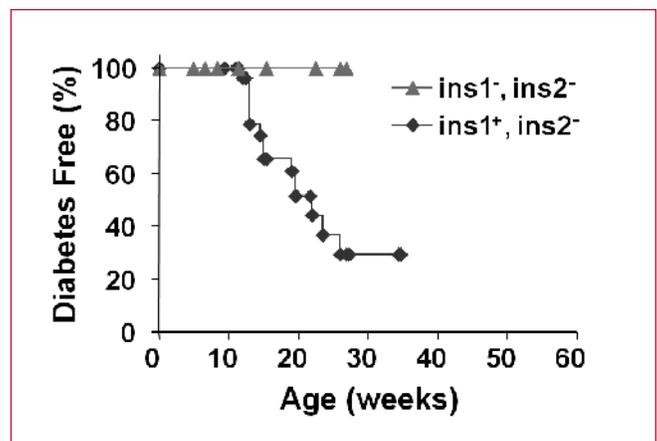
Anticipated Outcomes

The Prevention Centers support a multidisciplinary collaborative network of investigators focused on understanding the immune mechanisms that underlie autoimmunity and autoimmune diseases, approaches to modulation of the immune system, and the application of this knowledge toward the prevention of these chronic, debilitating diseases. The immune system is enormously complex, with the capacity to attack an extraordinary number of different types of substances. As the immune system targets and fights numerous types of infectious agents, it also produces cells and antibodies that recognize parts of the body, or “self.” Normally, the immune system employs mechanisms, not yet completely understood, for eliminating self-reactive components; this process is referred to as “tolerance.” In type 1 diabetes, however, the immune system goes awry and attacks insulin-producing beta cells. Any medical intervention to prevent type 1 diabetes should ideally be as selective as possible to squelch autoimmunity without impairing the immune system’s capacity for fighting infection. An example of one area of the Prevention Centers’ research is thus to identify and characterize cells of the immune system, such as certain types of T cells that attack and destroy the body’s beta cells. Another research area is to define the beta cell-derived molecules that are targeted for autoimmune attack. The Prevention Centers are also investigating how other aspects of the immune system, or experimental manipulations that alter the immune system, may confer protection against autoimmunity. Results of this research will open avenues for the design and testing of new therapeutic strategies for modulating the immune system to prevent autoimmune destruc-

tion and for developing markers for disease progression or regulation.

External Evaluation by Expert Panel

To supplement evaluation and guidance received from external advisors who attended the Prevention Centers’ 2005 all-investigator meeting, leading scientific and lay experts were asked to evaluate the progress of the Prevention Centers at an *ad hoc* planning and evaluation meeting convened by the NIH



Insulin is a key autoantigen in the development of type 1 diabetes: Mice have two insulin genes—insulin 1 (ins1) and insulin 2 (ins2). To address the role of insulin in type 1 diabetes development, researchers in the Cooperative Study Group for Autoimmune Disease Prevention genetically engineered mice to make a special insulin molecule that lowered glucose but was not recognized by the immune system, while at the same time, “knocking out” one or both of the mice’s own insulin genes. Mice that lacked both of their insulin genes did not develop diabetes (▲). In contrast, most of the mice that expressed ins1 developed diabetes (◆). These data suggest that insulin may be the critical initiator of the autoimmune destruction of insulin-producing pancreatic beta cells that leads to type 1 diabetes. (Figure courtesy of Dr. George Eisenbarth and adapted by permission from Macmillan Publishers Ltd: Nature. 435: 220-223, 2005.)

in January 2005 (see Appendix 3). Comments from the panel review included:

- ▶ This research effort is important to pursue because understanding autoimmunity in general is a cornerstone of type 1 diabetes research.
- ▶ Research supported through this Group is carried out through two major arms: (1) the five members; and (2) pilot and feasibility projects.
- ▶ This research Group has made progress toward all of its major goals.
- ▶ An interesting project is the “NOD Roadmap,” which aims to study the life history of the NOD mouse (a model for type 1 diabetes) from 1-20 weeks of age. The Group has made progress in this research endeavor.
- ▶ Expansion of regulatory T cells is a promising area for future investigation.
- ▶ Future research opportunities include: (1) increasing synergy by tackling large scientific projects; and (2) identifying ways to translate research studies from mice to humans.
- ▶ The panel stressed that the existence of a Cooperative Study Group is crucial to advancing the autoimmunity research field. Increased interaction among individual researchers in this Group would help to achieve synergistic scientific progress “over and above” what could be supported through regular investigator-initiated research projects.
- ▶ The panel endorsed the Group’s pilot and feasibility award mechanism as a venue to attract new research talent. Awards are made to participants within and outside the Group. The NIH and the Group should identify ways to advertise the pilot and feasibility program widely and make it available to the broader research community. For example, the Group could develop a website, and funding opportunities could be announced on relevant NIH websites.

Actions Taken in Response to Expert Panel Recommendations

The Prevention Centers took the following actions in response to recommendations of the expert panel at the *ad hoc* planning and evaluation meeting convened by the NIH in January 2005:

Recommendation: Promote Research in the Expansion of Regulatory T Cells

- ▶ Four new pilot projects on the expansion and function of regulatory T cells were initiated since January 2005. Three focus directly on the generation of regulatory T cells; one of these and the fourth project also include deeper mechanistic studies of regulatory T cell function.

Recommendation: Increase Sharing of Data and Information Between Prevention Centers

- ▶ The generation of complex amounts of data from the NOD Roadmap project emphasized to the individual Prevention Center investigators the importance of establishing data-sharing mechanisms. It has been agreed that these data will be initially hosted by the Barbara Davis Center website and mirrored at other locations. Further sharing of data and information will be facilitated by implementation of a dedicated Prevention Centers website in the next grant year (see below).

Recommendations: Increase the Synergy of the Prevention Centers by Tackling Large Scientific Projects and Increase Interaction Among Individual Researchers in the Group

- ▶ In recognition of the need to increase synergy within the limitations of a budget that would allow only a few projects the size of the NOD Roadmap, the Prevention Centers have recently focused on projects that will promote synergy through use of common research platforms and datasets. Examples include:

- A pilot project on generation of genetically-engineered mouse models, in which retroviral vectors are used to drive the expression of autoreactive T cell receptors. This experimental system could promote synergy among the Prevention Centers. Identical retroviral constructs can be used in different experimental settings, thereby permitting useful comparison and verification of results, and facilitating group planning and future directions.
- A project focused on identification of mechanisms of heritable immune trait variations by combining the HapMap with high-throughput cellular and immunologic phenotyping of Human Genome Diversity Cell Line Panel (HGDP). This project will begin to bridge—in an unbiased way—the gap between genetic susceptibility to autoimmune disease, as defined by gene association and family studies, and the biochemical mechanisms of susceptibility or resistance to disease. The tools developed in this project will provide a common technological platform and phenotype dataset that will not only promote synergy among the Prevention Centers investigators, but should also be of great use to other investigators.

