

# NIDDK

## Recent Advances & Emerging Opportunities

January 2022



U.S. Department of Health and Human Services  
National Institutes of Health  
National Institute of Diabetes & Digestive & Kidney Diseases

The NIDDK mission encompasses research on a broad array of diseases and conditions affecting people of all ages. This “word cloud,” built from a set of terms representing NIDDK research mission areas and efforts, illustrates how scientific inquiry and disciplines, the people who conduct and participate in research, and the dissemination of discoveries all contribute to the NIDDK research enterprise. Highlights of these activities are presented in this annual publication.

*Note: The relative size of words in this cloud is a design choice and does not imply differences in importance and/or funding levels.*

# NIDDK

## Recent Advances & Emerging Opportunities

January 2022



U.S. Department of Health and Human Services  
National Institutes of Health  
National Institute of Diabetes & Digestive & Kidney Diseases

## TABLE OF CONTENTS

### MESSAGE FROM THE DIRECTOR

### CROSS-CUTTING SCIENCE..... 1

COVID-19: A Continuing Story ..... 2

Progress in Understanding Multiple Impacts  
of COVID-19 on the Body..... 3

New Insights into Links Between  
SARS-CoV-2 Infection and Diabetes ..... 3

COVID-19 and Liver Injury ..... 4

Blood-based Biomarker May  
Both Predict COVID-19 Severity  
and Explain Tissue Injury..... 4

The NIDDK Strategic Plan for Research:  
Pathways to Health for All..... 6

NIDDK Efforts To Promote  
Scientific Workforce Diversity..... 8

### DIABETES, ENDOCRINOLOGY, AND METABOLIC DISEASES ..... 10

Research on Type 1 Diabetes..... 13

Testing a Next-generation Artificial Pancreas  
Device for Managing Type 1 Diabetes ..... 13

Large Study Sheds New Light on  
the Complex Type 1 Diabetes  
Genetics Landscape and Potential  
Drug Targets ..... 14

Research on Type 2 Diabetes..... 14

Determining What Prediabetes  
Means in Older Adults ..... 14

Young People with Type 2 Diabetes Are  
at High Risk for Having Complications  
in the Prime of Their Lives ..... 15

Treatment for Teenagers with  
Type 2 Diabetes Can Help Keep  
the Disease from Getting Worse ..... 15

Improving Type 2 Diabetes Self-care  
by Focusing on Friends and Family..... 16

Multi-ethnic Genetic Study  
Greatly Expands Knowledge About  
Regulation of Glucose Levels ..... 16

Protein Identified That Governs Adaptive  
Increase in Beta Cell Production and  
Function in Obesity..... 17

Research on Diabetes Complications ..... 17

Alterations in Brain Structures  
in Type 1 Diabetes ..... 17

Risk Factors Associated with  
Cognitive Decline in Older People  
with Type 1 Diabetes ..... 18

New Analysis of Diabetic Foot Ulcers  
Reveals Reduced Activation of  
Wound-healing Immune Response..... 18

Metabolic Regulators of Health and Disease ..... 19

Selective Fat-cell Elimination in Mice Leads to a Profound Increase in Bone Mass .....	19
Individual Genetic Variations in Fat and Liver Cells Predict Adverse Drug Response.....	20
Toward Precision Medicine in Diabetes Treatment: Anniversary Symposium Brings Together Research Leaders To Propel Progress.....	21
RADIANT: The Rare and Atypical Diabetes Network.....	22
The Diabetic Foot Consortium.....	23
PERSONAL PERSPECTIVE: Making Extraordinary Contributions to Type 1 Diabetes Prevention Research .....	24
PERSONAL PERSPECTIVE: Contributing to Research on Type 2 Diabetes in Youth.....	29
<b>OBESITY .....</b>	<b>33</b>
The Diet Debate .....	35
Weighing in on the Diet Debate: Low-carbohydrate Versus Low-fat .....	35
Genetic Underpinnings of Obesity.....	35
Large-scale Genetic Study Identifies Genes That Increase Risk of Obesity While Protecting Against Other Metabolic Diseases .....	35

Molecular Underpinnings of Exercise .....	36
Succinate Regulates Muscle Adaptations to Exercise in Mice and Humans .....	36
The Physiology of the Weight Reduced State .....	37
NIH Seminar Series: Obesity and COVID-19 .....	39
<b>DIGESTIVE DISEASES AND NUTRITION.....</b>	<b>41</b>
Gut Microbiome and Nutrition.....	44
Designer Foods Target Microbiome To Boost Host Health.....	44
Identifying Therapeutic Targets for Inflammatory Bowel Disease .....	45
New Potential Therapeutic Target Identified for Crohn’s Disease.....	46
Identifying Therapeutic Targets for Intestinal Disease in Children .....	46
Potential Therapeutic Target Identified To Prevent Severe Diarrhea in Children .....	46
New Insight into Intestinal Biology.....	47
Mucus Keeps Gut Bacteria at Bay To Prevent Inflammation.....	47
Liver Maintenance and Regeneration .....	47
Location, Location, Location—Cell Position in Maintaining and Regenerating Liver.....	47

Developing New Liver Disease Therapies.....	48
Lipid Nanoparticles Deliver Promise for Liver Disease Therapies .....	48
Targeted Hormone Treatment Transforms Liver Cells into Fibrosis-fighting Superheroes.....	48
Potential Regeneration Therapy for Liver Injury Activates Timely Burst of Growth Factors.....	49
Antiviral Drug Shows Promise Against Hepatitis C in Animal Models .....	49
Dietary Supplements and Metabolic Health.....	50
A Treatment That May Improve Muscle Insulin Sensitivity in Postmenopausal Women .....	50
Understanding the Role of the Nervous System in Gastrointestinal Health .....	51
NIH Meetings Explore Possible Role of Gut-brain Connections in Parkinson’s Disease .....	52
The NIDDK-supported Research Community and the CCF’s IBD Plexus – A Fruitful Collaboration .....	53
PERSONAL PERSPECTIVE: Advancing Treatments for Inflammatory Bowel Diseases in Children .....	55

## KIDNEY, UROLOGIC, AND HEMATOLOGIC DISEASES..... 59

Clinical Research on Kidney Disease.....	62
Robust Research Pipeline Ensures Exquisite Data Quality Control, Paving the Path Toward Personalized Kidney Care.....	62
Multidisciplinary Approach Uncovers Potential New Biomarker for Kidney Function.....	63
Identifying Therapeutic Targets for Kidney Diseases.....	63
Zeroing in on the Role of KIM-1 in Diabetic Kidney Disease .....	63
Exploring the Role of Exosomes in the Progression of Polycystic Kidney Disease.....	64
Research on Lower Urinary Tract Symptoms and Disorders.....	64
A New Approach to Studying the Links Between Bladder and Brain Activity in People .....	64
Potential Therapeutic Target for Erectile Dysfunction Following Prostate Cancer Surgery .....	65
Blood Stem Cells and the Bone Marrow.....	66
Elucidating New Mechanism for Blood Stem Cell Migration from the Bone Marrow.....	66

New Insight into the Mouse Blood Stem Cell Aging Process .....	66
Developing Vascular Access Technologies To Improve Kidney Disease Treatment—Highlights of Small Business Innovation Research .....	68
CCEH: Increasing Collaboration and Access to Critical Research Resources for Research To Combat Nonmalignant Blood Diseases .....	70

STORY OF DISCOVERY: The NIDDK Provides Foundational Research Support for First FDA-approved Therapy To Treat the Metabolic Disease Primary Hyperoxaluria Type 1 .....	70
--	----

PERSONAL PERSPECTIVE: Helping People Hydrate To Prevent Kidney Stones.....	73
---	----

<b>ACKNOWLEDGMENTS.....</b>	<b>79</b>
-----------------------------	-----------

# Message from the Director



As the Director of the National Institute of Diabetes and Digestive and Kidney Diseases (NIDDK), I am pleased to present this annual report highlighting the research efforts and programs supported by the Institute. The NIDDK has a broad research responsibility that includes some of the most common, debilitating, and costly conditions affecting Americans. These conditions include diabetes and other endocrine and metabolic diseases; liver disease and other digestive diseases and conditions, such as inflammatory bowel disease and irritable bowel syndrome; nutritional disorders; obesity; kidney diseases, such as polycystic kidney disease and glomerular disease; urologic diseases and conditions, such as interstitial cystitis/bladder pain syndrome, prostatitis, and urinary tract infection; and blood diseases.

In late 2021, we released the *NIDDK Strategic Plan for Research*, which presents a broad vision for accelerating research over the next 5 years to improve the health of people who have or are at risk for diseases within its mission. The *Plan* reflects NIDDK's commitment to empowering a multidisciplinary research community, engaging diverse stakeholders, and improving pathways to health for all through prevention, treatment, and health equity. The scientific goals of the *Strategic Plan* are detailed in the "Cross-Cutting Science" chapter of the 22nd edition of this report. In addition, this report describes recent NIDDK-supported scientific advances on topics such as:

- The multiple ways that SARS-CoV-2 infection can influence the development and course of organ

damage and disease—including diabetes, liver injury, and kidney injury—yielding potential targets for therapeutic intervention;

- A next-generation artificial pancreas device that helps adolescents and young adults with type 1 diabetes keep their blood glucose (sugar) levels in a healthy range;
- A finding that youth with type 2 diabetes are likely to experience serious complications of the disease by the time they are young adults;
- A wealth of discoveries about genes affecting blood glucose levels from a large study of people with different ancestries;
- Demonstration that blood glucose control and blood pressure management might preserve cognitive function in people with type 1 diabetes as they age;
- Discovery that the healing of diabetic foot ulcers may be impaired by a reduced immune response at the wound;
- Demonstration that a plant-based, low-fat diet leads to less caloric intake and a significant loss of body fat compared to an animal-based, low-carbohydrate diet, with each diet followed for 2 weeks;
- Identification of regions of the genome that are linked to both elevated levels of body fat and protection from some of the negative health impacts of obesity;
- A finding that a dietary supplement containing ingredients from locally available, nutrient-dense foods provided to malnourished children greatly improved growth and markers of development;
- Discovery of how a gene variant may contribute to intestinal inflammation, offering new possible therapeutic approaches to treat Crohn's disease;
- A study of signals passed between intestinal cells during rotavirus infection that has uncovered a potential therapeutic target for this common cause



of diarrhea, dehydration, and death in children around the world;

- A test of a new combination of drugs in animal models infected with different hepatitis C viral subtypes, which could represent the next generation of treatments with a range of benefits;
- Discovery that small packages of cellular components, called exosomes, may play a role in autosomal dominant polycystic kidney disease, in studies of mice and kidney cells grown in the lab;
- Demonstration of the efficacy of a new, more easily implemented, and natural approach to studying bladder function in people; and
- The role of pain sensory nerves in the migration of blood stem cells from the bone marrow into the blood circulation, a finding that may help to improve stem cell-based therapeutic protocols.

This report also includes personal perspectives of those who have given time and effort to participate in NIDDK-sponsored clinical research or who have worked with participants as part of a study team. A mother describes how enrolling her newborn daughter in a clinical study, which aims to identify the environmental factors involved in type 1 diabetes, helped accelerate the diagnosis of celiac disease when she was a toddler. A young woman tells of her participation, which began during childhood, in the longest-running study of its kind

comparing the effectiveness of medications, lifestyle modifications, or combinations of the two in youth with type 2 diabetes. A young man shares his story of participating in clinical research as a child to determine whether specific tests could predict response to standard medical therapy for children newly diagnosed with a type of inflammatory bowel disease called ulcerative colitis. Two study coordinators describe their experiences helping kids and adults navigate the logistics of participation in a clinical trial aimed at preventing kidney stones through improved hydration, including challenges presented by the COVID-19 pandemic.

The NIDDK continues efforts to ensure that knowledge gained from its research is disseminated to health care providers, patients, and the public. We develop science-based information on diseases and disorders within the NIDDK mission and distribute it through our information and outreach programs and our website. I invite you to visit us at [www.nidDK.nih.gov](http://www.nidDK.nih.gov). Health information, news, and scientific advances related to NIDDK research are also available on our Twitter feed: @NIDDKgov.

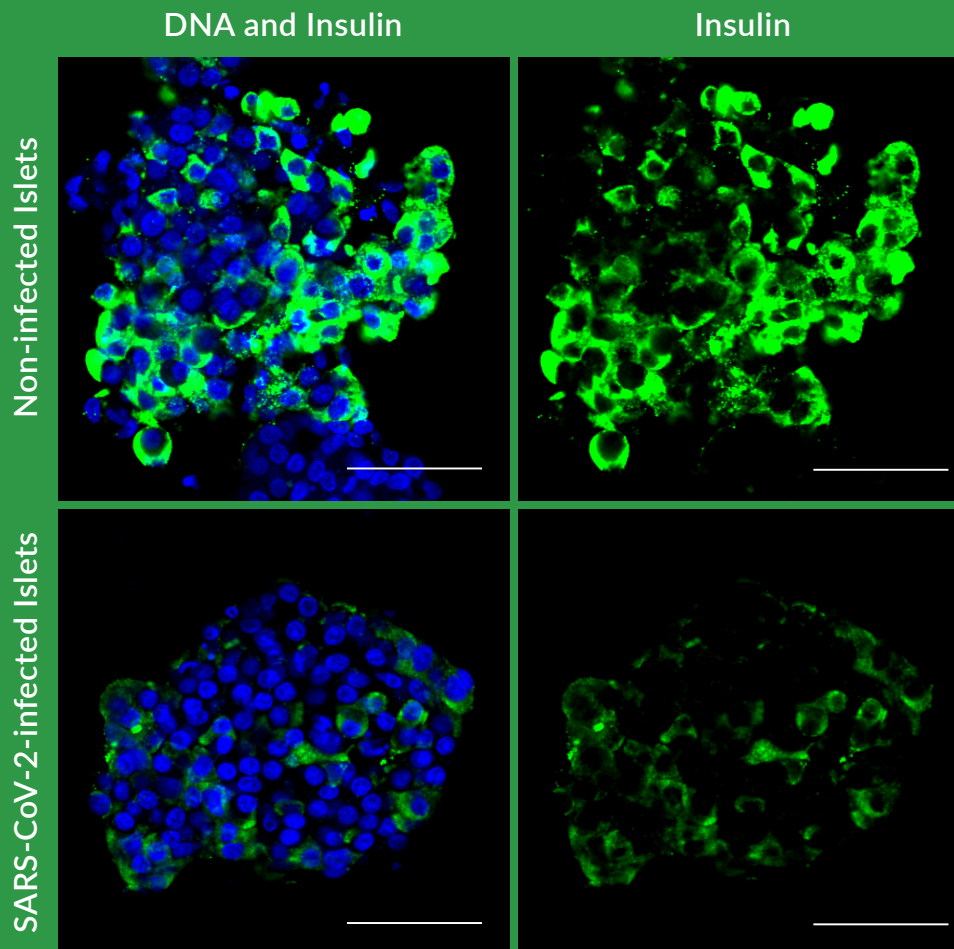
This report reflects only a fraction of the immense body of NIDDK-funded research across the country, performed by basic scientists, clinical investigators, and patient volunteers. Moving forward, we remain committed to supporting these important areas of research and translating scientific discoveries into improvements in the health and quality of life of all people.

The efforts featured in this publication reflect the core mission of the NIDDK, including the Director's guiding principles:

- Maintain a vigorous investigator-initiated research portfolio
- Support pivotal clinical studies and trials
- Promote a steady and diverse pool of talented new investigators
- Foster exceptional research training and mentoring opportunities
- Ensure knowledge dissemination through outreach and communications

## **Griffin P. Rodgers, M.D., M.A.C.P.**

Director  
National Institute of Diabetes and Digestive and Kidney Diseases  
National Institutes of Health  
U.S. Department of Health and Human Services



Previous research has shown that SARS-CoV-2 (the virus that causes COVID-19) uses its spike protein to anchor to another protein called ACE2 on the surface of human cells before infecting them. As described in this chapter, researchers have not only demonstrated SARS-CoV-2's ability to infect human pancreatic and liver cells but have also performed studies that are elucidating the impact of infection on diseases such as diabetes that are associated with greater health problems in people with COVID-19 and also may even be newly triggered or worsened by infection. For example, as shown in the images above, researchers found that the level of insulin (green) production detectable in beta cells of human pancreatic islets is much lower in islets newly infected with SARS-CoV-2 in the laboratory (bottom row) than in non-infected islets (top row). Combined with other findings, this significant change provides important insights into how COVID-19 can change the fate and function of cells important to the development and course of diabetes.

Panel images provided by Dr. Shuibing Chen, Weill Cornell Medicine. Originally published in Tang X, Uhl S, Zhang T,...Chen S. SARS-CoV-2 infection induces beta cell transdifferentiation. *Cell Metab* 33: 1577-1591, 2021. DOI: [10.1016/j.cmet.2021.05.015](https://doi.org/10.1016/j.cmet.2021.05.015) and reprinted under the terms of the [Creative Commons CC-BY license](https://creativecommons.org/licenses/by/4.0/).

# Cross-Cutting Science

*Medical advances are not usually achieved in great, intuitive leaps. More often, new prevention strategies, treatments, and cures result from a long, gradual accumulation of knowledge from years of scientific research. Insights into fundamental biologic building blocks and processes—genes, the proteins they encode, the inner workings of cells, and the ways cells communicate with each other—can have broad and far-reaching implications. Indeed, many significant advances in our understanding and treatment of disease can be traced to laboratory studies whose relevance to health could not have been fully known or appreciated at the time they were conducted.*

There are also moments when the biomedical research enterprise is called upon to rapidly harness knowledge, resources, and expertise across many fields to meet extraordinary and urgent challenges that threaten the public health. The past year has seen triumphs in research to combat COVID-19, as effective, FDA-authorized vaccines developed by the NIH and others were quickly deployed across the Nation to protect people against the worst outcomes of infection. However, the battle with COVID-19 is not yet won, and research on treatments and on the pandemic's short- and long-term human health consequences continues.<sup>1</sup> At the same time, in the wake of sobering pandemic statistics and heart-wrenching events across the Nation, researchers have joined forces with diverse communities to find ways to identify, address, and ultimately overturn health disparities and promote health equity in the United States. Described in this chapter are examples of NIDDK efforts to overcome these critical challenges through research and scientific workforce development for the betterment of public health today and into the future.

## COVID-19: A CONTINUING STORY

Since its recognition as a pandemic disease in early 2020, COVID-19 has brought illness, disability, and death to millions of people in the United States and around the world, while upending lives and normal activities for many millions more. People who contract the disease, which is caused by SARS-coronavirus-2, or SARS-CoV-2, can find themselves facing anything from a mild, cold-like illness to life-threatening symptoms that land them in a hospital intensive care unit. While the most obvious symptoms experienced by the majority of people with severe COVID-19 disease are respiratory, SARS-CoV-2 infection can cause damage to organs and tissues throughout the body.<sup>2</sup> Moreover, although people of any age or state of health can contract the virus, those

with chronic diseases such as type 2 diabetes, obesity, chronic kidney disease, and chronic liver disease are at greater risk of developing severe disease leading to hospitalization and death.<sup>3</sup>

*Critical findings about COVID-19 and its relationship to diseases such as diabetes and liver and kidney disease have already emerged from NIDDK-supported studies.*

The NIH has pursued a multi-pronged approach to balance the enormous and acute research needs imposed by COVID-19 with the research it supports to benefit the health of people with or at risk of many other diseases and conditions. Thus, throughout the course of the pandemic, the NIDDK and other NIH Institutes and Centers have sought to mitigate its impact on biomedical research and the scientific workforce. These efforts have included new flexibilities in the receipt, review, and funding of research and researcher training grant applications, which have been made available to help

---

<sup>1</sup> See <https://covid19.nih.gov/> for comprehensive information on the NIH research response to COVID-19, including efforts focused on vaccine development and testing, improved COVID-19 testing strategies, more effective treatments, reducing disparities, better basic understanding of the virus that causes COVID-19, and the lingering effects of the disease in many individuals (sometimes referred to as "long-COVID" or Post-Acute Sequelae of SARS-CoV-2).

<sup>2</sup> Gupta A, et al. *Nat Med* 26: 1017-1032, 2020.

<sup>3</sup> Centers for Disease Control and Prevention: <https://www.cdc.gov/coronavirus/2019-ncov/hcp/clinical-care/underlyingconditions.html>. Accessed November 5, 2021.

sustain investigators in the face of pandemic-induced disruptions to research and research career advancement. For example, for scientists at early career stages—who are particularly vulnerable to deleterious career impacts—the NIDDK provided opportunities to extend their existing research training and career development funding for up to 1 year, providing a little more breathing room so these individuals could apply for their next grants or search for the next jobs in their careers. The NIDDK has also worked with its clinical trial investigators to ensure participant safety and research progress as studies reopened, and to launch several much-needed clinical trials and studies after delays imposed by COVID-19.

### **NIDDK Research and COVID-19**

Understanding and preventing COVID-19's deleterious effects on people who have diseases and conditions within the NIDDK's mission are important goals for the Institute. Alarming, there is also evidence that COVID-19 can increase people's risk for developing acute or chronic diseases. Thus, in 2021 the NIDDK supported multiple research efforts to address these areas of concern. For example, the NIDDK organized a meeting of the Diabetes Mellitus Interagency Coordinating Committee (DMICC) in March so that leading diabetes researchers and DMICC representatives from across the federal government could review what data had emerged to date about diabetes and COVID-19. They identified numerous challenges to address in such areas as health disparities, the effects of diabetes treatments on COVID-19 outcomes, and the influence of each disease on the course of the other. As co-chair of the NIH Obesity Research Task Force, the NIDDK also hosted a virtual symposium on "Obesity and COVID-19" in September (see feature in the Obesity chapter).

There have also been opportunities for NIDDK-supported research projects and programs to pivot some activities toward COVID-19. For example, the NIDDK Diabetes Centers program has actively pursued efforts to leverage infrastructure, facilities, and internal programs to address COVID-19 and diabetes. Researchers have also been able to leverage their NIDDK-supported projects toward COVID-19 through supplemental support from central NIH COVID-19 initiatives—for example, the NIH Rapid Acceleration of Diagnostics initiative has provided supplemental funding to an NIDDK-supported diabetes center to scale rapidly a COVID-19 testing program for people in Georgia at high risk due to diabetes and

other metabolic disease. NIDDK staff have also been active in helping to shape trans-NIH efforts that could help people with COVID-19 who are also affected by diseases within the NIDDK research mission both in the near term and in the future.

Importantly, critical findings about COVID-19 and its relationship to diseases such as diabetes and liver and kidney diseases, have already emerged from teams of intramural and extramural researchers supported in whole or in part by the NIDDK, some of which are summarized below. To continue this course of discovery, the NIDDK recently funded a variety of basic and clinical research projects focused on understanding mechanisms underlying the impact of COVID-19 on organs, tissues, and biological systems within the Institute's purview. These projects may also contribute to our understanding of the lingering symptoms experienced by many people who have had COVID-19.

### **Moving Forward in the Era of COVID-19**

The NIDDK is encouraged by the enormous achievements we have seen in a short span of time. In particular, the rapid development of effective vaccines has been critical to saving lives and preserving health. However, the overall impact of the pandemic has been extremely sobering. As is the case with many infectious diseases, COVID-19 may be with us for a long time, testing our endurance, ingenuity, and health. There are challenges imposed by emerging viral variants, health disparities, and the continued need for interventions that can protect people for whom COVID-19 vaccines are not fully effective—just to name a few. As the Nation continues its battle to emerge from the pandemic, the NIDDK will continue seeking to ensure the health and safety of researchers, study volunteers, and patients, while also helping to maintain research progress and a robust scientific workforce across all the areas within its research mission.

*In the era of COVID-19, the NIDDK continues to strive toward ensuring the health and safety of researchers, study volunteers, and patients, while maintaining research progress and a robust scientific workforce across all the areas within its research mission.*

## PROGRESS IN UNDERSTANDING MULTIPLE IMPACTS OF COVID-19 ON THE BODY

### New Insights into Links Between SARS-CoV-2 Infection and Diabetes:

Research has revealed that the virus causing COVID-19 can infect pancreatic  $\beta$  (beta) cells, changing their function—a critical discovery toward understanding a devastating relationship between COVID-19 and chronic disease. Despite SARS-CoV-2 being primarily a respiratory virus, it affects multiple organs and systems during acute illness and sets the stage for long-term symptoms in a significant proportion of those infected. Thus, understanding the mechanisms underlying COVID-19's many complications and comorbidities is important to reducing the tremendous burden this disease is already imposing on people and health care systems around the world. For example, people with diabetes are at higher risk for grave outcomes from COVID-19. Moreover, researchers are trying to figure out whether SARS-CoV-2 infection may also trigger new onset of diabetes, as there is increasing evidence with the number of new cases appearing after infection. One unanswered question is whether SARS-CoV-2 exerts such effects through direct infection of susceptible cells and tissues.

In an initial study, researchers demonstrated in laboratory models how SARS-CoV-2 infects certain human cells and tissues—including pancreatic  $\beta$  cells and liver cells. They also found that donated human pancreatic islets were susceptible to SARS-CoV-2 infection. The team then collaborated with additional researchers to investigate whether or not  $\beta$ -cell infection also occurred in people infected with SARS-CoV-2, and, if so, what effects this had on the cells. Comparisons of islets from deceased human donors showed molecular signatures of SARS-CoV-2 in the  $\beta$  cells of those who had died from COVID-19. Closer examination of islets infected with SARS-CoV-2 in the laboratory revealed that infected  $\beta$  cells produced less insulin and produced proteins normally made by other pancreatic cells—suggesting that infection was causing them to change their function, or "transdifferentiate." A similar pattern of changes was also seen in islets from deceased donors with COVID-19, confirming that SARS-CoV-2 infection can alter  $\beta$ -cell activity in people.

Further analysis suggested that an intracellular pathway activated in response to viral infection and other stressors was governing the observed  $\beta$ -cell changes. Using artificially transdifferentiated human cells, the scientists identified a chemical that in subsequent experiments

blocked the  $\beta$ -cell transdifferentiation process upon SARS-CoV-2 infection, most likely through inhibition of virus-induced changes to the identified stress-activated pathway. These results not only suggest that SARS-CoV-2 infection directly induces changes in  $\beta$  cells that could affect the course or onset of diabetes, but also provide hope that this process may be preventable.

Tang X, Uhl S, Zhang T,...Chen S. SARS-CoV-2 infection induces beta cell transdifferentiation. *Cell Metab* 33: 1577-1591, 2021.

Yang L, Han Y, Nilsson-Payant BE,...Chen S. A human pluripotent stem cell-based platform to study SARS-CoV-2 tropism and model virus infection in human cells and organoids. *Cell Stem Cell* 27: 125-136, 2020.

*Evidence for SARS-CoV-2 infection and reprogramming of  $\beta$  cells provides new insights into the multi-faceted impact of this virus.*

**COVID-19 and Liver Injury:** Researchers have identified a link between an immune system factor and promotion of blood vessel damage that may explain liver injury in people with COVID-19—and help to inform future research efforts to prevent and combat such liver damage. People with COVID-19 commonly have liver injury. This injury appears to be linked to vascular (blood vessel) damage, but the underlying mechanisms have been elusive. A hint came from the observation that people with COVID-19 have elevated levels of an immune system factor called IL-6 in their blood, along with factors that either promote damage to cells lining blood vessels and/or enhance blood clot formation. This suggested that COVID-19-induced liver injury could be mediated through cells called liver sinusoidal endothelial cells, or LSECs, highly specialized cells that line the tiniest blood vessels in the liver and are critical to proper liver function and health. LSECs are both prime targets for damage and producers of factors involved

*Liver injury is commonly seen in people hospitalized with COVID-19. A new study reveals a mechanism that could explain why and how this occurs, providing insights important to evaluating COVID-19 induced liver damage and finding ways to prevent and treat it.*

in inflammation and clotting—production that could be stimulated by IL-6.

In support of this hypothesis, researchers found that among people hospitalized with COVID-19, higher levels of a liver injury biomarker correlated with higher levels of factors that promote blood clotting—including one produced primarily by LSECs, called factor VIII—as well as biomarkers of inflammation and IL-6. Comparing liver specimens from deceased donors at a microscopic level, the scientists found a similar pattern: among donors who had had COVID-19, those who had higher levels of the liver injury biomarker showed a greater abundance of cells and factors involved in injury, inflammation, and clotting in the vicinity of LSECs. This finding also tracked with higher levels of IL-6, again suggesting a role for this immune system factor. To test the possible role of IL-6 directly, the researchers treated laboratory samples of human LSECs with a complex of IL-6 and a soluble form of its receptor protein. They found that IL-6-treated LSECs produced several of the proinflammatory and clotting factors seen in people with COVID-19 and liver injury—a response that could be blocked by further treating the cells with inhibitors of this IL-6 signaling pathway.

Together with other experimental results, these study findings provide evidence that liver injury in COVID-19 may be mediated by IL-6 acting on LSECs, a mechanism that could be targeted therapeutically to prevent and effectively treat liver damage.

McConnell MJ, Kawaguchi N, Kondo R, ... Iwakiri Y. Liver injury in COVID-19 and IL-6 trans-signaling-induced endotheliopathy. *J Hepatol* 75: 647-658, 2021.