Recommendation: Identify Ways To Translate Research Studies from Mice to Humans

- ▶ Although the overall mission of the Prevention Centers focused on pre-clinical studies, often using small animal models, the Centers are sensitive and responsive to the need for both human and animal studies, and for translation from animal to human studies (and vice versa). Thus, while only 3 of the 11 main projects originally funded to establish the Centers were focused on human studies, 20 of the 48 pilot projects funded to date (42 percent) are primarily or completely focused on human studies, reflecting the Centers’ emphasis on using discretionary funds to bridge gaps between animal and human studies.

Primary examples include:

- A project that aims to recapitulate the development of diabetogenic autoreactivity in a mouse model;
- A project that is adapting a genetic library to probe functions of human T regulatory cells;
- Ongoing discussions within the Group of ways to pursue a project similar to the NOD Roadmap project in humans (this project is not yet funded).

Recommendation: Identify Ways To Widely Advertise the Pilot and Feasibility Program To Make It Available to the Broader Research Community (Develop a Website)

- ▶ A website will be implemented after the award of the new Prevention Centers grants, currently scheduled for July 2006. This website will integrate all Prevention Centers activities, including availability of the pilot and feasibility program and sharing of data among the Centers and with the immunology community.

Ongoing Evaluation

Continued and ongoing evaluation of the study design and the progress of the Prevention Centers are ensured through Steering Committee meetings, annual all-investigator meetings, and external input. The Prevention Centers Steering Committee meets to discuss ongoing pilot projects and new pilot proposals as well as the overall progress of the group. External reviewers attended the 2005 all-investigator meeting to provide feedback on the accomplishments and direction of the program.

Prevention Centers Administrative History

Date Initiative Started	2001
Date <i>Special Program</i> Funding Started	2001
Participating Components	NIAID, NICHD, NIDDK, ORWH, JDRF
This Consortium consists of five centers in the United States.	

Standardization Programs: Diabetes Autoantibody Standardization Program (DASP); C-peptide Standardization; and Improving the Clinical Measurement of Hemoglobin A1c (HbA1c)

The purpose of these programs is to develop and implement standardization programs designed to improve the measurement of: (1) aberrant molecules called “autoantibodies,” which are predictive of type 1 diabetes; (2) C-peptide as an indicator of insulin production; and (3) hemoglobin A1c (HbA1c) as an indicator of glycemic control. Such improvements and standardization will greatly advance both research and patient care.

DASP

DASP seeks to improve the measurement of autoantibodies in blood that are predictive of type 1 diabetes, and to decrease laboratory-to-laboratory variation. Autoantibody production reflects abnormal and destructive immune system functioning. A normal immune system is designed to fight infections; one part of this complex process is the production of antibodies that target infectious agents. The immune system of a person who has—or is developing—type 1 diabetes, however, also makes “autoantibodies” that recognize insulin and other beta cell-derived molecules. Autoantibodies are currently the best predictors of the onset of type 1 diabetes before the appearance of clinical symptoms. In combination with genetic screening, autoantibody tests are used to identify individuals at elevated risk of developing type 1 diabetes and to characterize autoimmunity. DASP sets of serum samples are used as standards to evaluate the performance of diabetes laboratories throughout the world and serve as reference materials for developing new methods and technologies. DASP also provides training and information to guide other laboratories in improving their performance.

C-peptide Standardization Program

This program aims to establish reliability in measurements of C-peptide, which is a byproduct of insulin production by beta cells and thus useful as a marker of beta cell function. In clinical trials of agents designed to prevent the disease in at-risk persons, or to preserve beta cell function in individuals with new onset type 1 diabetes, C-peptide will be used as an outcome measure that indicates insulin production. Residual beta cell function is associated with better glycemic control, lower risk of hypoglycemia (discussed in Goal IV), and lower risk of long-term diabetic complications.

National Glycohemoglobin Standardization Program (NGSP; HbA1c Standardization Program)

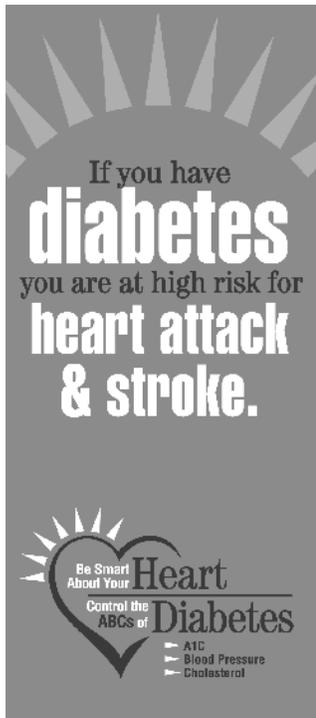
The purpose of the NGSP is to achieve standardization and reliability in measures of HbA1c, a component of blood that is a good surrogate measure of long-term blood glucose control and, as such, reflects risk of diabetic complications. The correlation between HbA1c levels and risk for complications was demonstrated in the Diabetes Control and Complications Trial (DCCT), as well as another trial, the United Kingdom Prospective Diabetes Study. Through efforts to improve HbA1c testing so that clinical laboratory results can be related directly to the results of the DCCT, this program will enable healthcare providers and patients to accurately and meaningfully assess glycemic control and risks for complications. The standardization of HbA1c measures is essential to public health efforts, such as those of the National Diabetes Education Program (NDEP), to improve diabetes control nationwide so that the public can reap the benefits of clinical trials proving that complications can be delayed or prevented. This effort also allows researchers to better evaluate a patient’s risk for complications and fosters comparison of results across

multiple studies worldwide. The NGSP consists of a Steering Committee and a Laboratory Network. The NGSP network interacts with manufacturers and laboratories to assist with calibration and to certify methods as traceable to the DCCT. The NGSP also works with the College of American Pathologists to assign HbA1c values to proficiency testing specimens for better evaluation of HbA1c results in clinical laboratories.