**Blood-based Biomarker May Both Predict COVID-19 Severity and Explain Tissue Injury:** Researchers have found that changes to the amounts and types of DNA present in the blood of people with COVID-19 not only correlate with disease severity but may also be mediating injury to kidneys and other organs—a discovery that could help tailor clinical management and advance treatment for COVID-19.

People normally have a small amount of DNA in their blood that isn't contained within cells. This so-called "cell-free DNA," or cfDNA, usually comes from the cells that give rise to immune system and blood cells. However, the source, amount, and characteristics of cfDNA can change in certain diseases and conditions—sometimes before symptoms are apparent—so cfDNA

*Researchers have identified a potential blood-based biomarker for COVID-19 disease severity and tissue injury that could aid efforts to improve disease outcomes for people.*

can be a useful tool in diagnosing and treating disease. Because COVID-19 severity varies so much from person to person, researchers wondered whether cfDNA could be an easy and useful way to predict COVID-19 outcomes. To find out, they evaluated blood samples voluntarily provided by 85 people with COVID-19 across the spectrum of disease severity, from non-hospitalized people with mild symptoms to hospitalized people requiring intensive care. They also evaluated blood samples from people without COVID-19 for comparison. The results showed that the people with COVID-19 not only had markedly higher levels of cfDNA overall early in the course of disease, but also that higher amounts of certain types of cfDNA correlated with more severe disease and even death. Moreover, the researchers found that multiple tissues and organs contributed to the elevated cfDNA, including lung, blood vessels, liver, heart, fat tissue, and kidney, and that in some cases tissue-specific cfDNA correlated with known markers of inflammation and tissue injury.

Based upon other research, the team also suspected that elevated cfDNA might not just be a marker of COVID-19 disease severity and tissue injury but also might incite some of that injury itself. Through experiments using laboratory grown mouse kidney cells, they found that exposure to the cfDNA from people with COVID-19 caused the cells to overproduce a potentially self-damaging molecule—a process that could largely be prevented by adding an inhibitor of the pathway thought to mediate this kidney cell response to cfDNA. Together, these findings—made possible by generous patient volunteers, some of whom did not survive COVID-19—not only suggest that cfDNA could be useful for predicting disease severity, but might also point the way to treatments that stave off some COVID-19-induced tissue injury.

Andargie TE, Tsuji N, Seifuddin F, ... Agbor-Enoh S. Cell-free DNA maps COVID-19 tissue injury and risk of death and can cause tissue injury. *JCI Insight* 6: e147610, 2021.

# The NIDDK Strategic Plan for Research: Pathways to Health for All

In December 2021, the NIDDK released the [Strategic Plan for Research](#), which presents a broad vision for accelerating research over the next 5 years to improve the health of people who have or are at risk for diseases within our mission. This overarching Strategic Plan complements the NIDDK's disease-specific planning efforts and will guide the Institute to build on its over 70 years of discovery, progress, and innovation.

The Strategic Plan is based on extensive input from leading researchers and patient advocates across the country who served on a strategic plan working group, numerous organizations and individuals who provided ideas in response to public Requests for Information, and members of the NIDDK's Advisory Council. It includes four major Scientific Goals, each with a set of research opportunities, around a unifying theme: to empower a multidisciplinary research community, engage diverse stakeholders, and leverage discoveries of connections among diseases to improve prevention, treatment, and health equity—pathways to health for all. Woven throughout the Plan are cross-cutting topics that are integral to all the Scientific Goals: reducing health disparities and increasing health equity among racial and ethnic minority populations

*To improve the health of people who have or are at risk for diseases in the NIDDK mission, the Institute is committed to empowering a multidisciplinary research community; engaging diverse stakeholders; and leveraging discoveries of connections among diseases to improve prevention, treatment, and health equity—pursuing pathways to health for all. This unifying theme is highlighted throughout the Strategic Plan.*

and others who are underserved; improving women's health; strengthening biomedical workforce diversity and training; and serving as an efficient and effective steward of public resources.

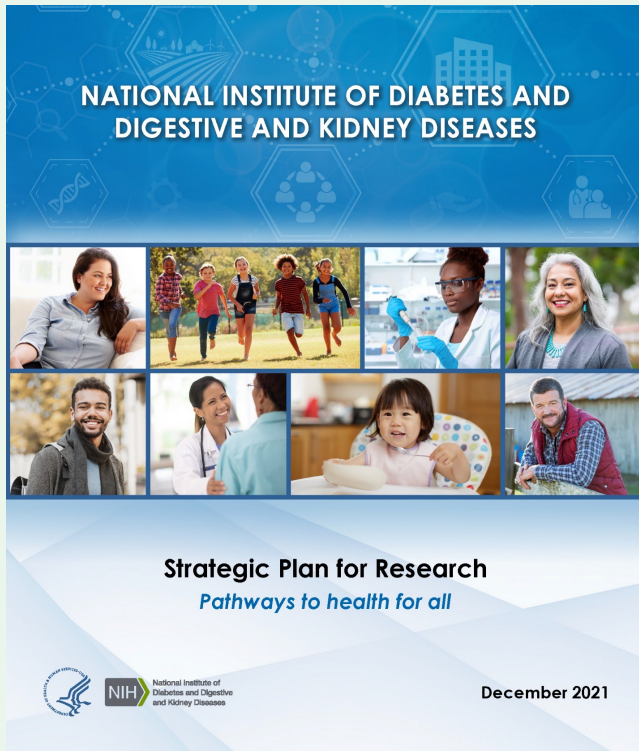
## The Strategic Plan's four major Scientific Goals are to:

1. Advance understanding of biological pathways and environmental contributors to health and disease;
2. Advance pivotal clinical studies and trials for prevention, treatment, and cures in diverse populations;
3. Advance research to disseminate and implement evidence-based prevention strategies and treatments in clinics and community settings, to improve the health of all people, more rapidly and more effectively; and
4. Advance stakeholder engagement, including patients and other participants as true partners in research.



*This image depicts the key components of the Strategic Plan, including four major Scientific Goals (shown in hexagons) and cross-cutting topics crucial to the NIDDK's mission (shown in circles).*

For each Scientific Goal, the Strategic Plan presents known challenges, recent advances and discoveries to build on, and a set of broad research opportunities. The Plan also highlights the NIDDK's commitment to serve as an efficient and effective steward of public resources. The



Institute will monitor the progress toward each Scientific Goal and identify areas to strengthen efforts, build on discoveries, and pursue emerging research opportunities. Progress will be shared with stakeholders in future editions of this publication and other venues. (Further information about the goals, research opportunities, stewardship, and the planning process is in the [Strategic Plan](#), available on the NIDDK's website.)

The NIDDK is grateful to all who contributed to the Plan's development, and looks forward to working with communities who share the NIDDK's interest in research to improve people's health and quality of life.



# NIDDK Efforts To Promote Scientific Workforce Diversity

While NIDDK researchers from every race and ethnicity are striving to help achieve health equity, the Institute believes that these vital efforts would be strengthened by having a scientific workforce that better reflects the diverse backgrounds and experiences of the U.S. population. While scientific talent is surely well represented across all groups, opportunity is not. The NIDDK is therefore committed to overcoming the dearth of minority scientists across its mission areas. The NIDDK's Office of Minority Health Research Coordination (OMHRC) works with the NIDDK extramural and intramural research divisions to lead these efforts.

One example of these efforts includes a collaboration, which began in 2003, between the NIDDK and the National Center for Research Resources to support the development and implementation of curriculum-based programs to train diverse doctoral and post-doctoral candidates in clinical research. In 2006, the collaboration ended, but the NIDDK continued the effort through several iterations, culminating in the current program, the Small Grants for New Investigators To Promote Diversity in Health-related Research, in collaboration with the National Human Genome Research Institute. From fiscal years 2010 through 2020, the program provided more than 60 such grants to mostly early career investigators from underrepresented minority groups, several of whom went on to compete successfully for traditional NIH research grants.

The NIDDK also provides support for diverse young investigators through partnerships with professional societies, which are uniquely positioned to work toward enhancing diversity in the biomedical research workforce. These organizations sponsor awards for promising young investigators and have a history of supporting the career development of their members—mechanisms they can harness to diversify the biomedical research workforce. Accordingly, the OMHRC created the Partnerships with Professional Societies to Enhance Scientific Workforce Diversity and Promote Scientific Leadership Program. This

initiative supports grants to societies with a focus on NIDDK mission areas to establish or expand training and career development programs for junior investigators from underrepresented backgrounds.

For example, one such grant enabled the American Gastroenterological Association (AGA) to establish a program called Fostering Opportunities Resulting in Workforce and Research Diversity (FORWARD) that supports underrepresented physician scientists to develop leadership skills, strengthen their research and management skills, and receive mentorship and training from top gastrointestinal investigators. FORWARD scholars participate in the AGA Leadership Development Conference, attend trainings for writing grant proposals and scientific manuscripts, and attend an academic skills workshop. Similarly, a grant to the Endocrine Society supports its Future Leaders Advancing Research in Endocrinology (FLARE) program for basic and clinical research trainees and junior faculty from underrepresented communities who have demonstrated achievement in endocrine research. The program provides structured leadership development and in-depth and practical training in topics ranging from grant writing to lab management. FLARE participants attend an annual workshop where they can network and develop skills such as: identifying and applying for funding, time and lab management, communication, and career development.

These initiatives reinforce other OMHRC programs that support research training for underrepresented minority students in high school and college and are part of the NIDDK's efforts to bring more and more talented individuals from an array of backgrounds into research.

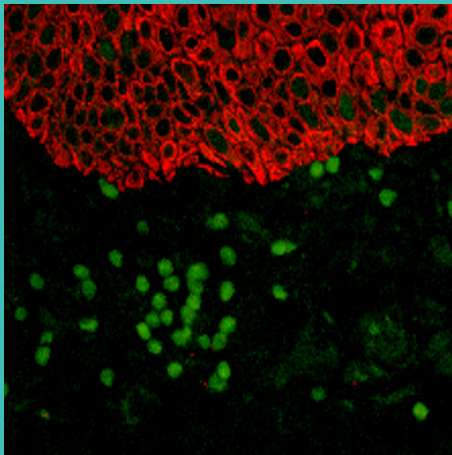
For example, the STEP-UP program provides high school and undergraduate students with biomedical research opportunities for the summer. At the graduate school level, the NIDDK provides scholarship support to students from underrepresented and underserved communities to complete their Ph.D. or M.D./Ph.D. degrees. For

individuals at various stages of their research careers post-doctoral and higher, the NIDDK established the Network of Minority Health Research Investigators to provide mentoring and other information and support for scientists from underrepresented groups and others interested in minority health. The NIDDK's Diversity Supplement Program provides support for promising researchers from backgrounds underrepresented in biomedical research as they

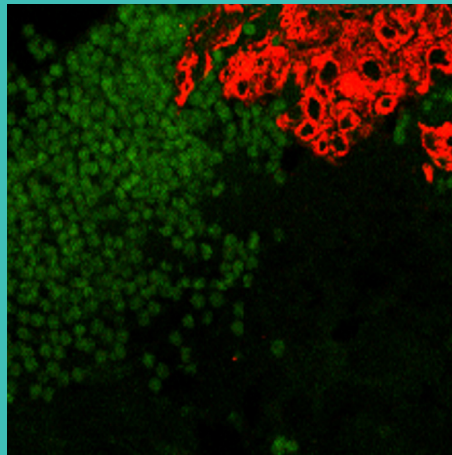
gather preliminary data to apply for their own independent research awards.

The NIDDK firmly believes that the overall biomedical research enterprise will be greatly strengthened by the scientific ideas and talent of people currently underrepresented in research and will help lead us toward health equity for all Americans.

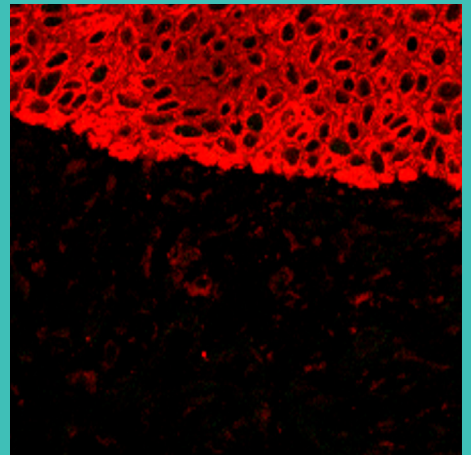
ORAL DAY 3



SKIN DAY 3



DIABETIC FOOT ULCER



As described in this chapter, a recent study described how healing of diabetic foot ulcers (DFUs) may “stall” due to a blunted immune response at the wound. The protein FOXM1 was known to promote the replication and survival of key immune cells involved in wound healing. Researchers found that levels of FOXM1 were reduced in human DFUs compared to levels in 3-day old acute wounds, both oral or non-oral skin wounds. This difference can be seen in the above microscopy image, where FOXM1 protein is indicated in green and keratin (a protein marking epithelial cells in oral, non-oral, and DFU skin) is indicated in red. Additional experiments in mice (not shown) confirmed that inhibiting FOXM1 reduced the wound-healing immune response and delayed wound closure. These results suggest that slow or delayed healing of DFUs could be due to reduced FOXM1 activity, a finding which opens new avenues to diagnose and treat these wounds.

Images provided by Dr. Maria Morasso, NIAMS/NIH and Dr. Marjana Tomic-Canic, University of Miami Miller School of Medicine. Originally published in Sawaya AP, Stone RC, Brooks SR,...Morasso MI, and Tomic-Canic M. Deregulated immune cell recruitment orchestrated by FOXM1 impairs human diabetic wound healing. *Nat Commun* 11: 4678, 2020. DOI: [10.1038/s41467-020-18276-0](https://doi.org/10.1038/s41467-020-18276-0) and reprinted under the terms of the [Creative Commons CC-BY](https://creativecommons.org/licenses/by/4.0/) license.

# Diabetes, Endocrinology, and Metabolic Diseases

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

Diabetes is a debilitating disease that affects an estimated 34.2 million people in the United States—or 10.5 percent of the total population—and is the seventh leading cause of death.<sup>1</sup> Although overall rates of diabetes-related complications have declined substantially in recent years, disease burden remains significant as the number of people with diabetes is still very high.<sup>2</sup> Diabetes can affect many parts of the body and is associated with serious complications, such as heart disease and stroke, blindness, kidney failure, and lower-limb amputation. In addition to these human costs, the estimated total financial cost for diagnosed diabetes in the United States in 2017—including costs of medical care, disability, and premature death—was \$327 billion.<sup>3</sup> Effective therapy can prevent or delay diabetic complications, but more than one-fifth of U.S. adults with diabetes are undiagnosed and therefore not receiving therapy.<sup>1</sup>

*Diabetes affects an estimated 34.2 million people in the United States—or just over 1 in every 10 people.<sup>1</sup> Another 88 million U.S. adults have “prediabetes,” which puts them at elevated risk of developing type 2 diabetes.<sup>1</sup> The estimated total financial cost for diagnosed diabetes in the United States in 2017 was \$327 billion.<sup>3</sup>*

Diabetes is characterized by the body’s inability to produce and/or respond appropriately to insulin, a hormone that is necessary for the body to absorb and use glucose (sugar) as a cellular fuel. These defects result in persistent elevation of blood glucose levels and other metabolic abnormalities, which in turn lead

to the development of disease complications. The most common forms of diabetes are type 1 diabetes, in which the body loses its ability to produce insulin, and type 2 diabetes, in which the body becomes resistant to insulin, with subsequent impaired insulin production. In addition, a significant proportion of pregnant women each year are diagnosed with gestational diabetes, a form of diabetes that develops during pregnancy, but in many cases resolves after pregnancy. However, women who develop gestational diabetes are at greater risk of developing type 2 diabetes later in life. Untreated, any form of diabetes during pregnancy increases the risk of serious complications for the mother and baby before, during, and after delivery. There are also rarer forms of diabetes associated with specific genes such as those known as monogenic diabetes.

*In addition to increasing the risk for complications of vision loss, kidney failure, and amputation, diabetes doubles the risk for heart disease, many forms of cancer, some forms of dementia, hearing loss, erectile dysfunction, urinary incontinence, and many other common diseases.<sup>2</sup>*

<sup>1</sup>Centers for Disease Control and Prevention. *National Diabetes Statistics Report, 2020*. Atlanta, GA: Centers for Disease Control and Prevention, U.S. Department of Health and Human Services, 2020.

<sup>2</sup>*Diabetes in America, 3rd edition*, Cowie CC, et al., Eds. Bethesda, MD, National Institutes of Health, NIH Pub No. 17-1468, 2018.

<sup>3</sup>American Diabetes Association. *Diabetes Care* 41: 917-928, 2018.

Type 1 diabetes affects approximately 5 percent of adults diagnosed with diabetes and the majority of children and youth diagnosed with diabetes.<sup>1</sup> It most often develops during childhood but may appear at any age. Type 1 diabetes is an autoimmune disease in which the immune system launches a misguided attack and destroys the insulin-producing  $\beta$  (beta) cells of the pancreas. Thus, people with type 1 diabetes require lifelong insulin administration—in the form of multiple daily injections or via an insulin pump—to regulate their blood glucose levels.

The NIDDK's landmark Diabetes Control and Complications Trial (DCCT)/Epidemiology of Diabetes Interventions and Complications (EDIC) study demonstrated that keeping blood glucose levels as near to normal as safely possible reduced the risk of eye, kidney, nerve, and heart complications associated with type 1 diabetes. These results underscore the importance of pursuing research to develop novel technologies to help people with type 1 diabetes manage their blood glucose levels with less burden, including new methods to improve blood glucose monitoring and insulin delivery. In this regard, NIDDK-supported research has contributed to the development or testing of new U.S. Food and Drug Administration-approved diabetes management technologies, including artificial pancreas devices that automatically link glucose monitoring and insulin delivery. Researchers are also working to further develop and enhance  $\beta$  cell replacement therapies, such as islet transplantation, to cure type 1 diabetes.

*NIDDK-supported research has contributed to the development and testing of new diabetes management technologies, including new artificial pancreas devices that automatically link glucose monitoring and insulin delivery.*

Type 2 diabetes is the most common form of the disease, affecting about 90 to 95 percent of adults diagnosed with diabetes in the United States.<sup>1</sup> The risk for developing type 2 diabetes is associated with older age, obesity, family history of diabetes, history of gestational diabetes, impaired glucose metabolism, physical inactivity, and race/ethnicity. Type 2 diabetes occurs at higher rates among racial and ethnic minority populations in the United States, including African Americans, Hispanic and Latino Americans, American Indians, some Asian Americans, and Native Hawaiians and Pacific Islanders.<sup>1</sup> Gestational diabetes is

also a risk factor: about half of women with gestational diabetes will develop type 2 diabetes within 5 to 10 years after giving birth.<sup>4</sup>

In people with type 2 diabetes, cells in muscle, fat, and liver tissue do not properly respond to insulin. As a result, the pancreas initially produces more insulin to compensate. Gradually, however, the pancreatic  $\beta$  cells lose their ability to secrete enough insulin to restore balance, and the reduction of insulin secretion, relative to the body's needs, results in elevated and abnormal blood glucose levels. Treatment approaches for managing glucose levels include lifestyle modification (i.e., diet and exercise), and oral and injected medications, with insulin often required as the disease progresses. There are also an estimated 88 million U.S. adults who have "prediabetes," in which blood glucose levels are higher than normal but not as high as in diabetes.<sup>1</sup> This population is at elevated risk of developing type 2 diabetes. Fortunately, the NIDDK-supported Diabetes Prevention Program (DPP) clinical trial has shown that people with prediabetes who make lifestyle changes to lose weight by adopting a healthy diet and increasing physical activity can dramatically reduce their risk of developing type 2 diabetes. To a more limited degree, the safe and well-tolerated drug metformin can also help prevent or delay type 2 diabetes.

*Type 2 diabetes is increasingly being diagnosed in children and adolescents, disproportionately affecting youth from racial and ethnic minority populations in the United States. NIDDK-supported research has shown that type 2 diabetes is more aggressive and difficult to treat in youth compared to adults.*

Previously called "adult onset" diabetes because it is predominantly diagnosed in older individuals, type 2 diabetes is increasingly being diagnosed in children and adolescents, and disproportionately affects youth from racial and ethnic minority populations in the United States. Results from NIDDK-supported research has shown that the disease may be more aggressive and difficult to treat in youth compared to adults. This is worrisome because those with early disease onset are at especially high risk for developing complications. In

<sup>4</sup> Kim C, et al. *Diabetes Care* 25: 1862-1868, 2002.

addition, increasing rates of type 2 diabetes in girls may lead to more women who enter pregnancy with diabetes, and maternal diabetes during pregnancy—either onset of type 2 diabetes before pregnancy or the development of gestational diabetes during pregnancy—confers an increased risk of type 2 diabetes in offspring. Thus, the rising rates of diabetes and prediabetes in young women could contribute to a cycle of ever-growing rates of diabetes, in addition to increasing risks for pregnancy complications. The advent of type 2 diabetes in youth has the potential to worsen the enormous health burden that diabetes already places on the United States.

The most common forms of diabetes, type 1 and type 2, are associated with variations in multiple genes. Some rare forms of diabetes, called monogenic diabetes, result from mutations in a single gene. Neonatal diabetes mellitus (NDM) and maturity-onset diabetes of the young (MODY) are the two main forms of monogenic diabetes. Many cases of monogenic diabetes may be incorrectly diagnosed, which may complicate management. For example, when high blood glucose is first detected, type 1 or type 2 diabetes may be considered the diagnosis instead of monogenic diabetes. A correct diagnosis allows for proper treatment and can lead to better glucose control and improved health. There are also unusual forms of diabetes that differ from known types, called “atypical diabetes.” People with atypical diabetes may be diagnosed with and treated for type 1 or type 2 diabetes, but not have a history or signs consistent with their diagnosis. In addition, individuals may have a condition called latent autoimmune diabetes in adults (LADA). Finally, more recently, an additional type of diabetes, referred to as type 3c, has been described that is associated with disease or deficiency of the exocrine pancreas, as may occur with pancreatitis. It is critical to discover and define rare and atypical forms of diabetes, which could lead to better diagnoses, improved treatments, and potential prevention of these diseases.

The NIDDK is supporting research to better understand metabolism and the mechanisms that lead to the development and progression of diabetes and the many other endocrine and metabolic diseases within its mission; such research ultimately will spur the design of potential new intervention strategies. In parallel, based on knowledge from past scientific research investments, the NIDDK is vigorously pursuing studies of prevention and treatment approaches for these diseases.

Highlights of recent advances from NIDDK-supported research on diabetes, endocrinology, and metabolic diseases are provided in this chapter.

## RESEARCH ON TYPE 1 DIABETES

**Testing a Next-generation Artificial Pancreas Device for Managing Type 1 Diabetes:** A clinical trial showed that a next-generation artificial pancreas device outperformed a commercially available first-generation device in adolescents and young adults with type 1 diabetes by increasing time that participants’ blood glucose (sugar) levels were in a healthy range without increasing episodes of dangerously low blood glucose (hypoglycemia). These results are important because glucose management is extremely challenging during adolescence and young adulthood. Artificial pancreas technology, or a closed-loop system, aims to automate type 1 diabetes management by measuring blood glucose levels using a continuous glucose monitor and automatically delivering insulin when needed using an insulin pump. In 2016, the U.S. Food and Drug Administration approved Medtronic’s MiniMed™ 670G as the first commercially available closed-loop system in the United States. However, research on real-world use of the 670G device shows a high discontinuation rate, especially in adolescents and young adults, suggesting that improvements are needed. In a recent clinical trial, researchers compared a next-generation, experimental closed-loop device from the same company with the 670G system. The experimental device had improvements such as advanced computer algorithms that control insulin delivery and easier operation.

The trial enrolled 113 female and male adolescents and young adults, ages 14 to 29 years, with type 1 diabetes. Participants were randomly assigned to use either the experimental device or the commercial 670G device for 12 weeks, and then were switched to the other device for 12 more weeks. The results showed that the experimental device improved the amount of time that participants’ blood glucose levels were in a healthy target range, both during the daytime hours and during the entire 24-hour day and night period, without increased episodes of hypoglycemia. The improvements translated to about 1 hour more per day in the target glucose range than achieved when using the 670G device. Participants also reported greater user satisfaction with the experimental device compared to the commercial device.

These trial results showed that the next-generation device outperformed the 670G device and was also more user-friendly. Identifying new and improved type 1 diabetes management technologies—particularly for groups for whom glucose management is challenging—could help people achieve recommended

blood glucose levels with less burden, toward improving their short- and long-term health.

Bergenstal RM, Nimri R, Beck RW,...Phillip M; For the FLAIR Study Group. A comparison of two hybrid closed-loop systems in adolescents and young adults with type 1 diabetes (FLAIR): A multicentre, randomised, crossover trial. *Lancet* 397: 208-219, 2021.

**Clinical trial results showed that a next-generation artificial pancreas device outperformed a commercially available device in helping adolescents and young adults with type 1 diabetes keep their blood glucose levels in a healthy range.**

### Large Study Sheds New Light on the Complex Type 1 Diabetes Genetics Landscape and Potential Drug Targets:

Conducting the largest and most ancestrally diverse study of type 1 diabetes genetics thus far, researchers have identified new regions of the genome associated with the disease and potential drug targets. In recent years, there has been tremendous research progress in understanding genetic contributors to type 1 diabetes, an autoimmune disease in which the insulin-producing cells in the pancreas are destroyed by the immune system. However, most studies have only included people of European ancestry. Less is known about genetic risk for type 1 diabetes in people with other ancestries, who are experiencing increasing rates of the disease. Additionally, little is known about how genetic variants associated with type 1 diabetes cause disease. This information has implications for understanding the disease process and developing future strategies to prevent or treat type 1 diabetes.

In new research, scientists analyzed genetic contributors to type 1 diabetes by studying over 61,000 participants (with and without the disease), including individuals from diverse ancestries. This approach led to the identification of 36 new gene regions associated with type 1 diabetes, some of which are also associated with other autoimmune diseases. Additionally, the scientists did “fine mapping” studies to pinpoint the specific disease-causing variants in genetic regions previously associated with type 1 diabetes, toward elucidating the role these variants may play in the disease process. In another set of experiments, they used their genetic association results with data about other autoimmune diseases to identify 12 genes that could potentially be

drug targets for type 1 diabetes. Drugs targeting these genes (by affecting the proteins the genes encode) have been studied in clinical trials for other autoimmune diseases, suggesting they could be repurposed for type 1 diabetes. Some of the targets are already being studied in type 1 diabetes clinical trials of various therapies, but others could potentially be explored in future trials to prevent type 1 diabetes.

This large research study with a diverse population has provided important knowledge about the complex type 1 diabetes genetics landscape, revealing new genetic regions associated with the disease, shedding light on how some genetic variants may influence disease, and identifying potential drug targets. Understanding what genes play a role in type 1 diabetes, and what role they play, paves the way to identify new targets to prevent or treat this autoimmune disease.

Robertson CC, Inshaw JRJ, Onengut-Gumuscu S,...Rich SS. Fine-mapping, trans-ancestral and genomic analyses identify causal variants, cells, genes and drug targets for type 1 diabetes. *Nat Genet* 53: 962-971, 2021.

## RESEARCH ON TYPE 2 DIABETES

### Determining What Prediabetes Means in Older Adults:

Recent research suggests that we need better ways to assess the risk of future type 2 diabetes in older adults. People with blood glucose (sugar) levels higher than normal but lower than the levels used to define diabetes are said to have “prediabetes,” because they are known to be at increased risk for the disease. Fortunately for those at high risk, the landmark, NIDDK-led Diabetes Prevention Program (DPP) clinical trial demonstrated that type 2 diabetes can often be prevented or delayed through diet and exercise changes designed to yield modest weight loss. This DPP lifestyle intervention was particularly effective in older DPP participants, and the infrastructure to provide a group-based adaptation of the approach has increased nationally in recent years. Targeting those programs to people most likely to benefit from them depends on knowing who has a high likelihood of developing diabetes. There are standard definitions for prediabetes using different measurements that work fairly well for individuals who are middle aged or younger, but less is known about how well they predict the development of type 2 diabetes among older individuals.

Researchers therefore studied a 3,412-person group of volunteers without diabetes (60 percent female; 17

percent Black; 83 percent White) who had an average age of about 75. The study tested different diagnostic criteria for prediabetes to compare how well they worked in older adults: using “HbA1c” levels; using fasting glucose levels; using either HbA1c or fasting glucose levels; or requiring the criteria for both measures to be met. The proportion of participants diagnosed with prediabetes differed greatly depending on which criteria were applied: 29 percent if using both HbA1c and fasting glucose, 44 percent based on HbA1c levels alone, 59 percent using fasting glucose levels alone, and 73 percent according to at least one of the two measures. As expected, a higher proportion of the individuals meeting one or more criteria for prediabetes developed type 2 diabetes than those who did not have prediabetes at the outset. However, for each of the individual prediabetes definitions, more of the volunteers saw their glucose or HbA1c levels improve to the normal range than progress to the diabetes range. Further, those considered to have prediabetes were also less likely to develop type 2 diabetes than they were to die of any cause. Taken together, these findings suggest the need for a better test to identify future diabetes risk in people over age 70.

Rooney MR, Rawlings AM, Pankow JS, ...Selvin E. Risk of progression to diabetes among older adults with prediabetes. *JAMA Intern Med* 181: 511-519, 2021.

### **Young People with Type 2 Diabetes Are at High Risk for Having Complications in the Prime of Their Lives:**

People with type 2 diabetes diagnosed during youth have a high risk of developing complications at early ages and are more likely than adults with the disease to develop multiple complications within 15 years after diagnosis. These findings are from a follow-up study of the Treatment Options for Type 2 Diabetes in Adolescents and Youth (TODAY) clinical trial, which tested 3 therapeutic strategies in participants between 10 and 17 years old.

The original study produced sobering results, showing that type 2 diabetes is harder to treat and worsens more rapidly in young people than in adults. In view of these findings, researchers recognized that it was important to learn more about the clinical course of pediatric type 2 diabetes, so they invited the TODAY participants to continue in a follow-up study dubbed TODAY2. As in the original study, the more than 500 participants who agreed to continue were predominantly from minority groups disproportionately affected by type 2 diabetes: 38 percent were Hispanic, and 36

percent were non-Hispanic Black, while less than 20 percent were non-Hispanic White. TODAY2 revealed that serious complications of diabetes arose more quickly among the study participants than would be expected in adults with the disease or in young people with type 1 diabetes: within 15 years of their initial diagnosis, at least 60 percent of the participants in the original TODAY study had one or more serious diabetic complications, such as kidney disease, peripheral nerve damage, or eye disease, and at least 28 percent had two or more. Participants of minority racial and ethnic groups and those whose levels of HbA1c—a measure of blood glucose (sugar) levels over time—were comparatively high during the follow-up period were more likely than other participants to develop one or more complications. These results reveal that children and teens who develop type 2 diabetes may potentially face burdensome complications of the disease for the entirety of their adult lives. Because approved methods for treating pediatric type 2 diabetes are frequently ineffective, the results also underscore the critical need to identify better therapies for those who have the disease, as well as better prevention strategies for those at risk.

Bjornstad P, Drews KL, Caprio S, ...Zeitler P. Long-term complications in youth-onset type 2 diabetes. *N Engl J Med* 385: 416-426, 2021.

*Youth with type 2 diabetes are likely to experience serious complications of the disease by the time they are young adults.*

**Treatment for Teenagers with Type 2 Diabetes Can Help Keep the Disease from Getting Worse:** Although a recent study did not identify a means to partially reverse pediatric type 2 diabetes and restore health, new results show that the interventions tested were helping to keep the disease from getting worse. Research in adults with recent-onset type 2 diabetes has shown that in some cases, early, aggressive treatment can partially restore the capacity of the pancreas to secrete insulin to control blood glucose (sugar) levels. Because type 2 diabetes tends to progress more rapidly when it occurs in young people, the Restoring Insulin Secretion (RISE) Pediatric Medication Study sought to determine whether early treatment similarly could improve natural blood glucose control in youth aged 10 to 19 years who either recently developed type 2 diabetes or who had elevated glucose levels close to that of diabetes. The original study tested whether treating youth with a long-acting form of insulin for 3 months followed by use



of the oral diabetes medication metformin for 9 months was more effective than simply using metformin for 12 months. Unfortunately, neither treatment approach succeeded in improving natural insulin secretion by the pancreas for blood glucose control: on average, the participants' blood glucose levels neither improved nor declined significantly during treatment. In follow-up, the researchers sought to determine whether the year of treatment was sufficient to stabilize blood glucose levels or whether longer-term medication may be needed. Thus, they continued measuring blood glucose levels for 9 months after the conclusion of the treatment phase. They found that type 2 diabetes progressed significantly more rapidly once the study participants stopped taking medication. These results suggest that it is valuable for young people with type 2 diabetes to use insulin or metformin to control their blood glucose levels until better, more durable means of treating pediatric diabetes are identified.

Hannon TS, Edelstein SL, Arslanian SA,...Nadeau KJ; for the RISE Consortium. Withdrawal of medications leads to worsening of OGTT parameters in youth with impaired glucose tolerance or recently-diagnosed type 2 diabetes. *Pediatr Diabetes* 21: 1437-1446, 2020.

**Improving Type 2 Diabetes Self-care by Focusing on Friends and Family:** Researchers have found that type 2 diabetes self-care may be improved by addressing the role of family and friends. Effective treatment of diabetes requires a long-term effort by the individual to take medications correctly and on time, and to manage diet and physical activity in a health-promoting way. Family and friends can be powerful allies in facing these challenges, but they can also have negative effects, such as by encouraging unhealthy meal choices or discouraging exercise. In a study of about 500 female and male adults with type 2 diabetes, researchers were investigating whether an intervention called REACH, which involves sending individualized text messages about type 2 diabetes self-care to participants' mobile phones, could improve diabetes self-care behaviors. A large percentage of the study participants were from racial and ethnic minority populations, and many had low income and were underserved. Half of the participants were assigned to the REACH group, and half received standard care (the control group). This study demonstrated that REACH resulted in short-term improvements to medication adherence and blood glucose control.

In an ancillary study, researchers further divided the REACH group into two groups, one of which continued

in the original REACH program. The other, designated REACH+FAMS, received one-on-one coaching to help them set self-care goals along with suggestions for ways both to achieve more helpful support from friends and family and to reduce harmful interactions. This group also received individualized text messaging support related to the goals established in the coaching sessions, so their text messages were different from those sent to the REACH-only group. The REACH+FAMS participants also were invited to nominate a friend or family member to serve as an ally in their diabetes self-care; this part of the intervention was optional, and as such, only 42 percent of the REACH+FAMS group had allies involved in the study who received text messages designed to help the study participant achieve self-care goals. After 6 months, the REACH+FAMS participants had improved family/friend involvement in their self-care, did better in some measures of self-care, and had a better diet, on average, than participants in either the control or the REACH-only groups, although there were no statistically significant differences in exercise between the three groups. These results suggest that coaching and mobile technology might be valuable for helping people with type 2 diabetes take better care of themselves and adopt a better diet by improving interactions with family and friends.

Mayberry LS, Berg CA, Greevy RA,...Elasz TA. Mixed-methods randomized evaluation of FAMS: a mobile phone-delivered intervention to improve family/friend involvement in adults' type 2 diabetes self-care. *Ann Behav Med* 55: 165-178, 2021.

**Multi-ethnic Genetic Study Greatly Expands Knowledge About Regulation of Glucose Levels:** A new analysis of six genetic data sets from people in different parts of the world has yielded a wealth of discoveries about the wide array of genes affecting control of glucose (sugar) levels in human beings. The study of more than 280,000 people, 30 percent of whom had non-European heritage and none of whom had diabetes, looked at whether genetic variations across the genome were associated with 4 different measures of glucose control or insulin levels. In this way, the researchers identified 99 more regions of the genome affecting glucose levels than had been known previously, 24 of which could not have been identified by simply enlarging the European data set to match the size of the multi-ethnic collection. Data from other studies conducted about the same time show that 27 of the 99 regions are linked to type 2 diabetes risk in East Asian and trans-ancestry studies; it remains to be determined whether the remaining 72 gene regions affect the risk

of diabetes or simply contribute to normal variation in glucose levels among people without diabetes. Because these regions of the genome are large enough to contain multiple genes, the researchers sought to narrow these down to identify the most likely genes affecting glucose levels. Importantly, including genetic data from varied groups was instrumental in helping identify potential candidate genes within the genetic regions. Among the other key findings of the study, 80 percent of the glucose-level affecting genetic variations were found in most or all of the lineages, suggesting significant commonality in the genetics of blood glucose control. However, genetic risk scores for type 2 diabetes that had been previously developed based primarily on research with people of European descent proved significantly less predictive in other populations, demonstrating another key reason for studying the biology of glucose control in people with a wide array of differing ancestries, an approach that also may accelerate the search for new and better ways to treat or prevent the disease.

Chen J, Spracklen CN, Marenne G,...Barroso I for the Meta-analysis of Glucose and Insulin-related Traits Consortium. *The trans-ancestral genomic architecture of glycemic traits.* *Nat Genet* 53: 840-860, 2021.

**A large study of people with different ancestries has yielded a wealth of discoveries about the genes affecting blood glucose (sugar) levels.**

### **Protein Identified That Governs Adaptive Increase in Beta Cell Production and Function in Obesity:**

Researchers studying insulin-producing  $\beta$  (beta) cells in a mouse model have identified a protein involved in increasing  $\beta$  cell numbers in mice with obesity. This finding, discovered by a team led by scientists in the NIDDK's Intramural Research Program, may open new avenues for treatment of type 2 diabetes, a disease associated with obesity. People with obesity and type 2 diabetes often have insulin resistance, a condition where tissues do not respond to insulin normally. To try to compensate for insulin resistance, the body often triggers an increase in  $\beta$  cell production that can delay or even prevent diabetes by increasing insulin levels. However, our understanding of the underlying cellular signals that trigger this adaptive increase in  $\beta$  cell numbers has been limited.

To further examine how  $\beta$  cells are regulated in obesity, researchers investigated the protein  $\beta$ -arrestin-1.

$\beta$ -arrestin-1 is part of the signaling machinery in many cells, including  $\beta$  cells, and was previously implicated in  $\beta$  cell activity. To explore the role of  $\beta$ -arrestin-1 in obesity, researchers generated male mice lacking  $\beta$ -arrestin-1 in their  $\beta$  cells. Both these mice and a group of normal mice were fed a high-fat diet so they would gain excess weight, and the mice's  $\beta$  cells were then compared. The scientists found that  $\beta$ -arrestin-1 was required for the increased  $\beta$  cell numbers seen in obesity. Mice without  $\beta$ -arrestin-1 in their  $\beta$  cells also had poorer metabolic health than normal mice of similar weight fed a similar diet. In comparison, obese mice with more  $\beta$ -arrestin-1 than usual in their  $\beta$  cells had better metabolic health than similar mice with normal levels of  $\beta$ -arrestin-1. Further investigations into how  $\beta$ -arrestin-1 was causing these differences revealed that a lack of  $\beta$ -arrestin-1 reduced production of the protein Pdx1, which regulates  $\beta$  cell function and overall  $\beta$  cell numbers. Similar poor outcomes on metabolic health and Pdx1 levels were also seen when  $\beta$ -arrestin-1 levels were reduced in human  $\beta$  cell lines in the laboratory.

Further research would be required to determine if  $\beta$ -arrestin-1 plays a similar adaptive role in people (both males and females) who have obesity and/or type 2 diabetes. If so, therapies to increase  $\beta$ -arrestin-1 activity might increase  $\beta$  cell numbers and thus might help treat or prevent diabetes.

Barella LF, Rossi M, Pydi SP,...Wess J.  *$\beta$ -arrestin-1 is required for adaptive  $\beta$ -cell mass expansion during obesity.* *Nat Commun* 12: 3385, 2021.

## **RESEARCH ON DIABETES COMPLICATIONS**

**Alterations in Brain Structures in Type 1 Diabetes:** In a study of people who have had type 1 diabetes for many years, researchers found structural alterations in regions of the brain involved in cognition and voluntary muscle movement—results that provide much-needed new insights into how the disease affects the brain. In type 1 diabetes, the complications to organs such as the heart, kidney, and central nervous system are associated with elevated blood glucose (sugar) levels and episodes of low blood glucose levels. Although the risk of impairment to the central nervous system in type 1 diabetes is not completely understood, several studies have reported subtle differences in cognitive ability between people with and without the disease.

To address the question of how type 1 diabetes may affect brain structure, the researchers analyzed magnetic

resonance imaging (MRI) brain scans of 61 women and men with long-duration (average of 21 years) disease compared to a control group of 54 women and men of similar ages who did not have type 1 diabetes. They found significant structural differences in several brain regions (e.g., striatum, thalamus, and mesial temporal cortex) between those with the disease and the control group. These brain regions are largely associated with cognition and motor functions. The findings of this study identify structural brain alterations in people with long-duration type 1 diabetes. Future studies could assess whether such changes in the brain correlate with differences in cognition, or other functions, and pave the way toward a longer-term goal of identifying ways to intervene and protect the brain.

Filip P, Canna A, Moheet A,...Mangia S. Structural alterations in deep brain structures in type 1 diabetes. *Diabetes* 69: 2458-2466, 2020.

**Risk Factors Associated with Cognitive Decline in Older People with Type 1 Diabetes:** Researchers identified risk factors that contribute to cognitive decline in people with type 1 diabetes as they age—findings that could inform strategies to preserve cognitive function over the lifespan. Because multiple avenues of research have demonstrated the importance of controlling blood glucose (sugar) levels and led to the development of improved diabetes management technologies, people with type 1 diabetes are living healthier and longer lives. In further research to help people maintain good health as they age, scientists sought to determine whether type 1 diabetes affected the cognitive decline that is seen as people age, as well as to identify risk factors associated with cognitive decline. Understanding any loss of cognitive function is important to ensuring that high quality of life and diabetes self-management can be maintained as people with type 1 diabetes age.

The NIDDK's Epidemiology of Diabetes Interventions and Complications (EDIC) study has followed over 1,000 participants from the landmark Diabetes Control and Complications Trial (DCCT) for over 30 years from a median age of 27. As part of the follow up, participants completed cognitive assessments at the start of the DCCT and 2, 5, 18, and 32 years later (median age of 59), as well as other assessments. Overall, the researchers found that, as the group aged, they performed less well on assessments of memory and psychomotor and mental efficiency. These cognitive declines were associated with higher hemoglobin A1c (HbA1c) levels (a measure of blood glucose levels

over time), more episodes of severe hypoglycemia (dangerously low blood glucose levels), and elevated systolic blood pressure levels. When combined, the presence of these three risk factors was associated with a cognitive decline equivalent to an additional 9 years of age, suggesting premature aging.

Participants who maintained better control of these risk factors, however, showed fewer changes in cognition. Lower average HbA1c, fewer episodes of severe hypoglycemia, and lower blood pressure were each associated with better performance on the cognitive assessments, suggesting that blood glucose control and blood pressure management could help to preserve cognitive function in people with type 1 diabetes as they age. These results add to the wealth of information that has come from the DCCT/EDIC demonstrating the long-term health benefits of maintaining blood glucose levels as close to those of a person without diabetes as safely possible.

*Research findings suggest that blood glucose (sugar) control and blood pressure management might preserve cognitive function in people with type 1 diabetes as they age.*

Jacobson AM, Ryan CM, Braffett BH,...Lachin M for the DCCT/EDIC Research Group. Cognitive performance declines in older adults with type 1 diabetes: Results from 32 years of follow-up in the DCCT and EDIC Study. *Lancet Diabetes Endocrinol* 9: 436-445, 2021.

**New Analysis of Diabetic Foot Ulcers Reveals Reduced Activation of Wound-healing Immune Response:** New research has revealed how diabetic foot ulcers (DFUs) may “stall” in their healing due to a blunted immune response at the wound. These findings may open new avenues for preventing or treating DFUs, a devastating complication of diabetes that can lead to lower limb amputation.

Little is known about how diabetes affects the skin or why some DFUs heal slowly or not at all. To better understand the various factors at work in DFUs, researchers compared the tissue samples collected from 21 individuals with diabetes, including DFUs, to wounded skin from people without diabetes. Scientists compared the genes active in these skin samples and identified several key differences between DFUs and wounded skin from people without diabetes. In general, they found that key molecular players in the immune response necessary for wound healing were dysregulated

in DFUs. In wounded skin from people without diabetes, there was increased activity of a network of genes responsible for recruitment, activation, replication, and survival of immune cells, but the same gene activity was markedly reduced in DFUs. This difference could be responsible for the decreased ability of certain immune cell types to replicate or survive at DFU wound sites, thus reducing their effectiveness during healing. In particular, the activity of the gene encoding the FOXM1 protein, which promotes the replication and survival of key immune cells involved in wound healing, was reduced in DFUs. Further experiments involving FOXM1 in male and female diabetic mouse models confirmed that inhibiting this protein reduced the immune response in wound healing and delayed wound closure, similar to what was found in patients with DFUs.

These results support the idea that DFUs are “stalled” wounds, where the immune system is not stimulated to the level needed to close the wound promptly. This study used samples from a limited number of participants, so further experiments will be needed to confirm that these findings are applicable to a larger number of DFUs. Nonetheless, this new knowledge about immune system activation in normal and chronically unhealing wounds has provided clues as to why some DFUs heal slowly or not at all. Because these findings also shed light on underlying pathways involved in this process, such as the identification of a key role for FOXM1, they also suggest possible new targets for ways to treat and predict healing in DFUs.

This study exemplifies a team science approach that was conducted as an ancillary study of the recently formed NIDDK Diabetic Foot Consortium (see feature later in this chapter) and funded, in part, by the NIH Bench-to-Bedside and Back Program. The goals of the NIH Bench-to-Bedside and Back Program are (1) to fund research teams seeking to translate basic scientific findings into therapeutic interventions for patients, and (2) to increase understanding of important disease processes by addressing barriers, such as the traditional silos between basic and clinical researchers in biomedical research, which can hinder progress toward finding new therapeutics for patients in need.

*Sawaya AP, Stone RC, Brooks SR, Morasso MI, and Tomic-Canic M. Deregulated immune cell recruitment orchestrated by FOXM1 impairs human diabetic wound healing. Nat Commun 11: 4678, 2020.*

*New research reveals how healing of diabetic foot ulcers may be impaired by a reduced immune response at the wound.*

## METABOLIC REGULATORS OF HEALTH AND DISEASE

**Selective Fat-cell Elimination in Mice Leads to a Profound Increase in Bone Mass:** By studying how fat cells in the bone marrow of mice affect bone growth, researchers have discovered potential therapeutic targets that may profoundly increase bone mass to prevent or restore bone loss. Osteoporosis, a bone disease that occurs when the body loses bone mass, is endemic in Western and Asian societies and predisposes older adults to weak bones, fractures, and premature mortality. Recent evidence that fat-cell depletion can trigger bone growth hints at the potential for new osteoporosis therapeutic strategies.

In the current study, researchers generated male and female mice in which administering a chemical could eliminate fat cells, including in bone marrow, which contains fat cells, and in other body fat tissue. They found that within 10 days of being given the chemical, bone growth increased 10-fold in mice without fat cells compared to mice with intact fat cells. New bone mass was markedly increased in older mice but to a lesser extent in young mice, and the new bone had enhanced strength and function compared to bones of mice with normal fat distribution. Moreover, researchers found that while removing the ovaries of normal female mice results in osteoporosis, those in the group that received the chemical and lacked fat cells were protected from bone loss. The researchers next investigated whether the bone growth was caused by depletion of fat cells in bone marrow or in other body fat tissues, in part by seeing whether the effects changed if they restored some fat tissue. When fat tissue from normal mice was transplanted under the skin of mice lacking fat, there was no effect on bone growth. This and other experiments suggested that bone formation is specifically inhibited by fat cells in the bone marrow, rather than by fat in other tissues. Further analysis indicated that marrow fat cells might be tamping down a signaling pathway previously associated with bone growth, providing potential therapeutic targets that may be considered to treat osteoporosis in humans. Because chronic activation of this pathway can lead to excessive growth of several other cell types, the use of

this strategy for treating bone disease may result in yet unknown effects that may present roadblocks.

Moreover, further research is needed to determine if bone growth in humans is similarly affected by marrow fat cells and, if so, to identify strategies for stimulating bone growth that may one day improve treatment or prevention of osteoporosis.

Zou W, Rohatgi N, Brestoff JR, ...Teitelbaum SL. Ablation of fat cells in adult mice induces massive bone gain. *Cell Metab* 32: 801-813, 2020.

### Individual Genetic Variations in Fat and Liver Cells

**Predict Adverse Drug Response:** Researchers have uncovered cell-type specific and individual-specific genetic variations that predispose people to adverse metabolic side effects of commonly prescribed steroid hormones called glucocorticoids. Glucocorticoids are widely used as anti-inflammatory drugs to treat a variety of health problems including rheumatoid arthritis and inflammatory bowel disease. Moreover, the powerful glucocorticoid dexamethasone is now widely used to treat people with advanced COVID-19. However, the long-term use of glucocorticoids often can have severe side effects such as weight gain and elevated blood glucose (sugar) and lipid (fat) levels, since they play an important role in regulating metabolism. Thus, identifying ways to determine who is likely to experience adverse metabolic side effects could help guide and personalize therapy decisions.

In this study, researchers examined changes in gene activity, or gene “expression,” after treating cells in the laboratory with dexamethasone. They studied fat cells and liver cells—two cell types responsible for many of the negative metabolic effects of glucocorticoids. The fat cells used in the experiments were generated from fat tissue samples from eight healthy females undergoing elective abdominal surgery. To generate liver cells, researchers first derived stem cells from blood donated by 11 healthy males and females, and then induced the stem cells to become liver cells. The

researchers found a high degree of variation in the number of dexamethasone-regulated genes between fat cells and liver cells, as well as variation between different individuals. It is known that glucocorticoids regulate gene expression by binding to and activating the glucocorticoid “receptor”—a protein that binds to DNA and regulates the activity of genes. Thus, the researchers examined the individual-specific and cell-type-specific variation in glucocorticoid receptor binding sites on DNA to assess whether that could explain the observed variation in gene expression. Indeed, a high degree of variability was observed in the number and strength of receptor binding sites between individuals and between cell types in response to glucocorticoid treatment. This variation in receptor binding could be traced to single genetic mutations that alter receptor binding and regulation of genes by glucocorticoid drugs.

Remarkably, when they examined people undergoing glucocorticoid therapy for different conditions, they found that these genetic variants could predict a person’s likelihood of adverse metabolic effects such as increases in blood glucose, lipids, and weight.

These findings provide new insight into the genetic variants that predispose people to metabolic side effects and pave the way for a future personalized-medicine approach to the clinical use of glucocorticoid therapeutics.

Hu W, Jiang C, Kim M, ...Lazar MA. Individual-specific functional epigenomics reveals genetic determinants of adverse metabolic effects of glucocorticoids. *Cell Metab* 33: 1592-1609, 2021.

*Researchers have uncovered genetic variations that predispose people to adverse metabolic side effects of commonly prescribed steroid hormones called glucocorticoids.*

# Toward Precision Medicine in Diabetes Treatment: Anniversary Symposium Brings Together Research Leaders to Propel Progress



The year 2021 marked the 100<sup>th</sup> anniversary of the discovery of the hormone insulin at the University of Toronto—a lifesaving discovery for people with type 1 diabetes. To commemorate this anniversary and global advances in diabetes research over the last 100 years, the NIDDK and the Canadian Institutes of Health Research’s Institute of Nutrition, Metabolism and Diabetes partnered to host a virtual symposium titled, “Heterogeneity of Diabetes: Beta Cells, Phenotypes, and Precision Medicine.” The 2-day symposium brought together leaders in diabetes research for thought-provoking presentations and insightful discussions about opportunities and challenges in diabetes research.

Although diabetes has been traditionally classified as either type 1 or type 2, other subtypes of diabetes have been characterized and are now recognized. An even greater range of unrecognized subtypes likely exists, given the considerable variation in the development and clinical presentation of diabetes. Understanding this heterogeneity and providing a better understanding of the pathophysiology is the next frontier in diabetes research with the goal of tailoring prevention and treatment approaches and possibly offering a reclassification of the disease.

The symposium was organized around three major sessions. The first session on “Islet Biology in Health and Diabetes” highlighted research progress in

understanding the biology of islets, the clusters of insulin-producing  $\beta$  (beta) cells and other cell types found in the pancreas. Scientists discussed research to elucidate how islet function is influenced by its neighboring cells in the pancreas; to engineer functional islets in the laboratory as a potential diabetes therapy or cure; and to understand how various types of stress influence the health and function of  $\beta$  cells.

A second session on “Heterogeneity of Diabetic Phenotypes Before and After Diagnosis—Impact on Management and Treatment” probed how diabetes varies in different individuals and the need to move away from a “one-size-fits-all” approach to management. Speakers presented research to characterize the heterogeneity in common and rare forms of diabetes; this led into discussions of efforts and ideas to bring precision medicine—designed to provide the most appropriate treatment to an individual based on genetic, metabolic, physiologic, and phenotypic factors—to diabetes in the third and final session “Precision Medicine in Diabetes.”

Understanding the heterogeneity of diabetes paves the way for new approaches to prevention and treatment, helps fulfill the promise of precision medicine, and is a fitting extension of the research stemming from the seminal discovery of insulin. The NIDDK and the scientific community have made great progress in diabetes research, but the work is not done. As the century since the discovery of insulin comes to a close, the NIDDK will continue working toward finding prevention methods, better treatments and, one day, a cure for all types of diabetes.

# RADIANT: The Rare and Atypical Diabetes Network

A nationwide, NIDDK-funded study will seek to discover the cause of several unusual forms of diabetes. For years, doctors and researchers have been stymied by cases of diabetes that differ from known types. Through research efforts at 20 U.S. research institutions, the Rare and Atypical Diabetes Network (RADIANT) aims to discover new forms of diabetes, understand what makes them different, and identify their causes.

A person with atypical diabetes may be diagnosed and treated for type 1 or type 2 diabetes, but not have a history or signs consistent with their diagnosis. For example, they may be diagnosed and treated for type 2 diabetes but may not have any of the typical risk factors for this diagnosis, such as being overweight, having a family history of diabetes, or being diagnosed as an adult. Alternately, a person with atypical diabetes may respond differently than expected to the standard diabetes treatments. For these reasons, diabetes management can be extremely frustrating for people with atypical forms of the disease.

RADIANT therefore provides these individuals hope for a treatment that better supports their health. Study scientists plan to screen about 2,000 people

with such unknown or atypical forms of the disease. RADIANT researchers will collect detailed health information from participants using questionnaires, physical exams, genetic sequencing, and other tests. People found to have unknown forms of diabetes may receive additional testing. Some participant family members may also be invited to take part in the study. These efforts will help provide a comprehensive description of the genetic and clinical characteristics of rare forms of diabetes, and underscore diabetes as a disease with many forms. Ultimately, the data may provide a framework to establish new diagnostic criteria for diabetes, find new markers for screening, or identify drug targets for new therapies that could ultimately bring precision medicine to diabetes.

The study opened recruitment on September 30, 2020, for people with atypical diabetes or a form of diabetes that seems different from known types of diabetes. RADIANT may one day provide critical understanding of the spectrum of diabetes and improve lives of people with rare forms of the disease and of everyone who cares for them.

Visit [www.atypicaldiabetesnetwork.org](http://www.atypicaldiabetesnetwork.org) for more information on the study and how to join.

# The Diabetic Foot Consortium

Funded by the NIDDK, leading U.S. research institutions have launched the first-ever multi-center network to study diabetic foot ulcers, a common and burdensome complication of diabetes and the leading cause of lower limb amputations in the United States. Up to 34 percent of people with diabetes will develop a foot ulcer in their lifetime, and half of foot ulcers become infected. Each year, about 100,000 Americans with diabetes will lose part of a lower limb because a foot ulcer becomes infected or does not heal. People with diabetic foot ulcers must manage careful at-home foot care over a long time to avoid infection until the wound heals. The Diabetic Foot Consortium (DFC) aims to lay the foundation for a clinical trial network that will address a major research gap to find ways to treat diabetic foot ulcers effectively and to prevent the risk of complicated infections and potential amputation among the more than 34 million American adults with diabetes. The DFC consists of clinical research sites that recruit patients who are undergoing foot ulcer treatment or follow-up care.

The first studies are focusing on finding biological clues, called biomarkers, that can guide treatment in people with diabetic foot ulcers and predict how the ulcer will heal or the likelihood of it returning. For example, a study led by the Indiana University School of Medicine is testing whether body fluid leaking through the skin on a newly healed ulcer can predict the likelihood of its recurrence. The DFC is also photographing the ulcers, collecting wound fluid, blood, urine, and the medical histories of patients with diabetic foot ulcers when they first come to the clinic and during their treatment. These biological samples and information will be available for future research studies to understand diabetic wound healing. An advantage of a consortium such as the DFC is the collaboration among leading investigators in the field of diabetic wound healing and clinical research, working together to participate in the same clinical study. In addition, the DFC will build a roadmap

and framework that will provide an opportunity for researchers to follow up on interesting leads or pursue new studies.

For people with diabetes, foot ulcers can be complex and difficult to manage and can lead to devastating amputations. They affect quality of life and cost the United States up to \$13 billion a year in care. Major obstacles to progress in the field are the diversity of wounds and complexity of causes. Finding biological clues from these ulcers to help tailor treatment to the individual will provide much-needed relief and could prevent future diabetic foot injuries.





## Making Extraordinary Contributions to Type 1 Diabetes Prevention Research

To achieve the goals of preventing, treating, and curing type 1 diabetes, it is imperative to understand the causes of this autoimmune disease. There has been tremendous research progress in understanding genetic contributors to type 1 diabetes. However, genes do not tell the whole story. Intriguingly, not everyone with high-risk genes develops type 1 diabetes, and most people with type 1 diabetes do not have a family history of the disease. Thus, it is believed that environmental factors represent another important piece of the complex type 1 diabetes puzzle. In some people genetically predisposed to the disease, a “triggering” environmental agent may prompt the body’s immune system to destroy the insulin-producing cells in the pancreas, leading to type 1 diabetes. Research to uncover possible environmental triggers of type 1 diabetes and other autoimmune diseases is underway in the NIDDK’s long-standing The Environmental Determinants of Diabetes in the Young (TEDDY) study. The dedicated contributions of over 6,000 families who enrolled their newborns in TEDDY have made the study possible. (See inset for the story of a family participating in TEDDY.)