Highlights of Progress

The progress that the Standardization Programs have made as of March 1, 2006, includes:

- DASP conducted and presented workshop evaluations of key international diabetes laboratories in 2000, 2002, 2003, and 2005.
- DASP validated improvement of two different technologies for measuring autoantibodies.
- DASP documented improvement in performance of the insulin autoantibody assay for laboratories with consistent participation in the DASP Training Program.
- DASP created laboratory reference materials (blood samples) from type 1 diabetes patients and healthy people that are available to ensure assay quality and to support further technology development.
- The C-peptide program evaluated the stability of C-peptide and effects of common interferences. The program also coordinated an international laboratory comparison of C-peptide measurement. The results of the first inter-laboratory comparison trial are being prepared for publication. This research is crucial for optimizing measurement techniques and standardizing results.
- The CDC HbA1c laboratory and the NGSP have participated in efforts of the International Federation of Clinical Chemistry and Laboratory Medicine (IFCC) to develop a “higher level” reference method for measuring HbA1c. This reference method was approved by the IFCC and is now the basis for uniform standardization of HbA1c assays worldwide. The IFCC Working Group also developed a mathematical equation to facilitate comparison among results obtained by this IFCC reference method and the NGSP, as well as with methods in Sweden and Japan.
- For HbA1c measurements, between 1996 and 2006, there was an increase in the number of methods and laboratories certified by the NGSP as traceable to the DCCT. Methods and laboratories are certified each year. In 2005, the NGSP certified 63 HbA1c diagnostic methods and 41 laboratories. Nearly all laboratories worldwide are using methods that are traceable to and certified by the NGSP.
- Proficiency testing data also showed a decrease in the variability of HbA1c measurements among laboratories.



Standardization of the HbA1c test supported the development of a national education campaign based on “knowing your HbA1c number.” Most people with diabetes are encouraged to keep their HbA1c level below 7 percent to prevent or delay the development of diabetes complications, including heart attack and stroke. The campaign is sponsored by the National Diabetes Education Program, which is a partnership of the NIDDK and CDC (www.ndep.nih.gov).

Anticipated Outcomes

The autoantibody, C-peptide, and HbA1c standardization programs are extensive efforts to improve laboratory measures of critical markers for type 1 diabetes risk and disease progression. Such improvements are important to the success of multicenter clinical trials as different participating laboratories must be able to obtain measurements that are comparable and can be meaningfully analyzed together. Research progress will also be enhanced when the results of different trials are based on standardized measures to facilitate comparison. Patients and their healthcare practitioners will be better able to ascertain what a given blood test means in terms of health risks and treatment plans when test results are sufficiently reliable for comparison with relevant research studies. As a result of research toward standardizing autoantibody testing and identifying new biomarkers for predictive assays, those at risk for type 1 diabetes may be diagnosed earlier, permitting earlier

intervention to diminish disease severity. Improved measurement techniques for C-peptide will impact research on agents that can preserve beta cell function, particularly in those with new onset diabetes. Improvements in HbA1c testing will facilitate implementation of a national and global strategy to reduce the complications associated with diabetes through proper blood glucose control. The standardization programs will thus have wide-reaching implications for researchers, clinicians, and patients.

External Evaluation by Expert Panel

To supplement ongoing evaluation and guidance received from participants at workshops and meetings of the Immunology of Diabetes Society (IDS) and others with relevant expertise, leading scientific and lay experts were asked to evaluate the progress of the Standardization Programs at an *ad hoc* planning and evaluation meeting convened by the NIH in January 2005 (see Appendix 3). Comments from the panel review included:

- ▶ These programs are key for advancing research on predicting susceptibility to type 1 diabetes and preventing the disease. For example, autoantibodies are used to predict if a person may develop type 1 diabetes, and C-peptide measurements are used to determine if strategies to prevent or reverse the disease are successful.
- ▶ The programs have had many accomplishments, have been managed well, and can be considered a “success story.”
- ▶ An important component of the programs is the training that is provided to other laboratories and researchers.
- ▶ These studies would not be successfully funded through an NIH R01 grant mechanism, so these programs are an appropriate mechanism for conducting this type of research.
- ▶ This investment of type 1 diabetes *Special Funds* could have a large payoff in terms of the importance of these assays to future clinical research efforts.

Actions Taken in Response to Expert Panel

Recommendations

The Standardization Programs took the following actions in response to recommendations of the expert panel at the *ad hoc* planning and evaluation meeting convened by the NIH in January 2005:

Recommendation: Pursue Research To Identify New Surrogate Markers That Can Predict Disease Onset or Monitor Disease Progression

- ▶ In addition to providing samples for research for several individual groups, a collaboration with the NIDDK-funded Proteomics Consortium was established. Studies are under way using DASP samples to evaluate the potential for proteomics to improve the sensitivity and specificity of the prediction and diagnosis of type 1 diabetes.
- ▶ A new request for applications was issued entitled, “Biomarkers of Autoimmunity of Type 1 Diabetes” for first funding in FY 2006. This initiative will fund grants focused on biomarker identification/validation in humans.

Recommendation: Bolster the Research Efforts on C-Peptide Measurement and Standardization

- ▶ The C-peptide standardization program completed an initial stability study and laboratory comparison study in 2005. In early 2006, another laboratory comparison study was initiated to evaluate improvement in C-peptide results since the initial comparison.

Recommendation: Encourage International Research Laboratory Participation in Standardization Programs

- ▶ Participation in DASP is encouraged by the efforts of the IDS and by publication manuscript reviewers. DASP data play a key role in harmonizing the autoantibody assays in type 1 diabetes consortia. Forty-seven key diabetes laboratories in 18 countries participated in DASP 2005.

- ▶ Over 70 percent of all laboratories certified by the NGSP are outside the U.S. The NGSP certified laboratories are located in the U.S., Europe, Asia, South America, and Australia. Laboratories in the U.S., Europe, and Asia are currently participating in C-peptide comparison studies.

Ongoing Evaluation

Continued and ongoing evaluation of the research and progress of the Standardization Programs is carried out as described below.

DASP: DASP efforts are managed by the IDS Autoantibody Standardization Committee and the CDC collaborator. The activities and progress are reviewed by IDS participants at the workshop presentations at the IDS meetings, and additional input is periodically sought from the IDS president and other prominent scientists in the field.

C-peptide: The C-peptide standardization program has project oversight from the CDC. In addition, a C-peptide Standardization Advisory Committee makes recommendations for research studies and assists in evaluation of results.

HbA1c: The effort to improve and standardize the measurement of HbA1c is divided between the CDC and the NGSP (with CDC support) at the University of Missouri. The CDC and the NGSP Laboratory also participate as members in the International Federation of Clinical Chemistry Reference Laboratory Network for HbA1c Measurement.

Coordination with Other Research Efforts

The Standardization Programs coordinate their efforts with multiple other type 1 diabetes research consortia and networks supported by the *Special Funding Program*. Collaboration, coordination, and resource sharing serve to synergize research efforts and accelerate research progress. Examples of coordination with other consortia are given below. For a

full description of ongoing collaborative efforts, please see Appendix 2.