### STUDYING ENVIRONMENTAL FACTORS INVOLVED IN TYPE 1 DIABETES

Environmental factors that may trigger type 1 diabetes in those at genetic risk include dietary components, environmental toxins, infectious agents, stress, or other factors. Finding these factors is like looking for a needle in a haystack, as people are exposed to large numbers of environmental influences each day. However, identifying these factors is critical to designing new prevention approaches. For example, if a dietary factor is found to play a role in type 1 diabetes onset, or shown to decrease the risk for the disease, then modifying one’s diet could be recommended.

Recognizing the importance of investigating environmental factors that trigger or protect against type 1 diabetes in genetically susceptible children, the NIDDK—with support from the Special Statutory Funding Program for Type 1 Diabetes Research—began the TEDDY study in 2002. Since then, TEDDY has been providing a coordinated and multidisciplinary approach to investigating the possible environmental causes of type 1 diabetes.

### TEDDY—AN EXTRAORDINARY COMMITMENT FROM CHILDREN AND FAMILIES

The ongoing TEDDY study is an international, long-term, ambitious effort involving many investigators and other study personnel at six research centers and a data coordinating center. During enrollment, researchers needed to screen about 425,000 newborns to find infants with genes that predicted an increased risk of type 1 diabetes. The researchers screened newborns who had a parent or sibling with type 1 diabetes (for the “first-degree relative” group), as well as newborns who did not (for the “general population” group). Since then, the scientists have been following over 6,000 of those high-risk children from birth until age 15. These “TEDDY children” are routinely monitored for the development of autoantibodies (proteins in the blood that are early markers of the autoimmune attack in type 1 diabetes) and type 1 diabetes.

The TEDDY study requires an extraordinary commitment from the participating children and their families, and provides for a comprehensive assessment over many years. As such, for 15 years, families are asked to provide regular information on their child’s diet, illnesses, vaccinations, and psychosocial stresses.

# PERSONAL PERSPECTIVE

They are also asked to visit the TEDDY clinic at regular intervals—every 3 or 6 months depending on the child’s age and autoantibody status—where the scientists draw the child’s blood, collect urine, take other measurements (*e.g.*, weight and height), and interview parents about the child’s life events. The parents also collect other biological samples at home, such as regular stool samples. Over the course of the study, the families have contributed over 3 million samples. The hope is that this treasure trove of information will enable scientists to find that needle in a haystack—environmental factors that could trigger or protect against type 1 diabetes.

TEDDY is also studying environmental triggers of other autoimmune diseases in addition to type 1 diabetes. For example, type 1 diabetes shares many risk genes with celiac disease—an autoimmune disease where the immune system responds abnormally to dietary gluten. TEDDY begins testing children for blood markers of celiac disease at age 2 and annually thereafter, so the TEDDY children are regularly monitored for both celiac disease and type 1 diabetes while researchers look for environmental triggers of both diseases.

## NEW INSIGHTS EMERGING FROM TEDDY

The TEDDY study is still ongoing, but important data has already been obtained and participating children have already benefitted. For example, undiagnosed type 1 diabetes can result in a condition called diabetic ketoacidosis, a potentially fatal acute complication. TEDDY researchers demonstrated that children who develop type 1 diabetes while in TEDDY are diagnosed earlier and have lower rates of diabetic ketoacidosis

compared to children diagnosed with type 1 diabetes outside of TEDDY. This earlier diagnosis is due to TEDDY’s close follow-up of children, the heightened awareness of onset of diabetes, and the education the study provides to families about diabetes risk.

Additionally, TEDDY scientists have already started analyzing the precious samples provided by TEDDY families, using sophisticated “-omics” technologies to examine the gene activity, proteins, and metabolites in cells. They are also analyzing the microbes in the children’s gut (*i.e.*, the gut microbiome), which have been shown to be associated with metabolism, and have reported some intriguing results about how environmental factors affect the gut microbiome as children age. For example, they identified three distinct phases of gut microbiome development: a developmental phase (3 to 14 months of age), a transitional phase (15 to 30 months of age) when the microbiome diversifies, and a stable phase (31 to 46 months of age) when the microbiome’s composition is largely established. Through these and other analyses, TEDDY is expanding our understanding of child development and the human microbiome.

Knowledge already gained from TEDDY—and new findings expected in the future—would not be possible without the unwavering and long-term dedication of the TEDDY children and their families, as well as TEDDY researchers. With continued research, TEDDY’s goal is to revolutionize our ability to prevent type 1 diabetes and potentially other autoimmune diseases.

## JENAE'S AND KATELYN'S STORY



Katelyn (front right) pictured with her father Rod, mother Jenae, and sister Madison

Jenae and her husband, Rod, first heard of The Environmental Determinants of Diabetes in the Young (TEDDY) study when their second-born daughter, Katelyn, was only a few hours old. They were unfamiliar with type 1 diabetes when they agreed to have their newborn's blood tested in the hospital as part of TEDDY and found out that she had genes that increased her risk for developing the disease. TEDDY is examining environmental factors that trigger or protect against type 1 diabetes in genetically susceptible children—those with a family history and those without—so the TEDDY researchers invited the family to join the study. Katelyn was eligible to enroll as part of the “general population” group because she had high-risk genes but did not have a family history of type 1 diabetes.

The invitation to join TEDDY was a big ask: requesting that a family without a personal connection to type 1 diabetes enroll their newborn in a 15-year study that would require a lot of work but could help identify ways to prevent this disease. Because most people with type 1 diabetes do not have a family history of the disease, it was critical to include children

like Katelyn in TEDDY. Even though it was a huge commitment, Jenae says, “It really didn't take us long to make the decision [to enroll]. We just felt like ... we can be helpful, and if she ever gets diabetes, they're going to find it faster. We felt, actually, almost more safe about being involved.” They did not know that their decision would have a profound impact on the health of Katelyn, now 12 years old, and on the family's lives.

### KATELYN'S EARLY YEARS IN TEDDY

With Katelyn enrolled in TEDDY, the family started following the demanding protocol. “The first few years of her life, all I remember is poop,” Jenae says with a laugh, referring to the fact that they had to collect monthly stool samples until Katelyn was 4 years old and then every 3 months until she was 10 years old as part of the study. They also visited the TEDDY clinic in Seattle, Washington, every 3 months until Katelyn was 4 years old and then every 6 months thereafter. Jenae shares that it was difficult watching Katelyn have her blood drawn at these visits, but they knew that the blood samples enabled the scientists to test if Katelyn had early markers (autoantibodies) of the autoimmune attack in type 1 diabetes.

*Regarding participating in a clinical study, Jenae's advice to others is: “Do not be afraid to take on things like this. The work is worth it.... It's empowering.”*

Another challenging part of the protocol was when Katelyn started eating solid food and Jenae and Rod had to track everything that Katelyn ate and drank over a 3-day period before study visits. “The first 3 years weren't that bad because as a toddler and baby, you control all of that [food] environment.... It got a little bit trickier as she got older and was in school,” Jenae

# PERSONAL PERSPECTIVE

explains, discussing how they had to adjust the food log if Katelyn did not eat all her packed school lunch.

*“We just felt like ... we can be helpful, and if she ever gets diabetes, they’re going to find it faster,” says Jenae, talking about why she and her husband enrolled their daughter, Katelyn, in the TEDDY study.*

Even during those busy early years, the family took everything in stride. “It really has not been a difficult thing,” Jenae shares. “[TEDDY] has been brilliant this whole time. They make it very easy.... They are very friendly and engaging.” Jenae also got personal satisfaction out of their hard work: “I felt scientific, like I was part of research. There was something fulfilling in that.”

## A DIAGNOSIS OF CELIAC DISEASE

Things drastically changed for the family when Katelyn was 3 years old. In addition to screening for type 1 diabetes, TEDDY also screens children for celiac disease—an autoimmune disease that is triggered by eating foods containing gluten and that shares the same risk genes as type 1 diabetes. Although Katelyn tested negative at her 2-year-old screening, Jenae reports that, “She scored off the chart” for celiac disease autoantibodies at her 3-year-old screening. Their personal doctor then diagnosed Katelyn with celiac disease after a blood test and a biopsy of her small intestine—the organ that is damaged by gluten consumption in people with celiac disease.

Looking back, Jenae recognizes that Katelyn was having some symptoms of celiac disease at the time of her diagnosis, but the symptoms were indistinguishable from common toddler behaviors and thus did not raise a red flag. For example, like many young children, Katelyn would say she was hungry but then not eat. Jenae says that it was only because of TEDDY that Katelyn’s celiac disease was promptly

diagnosed. “Every day, I am unbelievably grateful for TEDDY ... and that we knew [about the celiac disease] and we were immediately able to change course.”

However, changing course to implement a gluten-free diet was extremely difficult. “It was really life-altering,” Jenae recalls, “I remember standing in my kitchen and crying. I didn’t know what was going to poison her,” referring to the small intestine damage that results when people with celiac disease eat gluten. A particular challenge came shortly after Katelyn was diagnosed and the family had to live in a hotel for 6 weeks after a kitchen fire in their home. Jenae and Rod quickly had to learn how to advocate for their daughter at restaurants to ensure that the food was gluten-free. Even with such challenges and the incredible amount of work that Jenae and her husband have had to do to navigate the complexities of celiac disease, she focuses on the positives: “That Katelyn was healthy and that TEDDY had caught [celiac disease] so early so that we would be able to keep her safe.” Her advice to other parents of children with celiac disease is not to be afraid to advocate for your child and to ask for help, such as through support groups that Jenae says have been very helpful to her.

For Katelyn, the hardest part of having celiac disease is feeling left out—like when kids are eating food that she cannot eat. This was especially striking to Jenae after one of Katelyn’s basketball games about 5 years ago—Katelyn was hiding her gluten-free snacks from her teammates. “My heart sank, and I thought: ‘This is ridiculous. People should be able to have ... amazing gluten-free food that everybody could eat.’” That was a major reason why, in 2016, Jenae decided to quit her job in banking and open a gluten-free bakery out of her home. Because she has worked so hard to make her gluten-free baked goods taste like gluten-containing foods, her bakery has been a huge success. She is thrilled to see people’s joy when they have delicious birthday and wedding cakes that look and taste just like cakes they may have had before their celiac diagnosis. She loves that adults and children alike do not feel left out because of their

# PERSONAL PERSPECTIVE

celiac disease. Jenae adds that providing gluten-free wedding cakes brings her much happiness by greatly reducing her clients' stress on their wedding day and, importantly, improving the lives of people with celiac disease—the reason she started the bakery in the first place. Not only is Jenae bringing joy to her clients, she is also serving as a great entrepreneurial role model for Katelyn and her older sister, Madison.

## TEDDY TODAY—AND IN THE FUTURE

Today, Katelyn remains an active TEDDY participant, with the family's contributions mostly involving study visits twice a year. Jenae happily reports: "She's still negative for [type 1 diabetes] antibodies—yay!" Katelyn says that her favorite part of TEDDY is getting prizes at study visits, while the "pokes" (blood draws) are the hardest part. For the pokes, TEDDY staff give her things like a lollipop to distract her from the needle stick pain. "They do such a fabulous job of making it a kid-friendly experience," says Jenae. Katelyn also loves the TEDDY staff, many of whom have been working with them from day 1. "It feels like we're part of a big community," Jenae states. When asked if she would enroll in TEDDY again, Jenae enthusiastically says: "Yes!" Her advice to others is: "Do not be afraid to take on things like this. The work is worth it.... It's empowering."

*"Every day, I am unbelievably grateful for TEDDY," says Jenae, talking about how TEDDY facilitated her daughter's diagnosis of celiac disease.*

Katelyn, who loves camping, video games, playing guitar, hanging out with friends, and cheering on her beloved Seattle sports teams, will age out of TEDDY at 15 years old and will no longer be monitored for type 1 diabetes autoantibodies. Jenae admits that she is a little worried about the coming transition, as the regular monitoring has been a great comfort to the family, but she also adds: "It is fascinating to see how much is being discovered, and to be a tiny little piece of sand in that feels great."

When asked how she would feel if TEDDY found ways to prevent type 1 diabetes and celiac disease because of her participation, Katelyn's response is, "Cool."

Very few people could say that they have been active participants in a research study since birth—what an incredible legacy for Katelyn, her proud parents, and the other TEDDY participants.

## Contributing to Research on Type 2 Diabetes in Youth

Once considered an adult-onset disease, type 2 diabetes is increasingly diagnosed in children and adolescents—especially in racial and ethnic minority populations—largely as a consequence of the increase in childhood obesity. To address the rising trend of type 2 diabetes in young people and develop better treatment strategies, the NIDDK launched the multi-center trial Treatment Options for Type 2 Diabetes in Adolescents and Youth (TODAY) in 2004. It was the first NIDDK-sponsored trial to focus on this challenging problem. The TODAY trial compared 3 treatment options for type 2 diabetes in children and teens in 15 medical centers and their affiliated sites across the United States to identify the best therapeutic strategies to combat this disease in young people. At the conclusion of the TODAY clinical research trial in 2012, participants were transitioned to an observational follow-up study, TODAY2, to assess diabetes complications. Through the TODAY and TODAY2 studies, NIDDK-supported scientists, together with hundreds of research participants, aimed to change the health trajectory of at-risk youth and positively impact the course of young people's lives. (See inset for the story of a TODAY participant.)

### A RISING TREND: TYPE 2 DIABETES IN YOUTH

More than 34 million Americans—just over 1 in 10—have diabetes. It is the main cause of kidney failure, lower limb amputations, and new-onset blindness in adults, and is a major cause of heart disease and stroke. In diabetes, the body cannot keep blood sugar (glucose) levels from getting too high. Normally, the hormone insulin, which is made by the pancreas, acts in the tissues of the body (e.g., muscle) to promote absorption of sugar from the blood for use as fuel. In some people, their bodies become resistant to insulin, requiring the pancreas to produce more of the hormone to keep blood

sugar at a healthy level. Type 2 diabetes develops when the pancreas loses its capacity to produce enough insulin to compensate for insulin resistance.

The number of people with diabetes has risen dramatically during the last 30 years. Importantly, type 2 diabetes, which previously only affected older adults, has been rising in youth 10 to 19 years of age. The most recent data show that the growing number of young people with type 2 diabetes stems from rising incidence among racial and ethnic minority groups. Type 2 diabetes in both adults and youth is closely linked to having overweight/obesity, being inactive, and having a family history of diabetes.

### IMPLICATIONS OF EARLY ONSET TYPE 2 DIABETES

The longer a person has diabetes, the greater the chances of serious damage to the eyes, nerves, heart, kidneys, and blood vessels. This aspect of diabetes makes the growing burden of type 2 diabetes in children particularly alarming, as children with this diagnosis have a greater chance of developing medical complications during their lifetimes. Prevention of type 2 diabetes in youth is therefore a key public health goal. However, optimizing type 2 diabetes treatment options is equally critical to stall the onset of complications in children who already have the disease. Given the physiological changes that occur during adolescence, it is unclear whether treatments found to be effective for controlling diabetes in adults will work as well in youth.

### THE TODAY TRIAL

The TODAY trial tested how well 3 treatment approaches controlled blood sugar levels in ethnically

# PERSONAL PERSPECTIVE

and racially diverse youth who were 10 to 17 years of age, had overweight or obesity, and had been diagnosed with type 2 diabetes no more than 2 years before enrollment in the study. All participants received metformin, the first-line drug of choice among adults with type 2 diabetes and the only oral diabetes medication approved for use in children. Participants were randomly assigned to receive metformin alone; metformin plus another diabetes drug, rosiglitazone; or metformin plus a program of intensive lifestyle changes aimed at helping participants lose weight and increase physical activity.

Results showed that metformin alone failed to control blood sugar in 51.7 percent of participants, and metformin plus the lifestyle intervention failed 46.6 percent of the time. Although blood sugar levels remained healthier, on average, in participants who received both metformin and rosiglitazone than in the other groups, the two-drug combination still failed an alarmingly high 38.6 percent of the time over the course of the study.

To better understand the way the disease progressed in youth in the three TODAY treatment groups, study scientists analyzed changes in participants' insulin resistance and capacity for pancreatic insulin production over the course of the trial. They found that average insulin sensitivity gradually worsened for the metformin and metformin plus lifestyle groups, while for the metformin plus rosiglitazone group it improved significantly in the first 6 months of the trial, but later gradually fell back to initial levels. Thus, at the end of the trial, average insulin sensitivity among those youth getting both medicines was about where it began, but it had worsened among those receiving just metformin or metformin plus lifestyle.

In contrast, insulin-production capacity fell similarly in all three groups. Importantly, TODAY scientists found that in all the treatment groups, the participants with the poorest blood sugar regulation and insulin production at the beginning of the trial were the ones most likely to have higher than recommended blood sugar levels when the study ended. This points to the importance of beginning treatment

for pediatric type 2 diabetes before significant loss of insulin production capacity occurs and the body can no longer control blood sugar levels sufficiently. TODAY demonstrated that the development of insulin resistance and the deterioration of insulin-producing beta cell function progress more rapidly in youth-onset type 2 diabetes than in adult-onset diabetes.

## THE TODAY2 FOLLOW-UP STUDY

After completion of the TODAY study, participants were transitioned to treatment with metformin and/or insulin and enrolled in TODAY2, an observational follow-up. (In 2010, the U.S. Food and Drug Administration significantly restricted use of rosiglitazone for the treatment of type 2 diabetes due to increased risk of cardiovascular events.) Study scientists assessed participants for high blood pressure, elevated blood lipid levels, and early signs of diabetic kidney disease or nerve disease annually; eye (retina) health was assessed twice. In 2020, the 500 TODAY participants who enrolled in TODAY2 were on average 27 years old with diabetes duration of 13 years. Over the course of the studies, about 67.5 percent of participants developed high blood pressure and 51.6 percent developed elevated blood lipids. Diabetic kidney disease occurred in 54.8 percent and nerve disease in 32.4 percent of participants. The prevalence of retinal disease increased from 13.7 percent to 51.0 percent over 7 years, including more advanced stages. Sixty percent had at least one complication. Although these are staggering statistics, these studies have been instrumental in furthering our understanding of type 2 diabetes in children and adolescents.

## THE PROMISE OF FUTURE RESEARCH

The TODAY and TODAY2 studies had several strengths, including a large cohort of youth-onset type 2 diabetes participants and up to 15 years of prospective, comprehensive, and rigorous assessment. In addition, the cohort's diversity is representative of the general U.S. population with youth-onset type 2

# PERSONAL PERSPECTIVE

diabetes. The studies indicate that more research is needed to develop effective strategies to prevent and treat this disease in vulnerable young people. Understanding what makes type 2 diabetes more

difficult to treat in youth is critical to combatting the disease when it occurs in young people and preventing them from developing complications that often exact a heavy burden.

## KATHRINE'S STORY

Thirty-year-old Kathrine works for a non-profit organization whose mission is to create opportunities for people to live in affordable homes, improve lives, and strengthen communities. She grew up in Cleveland under the guardianship of her grandparents. Kathrine's grandparents both have type 2 diabetes, and as a child she was used to seeing them treat themselves with medication. She describes herself as having been an overweight child, but nonetheless very active. "I never felt limited by my physical health," she says.

*"These medical problems happen more often in marginalized communities because we experience the health care system differently. If my grandparents hadn't been paying attention, I could have become another statistic," says Kathrine, explaining how her grandparents noticed she was having diabetes symptoms at age 11.*

However, her grandmother noticed that Kathrine appeared thirstier and more tired than normal and recognized there might be a problem. So, she took Kathrine to the hospital to have her tested—she was diagnosed with type 2 diabetes at age 11. Shortly thereafter, Kathrine and her family were informed of a new clinical trial that was about to begin that would test treatment approaches for type 2 diabetes in youth—the Treatment Options for Type 2 Diabetes in Adolescents and Youth (TODAY) study. As it turned out, a study site was located at Rainbow Babies & Children's Hospital right there in Cleveland. "These

medical problems happen more often in marginalized communities because we experience the health care system differently. If my grandparents hadn't been paying attention, I could have become another statistic. But they jumped into action to make the conversation easier. It became a no-brainer for me to participate in the study," remembers Kathrine. And so, her journey began.

Kathrine became the first participant to be placed in the TODAY study's metformin and intensive lifestyle group. (Each of the participants was randomly assigned to receive either metformin alone or metformin in combination with another treatment strategy.)

The doctors and nurses involved in the study explained to Kathrine that she could make lifestyle changes such as incorporating healthy eating habits and increasing her physical activity to help control her blood sugar. This was appealing to her because she knew she did not want the burden of being dependent upon injected insulin to control her blood sugar. She thought the lifestyle changes would be helpful to achieving her many goals in life, including attending a prestigious private high school and going to college.

Kathrine was determined to take these measures to regain control of her own health, a resolution strengthened by meeting her TODAY lifestyle coach—a Black woman who happened to be from the same neighborhood in Cleveland. "I knew this was going to change my life because I have this Black woman I can relate to, looking out for me." Her coach turned her on to cooking; Kathrine had never cooked healthy foods before, and she grew to enjoy



# PERSONAL PERSPECTIVE

it. Learning new skills in the kitchen, Kathrine was well on her way to improving her health. She lost 15 pounds and saw her blood sugar levels improve. “So much of the education I got from TODAY was about nutrition and that I can make the choice to eat well.”

*“I knew this was going to change my life,” says Kathrine about meeting her TODAY lifestyle coach—a Black woman from her local Cleveland neighborhood.*

After graduating from an elite private high school in Cleveland, Kathrine was accepted into and attended a university in Atlanta. The TODAY study team ensured she would have continuity of care in the trial and arranged for quarterly visits in Cleveland. As for many college students, Kathrine faced challenges during this time. She was no longer able to cook for herself in this setting—grabbing a bag of chips on the go was cheap and easy. There were also the additional stressors of college studies. Her weight fluctuated and her blood sugar spiked. Kathrine realized that she was neglecting her health during this time and needed to regain control. “Twenty-year-old me didn’t realize that this is a journey, not a destination,” she recalls.

Kathrine graduated with degrees in psychology and sociology and continued in the TODAY2 follow-up study. She returned for a time to Cleveland to work in grassroots community development, establishing farmers’ markets and community gardens in the neighborhood where she grew up, before ultimately returning to Atlanta for work and to pursue graduate studies. Through the years, Kathrine has relied upon the lifestyle education she received in the TODAY study

to help guide her choices. She joined a running club, runs several miles per day now, and cooks healthful meals for herself daily. Today, Kathrine maintains a healthy weight and, quite impressively, her blood sugar levels are below that of even being considered to have prediabetes. This, along with the fact that she no longer requires diabetes medication, is a testament to her incredible determination and dedication. This past August, Kathrine received her master’s degree in communications and public policy, and she’s excited to continue her career in community development and confident her future is bright and healthy.

Reflecting upon her experience in the TODAY study, Kathrine expresses extreme gratitude. She says the study team watched her grow up, tracked her progress, and provided continuity of care. “TODAY didn’t complete me. It complemented me. It gave me the tools to be a better me.”

*Reflecting upon her experience in the TODAY trial, Kathrine expresses extreme gratitude: “TODAY didn’t complete me. It complemented me. It gave me the tools to be a better me.”*

Diabetes does not define Kathrine. She knows now that she is in control of her own health. She’s not Kathrine, the person with diabetes. She is Kathrine, the student, the professional, the runner, the home cook, the community activist—who happens to be managing risk for type 2 diabetes. She exclaims proudly: “I am not one thing. I am everything!”



The trans-NIH Obesity Research Task Force was established to accelerate progress in obesity research across the NIH, given the importance of the obesity epidemic as a major public health problem and its relevance to the missions of most of the NIH Institutes, Centers, and Offices. The Task Force is co-chaired by the Director of the National Institute of Diabetes and Digestive and Kidney Diseases, Dr. Griffin P. Rodgers; the Director of the National Heart, Lung, and Blood Institute, Dr. Gary H. Gibbons; and the Director of the *Eunice Kennedy Shriver* National Institute of Child Health and Human Development, Dr. Diana W. Bianchi. The Task Force holds two to three seminars each year, covering a broad range of topics to accelerate research to develop new and innovative prevention and treatment strategies for obesity and to close knowledge gaps.

On September 3, 2021, the Task Force convened a symposium on obesity and COVID-19 where 4 distinguished scientists highlighted their research on obesity, vaccine response, SARS-CoV-2 infection, and the resulting disease of COVID-19. A summary of this seminar is in this chapter.

# Obesity

***Obesity has risen to epidemic levels in the United States. Individuals who have obesity may develop devastating health problems, face reduced life expectancy, and experience stigma and discrimination. Obesity is a strong risk factor for type 2 diabetes, fatty liver disease, and many other diseases and disorders within the NIDDK's mission. More than 40 percent of U.S. adults are considered to have obesity based on body mass index (BMI), a measure of weight relative to height.<sup>1</sup> More than 19 percent of children and adolescents also have obesity, and thus are at increased risk for developing serious diseases both during their youth and later in adulthood.<sup>2,3</sup> Obesity disproportionately affects people from certain racial and ethnic groups and those who are socioeconomically disadvantaged.***

The high prevalence of obesity in the United States is thought to result from the interaction of genetic susceptibility with behaviors and factors in the environment (social determinants of health) such as a lack of healthy, affordable food and places to exercise in many communities, sedentary jobs, and other conditions that influence what, when, and how much people eat. Diet, activity, and aspects of our environment also may modify biologic factors in ways that promote obesity. Research is providing the foundation for actions to address this major public health problem by illuminating the causes and consequences of obesity, evaluating potential prevention and treatment strategies, and providing an evidence base to inform policy decisions.

***Individuals who have obesity may develop devastating health problems, face reduced life expectancy, and experience stigma and discrimination.***

The NIDDK supports a multi-dimensional research portfolio on obesity, spanning basic, clinical, and translational research. NIDDK-funded studies investigate a variety of approaches for preventing and treating obesity. These span behavioral and environmental interventions for children and adults in health care, home, community, and other settings using a variety of approaches and technologies; surgical interventions; and combinations of strategies. In parallel, NIDDK-supported investigations into the biologic processes associated with body weight have continued to spark new ideas for intervention approaches.

***More than 40 percent of U.S. adults and more than 19 percent of children and adolescents have obesity.<sup>1,2,3</sup>***

The NIDDK also continues to play a leading role in the NIH Obesity Research Task Force. The NIDDK Director co-chairs the Task Force along with the Directors of the National Heart, Lung, and Blood Institute and the Eunice Kennedy Shriver National Institute of Child Health and Human Development. The Task Force includes representatives from these and numerous other NIH Institutes, Centers, and Offices to promote collaboration and enhance obesity research across the NIH.

Highlights of recent advances from NIDDK-supported research on obesity are provided in this chapter.

***The NIDDK supports basic, clinical, and translational research to discover how body weight is regulated and to design and evaluate approaches for preventing and treating obesity.***

<sup>1</sup> Hales CM, et al. 2020. CDC. National Center for Health Statistics Data Brief No. 360.

<sup>2</sup> Fryar CD, et al. 2020. CDC. Prevalence of overweight, obesity, and severe obesity among children and adolescents aged 2–19 years: United States, 1963–1965 through 2017–2018. NCHS Health E-Stats.

<sup>3</sup> For children and adolescents, obesity refers to a BMI at or greater than the 95th percentile on growth charts (which are based on previous national surveys).

## THE DIET DEBATE

### Weighing in on the Diet Debate: Low-carbohydrate

**Versus Low-fat:** By comparing the effects of a plant-based, low-fat diet for 2 weeks to an animal-based, low-carbohydrate diet for 2 weeks in adult men and women, researchers have shown that while each diet had benefits, the low-fat diet led to less caloric intake and a significant loss of body fat. Two competing models of obesity contrast the roles of dietary fat and carbohydrate. According to the carbohydrate-insulin model, a diet high in carbohydrates results in elevated insulin levels after a meal, which is thought to cause increased hunger and calorie consumption. Alternatively, the passive overconsumption model predicts that high-fat, energy-dense (calorie-dense) foods promote increased caloric intake and weight gain.

To determine the impact of each diet on caloric intake, hormone levels, and body weight, 20 adult volunteers without diabetes were admitted for 4 continuous weeks to the National Institutes of Health Clinical Center and randomly assigned to receive either a plant-based, low-fat diet or an animal-based, low-carbohydrate diet for 2 weeks followed immediately by the alternate diet for 2 weeks. They received three meals a day plus snacks and could eat as much or as little as desired. Both diets were minimally processed and contained approximately the same amount of protein. When on a low-fat diet, participants ate 550-700 fewer calories per day than when on the low-carbohydrate diet. Despite the large differences in calorie intake, they reported no differences in hunger, enjoyment of meals, or fullness between the two diets. People lost weight on both diets, but only the low-fat diet led to a substantial loss of body fat. However, the low-carbohydrate diet resulted in lower blood glucose (sugar) and insulin levels compared with the low-fat diet.

Since the low-fat diet led to less caloric intake and the low-carbohydrate diet did not result in weight gain, the validity of both competing models of obesity is challenged. However, the study is limited by the tightly controlled clinical environment, which makes it difficult to generalize results to real-world settings where factors such as food costs, food availability, and meal preparation constraints can make adherence to a diet challenging. These findings suggest that regulation of calorie intake is more complex than these models propose. More research is needed to determine the long-term effects of both diets.

Hall KD, Guo J, Courville AB,...Chung ST. Effect of a plant-based, low-fat diet versus an animal-based, ketogenic diet on ad libitum energy intake. *Nat Med* 27: 344-353, 2021.

*When on a low-fat diet, participants ate 550-700 fewer calories per day than when on the low-carbohydrate diet.*

## GENETIC UNDERPINNINGS OF OBESITY

### Large-scale Genetic Study Identifies Genes That Increase Risk of Obesity While Protecting Against Other Metabolic Diseases:

A new analysis of genetic data sets has revealed regions of the genome that are linked to both elevated levels of body fat and protection from some of the negative health impacts of obesity. Obesity is a major risk factor for cardiometabolic diseases such as type 2 diabetes, heart disease, and related conditions, and people living with obesity tend to have unhealthy glucose (sugar) and lipid levels in their blood and high blood pressure. But scientists have observed that up to 45 percent of people with obesity have healthy blood pressure and blood glucose/lipid levels, and therefore may not be at high risk of diabetes and cardiovascular disease. The reasons why this group of people remains healthy have been poorly understood.

To determine whether genetics may play a role, a team of researchers analyzed data from hundreds of thousands of people, mainly of European descent, who had previously been assessed for body fat and disease risk markers. They identified 62 regions of the genome that have a seemingly paradoxical association of increased risk of obesity and favorable effects on cardiometabolic outcomes. Genes identified within these regions point to both known and novel ways in which excess body fat can become uncoupled from cardiometabolic diseases. For example, some of these body fat-increasing genes are associated with storage of the excess fat beneath the skin, as opposed to storage around the internal organs where fat is metabolically harmful. Further analyses identified genes that are functionally implicated in improved blood glucose levels, insulin signaling, regulation and development of fat cells, and energy (calorie) expenditure. Moreover, genes were identified that are linked to both increased body fat and changes in the nature of some of the fat tissue, from calorie-storing “white” fat to a form called “brown” or “beige” fat, a process that can increase calorie burning and promote

healthy metabolism. These results are helping to clarify the complex genetic underpinnings of obesity, and the genes identified may represent targets for new therapies to reduce cardiometabolic risk associated with excess body fat.

Huang LO, Rauch A, Mazzaferro E,...Loos RJF. *Genome-wide discovery of genetic loci that uncouple excess adiposity from its comorbidities.* *Nat Metab* 3: 228-243, 2021.

*A new analysis has revealed regions of the genome that are linked to both elevated levels of body fat and protection from some of the negative health impacts of obesity.*

## MOLECULAR UNDERPINNINGS OF EXERCISE

**Succinate Regulates Muscle Adaptations to Exercise in Mice and Humans:** Researchers have identified succinate, a small molecule produced through metabolism, as a signal in both mice and humans that triggers muscle remodeling and strength building in response to exercise. Muscle remodeling is the mechanism by which we get stronger in response to exercise, and it is thought to have other health benefits as well. Muscle adaptations require rapid cell-to-cell communication, but the signals originating from the contracting muscle that drive these processes are unknown.

In this study, researchers used small-molecule analysis to track the accumulation of metabolites secreted into the fluid between cells and into the circulation from acutely exercised muscle in both mice and humans. First, mice were exercised on a treadmill, and muscle samples were isolated and analyzed. In addition, the researchers

recruited young, healthy men of normal weight and collected blood samples (which can be used to study the fluid composition between muscle cells) while the men exercised on a bicycle for 1 hour and during recovery. Notably, the researchers discovered that succinate was selectively and rapidly released by both mouse and human muscle during exercise—an unexpected finding since succinate was not previously thought to be able to move freely out of cells. However, during exercise the interior of muscle cells becomes transiently and mildly acidic and in this acidic environment, succinate is converted into a form that can be transported out of the muscle cells by acid-dependent succinate transporters. Once released from the contracting muscle cells, succinate binds to the protein SUCNR1, which is present on non-muscle cells also located in the muscle (tissue or fiber). Following acute exercise, the succinate-SUCNR1 binding then sends a signal that is critical to induce muscle remodeling and muscle strengthening. Importantly, the succinate-SUCNR1 signaling also appears to be associated with another benefit of exercise—improved insulin sensitivity—and the research team found that the extent of this effect correlated with the peak exercise succinate concentration in humans.

This study suggests that succinate may play a key role as a signal that mediates muscle adaptation and remodeling responses to exercise. Further research could provide insight into whether dietary succinate supplementation or other therapeutic approaches could trigger the succinate-SUCNR1 signaling pathway to initiate muscle adaptations similar to those seen through exercise, and whether it is safe to do so.

Reddy A, Bozi LHM, Yaghi OK,...Chouchani ET. *pH-gated succinate secretion regulates muscle remodeling in response to exercise.* *Cell* 183: 62-75, 2020.

# POWERS

Preventing regain of lost weight is the most difficult challenge in the treatment of obesity. Individuals with overweight/obesity can alter dietary intake and physical activity to lose a significant amount of excess weight, but they will usually experience weight regain despite their best efforts. There is considerable evidence that various physiological adaptations occur that counter lifestyle attempts to maintain reduced weight.

The NIDDK convened a workshop, “Physiology of the Weight-reduced State,” in June 2019 to explore the potential biological mechanisms that account for adaptations in appetite and energy (calorie) expenditure. The workshop was chaired by Drs. Rudolph L. Leibel of Columbia University and Kevin D. Hall of NIDDK, and it brought together researchers who study the regulation of eating behavior and energy expenditure and clinical

scientists who are experienced in weight loss and weight maintenance interventions. Sessions focused on describing physiologic and metabolic changes that occur during the weight-reduced state in humans; studies examining the altered regulation of energy intake and energy expenditure after weight loss; and experimental approaches applied to the study of the weight-reduced state, particularly those such as metabolomics and proteomics, new genetics strategies, and neuroimaging. The goals of the presentations and discussions were to identify the state of knowledge regarding the biological mechanisms that oppose weight-loss maintenance, gaps in that knowledge, strategies to identify those pathways that are altered by weight loss, and new therapeutic targets for improving success in weight maintenance after weight loss.



## Physiology of the **Weight Reduced State**

Outcomes from the workshop included: (1) there is an equilibrium weight “set point” that is a function of physiology, genes, and environment; (2) changes in hunger and energy expenditure are the balancing factors that drive weight back to its set point; (3) increased appetite and reduced energy expenditure will likely persist until weight is restored to a set point; (4) variability in adaptation may account for some of the individual variability in weight loss maintenance; and (5) although current therapies have improved adherence and response to weight-loss and weight-maintenance interventions, new therapies are needed to prolong the weight-loss phase, reduce appetite and metabolic adaptation during the weight-maintenance phase, and alter the weight set point.

As a result of the workshop, the NIDDK launched the Physiology of the Weight-reduced State (POWERS) clinical trial consortium, which will seek to characterize the physiological mechanisms underlying individual variability in maintenance of reduced weight over time. This could reveal new therapeutic targets for interventions aimed at maintaining weight loss. Investigators will study adults with obesity before and after weight loss achieved through a behavioral/lifestyle intervention to explore the physiologic mechanisms that regulate energy intake and metabolic adaptation.

The clinical sites are at Columbia University and Drexel University (the Drexel award is comprised of investigators from Drexel, Tufts, and the University of Pennsylvania). The data coordinating center is at the University of Pittsburgh. The primary governing body of the consortium is a Steering Committee comprised of investigators from the clinical sites and coordinating center as well as an NIDDK project scientist. The Committee will employ common protocol elements that ensure enough participants can be enrolled and extensively studied to provide insight into the duration and extent of metabolic adaptation following weight loss. In accordance with NIDDK data sharing policies, all data produced by the consortium will be made publicly available.

Through this bold new initiative, novel approaches to achieving and maintaining weight loss based upon a better understanding of the interaction of genes and environment can be realized.

*Credit to Drs. Laughlin, Osganian, Yanovski, and Lynch for sampling from their manuscript: Laughlin MR, Osganian SK, Yanovski SZ, and Lynch CJ. Physiology of the weight-reduced state: A report from a National Institute of Diabetes and Digestive and Kidney Diseases Workshop. Obesity (Silver Spring). 29 Suppl 1: S5-S8, 2021.*

# NIH Seminar Series: Obesity and COVID-19

Since early 2020, the world has been in the midst of a deadly pandemic due to the SARS-CoV-2 virus and the resulting disease, COVID-19. COVID-19 has claimed millions of lives and changed the ways in which each of us relates to and navigates the world. For more than a year, many people were sheltered at home, where eating and physical activity routines were disrupted. Many people also encountered the added stress of loss of employment or dealt with illness in themselves or loved ones. These factors can contribute to an increase in obesity-promoting behaviors. Obesity, among many other diseases such as type 2 diabetes, is a known risk factor for severe illness and even death from COVID-19 in people of any age. Obesity trends continue to rise in the United States despite recognition of its many adverse health effects. The highest obesity prevalence has been in certain racial and ethnic minority populations, further contributing to the disparate outcomes seen in COVID-19. While we now have several vaccines available to help protect us against this deadly virus, obesity has been shown to impact an individual's immune response to certain vaccines; this is an important consideration for COVID vaccination as well. Given the global public health significance of obesity and COVID-19, the research community recognizes the need to accelerate development of new and innovative prevention and treatment strategies for obesity and to close knowledge gaps with the goal of translation into more effective patient care. To that end, four leading scientists highlighted their research at a September 2021 virtual symposium organized as part of the NIH Obesity Research Task Force Seminar Series. The research presented was supported by NIDDK and other NIH Institutes.

Dr. Barry Popkin from the University of North Carolina at Chapel Hill presented a global perspective of obesity and COVID-19. An analysis of multiple scientific studies revealed that individuals with overweight and obesity face a greater risk of severe consequences from COVID-19, including hospitalization, intensive clinical care, and death. Globally, there have been

substantial decreases in physical activity due to virus control measures and isolation related to loss of employment. In most countries, there have been rapid increases in consumption of ultra-processed, less nutritious, and less expensive foods. Increased weight has been documented, and COVID-19-associated changes in diet and exercise patterns could continue to result in greater weight gain and risk of obesity worldwide. In addition, economic stressors and food insecurity in some countries could lead to undernutrition, which is also associated with reduced immune function and poor outcomes from COVID-19. Dr. Popkin noted that this research was done prior to the delta variant of SARS-CoV-2 becoming the dominant cause of COVID-19 worldwide.

Dr. Melinda Beck from the University of North Carolina at Chapel Hill presented her research on the immune response to influenza vaccination in the context of obesity and potential implications for human responses to COVID-19 vaccines. Obesity is a risk factor for poor outcomes from both influenza and SARS-CoV-2 infection. Using an animal model, her team found that influenza-infected mice with diet-induced obesity showed greater lung inflammation, immune dysfunction, and a higher mortality rate compared to influenza-infected lean mice. In a human influenza vaccine study, adults with obesity who received an influenza vaccine experienced a steeper decline in vaccine-induced, influenza-specific antibodies over time compared to vaccinated, healthy weight adults. Moreover, a certain type of immune cell called "T cells" that typically recognize and fight viral infections were also relatively impaired in adults with obesity. Compared to vaccinated, healthy weight adults, vaccinated adults with obesity were twice as likely to become infected with influenza. This is similar to what is observed in vaccinated elderly individuals. Based upon these results, there is recognition in the scientific community that metabolic disease can influence the immune response and that perhaps the response to COVID-19 vaccines in people with obesity may not be as robust compared to healthy weight adults.

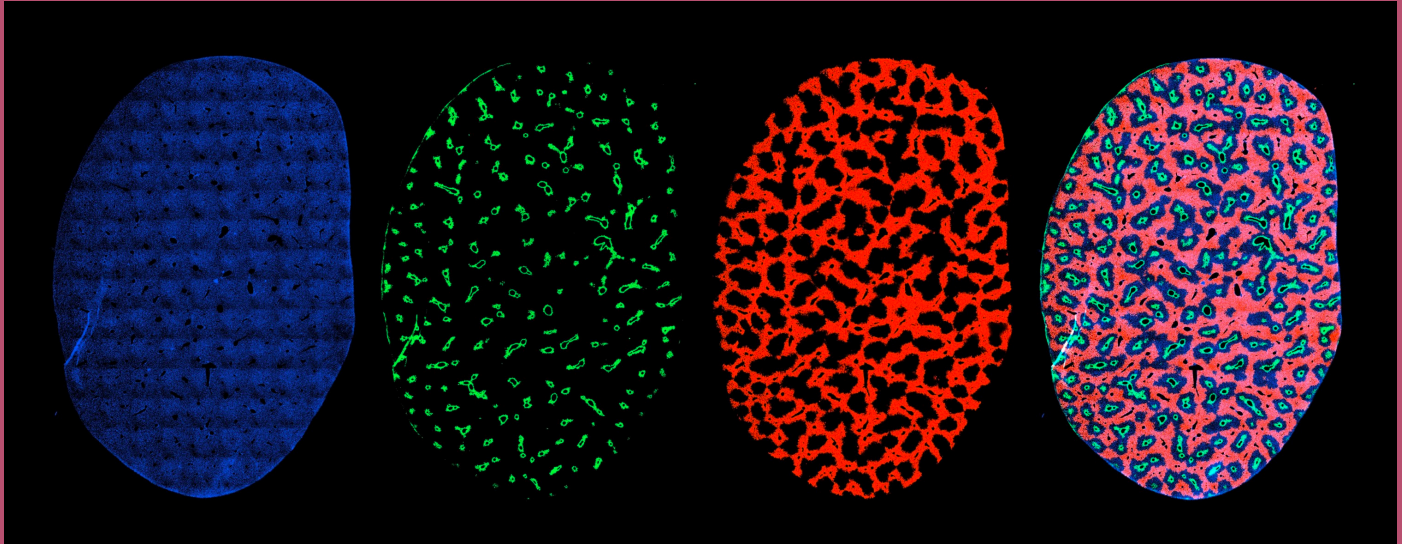


Dr. Dirk Homann from the Icahn School of Medicine at Mount Sinai presented his research on diabetes and COVID-19. Preexisting diabetes is associated with an increased risk of severe COVID-19. However, new-onset diabetes has also been observed in patients with COVID-19, raising the question of whether a more complex connection between COVID-19 and diabetes exists. In laboratory studies using human pancreatic cells in culture, Dr. Homann's group saw no evidence of enhanced cell death or pronounced insulin depletion of SARS-CoV-2 infected cells. Although they did find that the protein "receptor" to which the SARS-CoV-2 virus binds is required for productive infection of pancreatic cells, they found no indication that the virus affects pancreatic cell functionality, thereby inducing diabetes. Dr. Homann's group's results thus far suggest that SARS-CoV-2 infection of pancreatic cells is unlikely to induce diabetes. This is a rapidly evolving area of research, with different laboratories finding different results under different conditions that together are helping to build a comprehensive picture of the impact of SARS-CoV-2 on the pancreas. Research remains ongoing to better understand the relationship of COVID-19 and diabetes.

Dr. Dana Dabelea from the University of Colorado concluded the series of presentations by describing her work through the NIH-supported Environmental influences on Child Health Outcomes (ECHO) program on obesity-related behaviors in children and families during the COVID-19 pandemic. Through a consortium of nationwide pediatric cohorts, Dr. Dabelea's group

aimed to describe obesity-related behaviors in children during the pandemic, to identify socio-demographic groups who are at high risk for obesity-related behaviors, and to explore the extent to which behaviors are modified by parental coping strategies. They found that in the first 7 months of the pandemic, a substantial proportion of children did not meet recommended guidelines for healthy behaviors, including those for diet, physical activity, and screen time. Consistent with observed pre-pandemic disparities, healthier behaviors were seen among children who were younger, non-Hispanic White, and whose mothers had higher education levels. The vast majority of parents reported pandemic-related financial strain. Financial concerns and access to food as sources of stress were associated with increased intake of unhealthy foods, decreased exercise, and shorter sleep duration. Time-sensitive, ECHO-wide, ongoing studies are being conducted to examine changes in children's obesity-related behaviors that are occurring in tandem with societal changes related to the COVID-19 pandemic. Continued research is needed to understand the long-term effects of the pandemic on obesity-related behaviors and implications for children's health during COVID-19 and beyond.

The seminar included a lively discussion among speakers and participants on current challenges and opportunities. Continued research could reveal better strategies to prevent and treat obesity, thereby preventing many adverse health outcomes in general, as well as during the current and potential future pandemics.



The liver's unique ability to regenerate is legendary, and was first noted in Greek mythology and the story of Prometheus. Scientists are still working to understand which cells are responsible for restoring the organ under normal conditions and after injury. As described in this chapter, one group of researchers has mapped the liver's growth and regenerative potential by examining three zones within the functional unit of the liver called the lobule. They used fluorescent markers in mice to tag cells in these zones and track their contributions to liver growth and regeneration after injury from toxins. These images are from one strain of mouse whose cells have been fluorescently tagged to trace them to certain zones (first three images left to right); the rightmost image shows a composite image of all the tagged cells together. Mouse strains like these allowed the researchers to study the regenerative potential of the different zones. Future studies could elucidate the role that cells in different zones might play in liver disease and provide the basis for therapies that boost liver regeneration.

*Images provided by Drs. Hao Zhu and Yonglong Wei, University of Texas Southwestern Medical Center. From Wei Y, Wang YG, Jia Y, Li L, Yoon J, Zhang S, Wang Z, Zhang Y, Zhu M, Sharma T, Lin Y-H, Hsieh M-H, Albrecht JH, Le PT, Rosen CJ, Wang T, and Zhu H. Liver homeostasis is maintained by midlobular zone 2 hepatocytes. *Science* 371: eabb1625, 2021. Reprinted with permission from AAAS.*

# Digestive Diseases and Nutrition

***Digestive diseases are among the leading causes of doctor visits, hospitalizations, and disability in the United States each year. These conditions span a wide spectrum of disorders that affect the gastrointestinal (GI) tract, liver, gallbladder, and pancreas, as well as obesity and other nutrition-related disorders. NIDDK-supported scientists are pursuing research aimed at reducing the burden of digestive diseases. These efforts focus on better understanding how widespread these diseases are across the United States and in specific population groups, identifying their causes and how they progress, and testing new interventions for prevention and treatment, including drugs, surgery, and behavior modification.***

Digestive diseases affect individuals across the lifespan, exacting a significant toll in terms of their effects on quality of life, years lost due to premature death, and costs associated with hospitalization and pharmaceutical and surgical interventions. The burden of digestive diseases in the United States is substantial. Based on recent data, it is estimated that digestive disease is the primary diagnosis in a total of 66.4 million ambulatory care visits to physicians' offices and hospital emergency and outpatient departments in the United States each year.<sup>1</sup> Similarly, analyses with 2018 national inpatient samples identified 4.0 million hospitalizations with a primary diagnosis of digestive diseases and 16.5 million hospitalizations with a primary or secondary diagnosis of digestive diseases.<sup>2</sup> In addition, analyses focusing specifically on the clinical and economic burden of emergency department visits identified 18.7 million emergency department visits with a primary diagnosis of digestive diseases and costs totaling \$107.6 billion in 2018.<sup>3</sup>

Inflammatory bowel diseases (IBDs), which include Crohn's disease and ulcerative colitis, are marked by damaging intestinal inflammation leading to rectal bleeding, diarrhea, nutritional deficiencies, and other serious complications. These diseases often strike early in life, with a peak age of onset in adolescence or young adulthood. Treatment frequently requires prolonged use of multiple drugs and may require surgical removal

***Annual estimates of the burden of digestive diseases list these diseases as the primary diagnosis in 66.4 million ambulatory care visits to physicians' offices and hospital emergency and outpatient departments.<sup>1</sup>***

of the affected portion of the intestine. Scientists are investigating the complex interactions among the genetic, environmental, immune, microbial, and other factors that contribute to, or protect against, the development of IBD. The continued discovery of predisposing genetic variations, potential autoimmune and microbial influences, and new methods to repair damaged intestinal tissue will help catalyze the design of novel, more personalized therapeutic strategies. Research on controlling intestinal inflammation has potential benefits not only for patients with IBD, but also for those at risk of developing colorectal cancer.

Diseases of the stomach and intestines include some of the most common digestive diseases, such as peptic

***Scientists are investigating the complex interactions among the genetic, environmental, immune, microbial, and other factors that contribute to, or protect against, the development of inflammatory bowel diseases.***

<sup>1</sup> National Ambulatory Medical Care Survey (NAMCS) and National Hospital Ambulatory Medical Care Survey (NHAMCS), U.S. Centers for Disease Control and Prevention; available at: [www.cdc.gov/nchs/ahcd/index.htm](http://www.cdc.gov/nchs/ahcd/index.htm).

<sup>2</sup> Healthcare Cost and Utilization Project (H-CUP) National Inpatient Sample (NIS), Agency for Healthcare Research and Quality; available at: [www.hcup-us.ahrq.gov/nisoverview.jsp](http://www.hcup-us.ahrq.gov/nisoverview.jsp).

<sup>3</sup> Healthcare Cost and Utilization Project (H-CUP) Nationwide Emergency Department Sample (NEDS), Agency for Healthcare Research and Quality; available at: [www.hcup-us.ahrq.gov/nedsoverview.jsp](http://www.hcup-us.ahrq.gov/nedsoverview.jsp).

ulcer disease, which is typically caused by infection with the bacterium *Helicobacter pylori* or use of nonsteroidal anti-inflammatory drugs. Stomach and intestinal disorders also include functional gastrointestinal disorders, which can cause symptoms of abdominal pain and altered bowel habits. For example, irritable bowel syndrome (IBS) causes pain and constipation or diarrhea. IBS more frequently affects women, who may have a different range of symptoms and respond differently from men to pharmacologic treatments for the disease. While diet and stress contribute to this disorder, its underlying causes are unknown. Gastroesophageal reflux disease, in which stomach acids rise up into the esophagus, is a common functional gastrointestinal disorder that can lead to a condition known as Barrett's esophagus. This condition, in which cells lining the esophagus turn into an intestinal type of cell, is associated with a heightened risk of esophageal cancer—one of the cancer types still on the rise in the United States. Gastroparesis, another type of functional gastrointestinal disorder, is characterized by delayed emptying of food from the stomach, resulting in nausea, vomiting, and abdominal discomfort. Most cases of gastroparesis are of unknown origin, which makes it difficult to treat. Current therapies are directed toward helping people manage this chronic condition so they can be as comfortable and active as possible. Fecal incontinence, or impaired bowel control, is a disorder that poses a major public health burden. Although fecal incontinence is more common in older adults, it can affect people of any age. Because it is difficult to talk about, many people suffer without seeking professional treatment for this surprisingly prevalent condition. Researchers aim to examine barriers in addressing fecal incontinence and to develop improved treatment strategies. Scientists continue to strive for a deeper understanding of the causes of gastrointestinal disorders, which will lead to improvements in diagnosis and management.

*Stomach and intestinal disorders include functional gastrointestinal disorders, such as irritable bowel syndrome and gastroparesis, which can cause symptoms of abdominal pain and altered bowel habits.*

Some digestive diseases can be triggered by the body's reaction to certain foods. For example, in individuals with celiac disease, the immune system reacts to ingestion of gluten—a protein component of wheat, barley, and rye—

and damages the small intestine. This damage interferes with the ability of the intestine to absorb nutrients from foods and can result in chronic diarrhea, bloating, anemia, and, in children, slower growth and short stature. The only current treatment for celiac disease is maintenance of a strict gluten-free diet, which is difficult for many people. Recent and continued research advances in the understanding of genes and environmental triggers that are involved in the development of celiac disease may contribute to improved diagnosis and new ways to treat this condition in the future.

*Research to advance understanding of genes and environmental triggers involved in the development of celiac disease may contribute to improved diagnosis and therapy.*

The microbes that inhabit the GI tract are important factors in maintaining the balance between digestive health and disease. These bacteria, viruses, and other microorganisms can affect long-term health and nutritional status in some surprising ways, depending on their interactions with each other, with intestinal cells, and with nutrients ingested by their human host. Disruptions in this microbial ecosystem are associated with diseases such as IBD or infections by the harmful bacterium *Clostridium difficile*. Scientists are gaining insights into the ways these GI microbes influence the development and function of the digestive tract and other systems throughout the body, such as those with immune and metabolic functions, as well as how the composition of the GI microbial community changes with factors such as age, geography, diet, and antibiotic usage.

*Microbes that inhabit the gastrointestinal tract are important factors in maintaining the balance between digestive health and diseases, such as inflammatory bowel diseases.*

The exocrine pancreas, which secretes enzymes required for digestion, is vulnerable to disorders such as acute and chronic pancreatitis and their complications. Common causes of pancreatitis include gallstones, heavy alcohol use, inherited genetic factors, and some medicines. In all forms of pancreatitis, digestive enzymes attack the pancreas from within, causing inflammation, loss of function, and severe pain. Advanced pancreatitis can be debilitating and may lead to cancer or diabetes, but

because pancreatitis is difficult to detect in its early stages, many cases are advanced by the time they are diagnosed. Research has elucidated genetic and other factors contributing to pancreatitis that may lead to ways to treat or prevent this disorder.

The liver performs many critical metabolic functions within the digestive system, including processing and distributing of nutrients such as fats. When the liver is functionally compromised by disease, serious adverse health effects can occur, which sometimes leads to complete liver failure, called end-stage liver disease. Some liver diseases primarily affect children, such as biliary atresia (a progressive inflammatory liver disease), while others generally affect adults, such as nonalcoholic fatty liver disease (NAFLD) or its more severe form of nonalcoholic steatohepatitis. In recent years, however, NAFLD has been increasingly diagnosed in children in the United States as well, concurrent with rising overweight and obesity. Some forms of liver disease are caused by viral infection, as in most cases of hepatitis, or by genetic mutations such as alpha-1 antitrypsin deficiency; others arise from diverse factors such as autoimmune reactions, drug toxicity, bile duct obstruction, and other triggers, some of which are unknown. Many liver diseases, such as chronic hepatitis B and C, place individuals at elevated risk for developing liver cancer. When liver disease reaches the end stage, the only treatment is a liver transplant. Because the number of livers available for transplant from deceased donors is limited, sometimes a healthy living person will donate part of his or her liver. The living donor's liver eventually regenerates and grows back to normal size, as does the part of the liver that is donated. Research is critical to identify liver disease early, find methods to preserve liver function in people with liver disease, and develop and further study new treatment options, including experimental, cell-based approaches to liver regeneration.

*In recent years, nonalcoholic fatty liver disease has been increasingly diagnosed in children and adults in the United States, concurrent with rising overweight and obesity.*

The number of Americans with overweight or obesity has risen dramatically in recent decades and is now at epidemic levels. Obesity is associated with numerous diseases, including type 2 diabetes, heart disease, and cancer. Multiple factors contribute to obesity. As scientists elucidate the molecular, genetic, microbial,

behavioral, and environmental factors that influence obesity, they are identifying potential avenues for the development of new intervention strategies to promote safe, long-term weight loss. In addition to new pharmacologic interventions for obesity that may arise from research, existing bariatric surgical techniques are being evaluated for their long-term impacts on weight loss, obesity-associated disease, and well-being. Investigators are also continuing research to help people achieve healthy lifestyles that include physical activity and improved diet. (Additional information on NIDDK-supported research endeavors focusing on obesity is provided in the "Obesity" chapter.)

Other nutrition-related disorders under investigation involve specific, inherited alterations in nutrient metabolism. NIDDK-supported research has enhanced knowledge of how these nutritional disorders develop and how they can best be treated. Investigators also conduct basic, clinical, and translational research on the requirements, bioavailability, and metabolism of nutrients and other dietary components to understand dietary needs in health and disease. NIDDK staff works collaboratively with representatives from across the NIH, including in the NIH's Office of Nutrition Research, to advance nutrition research efforts.

*The NIDDK works to advance nutrition research efforts, both within the Institute and collaboratively with others across the NIH, including the NIH's Office of Nutrition Research.*

## GUT MICROBIOME AND NUTRITION

**Designer Foods Target Microbiome To Boost Host Health:** Researchers have developed foods designed to alter the gut microbiome, with an eye toward moving the needle on two immense public health challenges: malnutrition and obesity. These conditions represent opposite types of energy imbalance that co-exist in our modern world, sometimes within the same household. Decades of research has documented how the state of the community of microbes that inhabit the gut, or gut microbiome, reflects their human host's health. The gut microbiome also changes in response to dietary components that are metabolized by specific microbial species, such as fiber. Two recent studies explored the impacts of foods customized to shape the gut microbial landscape in children with malnutrition and adults with obesity.

One study represented the latest in an ongoing series of investigations supported by NIDDK, the Bill and Melinda Gates Foundation, and others of microbiome-based dietary interventions in malnourished children in Bangladesh. These children show signs of an immature gut microbiome for their age, which in turn impacts their response to therapeutic food interventions. Previously, this group of researchers from the United States and the International Centre for Diarrhoeal Disease Research, Dhaka, designed a “microbiota-directed complementary food,” or “MDCF,” containing flours and oils from nutrient-dense, locally available foods such as chickpeas, nuts, and bananas. In the most recent study, they compared the effects of their MDCF with the standard ready-to-use supplementary food given to malnourished children. Bangladeshi girls and boys ages 12 to 18 months with moderate malnutrition received either the standard supplementary food or MDCF twice daily for 3 months, and were monitored for changes in growth, levels of proteins linked to growth and neural development, and gut microbial species present. The results demonstrated that the MDCF dietary supplement not only supported a more age-appropriate gut microbiome, but also resulted in greater improvements in growth and markers of neural development in these children, compared to those given the standard ready-to-use supplementary food.