Enhancing Quality and Standardization of Laboratory Measures in Multicenter Clinical Trials:

- ▶ DASP interacts with the TEDDY, T1DGC, and TrialNet autoantibody labs, by providing laboratory materials and proficiency testing to facilitate their autoantibody measurements.
- ▶ The C-peptide program included two laboratories from TrialNet in an international comparison effort, the results of which illustrated the need to identify and minimize the major sources of variation in C-peptide measurements in multicenter, multi-laboratory clinical studies.
- ▶ TrialNet, EDIC, and other clinical studies supported by the *Special Funding Program* use laboratories certified through the NGSP.

Improving and Developing Technology:

- ▶ Because of limitations associated with autoantibody testing, DASP is working with NIDDK-supported investigators studying proteomics and type 1 diabetes, and collaborating with the Pacific Northwest National Laboratory, to find new biomarkers to improve diagnosis of and prediction of risk for this disease. This collaborative project will use blood samples collected by DASP from newly diagnosed type 1 diabetes patients and healthy people. The samples will be analyzed with proteomic and metabolomic technologies: that is, large-scale profiling and characterization of the component proteins and small molecules, respectively. Differences identified between samples from patients and healthy individuals can be further investigated for potential predictive or diagnostic value.

Standardization Programs Administrative History

DASP

Date Initiative Started	1998
Date <i>Special Program</i> Funding Started	1998
Participating Components	CDC, NIDDK, Immunology of Diabetes Society
Website	www.idsoc.org/committees/antibody/dasphome.html

C-peptide

Date Initiative Started	2002
Date <i>Special Program</i> Funding Started	2003
Participating Components	CDC, NIDDK, C-peptide Standardization Advisory Committee, and University of Missouri

NGSP (HbA1c)

Date Initiative Started	1996
Date <i>Special Program</i> Funding Started	1998
Participating Components	CDC, NIDDK, NGSP
Website	www.ngsp.org

The HbA1c program is carried out at the CDC-supported National Glycohemoglobin Standardization Program (NGSP), as well as the National Diabetes Laboratory at CDC, both members of the International Federation of Clinical Chemistry (IFCC) Reference Laboratory Network for HbA1c; the NIDDK also funds this effort.

Trial To Reduce IDDM in the Genetically at-Risk (TRIGR)

TRIGR is an international clinical trial to determine, for infants at risk for type 1 diabetes, whether weaning to extensively-hydrolyzed formula, as compared to standard cow's milk formula, will reduce the risk of developing diabetes-predictive autoantibodies and, ultimately, type 1 diabetes. Environmental factors, such as exposure during infancy to foreign proteins from food, may interfere with normal immune system development in genetically-susceptible individuals, and formula is usually the first foreign food given to infants as they are weaned from human breast milk. Standard cow's milk formula contains proteins that are intact and thus capable of inciting the immune system. Hydrolyzing proteins breaks them into very small pieces, which are much less likely to elicit an immune response, and prior research has suggested that weaning to hydrolyzed (versus intact-protein) formula may reduce risk of type 1 diabetes. The first phases of TRIGR are extensive, multi-national efforts to identify several thousand infants at risk for type 1 diabetes by recruiting pregnant women who have the disease, or an affected family member, and subsequent screening of the infants for diabetes-associated variants of certain immune system genes (HLA genes). As part of the study, exclusive breastfeeding will be encouraged, but once this is no longer possible, babies will enter the intervention portion of the study by being randomly assigned to receive either standard or extensively-hydrolyzed formula (up to age 8 months). Follow-up monitoring will assess autoantibody development and diabetes incidence up to age 10 years.

Highlights of Progress

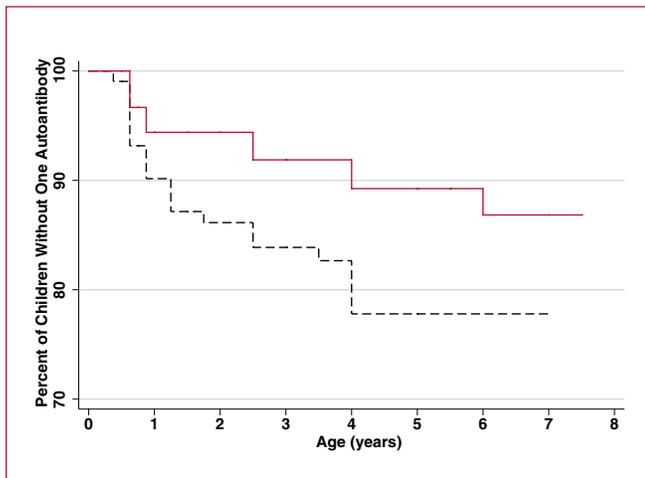
The progress that TRIGR has made as of March 1, 2006, includes:

- Recruited over 4,740 pregnant women toward the goal of 4,936.
- Screened over 4,220 infants for diabetes-related HLA genotype.
- Entered 1,770 eligible infants into the intervention portion of the study (began receiving hydrolyzed or intact-protein formulas).
- Collected over 70,280 blood samples for antibody analyses.
- Met all study milestones in terms of recruitment and protocol compliance with forms, tests, and visits.
- Successfully applied for continuation of the study in competitive NIH peer review.

Anticipated Outcomes

TRIGR is a large-scale, well-coordinated clinical trial to test the effect of a dietary intervention during infancy on the development of type 1 diabetes in genetically-susceptible individuals. If the results of this trial show that weaning to hydrolyzed infant formula, as compared to standard formula, reduces incidence of type 1 diabetes, then it will have validated a practical way to alter the course of autoimmunity development and reduce type 1 diabetes incidence in young children. TRIGR builds on prior research in animals and on a pilot study in humans that investigated the association of different

infant formulas with autoantibody appearance. It has been hypothesized that diabetes-related autoimmunity may be triggered when the immature gut of an at-risk infant encounters foreign dietary proteins. The use of extensively-hydrolyzed formula during weaning would delay the introduction of more complex, intact foreign proteins. Thus, TRIGR may also shed further light on the role of the gut and its immune system in the development of type 1 diabetes. Additionally, upon completion of recruitment, TRIGR will have the largest international experience in the identification, recruitment,



Results of a pilot study preceding the ongoing Trial to Reduce IDDM in the Genetically At Risk (TRIGR), which randomized newborns receiving formula to either an intervention formula (hydrolyzed proteins; solid red line) or standard cow's milk formula (dashed line). Follow-up analysis showed that 22 percent of children who received the standard formula developed at least one autoantibody predictive of type 1 diabetes, while only 13 percent of children who received the intervention formula developed at least one autoantibody. This finding in the small pilot study is now being tested in the full-scale TRIGR study, which is currently under way. (Figure courtesy of Dr. Hans Åkerblom and adapted with kind permission of Springer Science and Business Media from Åkerblom, HK et al. *Diabetologia*. 48: 829-837, 2005.)

and follow-up of newborn infants at risk. The blood specimen and information repository may permit potential contribution to other research on environmental determinants of diabetes, such as the TEDDY study (see Goal I). The potential for a dietary modification in infancy to reduce type 1 diabetes (along with biological data on the very large number of genetically susceptible infants being recruited for study) makes the TRIGR study enormously beneficial to families at risk.

External Evaluation by Expert Panel

To supplement ongoing evaluation and guidance from an External Data Safety Monitoring Board/Advisory Panel focused on TRIGR, leading scientific and lay experts were asked to evaluate the progress of TRIGR at an *ad hoc* planning and

evaluation meeting convened by the NIH in January 2005 (see Appendix 3). Comments from the panel review included:

- ▶ This study has made excellent progress.
- ▶ If the results of the trial are affirmative and show a decrease in autoantibodies in children first exposed to an extensively-hydrolyzed formula, then the study has the potential for making a positive impact on patient care.
- ▶ The natural history aspect of the study could provide insights into disease pathogenesis, particularly if it is integrated with the TEDDY study.
- ▶ A major strength of this study is that the participant retention rate is extremely high.

Actions Taken in Response to Expert Panel Recommendations

TRIGR took the following actions in response to recommendations of the expert panel at the *ad hoc* planning and evaluation meeting convened by the NIH in January 2005:

Recommendation: Increase Collaboration, Integration, and Coordination with the TEDDY Consortium

- ▶ Representatives from both TRIGR and TEDDY participated in a type 1 diabetes consortia coordination meeting that the NIDDK convened in May 2005. The purpose of the meeting was to identify opportunities to enhance collaboration among all of the research consortia studying type 1 diabetes. Prior to the larger meeting, representatives from consortia studying newborns (TEDDY, TRIGR, and Type 1 Diabetes TrialNet) met to discuss how to obtain the most useful information when looking at these studies as a group. Common data variables across the studies and future analytic strategies were discussed.
- ▶ TRIGR and TEDDY study investigators have met to consider using the TRIGR network to enhance accrual to TEDDY for first-degree relatives when TRIGR accrual ends.

- ▶ TRIGR and TEDDY laboratory investigators are participating together in harmonization efforts to bring uniform standards to autoantibody assays.

Recommendation: Combine Data from TEDDY and TRIGR To Foster the Future Conduct of Analyses

- ▶ TRIGR and TEDDY have implemented similar standards in data collection and entry, thus permitting direct comparison between results obtained in each study relevant to nutrition and to diabetes-associated variants of certain immune system genes (HLA genes).

Recommendation: Create Mechanisms To Make Results of the TRIGR Studies Generalizable to the U.S.

- ▶ TRIGR recruits from both the U.S. population as well as sites worldwide. Eligibility criteria are based upon HLA risk levels, thus the results of the study would be generally applicable to anyone in the U.S. population who has a risk-conferring HLA genotype. In addition, U.S. sites contribute the majority of minorities to the study from which study results can be assessed for these special populations. Also, U.S. infant feeding practices are readily identifiable and can serve as a basis for considering the generalizability of study results specifically to the U.S.

Ongoing Evaluation

To ensure continued and ongoing evaluation of the study design and the progress of TRIGR, the NICHD has established an External Data Safety Monitoring Board/Advisory Panel for this trial. Additional critical entities include the trial's International Coordinating Center, which integrates operations for all regions of the TRIGR Study Group, maintains and validates documents related to the operations of TRIGR, and is in charge of developing study forms and the Manual of Operations. A Data Management Unit is responsible for data management systems; monitoring the study for protocol

compliance, adverse events, and other issues; and data analysis and reporting. There are also a number of working committees focused on such topics as nutritional intervention, ancillary studies, and internal safety monitoring, among others.

Coordination with Other Research Efforts

TRIGR coordinates its efforts with multiple other type 1 diabetes research consortia and networks supported by the *Special Funding Program*. Collaboration, coordination, and resource sharing serve to synergize research efforts and accelerate research progress. Examples of coordination with other consortia are given below. For a full description of ongoing collaborative efforts, please see Appendix 2.

Coordinating Research Studies Involving Newborns:

- ▶ TRIGR investigators have met with investigators participating in other type 1 diabetes research studies involving newborns (TEDDY and TrialNet) to discuss opportunities for enhancing coordination and collaboration.
- ▶ TEDDY and TRIGR share the same Data Coordinating Center. This coordination has resulted in implementation of similar standards in data collection, entry, management of quality control, and analyses for both studies.
- ▶ TRIGR and TEDDY investigators are considering collaborative efforts on recruitment after TRIGR accrual ends. Both groups are also considering a follow-up intervention protocol.

Coordinating Patient Recruitment Efforts:

- ▶ Two SEARCH sites are assisting with TRIGR recruitment by providing brochures and other information about TRIGR.
- ▶ TRIGR, TrialNet, and TEDDY have coordinated recruitment efforts to ensure that they are not adversely competing for patient participants in their studies.

Enhancing Data Comparison Among Studies:

- ▶ As described previously, TRIGR and TEDDY have implemented similar standards in data collection and entry.
- ▶ TRIGR and the ITN are coordinating their efforts in the area of T cell assays.

TRIGR Administrative History

Date Initiative Started	2001
Date <i>Special Program</i> Funding Started	2001
Date TRIGR Successful Recompensation	2006
Participating Components	NICHHD, the Canadian Institutes of Health Research, the European Foundation for the Study of Diabetes, the European Union, JDRF, the Netherlands Diabetes Foundation, and industry
Website	www.trigr.org

TRIGR is taking place at 77 sites in 15 countries including the United States, several European countries, Canada, and Australia.

EVALUATION OF INVESTIGATOR-INITIATED RESEARCH

In addition to the research consortia previously described, the Special Funding Program supported investigator-initiated research projects addressing particular challenges and opportunities identified by the NIH with the aid of scientific experts at workshops and advisory meetings. Often these recommendations were disseminated to the research community in a Request for Applications (RFA) or Request for Proposals (RFP). (For a list of initiatives supported by the Special Funding Program, please see Appendix 1.) The NIDDK conducted a Grantee Survey (see Appendix 5) to evaluate the impact of the Special Funding Program on investigators with research project grants principally supported by the Special Funds. The survey was used as a tool to assess the research accomplishments (e.g., publications, resulting patents, impact on patients' health), research collaborations, and impact that the Special Program had on careers of investigators supported by it. Data from this survey are found in the "Assessment" chapter.