In another study, the scientists aimed to design snacks containing plant-based fiber to promote beneficial microbial responses in adults with overweight/obesity. Beginning with an animal model, male mice raised in a sterile environment free of microbes were colonized with gut microbes from women with obesity. The mice were then fed a fiber-deficient, high-fat diet supplemented sequentially with plant fiber from either peas, oranges, or barley. Over time, the abundance of microbial genetic material present in the stool increased dramatically for the enzymes needed to break down each unique fiber, and for microbial species that are associated with protecting against obesity such as *Bacteroides*. Pilot clinical trials tested whether this effect was translatable to people. Men and women with overweight/obesity were given prepared meals of a fiber-deficient, high-fat diet similar in nutritional composition to the one fed to the mice, supplemented with a snack composed of pea fiber, for several weeks. Similar to the results in mice, the study participants showed the same shift in microbial gene abundance toward a ramped up metabolic capacity for fiber. Follow-up studies testing snacks composed of multiple types of plant-based fibers, including those used in the mouse studies, demonstrated an even greater

microbial shift towards fiber metabolism. Consumption of the extra fiber in snacks was also associated with altered protein profiles in participants’ blood, indicating impacts on diverse physiological responses, such as metabolism and immune function. These findings provide compelling evidence of the strong connections among the gut microbiome, nutrition, and health impacts, and for the utility of mouse models of human response to food-based interventions that target the microbiome.

Chen RY, Mostafa I, Hibberd MC,...Gordon JI. A microbiota-directed food intervention for undernourished children. *N Engl J Med* 384: 1517-1528, 2021.

Delannoy-Bruno O, Desai C, Raman AS,...Gordon JI. Evaluating microbiome-directed fibre snacks in gnotobiotic mice and humans. *Nature* 595: 91-95, 2021.

Two recent studies explored the impacts of different foods designed to change the community of microbes that inhabit the gut, or gut microbiome, in children with malnutrition and in adults with obesity. This research shows the promise of this approach for helping to address these disparate public health challenges.

## IDENTIFYING THERAPEUTIC TARGETS FOR INFLAMMATORY BOWEL DISEASE

**New Potential Therapeutic Target Identified for Crohn’s Disease:** New research has shed light on how known genetic risk factors can contribute to Crohn’s disease and treatment response, opening the door to new treatment approaches. Crohn’s disease is a form of inflammatory bowel disease in which the digestive tract is marked by lesions of damaging inflammation. It can start at any age, causing lifelong episodes of cramping, diarrhea, and malnutrition. Medications that block a major component of the inflammatory response called tumor necrosis factor (TNF) are effective for many people, but in some cases the disease does not respond to these drugs. Among the scores of genetic variations that have been linked to a higher risk for developing Crohn’s disease, changes in a gene called *NOD2* that impair its function have been found to be a major risk factor. Exactly how these *NOD2* genetic variations could contribute to Crohn’s disease has been unclear, however, which has been a major roadblock for developing new therapies.

Researchers set out to answer this question by analyzing

intestinal samples from a well-characterized group of male and female children with Crohn's disease. They found that genetic variations inhibiting *NOD2* function were linked to changes in fibroblasts (cells that make up connective tissue) and immune cells in Crohn's disease lesions. Specifically, these cell types showed signs that they were "activated" and producing factors involved in inflammation. Importantly, activated immune cells and fibroblasts have also been found in lesions from people with refractory Crohn's disease that is resistant to anti-TNF therapy, suggesting that these activated cells provide an additional route to inflammation that is independent of TNF-mediated inflammation. Using cultured cells and a zebrafish model that effectively mimics human Crohn's disease, the researchers identified a protein known as gp130 that plays a critical role in activating these cells when *NOD2* is impaired. Data from women and men with Crohn's disease that did not respond well to anti-TNF therapy showed high levels of intestinal proteins in the cellular pathway used by gp130. Additionally, the researchers found that treating zebrafish or cultured cells with a gp130-blocking drug inhibits activation of inflammatory cells. More research is needed to determine if blocking gp130 will similarly reduce cellular activation in human intestinal lesions. However, this study suggests that drugs targeting gp130, when used in conjunction with anti-TNF therapy, might be effective treatments for people with Crohn's disease resulting from *NOD2* risk variants.

Nayar S, Morrison JK, Giri M,...Cho JH. A myeloid-stromal niche and gp130 rescue in *NOD2*-driven Crohn's disease. *Nature* 593: 275-281, 2021.

*Researchers have discovered how variants in the *NOD2* gene may contribute to intestinal inflammation, offering new possible therapeutic approaches to treat Crohn's disease.*

## IDENTIFYING THERAPEUTIC TARGETS FOR INTESTINAL DISEASE IN CHILDREN

**Potential Therapeutic Target Identified To Prevent Severe Diarrhea in Children:** New findings suggest that rotavirus-induced severe diarrheal disease in children may be prevented by targeting a cell-to-cell signaling pathway in the gut. Rotavirus infections of the small intestine are the leading cause of severe diarrhea, vomiting, and dehydration in children worldwide; while an extremely effective vaccine for the most common form of the virus is now available, not every child has access, so improving

therapeutic options remains a priority. A key question is how the virus causes severe disease even before damage to intestinal tissue is apparent. One theory has been that infected cells release a signal molecule that disperses and disrupts nearby uninfected cells, specifically by causing inappropriate increases in levels of calcium—a molecule critical to many cellular processes—within those cells. These increases in turn activate cellular pathways that lead to vomiting and diarrhea. Researchers studying rotavirus infections in both non-human primate epithelial cells and human intestinal epithelial cell models called "enteroids" have identified the molecule adenosine 5' diphosphate, or ADP, as the candidate signal. They found that ADP released from infected cells activates a specific cell-surface receptor, called P2Y1, to cause increased calcium levels in neighboring uninfected cells—so-called "intercellular calcium waves" that they could both visualize and measure experimentally. Importantly, rotaviruses also require elevated calcium for their replication, and the team found that blocking ADP-P2Y1 signaling significantly reduced the amount of rotavirus produced by infected cells.

To help determine whether what they observed in laboratory-grown cells is relevant to actual disease, the researchers tested their findings in an animal model. They found that two different, orally administered drugs that block P2Y1 receptor function were each able to reduce severity of diarrheal disease in young male and female rodent pups infected with rotavirus.

Although not yet well studied, intercellular calcium waves induced by ADP may be a normal route of communication for intestinal cells. While rotaviruses employ other mechanisms to initiate calcium increases when infecting a cell, the study findings suggest that by exploiting a preexisting ADP signaling pathway in the gut, the virus can simultaneously boost calcium in "instigator" infected cells while also causing disruptions in neighboring uninfected cells that will facilitate viral spread to new hosts via diarrhea and vomiting. By identifying ADP-P2Y1 as the likely signaling pathway, researchers can now see whether a safe therapeutic intervention targeting this pathway can be developed that could easily be administered to infected children to prevent or reduce symptoms and also possibly prevent spread of rotaviral disease.

Chang-Graham AL, Perry JL, Engevik MA,...Hyser JM. Rotavirus induces intercellular calcium waves through ADP signaling. *Science* 370: eabc3621, 2020.

*A study of signals passed between intestinal cells during rotavirus infection has uncovered a potential therapeutic target for this common cause of diarrhea, dehydration, and death in children around the world.*

## NEW INSIGHT INTO INTESTINAL BIOLOGY

### **Mucus Keeps Gut Bacteria at Bay To Prevent Inflammation:**

Research using a mouse model has provided a new understanding of how mucus coating the inner colon isolates bacteria from the gut wall, potentially offering new ways to diagnose and treat intestinal inflammation. A thriving community of bacteria (microbiome) lives in the human gut, helping with digestion and receiving nourishment from the food we eat. However, bacterial contact with the intestinal lining can trigger an inflammatory response (colitis), leading to chronic pain, bleeding, and diarrhea. One important way the body keeps inflammation in check is through a layer of mucus coating the inside of the colon, physically separating gut bacteria from the intestinal lining. A breakdown of this mucus layer has been implicated in several gastrointestinal diseases, but developing treatments to restore it has been challenging because of an unclear understanding of how it is produced.

To gain a better understanding of how the colon builds and maintains the mucus barrier, researchers conducted studies in male and female mice to develop a new way to visualize the mucus layer along the entire length of the colon. They found that the proximal area of the colon—where the colon joins with the small intestine—produces a thick layer of mucus that not only coats the intestinal lining, but also encapsulates fecal material and its associated bacteria. As the intestinal contents move further along the GI tract, they encounter a second, thinner, chemically distinct layer of mucus that is produced by the distal colon (the latter portion of the colon). The researchers generated mouse models with disrupted mucus production in either one or both areas of the colon and showed that both of the mucus layers were important for keeping bacteria sequestered in fecal pellets and separated from the intestinal lining. They also showed that both layers were critical for preventing spontaneous inflammation, with the most severe inflammation occurring when mucus production was turned off in both areas. The researchers also uncovered an intimate interaction between gut bacteria and the mucus layer: the presence of bacteria drove up mucus

production in the proximal colon, while turning off mucus production changed the makeup of the bacterial community. This suggests that the proximal colon mucus provides an environment that favors growth of healthy bacteria, and a defective mucus layer could be a sign of an unhealthy microbiome.

These results in mice present a new model for intestinal mucus function, whereby the mucus layer dynamically insulates the intestinal lining from bacteria. Further research would be needed to confirm whether human colonic mucus functions in a similar way. If so, the fecal mucus coating could serve as a diagnostic tool to detect gastrointestinal disease, and, ultimately, as a target for new approaches to restore the gut to a healthy state.

*Bergstrom K, Shan X, Casero D, ...Xia L. Proximal colon-derived O-glycosylated mucus encapsulates and modulates the microbiota. Science 370: 467-472, 2020.*

## LIVER MAINTENANCE AND REGENERATION

### **Location, Location, Location—Cell Position in Maintaining and Regenerating Liver:**

New research in mice has identified which cells of the liver contribute in large part toward maintaining the organ or regenerating it after injury. The liver is unique by virtue of its ability to regenerate and adjust its size in proportion to overall body size. At the microscopic level, the liver tissue is arranged in a honeycomb pattern with repeating hexagonal structures called lobules. These structures are made up of a central vein surrounded at each of the six outer points by a bundle consisting of a portal vein, hepatic artery, and bile duct. Lobule geography is mapped accordingly based on these landmarks into three areas, or “zones.” The same liver cell type can have unique capabilities depending on its zonal position, such as production of specific metabolic enzymes.

Scientists applied advanced genetic tools to answer a question that had conflicting results in the past: do cells in one zone contribute more to liver growth and regeneration than others? Using state-of-the-art gene editing technology, they generated 11 new strains of mice with modifications to switch on a fluorescent marker that selectively labels different groups of cells in the three zones of the liver lobule. In these mice and three other similar models, they tracked the abundance of new cells descended from the marked cells in these three zones. Under conditions of either normal day-to-day growth or regeneration after injury from



two different toxins, cells in zone 2 showed the greatest increases in number to maintain or restore the liver. The researchers hypothesize that these cells may be more protected from insults coming from the circulation or bile ducts. Next, the researchers further probed how the zone 2 cells are better able to help the liver grow and regenerate by identifying which genes were ramped up or suppressed to support liver cell growth. They identified a cellular signaling pathway that is involved in the growth of these zone 2 cells, which could be important for the development of potential therapeutics.

This work provides compelling evidence for the importance of cellular positioning within the liver lobule, particularly in zone 2, in maintaining equilibrium within the organ and replenishing it after injury. Future studies could help to define the role of these cells in liver disease and inform therapeutic strategies to boost liver regeneration.

Wei Y, Wang YG, Jia Y, ...Zhu H. Liver homeostasis is maintained by midlobular zone 2 hepatocytes. *Science* 371: eabb1625, 2021.

## DEVELOPING NEW LIVER DISEASE THERAPIES

### **Lipid Nanoparticles Deliver Promise for Liver Disease Therapies**

The success of messenger RNA (mRNA)-based vaccines, such as ones for COVID-19, depends on their delivery vehicle—a high-tech bubble of lipids (fats) called a lipid nanoparticle (LNP) encasing an inner package of fragile genetic material. This protective coating enables the mRNA to safely reach its destination inside target cells, where it produces viral proteins that train the immune system to resist infection. However, the potential medical applications of LNPs extend well beyond vaccines. Two groups of scientists have tested LNPs in mouse models to deliver treatments for multiple forms of liver disease, including liver fibrosis, nonalcoholic fatty liver disease, and drug-induced liver injury.

**Targeted Hormone Treatment Transforms Liver Cells into Fibrosis-fighting Superheroes:** New research in mice shows the potential for LNP-based therapies that incorporate a dynamic duo—the hormone relaxin and liver macrophage cells—on a mission to deactivate the liver's fibrosis-promoting cells. Liver fibrosis (scarring associated with tissue injury), and its advanced stage of cirrhosis, are common health problems that develop over the course of diseases such as fatty liver disease. A cast of characters play a role in either activating or repairing

liver fibrosis. Hepatic stellate cells are key players in liver fibrosis. When activated by liver injury, they participate in the wound healing response and contribute to scar tissue formation in the liver and subsequent fibrosis. Some other factors work to limit fibrosis, potentially by inactivating stellate cells, including the hormone relaxin, which is primarily known for its role in pregnancy-related changes in the female reproductive system. And finally, some cells act as a double agent, like the liver macrophage, a type of immune cell with the ability either to promote inflammation or to foster tissue repair.

Scientists used an animal model of liver fibrosis to unravel these interactions and unlock their therapeutic potential by employing the powerful LNP delivery technology. Male mice given a toxic chemical for several weeks developed liver injury and fibrosis. But this fibrosis could be reversed by 2 weeks of daily injection with LNPs delivering a gene-based therapy that boosted relaxin production in liver cells. However, when the researchers studied hepatic stellate cells in culture, they were surprised that the relaxin-treated cells remained active and fibrosis-promoting. This finding suggested that the cells in culture were missing essential cues from their environment to cause inactivation in response to relaxin. This prompted the scientists to identify relaxin receptors on the liver macrophages in mice and in samples taken from men with cirrhosis. Upon relaxin binding to its receptor, the macrophages transformed from fibrosis promoters to fibrosis-fighting cells that secrete small LNP-like packages of microRNAs (small pieces of genetic material that control the activity of other genes), which then targeted hepatic stellate cells and blocked their ability to induce fibrosis. The scientists engineered a different type of LNP to carry one of the microRNAs and delivered these along with the LNPs containing the relaxin gene. This combination treatment was even more powerful in reversing fibrosis, both in the liver fibrosis model and in a mouse model of nonalcoholic fatty liver disease.

These studies highlight the important role of macrophages in relaxin-mediated amelioration of liver fibrosis in diseases such as nonalcoholic fatty liver disease, through inactivating hepatic stellate cells. This work shows the potential for approaches combining gene therapy with LNP-based nanotechnology that directly target liver cell networks to provide new treatment options for liver fibrosis.

Hu M, Wang Y, Liu Z, ...Huang L. Hepatic macrophages act as a central hub for relaxin-mediated alleviation of liver fibrosis. *Nat Nanotechnol* 16: 466-477, 2021.

### **Potential Regeneration Therapy for Liver Injury**

**Activates Timely Burst of Growth Factors:** Scientists have showcased the ability of LNP-encapsulated mRNA to deliver controlled bursts of growth factors that boost regeneration as a potential therapy for acute and chronic liver injury. Effective therapies are lacking for common forms of liver disease, such as nonalcoholic fatty liver disease and cirrhosis. People with acute or chronic forms of liver injury can have a limited window of opportunity in which a therapy can curb damage and restore function of this vital organ. An ideal treatment would need to accomplish this feat of regenerating the liver during a narrow time frame and in a safe way.

Researchers designed LNP capsules containing mRNA that was modified to increase its stability to produce two growth factors, called hepatocyte growth factor and epidermal growth factor. When injected into female mice in these studies, the capsules were delivered to liver cells as their primary target, which manufactured the growth factors for approximately 3 days. In healthy mice, the short-term hepatic growth factor production boosted liver regeneration by ramping up liver cell production that typically maintains the organ. Furthermore, in mice fed a diet resulting in features of chronic liver injury similar to nonalcoholic fatty liver disease, treatment with LNPs containing mRNAs for both growth factors reversed fat deposits in the liver and enhanced liver function, more than in mice given a control LNP vehicle. Similarly, in a mouse model of an acute liver injury due to overdose of the drug acetaminophen, treatment with both growth factors in the mRNA delivery vehicles accelerated liver regeneration and lowered liver enzymes to normal levels.

This research shows the usefulness of LNP-encapsulated mRNA for delivering discrete bursts of growth factors as a possible, safe therapy for promoting liver regeneration in animal models of acute and chronic liver injury. Future studies will explore whether this new treatment approach can be translated to the clinic to prevent liver disease progression and restore function.

*Rizvi F, Everton E, Smith AR, ...Gouon-Evans V. Murine liver repair via transient activation of regenerative pathways in hepatocytes using lipid nanoparticle-complexed nucleoside-modified mRNA. Nat Commun 12: 613, 2021.*

### **Antiviral Drug Shows Promise Against Hepatitis C in**

**Animal Models:** Scientists in the NIDDK's Intramural Research Program showed that a new antiviral drug, called fluoxazolevir, effectively fights infection by many

*New research in mice showcases the ability of lipid nanoparticle-encapsulated messenger RNA to deliver controlled bursts of growth factors to safely boost liver regeneration. This may serve as a potential therapy for both acute and chronic forms of liver injury.*

types of hepatitis C virus, when given in combination with approved antiviral drugs in animal models. Hepatitis C is the most common chronic viral infection from a blood-borne pathogen in the United States. It can lead to damaging inflammation and liver disease, including cirrhosis, liver failure, and cancer. Current treatments with one or more drugs specifically targeting the virus are highly effective, with response rates of over 90 percent. Research efforts continue, however, to develop treatments for people infected with viral subtypes, or genotypes, that are less responsive to available drugs, and for viruses that develop drug resistance.

The researchers defined how fluoxazolevir blocks the hepatitis C virus from entering a human liver cell line in culture by blocking attachment with the outer membrane of the cell. The drug was effective in protecting the cells against infection with several viral genotypes. In male mice, rats, and dogs, the drug localized to the liver, an indication of potentially fewer side effects in other tissues. Additional testing in male mice transplanted with human liver cells showed that a 4-week treatment with the drug suppressed levels of multiple viral genotypes, without toxic side effects. When fluoxazolevir was given to the mice together with an antiviral drug already approved to treat hepatitis C, the combination treatment led to undetectable viral levels, indicating a sustained response to treatment that was not seen with the approved drug alone. This response was seen with infections by different viral genotypes, and no signs of viral drug resistance were observed. A longer 6-week treatment with fluoxazolevir in combination with two approved drugs for hepatitis C resulted in a sustained treatment response in mice infected with a drug-resistant strain of hepatitis C. Fluoxazolevir also showed promise at partially preventing hepatitis C, blunting viral levels when given prior to infection.

These studies demonstrate that the new drug fluoxazolevir, in combination with antiviral drugs already approved for hepatitis C, can achieve a sustained

response against different genotypes of hepatitis C virus, with a shorter and potentially less costly treatment duration compared to current regimens and less chance of developing viral drug resistance. In combination with approved antiviral drugs, fluoxazolevir may represent a next generation treatment for hepatitis C.

Ma CD, Imamura M, Talley DC,...Liang TJ. Fluoxazolevir inhibits hepatitis C virus infection in humanized chimeric mice by blocking viral membrane fusion. *Nat Microbiol* 5: 1532-1541, 2020.

*Animal models infected with different hepatitis C viral subtypes, including a drug-resistant one, responded well to a new combination treatment. This could represent the next generation of hepatitis C treatments, with benefits such as shorter treatment length, improved response across viral subtypes, and lower chance of developing viral drug resistance.*

## DIETARY SUPPLEMENTS AND METABOLIC HEALTH

**A Treatment That May Improve Muscle Insulin Sensitivity in Postmenopausal Women:** A small, short-term clinical trial in postmenopausal women with overweight or obesity, who also had prediabetes, found that dietary supplementation with nicotinamide mononucleotide (NMN) can improve insulin sensitivity in muscle. NMN is an essential ingredient in the production of other biomolecules that are involved in numerous

cellular processes, including those governing metabolism. NMN has therefore come under scrutiny in efforts to improve or boost metabolism. Previous research has shown that administering supplemental NMN to mice can partially overcome aging-related losses of insulin sensitivity, particularly in females. Based upon findings such as these, NMN has been marketed as a dietary supplement that can improve glycemic control, but no previous study with human participants has tested this assertion, and NMN has not been approved for this purpose by the U.S. Food and Drug Administration.

In new research, 25 postmenopausal women with overweight or obesity in addition to prediabetes were randomly assigned to receive either NMN supplementation or a placebo. The elevated blood glucose levels observed in people with prediabetes—detectable in a fasting blood test or through other measures—resulted largely from their impaired response to insulin in liver and muscle. After 10 weeks, study scientists found that the muscles of the women who had received NMN were more responsive to insulin than were those of the women who had received placebo. However, no difference was observed in the liver insulin response, and overall fasting glucose levels were not significantly different between the two groups. Further research will be needed to determine whether NMN has similar effects in younger women, in men, or in people with type 2 diabetes, whether its effects are safe and durable, and whether the supplement is therapeutically valuable for helping prevent or treat type 2 diabetes.

Yoshino M, Yoshino J, Kayser BD,...Klein S. Nicotinamide mononucleotide increases muscle insulin sensitivity in prediabetic women. *Science* 372: 1224-1229, 2021.

# Understanding the Role of the Nervous System in Gastrointestinal Health

The movement (motility) of food through the gastrointestinal (GI) tract is required for proper absorption of nutrients, elimination of waste, and overall health. Chronic difficulties with regulating GI motility can contribute to functional GI disorders (FGIDs) which have GI symptoms with no obvious cause, such as irritable bowel syndrome, functional dyspepsia (indigestion), and many instances of gastroparesis (delayed stomach emptying). GI dysmotility also underlies conditions for which the underlying cause can usually be identified, such as gastroesophageal reflux disease (GERD, or acid reflux). A better understanding of how GI motility is controlled could lead to better treatments for these conditions.

One particular focus of GI motility research is the role of the enteric nervous system (ENS). The ENS is located entirely in the wall of the gut and uses peripheral nerves to send and receive information from other organs. Often called the “little brain” in the gut, the ENS is critical for the control of normal GI function. Understanding how the ENS regulates GI function could reveal new ways to treat not only GI disorders but also conditions where the peripheral nervous system is affecting the function of other organs.

NIDDK’s participation in the Stimulating Peripheral Activity to Relieve Conditions (SPARC) program is one major aspect of NIDDK’s efforts to understand the ENS and treat GI motility disorders. A joint venture between NIH’s Common Fund and multiple NIH Institutes, SPARC is generating maps and tools to identify therapeutic targets within the neural circuitry of a wide range of organs and tissues. It is also funding high-risk, goal-driven research to accelerate development of therapeutic devices to control electrical activity in peripheral nerves. Such devices may ultimately facilitate treatments to improve the function of organs, including those in the GI tract. Current SPARC research projects include efforts to profile and map cells in the ENS to better understand how these cells control GI motility. SPARC is also

testing medical devices that communicate with the nerve cells of the ENS as potential treatments and research tools for motility and other GI disorders.

Complementing the studies supported through SPARC, NIDDK is also interested in the “brain-gut axis,” the bidirectional communication between the “little brain” of the ENS and the “big brain” that is part of the central nervous system. NIDDK is also supporting research into how the ENS develops and into how defects in brain-gut communication could lead to motility disorders. Such research could better illuminate how the brain affects gut motility and vice versa. NIDDK is also supporting research into how pain is transmitted through sensory nerves. For example, researchers are examining how nerves that interact with the esophagus sense pain due to acid reflux and how they signal to the rest of the nervous system, information that may lead to better ways to treat the pain associated with GERD.

Another area of NIDDK-supported research involves a possible link between the ENS and brain disorders such as Parkinson’s disease. Growing evidence has suggested that the misfolding of a protein called  $\alpha$  (alpha)-synuclein in the ENS could start a chain reaction that ultimately leads to changes in the brain seen in Parkinson’s disease. Several NIDDK-funded projects are studying  $\alpha$ -synuclein’s role in Parkinson’s disease. Collectively, these efforts to understand how the ENS regulates GI functions such as motility will increase our understanding of how the peripheral nervous system regulates organ function, which could lead to new treatment options for diverse diseases and conditions.

# NIH Meetings Explore Possible Role of Gut-brain Connections in Parkinson's Disease



The NIDDK, together with the National Institute of Neurological Disorders and Stroke (NINDS) and the National Institute of Environmental Health Sciences (NIEHS), co-sponsored two scientific meetings in 2021 focused on how connections between the gut and brain may drive Parkinson's disease and other neurodegenerative disorders.

The most well-known symptoms of Parkinson's disease are motor issues such as tremor (shaking), muscle stiffness, slowed movement, and impaired balance. But many people also experience gastrointestinal (GI) symptoms, such as constipation, nausea, and trouble swallowing, that can have significant impacts on quality of life. Intriguingly, these GI symptoms can precede diagnosis of Parkinson's disease by decades and, therefore, may be some of its earliest indicators. Competing theories exist as to the gut's role in Parkinson's disease development, or contributions from genetics, environmental chemicals, diet, gut microbes, or other factors. However, evidence has grown in recent years for the importance of bi-directional brain-gut communication in neurodegenerative diseases.

In March 2021, the three Institutes hosted a virtual symposium on "The Gut-Brain Axis as a Critical Element in the Development of Parkinson's Disease," designed as an introduction to current concepts of gut-brain communications in Parkinson's disease. This was followed by a more comprehensive, virtual workshop in September-October 2021 on "Neurodegenerative Disorders and the Gut-Brain Axis: Parkinson's Disease." This workshop addressed

research gaps and identified opportunities for research and collaboration among gut-brain investigators from the Parkinson's disease, GI, and neuroscience fields, both in the wider research community and within NIH. It concluded with recommendations for future research opportunities. A summary of the workshop and its recommendations is planned for publication in the scientific literature.

A person with Parkinson's disease had a key role in facilitating these meetings by serving on the organizing committee. Additional meeting participants represented the patient advocacy community and pharmaceutical industry. These activities reflect NIDDK's commitment to stakeholder engagement in research, including people living with diseases in our mission and others who share an interest in improving health.

The main goal of these meetings was to build future collaborations for research to elucidate the role of the GI tract in Parkinson's disease development, with an eye toward developing ways to potentially improve early diagnosis and prevention. Additionally, results from this research could have broader applicability to other forms of neurodegenerative disease with GI involvement. A related effort was the NIDDK's release in 2021 of a notice of special interest in supporting research on causes, diagnosis, prevention, and treatment of GI dysfunction in people with neurodevelopmental disorders, which may also provide insights of relevance to neurodegenerative disorders.

# The NIDDK-supported Research Community and the CCF's IBD Plexus – A Fruitful Collaboration

The NIDDK seeks collaborative opportunities with professional societies and disease-oriented volunteer organizations to advance research and ultimately improve the health of people with diseases within the Institute's mission.

One example of such an effort is in research on Crohn's disease and ulcerative colitis (inflammatory bowel diseases, IBD), with a resource developed by The Crohn's & Colitis Foundation (CCF). In 2015, the CCF launched the development of the IBD Plexus toward improving the quality of care of people with IBD. A cloud-based platform, IBD Plexus consolidates clinical, patient-reported, genetic, and other data from various IBD research cohorts, real-world clinical care settings, and the experiences of people living with IBD. The IBD Plexus includes pediatric and adult registries, a biobank, data and analytical platforms, high-performance computing, a centralized analytical lab, and a portal for researchers. Designed to support a variety of research activities, the platform provides data central to discovery (e.g., drug target discovery), clinical development (e.g., study feasibility), safety surveillance, and outcomes research (e.g., health care utilization). By optimizing and sharing data and samples across the research community, this resource could spur the development of new drug target treatments, biomarkers, and diagnostics. Access to IBD Plexus is available at no cost to qualified researchers.

NIDDK-supported investigators have made considerable use of the CCF-supported Pediatric RISK Stratification Study data available on the IBD Plexus to advance knowledge of diagnosis, progression, and treatment of IBD. This study, established in 2008, enrolled 1,800 participants from 28 clinics in the

United States and Canada. Over 900 children newly diagnosed with Crohn's disease generously provided collections of clinical, demographic, and biological samples every 6 months over the course of 36 months with continuing follow-up for 5 years.