Impact of Special Funding Program on Extramural Grantees

Principal investigators who received grants related to preventing or reversing type 1 diabetes responded to the survey that asked, in part, about the value of their grant or funding source. Representative remarks include:

- ▶ "The grant has had a huge impact on my career. Being my first extramural grant award, it enabled me to establish my laboratory and publish my first several papers, which became the basis of a successful R01 application. I also recently received tenure and promotion at my institution."
- ▶ "Support from this *Program* allowed me to develop an interest in diabetes and gain new expertise in autoimmunity. As a result, type 1 diabetes is presently the major focus of my research. Most importantly, this high-risk application allowed me to make a career shift from performing basic science to bench to bedside research."
- ▶ "Support by this *Program* has given me the opportunity to enter into and focus my research career on a new research problem, type 1 diabetes. It afforded me the opportunity to assemble a critical mass of investigators that work in a variety of disciplines, but often studying similar signaling mechanisms.... Support of these types of collaborations will significantly accelerate our understanding of the causes of type 1 diabetes, and will expedite the development of successful therapeutic treatments aimed at diabetes, as well as other diseases."
- ▶ "Type 1 diabetes has been a leading model in the field of complex disease genetics and this has occurred, in large part, due to the availability of this funding source."
- ▶ "This funding source was of critical value to the ongoing and future research in my laboratory. All ongoing research, and all grant applications currently pending, stem from projects that arose from this grant."
- ▶ "The grant enabled me to establish an innovative research program and international reputation in type 1 diabetes and organ-specific T cell tolerance research. From that base, I have continued to expand my research team in diabetes, recruiting several Ph.D. students, postdoctoral fellows, and a Senior Lecturer to the team to initiate both deeper analyses of NOD diabetes susceptibility and genome-wide strategies for discovering diabetes-suppressing genes and mechanisms."

IMMUNE TOLERANCE

The key to a healthy immune system is its remarkable ability to distinguish between the body's own cells—referred to as “self”—and foreign cells—“nonself.” The body's immune defenses normally coexist peacefully with cells that carry distinctive “self” marker molecules. But when immune defenders encounter cells or organisms carrying markers that say “nonself,” they quickly launch an attack. The tendency of the cells of the immune system to ignore the body's own tissues is known as “immune tolerance.”

Tolerance occurs through at least two processes. During their development, cells of the immune system are educated by the body to recognize foreign molecules while at the same time they are taught to ignore, or tolerate, normal cells and self molecules. This process, known as central tolerance, occurs during the development of disease-fighting immune cells.

However, maturing immune cells do not encounter every molecule in the body during their development, so they must also learn to ignore mature cells and tissues. In *peripheral tolerance*, circulating immune cells that happen to recognize a self molecule are unable to respond because some of the chemical signals required to activate the immune cell are absent. This requirement for more than one signal, therefore, keeps potentially harmful immune cells switched off.

Maintaining tolerance is important because it prevents the immune system from attacking its fellow cells. It is the safeguard that invariably fails in type 1 diabetes and other autoimmune diseases. The ability to induce, maintain, and monitor immune tolerance would be an important step forward in the treatment of type 1 diabetes. If initiated early, it could prevent destruction of insulin-producing beta cells. Inducing and maintaining immune tolerance in patients with advanced type 1 diabetes with transplanted islets would obviate the need for lifelong drug regimens to prevent rejection of the transplant and their attendant risks.

This sidebar was adapted from “Understanding the Immune System,” published by the NIAID and available at: www.niaid.nih.gov/publications/immune/the_immune_system.pdf

A Personal Interest in Curing Type 1 Diabetes

At age 49, Kevan Herold, MD, is highly accomplished. He is Professor of Immunobiology and Medicine at Yale University in New Haven, CT; an endocrinologist and groundbreaking medical researcher; a husband; a father to three daughters; and an avid tennis player.

He also has type 1 diabetes.

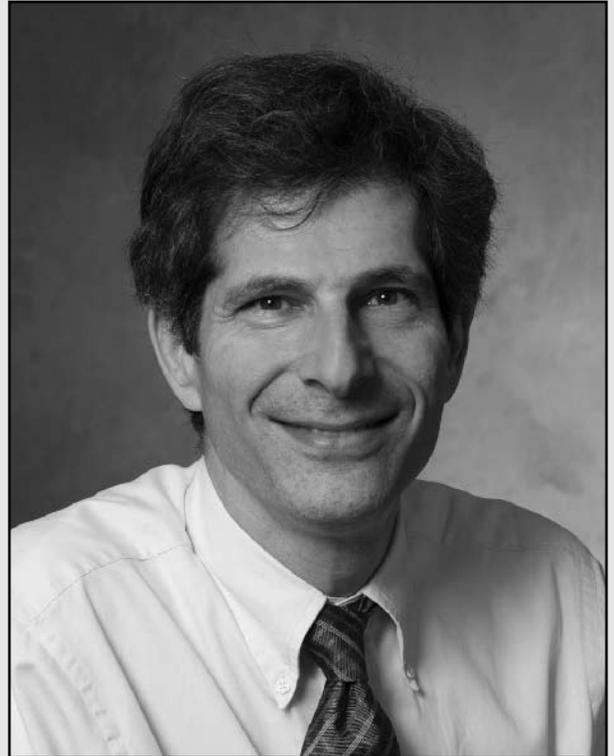
"I was 17 years old and in the first month of my freshman year at Penn State when I started experiencing classic symptoms of type 1 diabetes, including loss of weight, fatigue, and the need to urinate and drink a great deal of water," says Dr. Herold. He was diagnosed with the disease shortly thereafter.

Although his first medical interest, even before entering college, was cardiology, it's not surprising that today Dr. Herold is one of the nation's leaders in type 1 diabetes clinical research. His important work is supported by the *Special Statutory Funding Program for Type 1 Diabetes Research*, the NIDDK, the NIAID, the JDRF, and others.

"Living with diabetes is a great motivator for me to help find a cure for this disease," he says. Like his research, Dr. Herold's message to others with the disease is practical and important: "Avoid diabetes-related health complications by controlling blood sugar levels now, because clearly good, new treatments and technologies for those of us with diabetes are coming."