This wealth of data and biosamples has led to many exciting discoveries. For example, two different studies of disease progression found that rare variants in the *NOX1* gene predispose people to early onset IBD with progressive and severe disease, and these variants also convey resistance to conventional therapy. Other research showed that in older patients with Crohn's disease, variants in other genes are associated with a more aggressive course of disease. Adding knowledge regarding Crohn's disease complications, researchers identified a set of gene signatures in the small intestine that is associated with increased risk of stricture—a common complication in which there is a narrowing of the intestine that makes it difficult for stool to pass through and can lead to blockage. Toward predicting effective treatments for IBD, a recent study identified a combination of certain types of cells in people with Crohn's disease whose symptoms did not improve with a treatment referred to as anti-TNF therapy; this finding could help health care providers predict which therapies would be most effective.

The generous contributions of research participants, the development of this valuable resource, and the ingenuity of NIDDK-supported researchers has brought exciting advances to the field, and opportunities for further research to improve the lives of people with IBD.

Schwerd T, Bryant RV, Pandey S,...Uhlig HH. NOX1 loss-of-function genetic variants in patients with inflammatory bowel disease. *Mucosal Immunol* 11: 562-574, 2018.

Denson LA, Jurickova I, Karns R,...Kugathasan S. Clinical and Genomic Correlates of Neutrophil Reactive Oxygen Species Production in Pediatric Patients With Crohn's Disease. *Gastroenterology* 154: 2097-2110, 2018.

Martin JC, Chang C, Boschetti G,...Kenigsberg E. Single-Cell Analysis of Crohn's Disease Lesions Identifies a Pathogenic Cellular Module Associated with Resistance to Anti-TNF Therapy. *Cell* 178: 1493-1508. e20, 2019.

Haberman Y, Minar P, Karns R,...Denson LA. Mucosal Inflammatory and Wound Healing Gene Programs Reveal Targets for Stricturing Behavior in Pediatric Crohn's Disease. *J Crohns Colitis* 15: 273-286, 2020.

## Advancing Treatments for Inflammatory Bowel Diseases in Children

Inflammatory bowel diseases (IBDs), such as ulcerative colitis (UC) and Crohn's disease (CD), are marked by chronic inflammation in the intestines that causes debilitating symptoms of abdominal pain and cramping, diarrhea, blood in the stool, nausea, and weight loss. The symptoms typically appear in affected individuals early in life, meaning that the impact is often felt for most of their lifetimes. Treatments aim to reduce inflammation and pain and, if needed, surgically remove damaged tissue, but treatment responses vary greatly from person to person. NIDDK-supported research has focused on advancing discoveries that will fuel greater understanding of IBD and improve its care, including uncovering the biological underpinnings of these variable treatment responses. (See inset for the story of a participant in an NIDDK-supported clinical research study of pediatric UC.)

### ABOUT INFLAMMATORY BOWEL DISEASES

In both UC and CD, the chronic inflammation is caused by an autoimmune response where the immune system reacts inappropriately to a person's own intestinal lining. The exact causes are unclear, but the inflammation is likely driven by genetic factors and triggered by an immune response to friendly gut bacteria or environmental factors. Both of these forms of IBD are typically diagnosed in childhood or early adulthood, a period when the compromised nutrition caused by intestinal inflammation can be especially detrimental to growth. However, other distinguishing characteristics set the two IBD types apart. In UC, the inflammation affects the inner lining of the colon (large intestine), while CD can affect any portion of the gastrointestinal (GI) tract, typically with patchy lesions that traverse several layers of the GI tract wall.

Treatments include drugs to reduce inflammation, such as corticosteroids, though these have side effects that could be serious if used for a long period of time. The nonsteroidal anti-inflammatory drug mesalamine (also known as mesalazine) is another available treatment. However, even with these treatments, the health of many people with IBD does not improve, and they will eventually need to be treated with more potent drugs that suppress the immune response that is causing inflammation (immunosuppressive drugs). More recently, multiple different therapies, such as monoclonal antibodies that target specific immune system components, have been shown to benefit some patients. In some cases, surgery is also needed to remove the damaged portion of the intestine. Determining the best treatment approach for children with IBD has proven difficult, not only due to varying individual responses, but also because most treatments are based upon results from adult studies.

### PREDICTING TREATMENT RESPONSE IN CHILDREN

The variability from person to person in IBD symptoms and whether the disease responds to medications represents an opportunity to improve clinical care. Advance knowledge about which treatments will work for different children with IBD could enable them to be treated more effectively and undergo remission as quickly as possible.

The Predicting Response to Standardized Pediatric Colitis Therapy (PROTECT) study was conducted at 29 sites throughout North America and supported by the NIDDK, in collaboration with the Crohn's and Colitis Foundation and their Pediatric Resource Organization for Kids with Inflammatory Intestinal Diseases (PRO-KIIDS) network. (For information on



# PERSONAL PERSPECTIVE

additional IBD research collaborations between the NIDDK and the Crohn's and Colitis Foundation, see the feature in this chapter.) From 2012 to 2017, this study evaluated whether a combination of clinical, genetic, and immunologic tests could be used to predict response to standard medical therapy for children newly diagnosed with UC. The two standard therapies tested were drugs to reduce inflammation: mesalamine, derived from the same chemical as aspirin, and prednisone, a type of corticosteroid. Based on pre-established criteria, if these anti-inflammatory drugs did not elicit responses, treatment was escalated to a therapy that modulated the immune system, such as thiopurines, or an antibody-based therapy that blocked an inflammatory protein called TNF $\alpha$ . The researchers mainly sought to determine whether 1 year of receiving the study therapy would lead to remission (*i.e.*, decrease in or disappearance of symptoms) for the participants, without need for any additional steroid treatment.

Results from the PROTECT study found that higher amounts of an immunologic biomarker in the blood correlates with disease severity, suggesting that this biomarker, which also may be associated with resistance to standard therapy, potentially could be used as a diagnostic tool to help plan individualized treatments for children with the disease. More recent results from the study identified several other characteristics that can predict how well children with UC will respond to treatment, further enabling a more personalized approach to treating the disease. These characteristics—such as high hemoglobin levels, clinical remission after 4 weeks, and the makeup of the gut microbial community—were associated with achieving steroid-free remission, suggesting these characteristics can predict whether immunosuppressive drugs will be necessary.

The NIDDK recently initiated support of a planning grant for a study focusing on pediatric CD entitled ClinicAI, IMaging, and Endoscopic Outcomes of Children Newly Diagnosed with Crohn's Disease (or CAMEO). The investigators will test whether study participants' unique clinical, radiologic, genomic, and microbial features are associated with intestinal healing in response to treatment with a drug that inhibits the inflammatory protein TNF $\alpha$ . This study

will build on the progress made by the PROTECT study toward achieving more personalized IBD therapy.

## OTHER IBD RESEARCH EFFORTS

In addition to the PROTECT study, the NIDDK supports many other ongoing research efforts aimed at facilitating improved understanding and care for both forms of IBD. For example, the NIDDK-supported IBD Genetics Consortium works, in collaboration with the International IBD Genetics Consortium, to further knowledge of genetic contributors to IBD disease processes and inform better disease management.

NIDDK has sponsored research on microbial contributors to IBD, including a study as part of the larger NIH Integrative Human Microbiome Project that seeks to understand changes in gut microbes in adults and children with IBD. For example, NIDDK-supported studies in children and adults with IBD found changes in the gut microbes during disease flare-ups that could point to ways to predict these disease flares and treat them. Another NIDDK-supported study by an independent team of researchers showed that different treatments for CD have varying effects on the gut microbiome in children and teens.

The Intestinal Stem Cell Consortium, co-supported by the NIDDK and the National Institute of Allergy and Infectious Diseases, funds team-based science to develop novel therapies that target stem cells within the intestine and support intestinal regeneration after injury in diseases such as IBD. Recently, Consortium scientists developed “mini-intestines” using cells from people with UC that model many of the features of human disease and can enable greater insights into disease processes and interventions tailored to individuals.

In addition to these efforts, the NIDDK also supports many other studies initiated by investigators on advancing understanding, diagnosis, treatment, and prevention of IBD. The collective impact of these efforts to discover genetic, autoimmune, and microbial contributors to IBD, and to identify innovative ways to restore intestinal health, promises to facilitate new therapeutic approaches for young people with this disease.

## BEN'S STORY



Ben (center) with Jane (mother) and Rex (father)

The picture of a healthy, busy 19-year-old, Ben engages in sports such as golf, waterskiing, and boating whenever he can, works summers as a farm technician picking and packing vegetables, and attends college full-time during the school year. Contrast that with his experience just 8 years earlier—of the debilitating stomach pain, nausea, diarrhea, weight loss, and other symptoms that accompanied his diagnosis of ulcerative colitis (UC). It has not been an easy journey, but Ben and his family have persevered and found meaning through participating in clinical research as part of the Predicting Response to Standardized Pediatric Colitis Therapy (PROTECT) study and also volunteering their time to help others with this condition.

Ben and his family—mother Jane, father Rex, and their family pet, a Westie dog named Gigi—are based in the Greater Cincinnati area. When Ben started feeling sick with stomach pain, nausea, and diarrhea at age 11, his mother immediately took him to see the pediatrician, who suspected a stomach bug. But after several days of no improvement in his symptoms, he was referred to the nearby campus of Cincinnati Children's Hospital

Medical Center (CCHMC), where he was treated for dehydration and nausea. Tests for intestinal parasites and bacterial infections came back negative, though gut tenderness signaled that inflammation was present. Ben was sent home with instructions to stay hydrated and continue taking medications to manage the nausea and pain. However, the next evening he experienced diarrhea with blood and mucus, as well as frequent vomiting. By the time they returned to their pediatrician's office a few days later, Ben had lost 3 pounds over the course of 5 days, and the nausea and diarrhea still had not abated.

They were referred to Dr. Lee Denson, a gastroenterologist at CCHMC's Inflammatory Bowel Disease (IBD) center, who suspected Ben might have UC and admitted him for diagnostic tests such as abdominal x-ray, colonoscopy, and endoscopy. Those tests showed that his stomach and intestines were irritated and inflamed. Ben was too young and ill at the time to remember all the details of his ordeal, but Jane recalls from her careful notes documenting the disease severity ratings that "he was one sick little boy." He was diagnosed with severe UC and treated with intravenous corticosteroids.

*"Although I was really young when I was diagnosed with UC and participated in the PROTECT study, I would recommend that others participate in research studies if they have the opportunity," says Ben, who was a participant in the Predicting Response to Standardized Pediatric Colitis Therapy (PROTECT) study of ulcerative colitis (UC) treatment sponsored by the NIDDK.*

At that time, the CCHMC staff informed Ben and his family about the opportunity to participate in the PROTECT study. Dr. Denson was the Principal Investigator of the CCHMC PROTECT study site—

# PERSONAL PERSPECTIVE

1 of the 29 participating PROTECT sites throughout North America. Dr. Denson and his staff relayed to the family the details of the study process, how to take the mesalamine treatment used in the study, and how to track Ben's daily medications. The family decided to enroll Ben as a study participant, and the next day he was able to return home, where he continued taking the mesalamine study medication, as well as the steroid, anti-nausea, and antacid drugs to manage his symptoms. Ben had a difficult time swallowing some of the pills, so early on the family would mix the medication in with applesauce or yogurt, until a medical psychologist at CCHMC helped him learn to more easily swallow the pills. At follow-up appointments, the doctors found that his UC responded well to the mesalamine treatment, with continued gut healing, to the point that he was able to wean off the steroid after only 12 weeks—a primary goal of the study.

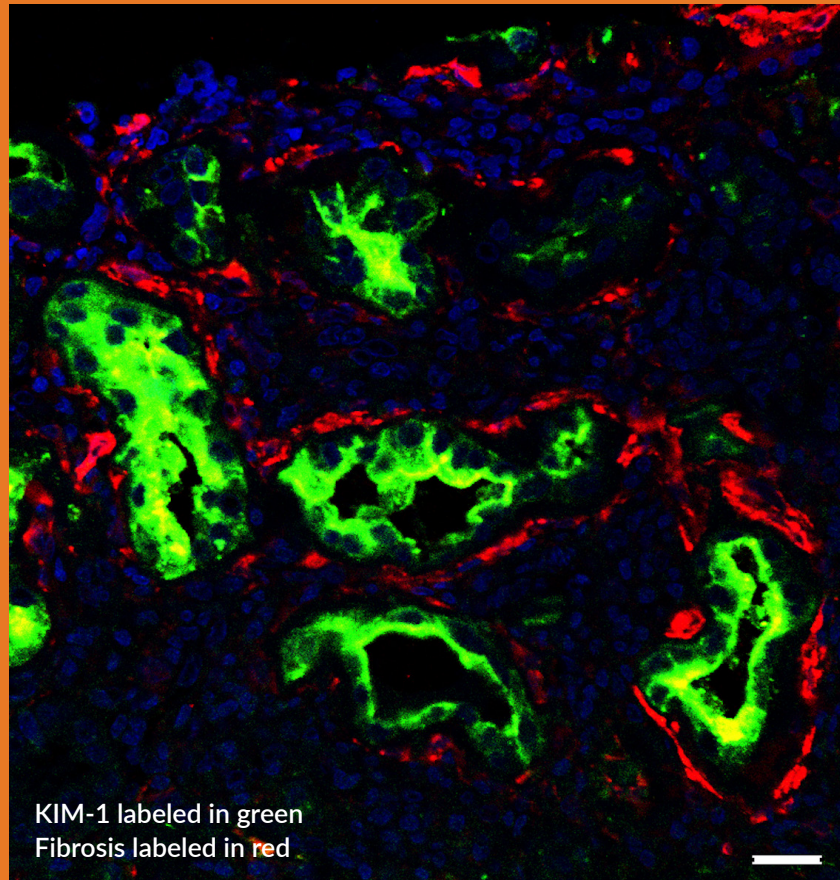
The family credits the staff at CCHMC who helped Ben return to good health for serving as a trusted resource and positive influence throughout this challenging time. Jane describes the PROTECT study staff as “helpful and so easy to work with ... they were diligent and dedicated to the success of the study.” Ben also participated in several other clinical studies at CCHMC and would recommend the experience. “Although I was really young when I was diagnosed with UC and participated in the PROTECT study, I would recommend that others participate in research studies if they have the opportunity,” says Ben. “I feel that by participating, you are helping with research that could really benefit others in the future.” Jane echoes this sentiment: “I always felt that participating in a study that might benefit other patients and families was very important.... Our hope is that other

kids and parents or caregivers will benefit from the research that is being done.”

*“Our hope is that other kids and parents or caregivers will benefit from the research that is being done,” says Jane, whose son Ben participated in a clinical research study of pediatric ulcerative colitis.*

In addition to participating in clinical research, Ben and his family have taken other actions that pay forward the support they received. Ben has volunteered as part of walks sponsored by the Crohn's and Colitis Foundation—the same patient advocacy organization that collaborates with the NIDDK on the PROTECT study and also hosts a summer camp for kids with IBD that he attended as a child. Jane has served on the board of the Foundation's southwest Ohio chapter and volunteered with its walks and fundraisers. Through these events, they are working to ensure better days ahead for those diagnosed with UC and other forms of IBD.

These days, Ben's overall health continues to be good, and his UC is considered to be in remission. The last few years of his schooling have been affected by the COVID-19 pandemic, with many cancelled senior year activities and events, and taking classes from his dorm room during his first year at college. But he is currently in his sophomore year at college studying finance. He continues to take the mesalamine medication and vitamins, and sees his doctor for routine colonoscopies to monitor his condition. The latest showed only mild intestinal inflammation. “We are very pleased with these results,” says Jane.



As described later in this chapter, the protein Kidney Injury Molecule-1 (KIM-1) is highly expressed in kidney proximal tubules of individuals with diabetic kidney disease (DKD), but not of people without DKD. Further analyses revealed that in kidneys from individuals with DKD, KIM-1-positive tubules were surrounded by fibrosis, which is the deposition of large amounts of collagen-rich connective tissue that can lead to organ damage. Inhibition of KIM-1 led to reduced fibrosis, suggesting that KIM-1 represents a novel target for developing treatments for DKD.

*Image provided by Drs. Joseph Bonventre, Amrendra Ajay, and Yutaro Mori, Brigham and Women's Hospital, Harvard Medical School. Reprinted from Cell Metabolism, 33/5, Mori Y, Ajay AK, Chang J-H, Mou S, Zhao H, Kishi S, Li J, Brooks CR, Xiao S, Woo H-M, Sabbisetti VS, Palmer SC, Galichon P, Li L, Henderson JM, Kuchroo VK, Hawkins J, Ichimura T, and Bonventre JV, KIM-1 mediates fatty acid uptake by renal tubular cells to promote progressive diabetic kidney disease. 1042-1061, 2021, with permission from Elsevier.*

# Kidney, Urologic, and Hematologic Diseases

***Diseases of the kidneys, urologic system, and blood are among the most critical health problems in the United States. They affect millions of Americans, and their impact is felt across the lifespan. To improve our understanding of the causes of these diseases, and to identify potential new prevention and treatment strategies, the NIDDK supports basic and clinical research studies of the kidney and urinary tract and of the blood and blood-forming organs. The overall goal of the NIDDK's research programs is to improve the health of people who have or are at risk for kidney, urologic, and hematologic (blood) diseases.***

Normal, healthy kidneys filter about 200 quarts of blood each day, generating about 2 quarts of excess fluid, salts, and waste products that are excreted as urine. Loss of function of these organs, either for a short period of time or as a consequence of a gradual, long-term decline in kidney function, is a life-threatening condition.

An estimated 37 million American adults have impaired kidney function—also called chronic kidney disease (CKD).<sup>1</sup> CKD has two main causes: high blood pressure and diabetes. The increases in obesity and type 2 diabetes in the United States in recent years—especially among children and adolescents—have grave implications for the Nation's health, as young people with these conditions are likely to face serious health complications at an earlier age than people who historically have developed these conditions later in life. CKD can also result from other factors, as noted below.

***An estimated 37 million American adults have chronic kidney disease.<sup>1</sup>***

CKD, especially if undetected, can progress to irreversible kidney failure, a condition known as end-stage renal disease (ESRD). People with ESRD require dialysis or a kidney transplant to live. In 2018, over 783,000 patients received treatment for ESRD: over 554,000 received either hemodialysis or peritoneal dialysis, and over 229,000 were living with a kidney transplant.<sup>2</sup> Racial and ethnic minority populations in the United States, particularly African Americans, Hispanic and Latino Americans, and American Indians and Alaska Natives, bear a disproportionate burden of CKD and ESRD. Compared to Whites, ESRD prevalence in 2018

was about 3.4 times greater in African Americans, 1.9 times greater in American Indians or Alaska Natives, and 1.3 times greater in Asian Americans.<sup>2</sup> Compared to the non-Hispanic population, Hispanic Americans had 1.5 times the risk for kidney failure.<sup>2</sup>

***In 2018, over 783,000 patients received treatment for end-stage renal (kidney) disease.<sup>2</sup>***

The NIDDK supports a significant body of research aimed at understanding the biology underlying CKD and developing treatment strategies. The chronic renal diseases program supports basic, translational, and clinical research on kidney development and disease, including the causes of kidney disease, the underlying mechanisms leading to progression of kidney disease to ESRD, and the identification and testing of possible strategies to prevent development or halt progression of kidney disease. In addition to research on kidney disease related to diabetes and high blood pressure, the NIDDK also supports studies of inherited diseases, such as polycystic kidney disease, congenital kidney disorders, and focal segmental glomerulosclerosis, and immune-related kidney diseases, such as IgA nephropathy and hemolytic

<sup>1</sup> Centers for Disease Control and Prevention. *Chronic Kidney Disease in the United States, 2021*. Atlanta, GA: U.S. Department of Health and Human Services, Centers for Disease Control and Prevention, 2021.

<sup>2</sup> United States Renal Data System. *2020USRDS Annual Data Report: Epidemiology of kidney disease in the United States*. National Institutes of Health, National Institute of Diabetes and Digestive and Kidney Diseases, Bethesda, MD, 2020.

uremic syndrome. One feature common to kidney diseases arising from varying causes is the deposition of fibrotic scar tissue in the kidney. Research supported by the NIDDK has enhanced our understanding of the origin of this scar tissue, how it can impair kidney function, and how it might be prevented or treated.

*Racial and ethnic minority populations in the United States, particularly African Americans, Hispanic and Latino Americans, and American Indians and Alaska Natives, bear a disproportionate burden of chronic kidney disease and end-stage renal disease.<sup>2</sup>*

Urologic diseases and conditions affect people of all ages, result in significant health care expenditures, and may lead to substantial disability and impaired quality of life. The NIDDK's urology research program supports basic and clinical research on the normal and abnormal development, structure, function, and injury repair of the genitourinary tract. Areas of interest include the causes of and treatments for urologic diseases and disorders, such as urinary tract infections and urinary stone disease, two of the most common and costly urologic conditions affecting people in the United States. Urinary incontinence is another prevalent problem. Based on national public health surveys conducted over several years, it is estimated that about 54 percent of women 20 years and older experience urinary incontinence each year. Urinary incontinence was self-reported by approximately 15 percent of men surveyed.<sup>3</sup> Many suffer in silence due to embarrassment and lack of knowledge about treatment options available.

*About 54 percent of women 20 years and older experience urinary incontinence each year.<sup>3</sup>*

Many people are also living with one of a cluster of disorders collectively called urologic chronic pelvic pain syndrome (UCPPS). The two most common examples of UCPPS are interstitial cystitis/bladder pain syndrome (IC/BPS)—also known as IC/painful bladder syndrome (PBS)—and chronic prostatitis/chronic pelvic pain syndrome (CP/CPPS). IC/BPS is a debilitating, chronic, and painful urologic disorder. Based on a recent large, national interview survey, it is estimated that among U.S. women 18 years or older, 3.3 million (2.7 percent) have pelvic pain and other symptoms, such as urinary urgency or frequency, that are associated with IC/BPS.<sup>4</sup> Using

a community-based epidemiologic survey, researchers have estimated that among U.S. men ages 30 to 79 years old, 1.6 million (1.3 percent) have persistent urologic symptoms, such as pain with bladder filling and/or pain relieved by bladder emptying, that are associated with IC/BPS.<sup>5</sup> NIDDK-supported basic and clinical research on IC/BPS and on CP/CPPS is focused on elucidating the causes of these conditions, identifying important subsets of patients to aid diagnostic stratification, and improving treatment and interventions.

*Among U.S. women 18 years or older, 3.3 million (2.7 percent) have pelvic pain and other symptoms, such as urinary urgency or frequency, that are associated with interstitial cystitis/bladder pain syndrome.<sup>3</sup>*

Research on UCPPS is one example of how the NIDDK is seeking a broad-based understanding of symptoms affecting the lower urinary tract (LUTS). LUTS—including pain, bladder leakage, and problems urinating—are not always associated with discrete conditions or tissue dysfunctions, different conditions can share the same symptoms, and symptom causes may actually lie outside the urinary tract. For example, urinary incontinence symptoms have been linked to anxiety disorders in some cases. For the wide range of LUTS, we still need to learn more about causes and contributing factors to improve management and treatment of symptoms. This is true even when a person's LUTS have been linked to a problem in a structure or function in the lower urinary tract, as there still may be other factors influencing the symptoms he or she is experiencing. Moreover, researchers now have a better appreciation that clinical end points for treatments do not always match up with individual preferences for a satisfactory outcome. Thus, the NIDDK is supporting multiple efforts to identify and understand different subgroups of people with LUTS through improved measurement of their symptom experiences that can inform future therapeutic strategies. Simultaneously, the NIDDK is supporting research to better understand factors that contribute to bladder health over the lifespan, with the ultimate goal of preventing LUTS to begin with.

<sup>3</sup> *Urological Diseases in America, 2018 Addendum. NIDDK, NIH Publication Number 12-7865, 2018.*

<sup>4</sup> *Berry SH, et al. J Urol 186: 540-544, 2011.*

<sup>5</sup> *Link CL, et al. J Urol 180: 599-606, 2008.*

*Blood diseases and disorders—some of which cause severe, debilitating pain, and premature death—affect millions of Americans.*

The NIDDK's hematology research program uses a broad approach to enhance understanding of the normal and abnormal function of blood cells and the blood-forming (hematopoietic) system in order to develop effective treatment strategies. Blood diseases and disorders—some of which cause severe, debilitating pain, and premature death—affect millions of Americans. These inherited and acquired diseases can affect red and white blood cells, platelets, bone marrow, or blood vessels. Research efforts include studies of a number of nonmalignant blood diseases, including sickle cell disease, the thalassemias, aplastic anemia, iron deficiency anemia, hemolytic anemias, thrombocytopenia, the anemia of inflammation and of chronic diseases, hemochromatosis, HIV-associated blood-related dysfunction, and bone marrow failure. The NIDDK also supports research on the basic biology of adult blood (hematopoietic) stem cells, which are used clinically in bone marrow transplants and may have broader application in gene therapy research.

Highlights of recent advances from NIDDK-supported research on kidney, urology, and hematology topics are provided in this chapter.

## CLINICAL RESEARCH ON KIDNEY DISEASE

### **Robust Research Pipeline Ensures Exquisite Data Quality Control, Paving the Path Toward Personalized Kidney Care:**

A large team of scientists has developed and tested a robust research pipeline for rigorously collecting and analyzing kidney biopsy samples; these analyses will be essential for catalyzing research toward personalized care for people with kidney diseases. Few therapies are currently available for treating chronic kidney disease and acute kidney injury—two relatively common types of kidney diseases. The NIDDK's Kidney Precision Medicine Project (KPMP) aims to identify previously elusive cells, molecules, and pathways involved in kidney disease progression by harnessing sophisticated research technologies. Ultimately, KPMP's goals are to construct a high-resolution three-dimensional kidney "atlas," define kidney disease subgroups, and identify new therapeutic targets, which together will provide a foundation for personalized approaches to kidney care.

A central component of KPMP is the procurement of kidney biopsies from people with and without kidney diseases across multiple U.S. locations. These biopsy samples are then divided and shipped to different laboratories for analysis to identify a range of molecules (e.g., genetic material, proteins, and metabolites) and cells in the kidney that are associated with health or disease states. Importantly, reproducibility of research results across KPMP sites requires standardized processes and high levels of quality control at each step, such as biopsy procurement, preservation, storage, and analysis, as well as data generation and validation. In a recent report, KPMP scientists detailed this enormous collaborative undertaking, in which they carefully developed a series of protocols to generate an integrated "follow the tissue" pipeline that will maximize the information gleaned from the precious biopsy samples. The researchers then tested the pipeline in a pilot experiment using adult human kidney tissue from a single source. The pilot experiment demonstrated that the pipeline was indeed robust and results were reproducible at five laboratories. The experiment also revealed some areas for improvement, such as inconsistencies in temperature states during shipping and the need for some additional surrounding tissue to optimize a particular imaging technique. These issues will be addressed and corrected as the KPMP study moves forward.

The establishment of this robust "follow the tissue" pipeline was essential for KPMP to begin generating rigorous and reproducible data that will accelerate research toward personalized kidney care. This framework could serve as a model for developing organ atlases with limited tissue to advance precision medicine research for other diseases.

*El-Achkar TM, Eadon MT, Menon R, ... Jain S; for The Kidney Precision Medicine Project. A multimodal and integrated approach to interrogate human kidney biopsies with rigor and reproducibility: guidelines from the Kidney Precision Medicine Project. *Physiol Genomics* 53: 1-11, 2020.*

*Kidney Precision Medicine Project (KPMP) scientists have developed a robust research pipeline for rigorous and reproducible analysis of human kidney biopsies—an essential foundation for paving the path toward personalized care for people with kidney diseases.*

### **Multidisciplinary Approach Uncovers Potential New Biomarker for Kidney Function:**

A new study has revealed a potential novel blood-based biomarker for assessing kidney health. Normally, the kidneys filter out a wide variety of molecules from the blood, preventing buildup of wastes and toxins. Currently available blood tests to assess kidney function rely on detecting higher levels of such molecules in the blood; however, it would be useful to have biomarkers of kidney health and function that are independent of the kidney's filtration process. Researchers initially identified a candidate biomarker, a protein called testican-2, when looking for proteins that are secreted by the kidneys into the blood rather than cleared from it. Using an advanced technique called aptamer-based profiling, they quickly assessed over 1,300 proteins present in blood samples available from 22 patients with cardiovascular disease and found six proteins present at higher levels in blood exiting the kidney than that entering it, with testican-2 showing the highest relative increase. To determine whether this discovery might have clinical significance, the team applied the same advanced technique to samples available from over 3,500 participants in two large clinical study populations—one African American, the other White—this time to assess associations between blood proteins and standard measures of kidney function. Among their findings was the observation that testican-2 levels correlated directly with standard measures of kidney function in both cohorts. Moreover, when they analyzed follow-up health information available from a subset of these participants, they found that having relatively higher levels of testican-2 at study entry was associated with a lower rate of decline in kidney function in both cohorts and a decreased risk of new-onset chronic kidney disease.

Given the potential of testican-2 as a biomarker for kidney health, the team also performed a series of molecular experiments to better characterize testican-2 and gain insight into its possible function in human kidneys. These experiments revealed that human testican-2 is encoded by the gene *SPOCK2* and is expressed exclusively by podocytes—highly specialized cells in the kidney that prevent loss of critical blood proteins into the urine during the filtration process. Additional experiments with laboratory-grown cells suggested that testican-2 may enhance formation of the tiny blood vessels involved in kidney filtration, but its actual function remains unknown.

Together with data captured about other kidney proteins during the course of the study, these findings lay the

foundation for future studies, such as targeted blood tests for testican-2 and direct studies of its association with kidney disease outcomes. Additional research could lead to new clinical tools for assessing kidney health and decline that could be a significant improvement over current methods.