A Focus on Diabetes

Type 1 diabetes is an autoimmune disease that results when the body's immune system turns on itself and destroys the pancreatic cells that produce insulin. People with type 1 diabetes typically require insulin injections three to four times a day to stay alive. Many type 1



Dr. Kevan Herold

diabetes patients grapple with the long-term complications of diabetes, including blindness, kidney failure, and heart disease.

Fortunately for Dr. Herold, after living with diabetes for more than 30 years, he has experienced no disease-related complications. But even for a doctor with diabetes, living with this disease has not been easy. "In 1974, when I was diagnosed, we had no idea what blood sugar control was like or all about; we could only measure sugar in the urine," says Dr. Herold. Since then, he adds, there has been enormous growth in the understanding and technology related to this disease.

"Our understanding and treatment of diabetes now is nothing like it was even 10 years ago," says Dr. Herold. "With new developments in cellular and immune therapies, I am sure this pace will continue. The recent developments suggest that even the most fundamental

approaches to treatment will change over the next several years and that the primary problem of beta cell deficiency will be solved.” To that end, Dr. Herold’s research, which includes both immune and cellular therapy, attacks type 1 diabetes on several fronts.

Manipulating the Immune System

Dr. Herold and other immunologists in two NIDDK-, NIAID- and *Special Program*-supported networks, TrialNet and the ITN, have been searching for ways to control autoimmunity to reverse autoimmune diseases such as type 1 diabetes. However, current therapies, including immunosuppression, could compromise the immune system’s ability to fight disease. The challenge is to find a way to manipulate the immune system to specifically target the disease-causing immune cells (T cells).

Dr. Herold and his colleagues have made significant progress toward the goal of reversing recently diagnosed type 1 diabetes with a promising new therapy that can effectively alter the clinical course of the disease. The drug, an anti-CD3 antibody developed by Dr. Jeffrey Bluestone, binds to the T cell receptor. In a groundbreaking NIDDK- and NIAID-supported clinical trial, Dr. Herold and colleagues treated patients with anti-CD3 for 2 weeks shortly after their diagnosis. The drug helped patients by reducing the decline in their ability to produce insulin for at least 2 years after diagnosis compared to a placebo treatment. These impressive results were confirmed in a second industry-supported clinical trial. “The new antibodies alter the signal that otherwise causes T cells to attack insulin-secreting cells,” explains Dr. Herold. He is now expanding these studies to find out what treatment regimens are the most effective at stopping the disease and how long the treatment can last before it requires a booster.

Beta Cell Regeneration

Dr. Herold is also searching for ways to naturally replenish beta cells through regeneration. In a *Special Program*-supported initiative on “Innovative Partnerships in

Type 1 Diabetes Research” (see Goal VI), Dr. Herold has collaborated with Dr. Virginia Papaioannou at Columbia University, to develop new approaches to stimulate beta cell regeneration in the setting of immune tolerance. These studies have involved strategies to stimulate the growth of the stem cells that develop into mature beta cells. In studies also supported by this *Special Program* initiative, preclinical studies pairing anti-CD3 therapy with the drug exenatide has led to a new clinical study that is planned to be conducted by TrialNet.

Disease Prevention

A successful prevention strategy needs to be both long-lasting and specific to the disease. Anti-CD3 provides systemic modulation of the entire immune system but may only confer temporary protection and may lead to side effects. Conversely, another strategy is to induce disease-specific tolerance by using molecules that cause the disease (antigens) to stimulate the expansion of beneficial immune system cells. However, type 1 diabetes antigens have not been clearly identified, and such targeted interventions have thus far only been successful in the prediabetic phase, thus limiting their therapeutic potential. Dr. Herold’s team has worked with Dr. Matthias von Herrath, at LaJolla Institute of Allergy and Immunology, to develop an exciting new strategy that combines these two therapies and has exhibited long-lasting success in reversing type 1 diabetes in two different animal models.

Improving the Health of Type 1 Diabetes Patients

“I’ve always liked science and research, and I’m always looking for ways to apply science to treating patients,” says Dr. Herold. “In my mind, all of these research efforts are related.”

As someone who lives with type 1 diabetes every day, Dr. Herold says that he can easily recognize and relate to what his patients are going through, right down to the daily details of what they need to carry with them in case their blood sugar levels should suddenly drop, often referred to

as hypoglycemic episodes. These episodes can be life-threatening. Despite checking his own blood sugar on average seven to eight times a day, Dr. Herold, too, experiences hypoglycemic episodes from time to time and personally looks forward to the development of new therapies that

will prevent them. Thus, in addition to all the people with type 1 diabetes that Dr. Herold has focused on helping all these many years, he too may end up reaping the benefits of his own research.

Mother and Daughter Living with Type 1 Diabetes

Katie Clark spent weeks denying her 5-year-old daughter's symptoms of type 1 diabetes. Up to that point, Katie thought that the fact that everyone wanted to touch Ellie's beautiful curly blond hair would be her daughter's burden to bear. She was wrong.

When sugar was found in Ellie's urine on what was supposed to be her first day at a new preschool, Katie learned that Ellie had type 1 diabetes. Katie was devastated. She spent her 30th birthday at the hospital and was deeply depressed for most of the next 2 weeks. She was also very angry. "Anger isn't the most common emotion at the beginning," Katie observes. "However, we're not new to the disease. I've had type 1 diabetes for 28 years."

At the time this profile was written, 10 months after being diagnosed, Ellie had already suffered many unwanted side effects from disease treatment. She had calluses on her fingers. Her bottom had scar tissue from her insulin pump sites. She had undergone 1,494 finger pricks and 98 pump site changes. Ellie's insulin pump site must be changed every 3 days. Ellie will ask, out of the blue, "Is it day three?" Katie laments, "I cannot tell you how heartbreaking it is for me to see my daughter worrying about an impending pump site change. There is relief on her face on those days when we can say, 'No honey, not today.' The devastation in her eyes is almost more than I can stand when we have to say, 'Yes, today is day three, sweetheart.'" Katie is concerned that "Ellie is spending her time worrying about diabetes when she should be playing with her baby dolls and learning to read."

One of the greatest difficulties Katie finds in dealing with Ellie's disease is knowing firsthand the challenges that Ellie will face as she grows up. Katie knows just how type 1 diabetes will affect every detail of Ellie's life. Katie



Ellie and Katie Clark

states, "There is no escape...there are no vacations from type 1 diabetes." Ellie will have to endure constant finger sticks and worry about when her next meal will be. Like Katie, Ellie is at risk of developing devastating disease complications, such as blindness, kidney disease, and heart disease, which could ultimately reduce her life span by approximately 15 years. Katie recalls, "I can very vividly remember reading a magazine article about the complications of diabetes when I was 8 years old. I was horrified. I can see Ellie will be going through these same thoughts and dealing with these same issues, and it's horrible. This is not the life I dreamt of for my precious daughter."

Other less common but very memorable events will leave their imprint as well, as they have during some of the happiest moments of Katie's life. Recalling the insulin reaction she had on her wedding day, Katie laments, "My newly styled hair got messed up, and orange juice I needed to take immediately to adjust my blood sugar level was spilled on my veil." For each of her pregnancies, Katie saw her high-risk pregnancy obstetrician once a week, and in the months leading up to the births, she saw her doctors twice a week. While in labor, Katie was forced to check her blood sugar every hour. After the births, the nurses whisked the babies away to check their blood sugar levels, because newborns of mothers with diabetes often have low blood sugar. The

nurses had to put a tube down their throats to pump sugar into their stomachs to normalize their blood sugar levels.

Ellie's diabetes hits Katie and her husband particularly hard when they're tucking Ellie into bed at night. That's when she asks questions such as; "Mommy, why do some people get diabetes and some people don't?" Or she says, "Daddy, I don't want diabetes anymore." Katie and her husband face a new challenge now that Ellie has begun school. "We have to teach Ellie's teachers how to take care of her," Katie observes.

The Clarks dream of giving Ellie back the life she was living before her diagnosis and having a future brighter than one clouded by diabetes. "I'd give everything I have—even my own life—for Ellie not to have to endure another day of this dreadful disease," Katie stresses. "We must do everything we can to find a cure. Our sweet little girl with the curls deserves it."

Hope Through Research

In type 1 diabetes, a genetic predisposition for the disease is believed to be triggered by environmental factors. Researchers have already identified genetic regions that play a key role in disease development. However, there are other important genes that have not yet been identified, and it is still unclear how gene-environment interactions may promote the disease. Therefore, the *Special Statutory Funding Program for Type 1 Diabetes Research* is supporting multifaceted research efforts to uncover important genes and environmental factors that promote the onset of, or confer resistance to, type 1 diabetes. For example, the T1DGC is a monumental effort to analyze the genetic makeup of families in which two or more siblings have type 1 diabetes to determine which genes confer disease susceptibility. Because not just a single gene "causes" type 1 diabetes, this

type of large-scale effort is crucial to understanding the complex genetic underpinnings of the disease. Another example is a multicenter, multinational, NIH-funded epidemiological study called TEDDY. In this study, researchers are following newborns who are known to be genetically at risk for developing type 1 diabetes until they are 15 years old, to see who develops the disease. Researchers will use the population to pinpoint the environmental factors that either triggered the disease or provided protection from it. An NIDDK website describes opportunities for patients and family members to enroll in these and other type 1 diabetes clinical research studies (www.T1Diabetes.nih.gov/patient).

How will new knowledge about genes and environmental triggers help people with type 1 diabetes, like Katie and Ellie Clark? The potential for this research to positively affect the lives of people with type 1 diabetes is far-reaching. Genetic and environmental factors identified through these research efforts could be used as targets for researchers to develop novel disease prevention strategies. If a certain virus were found to contribute to disease onset, researchers could pursue the development of a vaccine against the virus. In addition, knowledge about which genes are passed down from one generation to the next will allow researchers to more easily identify who is at high risk for developing the disease and therefore intervene earlier in the disease process, before the destruction of insulin-producing beta cells even starts. Preventing disease onset means that children like Ellie would never have to endure the thousands of finger sticks or pump changes/insulin injections that are now part of their everyday lives. Disease prevention also prevents the development of life-threatening complications. Therefore, pursuing research on the genetic underpinnings and environmental triggers of type 1 diabetes has great potential for allowing children, like Ellie, to live the life that their parents dreamt for them.

EMERGING RESEARCH OPPORTUNITIES RESULTING FROM THE SPECIAL STATUTORY FUNDING PROGRAM FOR TYPE 1 DIABETES RESEARCH

The Special Funding Program has fueled the emergence of a wide range of research opportunities. Opportunities that have largely been made possible by the Special Funding Program have been excerpted below from the Type 1 Diabetes Research Strategic Plan (see Appendix 6).

Risk Assessment

Identify and Optimize the Detection of Immunologic, Genetic, and Metabolic Markers of Type 1 Diabetes:

- ▶ Achieve accurate identification of those at risk in the general population by improved measurement of autoantibodies and other autoimmune markers.
- ▶ Achieve accurate type 1 diabetes risk assessments by exploiting additional genetic markers.
- ▶ Achieve accurate type 1 diabetes risk assessment using metabolic parameters.

Immunopathogenesis

Understand the Interplay Between Early Environmental Encounters and the Immunoregulatory Defects That Results in Beta Cell Destruction in Human Type 1 Diabetes:

- ▶ Improve understanding of the interplay between the environment and the immune system, which leads to the autoimmune destruction of beta cells in humans.
- ▶ Create a database of the genes expressed in the pancreas at sequential stages of type 1 diabetes development, as well as accessible tissues involved in the (auto)immune response.

Advance Basic Understanding of Facets of the Human Immune Response (e.g., Regulatory T Cells, Innate Immunity) That Have Recently Been Appreciated as Key Mediators of Beta Cell Destruction:

- ▶ Improve the understanding of the generation and function of regulatory T cells in type 1 diabetes.
- ▶ Develop better assays to measure the autoimmune response and to serve as biomarkers of response to therapy.

- ▶ Detect and measure the autoimmune response, as well as the mass and function of beta cells, at the level of the pancreatic islet.

Clinical Trials

Identify an Intervention Capable of Long-term Reversal of Recent Onset Type 1 Diabetes Without Concomitant Short- or Long-term Adverse Effects:

- ▶ Standardize trial design and outcome measures.
- ▶ Determine whether combination therapies offer improvements in terms of efficacy over monotherapies directed solely at the immune system.
- ▶ Identify novel therapeutic agents.
- ▶ Assess the safety of all immunomodulating or immunosuppressive therapies tested in type 1 diabetes.
- ▶ Enhance animal models for the study of relevant immune mechanisms and potential interventions.

Develop a Safe and Universal Means for the Primary Prevention of Type 1 Diabetes:

- ▶ Further investigate the potential utility of autoantigens as “vaccines” for prevention of anti-beta cell autoimmunity.
- ▶ Determine the importance of exposure to cow’s milk protein in the development of islet autoimmunity and type 1 diabetes via TRIGR.
- ▶ Begin the design and implementation of clinical trials aimed at reducing the impact of environmental factors that trigger islet autoimmunity and type 1 diabetes *in utero*, during early postnatal life, and later in development.