*Ngo D, Wen D, Gao Y,...Rhee EP. Circulating testican-2 is a podocyte-derived marker of kidney health. PNAS 117: 25026-25035, 2020.*

### **IDENTIFYING THERAPEUTIC TARGETS FOR KIDNEY DISEASES**

#### **Zeroing in on the Role of KIM-1 in Diabetic Kidney Disease:**

Researchers have found that the protein KIM-1 plays a critical role in progression of diabetic kidney disease (DKD) and thus may serve as a promising therapeutic target. Diabetes is the most frequent cause of chronic kidney disease. DKD generally has been thought to result from damage to the glomerulus, which is a specific segment in each of the kidney's roughly one million nephrons (functional filtration units). However, abnormalities in a different segment of the nephron called the proximal tubule (PT) may appear earlier than glomerular injury during DKD progression. Previously, researchers discovered that a protein called kidney injury molecule-1 (KIM-1) was associated with PT damage at early stages of DKD, and found that elevated levels of circulating KIM-1 predicted disease progression in people with type 1 diabetes. These and other findings suggested a PT-specific role for KIM-1 in DKD progression.

The scientists have now extended their previous research, reporting that KIM-1 protein levels in kidney PT cells were higher in biopsy samples from people with DKD (1 female, 6 males) than from people with other forms of kidney disease that do not exhibit tubule damage (3 females, 2 males). The scientists conducted a series of experiments with cultured cells, as well as with mouse models of diabetes, and showed that KIM-1 is required for PT cells to take up (*i.e.*, to bring materials from the outside to the inside of the cell) a protein called albumin only when it is bound by a specific fatty acid known as palmitic acid (PA). The scientists showed that PA-albumin uptake leads to PT cell injury, as well as other types of kidney damage. Furthermore, genetic deletion of a portion of KIM-1, resulting in a nonfunctional protein, prevented DKD in an experimental model of male mice, suggesting that KIM-1 is required for kidney injury and resulting disease. The



scientists then tested more than 14,000 small molecules for their ability to prevent KIM-1-mediated PA-albumin uptake in cultured cells, and identified TW-37 as a candidate molecule. Mice treated with TW-37 were protected from PA-induced kidney damage, as were human kidney cell aggregates tested in culture. Together, these findings strongly implicate KIM-1 as a potential therapeutic target in people with DKD, and TW-37 could be a promising candidate as a small-molecule drug. Additional studies are needed to explore these possibilities.

Mori Y, Ajay AK, Chang J-H,...Bonventre JV. KIM-1 mediates fatty acid uptake by renal tubular cells to promote progressive diabetic kidney disease. *Cell Metab* 33: 1042-1061, 2021.

**Exploring the Role of Exosomes in the Progression of Polycystic Kidney Disease:** A recent report described a previously unknown role of exosomes in the progression of polycystic kidney disease and identified a compound capable of delaying cyst growth in mouse models of the disease. Autosomal dominant polycystic kidney disease (ADPKD) is the most common genetic cause of chronic kidney failure; most people with the disease have mutations in the *PKD1* gene. ADPKD is characterized by the growth of numerous fluid-filled cysts in the kidneys. Over time, growth of these cysts results in enlarged kidneys in which normal tissue is displaced and kidney function is impaired, sometimes quite severely. The fundamental processes that control ADPKD progression remain elusive. The exosome is a tiny sac-like structure that is formed inside a cell and contains some of the cell's proteins, DNA, and RNA. Many types of cells contain exosomes which are released into the blood or urine. Exosomes, therefore, can serve as cellular messengers as they transfer proteins, DNA, and RNA into other cells. Exosomes have received much attention lately for their involvement in human diseases such as cancer, but the role exosomes may play in some other diseases, such as ADPKD, is currently unknown.

Researchers have now shown that exosomes obtained from the urine of people with ADPKD stimulated a significant increase in the number of mouse kidney cells (*i.e.*, increased proliferation) in cell culture compared to exosomes obtained from healthy people. These results suggested that ADPKD urinary exosomes contain a critical factor(s) capable of activating PKD-associated signaling pathways to ultimately increase kidney cell proliferation, which in turn could contribute to the growth of cysts. Intriguingly, mouse kidney cells treated with urinary exosomes from healthy people formed

kidney-like tubule structures in three-dimensional gels while mouse kidney cells treated with urinary exosomes from people with ADPKD formed cyst-like structures in gels. Treatment with exosomes obtained from mouse cells containing *Pkd1* gene mutations promoted cyst growth in a mouse model of ADPKD compared to the same mouse model not treated with exosomes. Furthermore, the exosome inhibitor GW4869, which blocks exosome release into the blood, delayed cyst growth in a mouse model of ADPKD. The results of this study suggest that targeting exosome secretion may be a potential therapeutic strategy to reduce or delay cyst formation in people with ADPKD.

Ding H, Li LX, Harris PC, Yang J, and Li X. Extracellular vesicles and exosomes generated from cystic renal epithelial cells promote cyst growth in autosomal dominant polycystic kidney disease. *Nat Commun* 12: 4548, 2021.

*For the first time, small packages of cellular components, called exosomes, have been shown to play a role in mice and in kidney cells grown in the laboratory and targeting their secretion may have therapeutic benefit for people with autosomal dominant polycystic kidney disease.*

## RESEARCH ON LOWER URINARY TRACT SYMPTOMS AND DISORDERS

**A New Approach to Studying the Links Between Bladder and Brain Activity in People:** Researchers have demonstrated the efficacy of a new, more natural approach to studying bladder function in people. Many studies of people with urologic problems, such as bladder pain or sudden urinary urgency, have involved artificially filling the bladder with liquid using a catheter and simultaneously visualizing brain activity using magnetic resonance imaging (MRI). Such studies have helped identify brain regions and networks involved in perceiving and responding to changes in bladder fullness, in the hope of finding ways to alleviate suffering in people with urologic symptoms that are as yet hard to explain or treat. However, this research approach, while having advantages such as control over the amount of liquid inserted, comes with potential drawbacks—for example, the procedure of catheterization itself can cause discomfort and anxiety in study participants, thereby influencing what is seen in brain scans.

To see whether a more natural bladder-filling strategy would be viable and possibly superior as a study method,

researchers in the Multidisciplinary Approach to the Study of Chronic Pelvic Pain (MAPP) Research Network studied the brain's response to natural bladder filling in study participants without urologic problems. Sixty-two healthy men and women were asked to drink about 12 ounces of water after first voiding their bladders; 40 minutes later, they underwent an MRI brain scan for 10 minutes, emptied their bladders into a urine collection container, and underwent a second 10-minute scan. Participants were also asked to report the degree of urgency they felt at different times during the procedure—just after drinking the water, 20 minutes later, and both before and after each MRI scan. The majority of participants responded to the test procedure with an increasing feeling of urinary urgency that peaked by the end of the first scan and was relieved when they voided. More intense feelings of urgency correlated with larger void volumes—a proxy measure of bladder fullness. The researchers found that activity in specific brain regions and networks associated with bladder filling and voiding—from sensory recognition to physical response—not only was detectable, but also correlated with perceived urinary urgency and with void volumes.

This study provides proof that a more natural bladder filling method that avoids invasive catheterization is effective for studying brain activities important to urologic function and correlating them with people's symptom experiences. Such an approach should be easier to implement and help researchers perform studies in larger and more diverse groups of people. Already, it is being used to study symptomatic participants in the MAPP Research Network, and should advance overall efforts to study people with underlying urologic diseases, disorders, and symptoms.

Mawla I, Schrepf A, Ichesco E,...Kutch JJ. Natural bladder filling alters resting brain function at multiple spatial scales: a proof-of-concept MAPP Network Neuroimaging Study. *Sci Rep* 10: 19901, 2020.

**A recent report suggests that researchers will be able to make new, clinically relevant discoveries about the role of the central nervous system in urologic problems using an approach that is much less invasive than has been used previously.**

### Potential Therapeutic Target for Erectile Dysfunction

**Following Prostate Cancer Surgery:** Researchers have identified a nerve regeneration pathway that could be targeted to reverse erectile dysfunction (ED) caused by surgery. ED is a condition in which one is unable to get or maintain a penile erection firm enough for satisfactory sexual activity. Notably, many men experience ED after prostate cancer surgery. ED may occur if nerves that regulate blood flow to the penis, called cavernous nerves, are inadvertently damaged or removed during surgery. Unfortunately, existing treatments are rarely effective for men with ED following prostate cancer surgery.

Investigators recently described a series of experiments in male rodents that highlight the important role of a protein called fidgetin-like 2 (FL2) in the regeneration of neurons, or nerve cells. One way to assess neuronal regeneration is to measure growth of the axon, a specialized cellular extension neurons use to transmit signals throughout the body. The researchers found that mouse neurons genetically engineered to no longer produce FL2 protein had significantly longer axon growth compared to neurons with normal levels of FL2. This finding suggested that FL2 acts as a negative regulator of axon growth and that its absence might facilitate neuron growth. To test this, the researchers used a technique employing small interfering RNAs (siRNAs)—short strips of genetic material—to effectively “turn off” the *FL2* gene in a rat model of cavernous nerve injury that mimics what can occur in human prostate cancer surgery. In this experiment, *FL2*-siRNAs, packaged in nanoparticles, were topically applied to crushed cavernous nerves immediately after injury. After 4 weeks, the erectile response of the rats treated with *FL2*-siRNAs was significantly improved compared to that of animals treated with controls (no *FL2*-siRNAs). In addition, applying *FL2*-siRNA treatment to rats with severed cavernous nerves resulted in seven of eight animals exhibiting visible nerve regeneration and erectile response 2 weeks later, whereas rats treated with scrambled siRNA had no visible nerve regrowth.

These findings demonstrate that depleting FL2 protein in neurons enhances their regeneration potential and suggest that approaches targeting production of this protein may lead to promising therapeutic strategies for ED caused by prostate cancer surgery.

Baker L, Tar M, Kramer AH,...Sharp DJ. Fidgetin-like 2 negatively regulates axonal growth and can be targeted to promote functional nerve regeneration. *JCI Insight* 6: e138484, 2021.

## BLOOD STEM CELLS AND THE BONE MARROW

**Elucidating New Mechanism for Blood Stem Cell Migration from the Bone Marrow:** Scientists recently reported that a type of nerve cell plays an important role in the migration of blood stem cells from the bone marrow and into the circulation—potentially facilitating their collection for therapeutic uses. Blood stem cells—also called hematopoietic stem cells, or HSCs—have the potential either to self-renew into two identical daughter stem cells or to give rise (mature) into specialized cell types: red blood cells, white blood cells, or platelets. HSCs reside in specialized microenvironments in the bone marrow from which they can be mobilized via physiological stressors, such as significant blood loss, to enter the blood circulation and mature into all the body's blood cells. However, the mechanisms by which HSCs migrate out of the bone marrow are largely unknown.

In new research using both female and male mice, scientists showed that the specialized microenvironments in the bone marrow also contain a high density of sensory nerves called nociceptive nerves. Found throughout the body, these nerves are normally important to pain perception and prevention of tissue damage, sending signals to the spinal cord and brain in response to a noxious (painful) stimulus. So what are nociceptive nerves doing in the bone marrow? It turns out that a molecule released by these specialized nerve cells, called CGRP, acts in concert with other signals in the bone marrow to drive a cascade of events ultimately instructing the HSCs to migrate and enter the blood circulation. In the mid-1990s, a report described how capsaicin, an ingredient in chili peppers, interacts with its cell surface receptor to “turn on” nociceptive nerves in the gastrointestinal tract to produce a burning sensation. With this knowledge in mind, the scientists asked whether capsaicin could affect HSC migration out of the bone marrow. Intriguingly, mice that consumed a capsaicin-containing spicy food diet were found to have significantly enhanced HSC migration into the bloodstream compared to mice fed a standard diet. Targeting the nociceptive nervous system could, therefore, represent a strategy to improve the yield of HSCs needed for stem cell-based therapeutic protocols.

Gao X, Zhang D, Xu C, Li H, Caron KM, and Frenette PS. *Nociceptive nerves regulate haematopoietic stem cell mobilization.* *Nature* 589: 591-596, 2021.

### New Insight into the Mouse Blood Stem Cell Aging

**Process:** New research has identified changes in the microenvironment of the bone marrow that contributes to the aging process of blood (hematopoietic) stem cells (HSCs). The HSC is a type of adult stem cell found in the bone marrow that can self-renew and develop into any type of white blood cell (e.g., B-cell, T-cell, and natural killer cell), oxygen-carrying red blood cells, and a variety of immune cells (e.g., megakaryocytes, monocytes). Because they can effectively regenerate any type of blood cell the body needs, HSCs are recognized as a new way to treat various diseases such as cancers, autoimmune diseases, and nonmalignant blood diseases such as sickle cell disease. However, previous research has shown that for unknown reasons, as people age, the capacity of their HSCs to develop into different types of blood cells diminishes.

*New research shows that pain sensory nerves play a role in the migration of blood stem cells from the bone marrow into the blood circulation, providing insights that could help to improve stem cell-based therapeutic protocols.*

In this study, investigators sought out changes in the aging bone marrow of mice that may contribute to the diminished capacity of HSCs to develop into different types of blood cells. They reported that characteristic hallmarks of aging HSCs in middle-aged female mice are linked to a reduction in the level of a hormone called insulin-like growth factor 1 (IGF1) in the bone marrow. Interestingly, IGF1 treatment of HSCs from middle-aged female mice restored characteristics of younger, healthier HSCs. These findings suggest that therapeutic strategies aimed at increasing IGF1 levels in the bone marrow may be able to halt or partially reverse HSC aging, and, thereby, boost HSC function through middle age and perhaps longer.

Young K, Eudy E, Bell R,...Trowbridge JJ. *Decline in IGF1 in the bone marrow microenvironment initiates hematopoietic stem cell aging.* *Cell Stem Cell* 28: 1473-1482.e7, 2021.

# Developing Vascular Access Technologies To Improve Kidney Disease Treatment—Highlights of Small Business Innovation Research

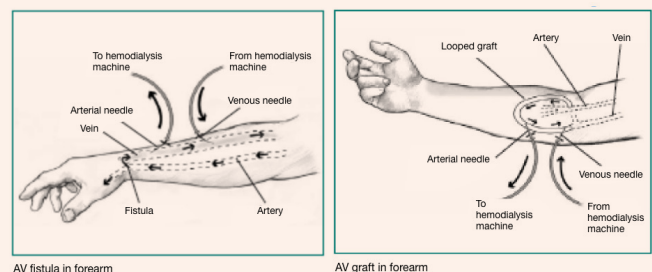
The Small Business Innovation Research (SBIR) and Small Business Technology Transfer (STTR) programs together are one of the largest sources of early-stage capital for technology commercialization in the United States. The SBIR program allows U.S.-owned and -operated small businesses to engage in federal research and development (R&D) that has a strong potential for commercialization. The NIDDK's program supports health and life science companies that are creating innovative technologies that align with the Institute's mission to improve health and save lives. A key objective of the program is to translate promising technologies to the private sector and enable life-saving innovations to reach consumer markets.

As mandated by U.S. law, SBIR and STTR applications—like other grant applications—undergo a two-step peer-review process. SBIR and STTR applications that are approved are then funded through a congressionally mandated set-aside share of the Institute's budget in the form of grants. (Additional information is available at <https://sbir.nih.gov>.)

## FEATURE HIGHLIGHT: SBIR PROJECTS ON VASCULAR ACCESS FOR KIDNEY DIALYSIS

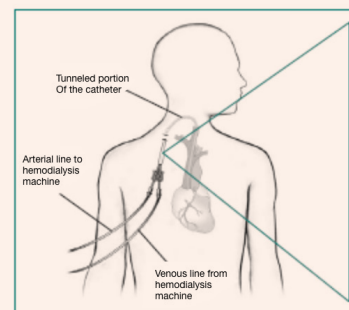
The NIDDK supports SBIR grants in many areas across its mission. This feature highlights just one of those research areas as an example—developing technologies to improve vascular access for kidney dialysis treatment. People whose kidneys no longer function sufficiently (kidney failure) require either a kidney transplant or dialysis treatment to live. One form of dialysis, hemodialysis, serves to filter waste products and water from the blood—imitating the function of healthy kidneys. Before starting

hemodialysis, a surgical procedure is performed to create what is called a “vascular access,” the point at which a connection can be made to the dialysis machine, for blood to flow into the dialysis machine to be filtered and then returned to the body.



AV fistula in forearm

AV graft in forearm



Tunneled venous catheter

There are several types of vascular access. The arteriovenous (AV) fistula and AV graft are two types designed for long-term use. A catheter is sometimes used as a temporary means of vascular access until a longer-term form of access can be performed surgically. Vascular access has been described as the “Achilles' heel” of hemodialysis because of complications that can arise, such as infection, poor blood flow, or blockage. But for people with end-stage kidney disease, it is their lifeline—without it they cannot receive the lifesaving treatment. During each hemodialysis session, a connection is formed between the patient's blood

and the filtration machine by inserting needles through the skin into the AV fistula or graft. This process of “cannulation” is also prone to several complications (e.g., balloon-like bulge in an artery, bleeding, and clotting). Researchers are thus continuing to develop ways to improve vascular access outcomes, as well as to improve ease of use and safety as a step toward making dialysis treatment more feasible for patients to receive in their homes, rather than thrice weekly in dialysis centers.

In recent years, the NIDDK’s SBIR program has supported several vascular access technology projects to promote graft and fistula maturation. The following are examples of these projects, which are at different stages of development:

*Addressing the need to reduce clotting events, infection, and blood vessel narrowing.*

Synthetic grafts are prone to clotting events, infection, and blood vessel narrowing. This project used a regenerative medicine approach to develop a “humanized” graft (i.e., a human protein scaffolding surrounding a biodegradable polymer). In multiple clinical trials, the humanized graft was reported to be safe and functional. The small business has raised non-NIDDK/NIH capital to evaluate the product in later-stage clinical trials.

*Addressing the need to reduce needle-related complications during cannulation.*

This project developed an innovative graft that was implanted into pre-clinical models (e.g., pig and sheep) and functioned as intended with no needle-related complications reported. One version of the graft was studied in a clinical trial; then improvements were made, and a newer version is being studied in a larger trial supported by this grant. This technology could facilitate home dialysis.

*Addressing the need to improve AV fistula utilization.*

Some patients are excluded from receiving an AV fistula for lack of vessels meeting size thresholds. Prior studies have observed that patients with larger initial vein diameters have better AV fistula outcomes. Thus, there is a need to develop a system designed to rapidly and permanently dilate peripheral veins. This project developed a new technology that, when tested in sheep, doubled the diameter of veins with no adverse effects reported. A first-in-human test was then successfully completed using non-NIDDK/NIH capital.

The NIDDK will continue to pursue technologies to improve the safety and efficacy of vascular access through the SBIR program. These SBIR grants complement other NIDDK research grants to fill long-standing gaps in optimal care that are essential to ensuring the best possible hemodialysis outcomes.

*Lawson JH, Glickman MH, Ilzecki M,...Niklason LE. Bioengineered human acellular vessels for dialysis access in patients with end-stage renal disease: two phase 2 single-arm trials. Lancet 387: 2026-2034, 2016.*

*Gage SM, Lawson M, Nichols C, Sycks D, Manson RJ, and Knight JA. An immediate access dialysis graft designed to prevent needle-related complications: Results from the initial pre-clinical studies. J Vasc Access 21: 328-335, 2020.*

*Gage SM, Illig KA, and Ross JR. Use of a novel immediate access dialysis graft designed to prevent needle-related complications: A first-in-man case report. J Vasc Access 22: 475-479, 2021.*

# CCEH: Increasing Collaboration and Access to Critical Research Resources for Research To Combat Nonmalignant Blood Diseases

To increase access to critical research resources, foster collaboration in blood (hematology) research, and increase the cost-effectiveness of this research, The NIDDK supports the Cooperative Centers of Excellence in Hematology (CCEH). The CCEH collaborate with the NIDDK Hematology Central Coordinating Center, collectively comprising the NIDDK Hematology Centers Program in a national multidisciplinary research effort to combat nonmalignant hematologic diseases and to study normal hematopoiesis (*i.e.*, production of blood or blood cells). The CCEH currently include five Centers, each of which involves integrated teams of investigators from a wide range of disciplines, shares specialized state-of-the-art equipment, and serves as a regional or national resource.

The Program serves as a national hub for nonmalignant hematology research by generating and providing resources and expertise to the broader research

community. In addition, the CCEH support career development of scientists in the field through national pilot and feasibility programs and local programs that fund small projects to generate preliminary data for inclusion in larger grant applications. The CCEH also sponsor short-term enrichment activities for the entire hematology research community, such as seminar series.

Examples of research areas currently under study by the CCEH include the biology of blood cells and the bone marrow niche in which blood stem cells reside, the use of blood stem cell transplantation as a potential cure for nonmalignant blood diseases, the role of iron in biological processes and its misregulation leading to deficiency or excess, and other areas.

For more information, see: <https://www.niddk.nih.gov/research-funding/research-programs/hematology-centers>.

# STORY OF DISCOVERY

## The NIDDK Provides Foundational Research Support for First FDA-approved Therapy To Treat the Metabolic Disease Primary Hyperoxaluria Type 1

### PRIMARY HYPEROXALURIA (PH)

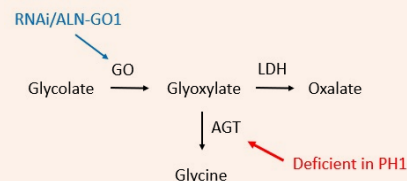
PH is a set of genetic metabolic disorders characterized by increased levels of oxalate in the kidneys, urine, and other organs of the body. The three types of PH (PH1, PH2, and PH3) are caused by a "protein deficiency" and distinguished by deficiencies in different proteins.

oxalate can build up and damage other organs, including the heart, bones, and eyes. Treatments for the disease have been limited, and the only effective treatment for most people is a combined liver-kidney transplant. The lack of effective, nonsurgical treatment options underscores the urgent need to develop new drug therapies for this serious disease.

### PRIMARY HYPEROXALURIA TYPE 1 (PH1)

- **Primary hyperoxaluria type 1 (PH1)** is a rare disorder that mainly affects the kidneys. It results from buildup of a substance called oxalate, which is normally filtered through the kidneys and excreted in the urine. In people with PH1, the accumulated oxalate is deposited in the kidneys and urinary tract and combines with calcium to form calcium oxalate—the main component of kidney stones. Symptoms of kidney stones include sudden abdominal or flank pain, blood in the urine, frequent urge to urinate, pain while urinating, and/or fever and chills.
- Indications and symptoms of PH1 vary in severity and may begin any time from infancy to early adulthood. People with PH1 may experience recurrent kidney stones, blood in the urine, and urinary tract infections. PH1 can result in end-stage kidney disease, which is life-threatening, and the need for dialysis. As kidney function worsens,

### GENETIC MECHANISMS OF PH1



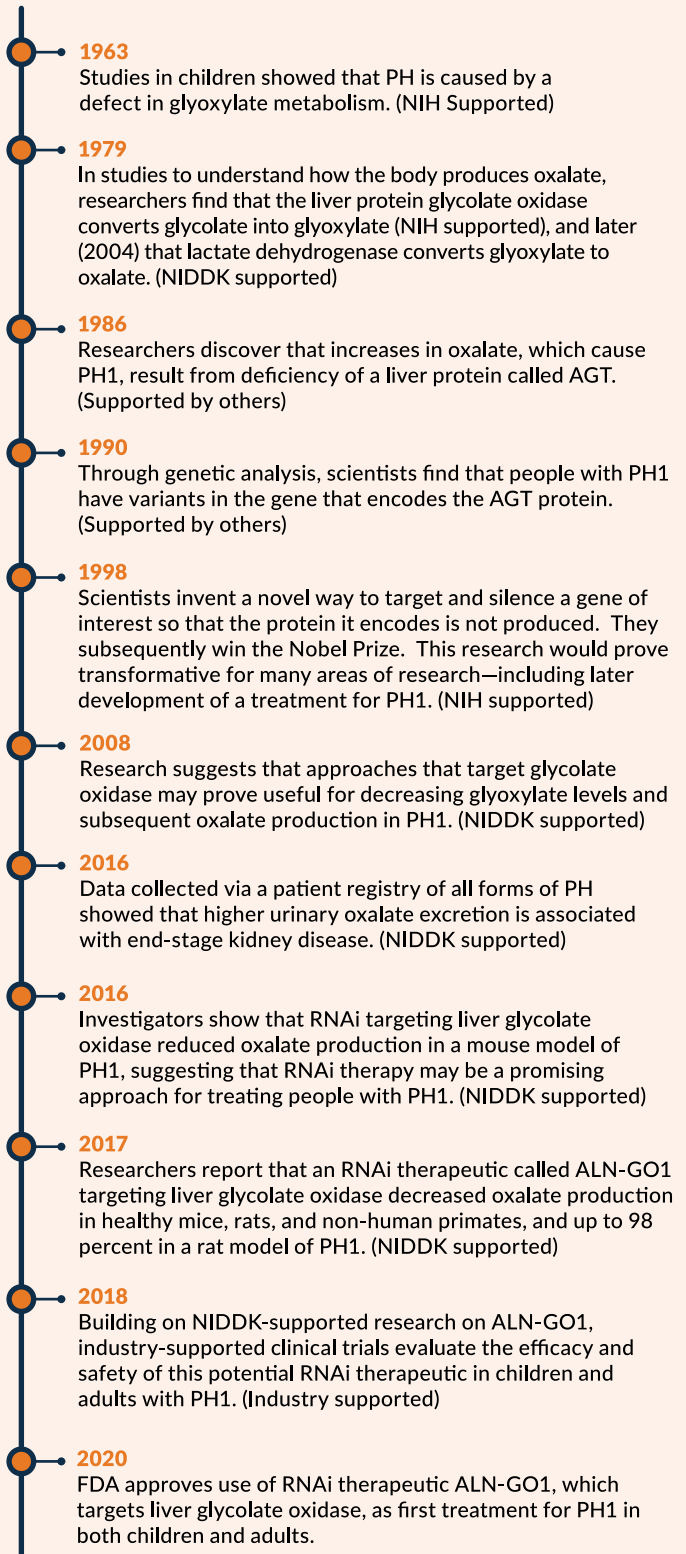
- PH1 is caused by mutations in a gene called *AGXT*. This gene gives the body instructions for producing a protein called alanine-glyoxylate aminotransferase (AGT). AGT is found in the liver and converts a compound called glyoxylate to the amino acid glycine.
- Mutations in the *AGXT* gene lead to a deficiency of AGT to convert glyoxylate to glycine. This, in turn, causes glyoxylate to accumulate, and it is ultimately converted to oxalate by a protein called lactate dehydrogenase. Excess oxalate that is not excreted from the body then combines with calcium to form calcium oxalate, leading to kidney damage.

# STORY OF DISCOVERY

- The metabolic pathway leading to oxalate production includes the conversion of glycolate to glyoxylate by the protein glycolate oxidase (GO).
- Thus, glycolate oxidase is an important player in the biological mechanism leading to high levels of oxalate in PH1. Researchers discovered that targeting and thus reducing levels of glycolate oxidase using RNA interference (RNAi) technology was a promising strategy to reduce oxalate levels and treat the disease.

## THE RESEARCH PATH TO AN FDA-APPROVED TREATMENT

- The accompanying timeline shows that, for many decades, the NIDDK and the NIH have supported foundational research to better understand the metabolic dysfunction underlying PH1 (1960s), including biological pathways involved in oxalate production (1970s), development of RNAi technology to target/silence genes (1990s, 2000s), identifying glycolate oxidase as a promising target to treat PH1 (2000s), and translational research using RNAi-based approaches to target GO and reduce oxalate production in animal models of PH1 (2010s). This research laid the foundation for industry-supported trials of the RNAi therapeutic ALN-GO1 (2010s), culminating in its recent U.S. Food and Drug Administration (FDA) approval as the first drug to treat PH1 in both children and adults (2020). This decades-long story elegantly showcases how basic research elucidated biological mechanisms of both health and disease and resulted in a new therapeutic that greatly improves the prognosis of people with PH1.
- The NIDDK continues its support of this research area to improve the lives of people living with PH1 as evidenced by a recent report describing three patients with PH1 and kidney failure who were able to regain kidney function and discontinue dialysis after treatment with pyridoxine (vitamin B6).





## Helping People Hydrate To Prevent Kidney Stones

Kidney stones are a common ailment in the United States. Most kidney stone research and treatments to date have focused on helping people who have a kidney stone and are having excruciating pain; more research is needed about how to prevent and effectively manage kidney stones. The NIDDK is currently supporting a clinical trial that hopes to identify ways to change behavior so a person is less likely to have another kidney stone, and help determine if drinking more water can help prevent kidney stone recurrence. As behavior change is not always easy—especially in times of challenge such as the current COVID-19 pandemic—teamwork, support, and flexibility among study volunteers, staff, and investigators have been especially important to successfully moving this important research forward. (See insets for perspectives on the clinical trial from study coordinators.)

### WHAT ARE KIDNEY STONES?

A key function of the kidneys is to filter out toxins and waste products from the blood. Together with water and salt, this filtration process forms a waste liquid called urine, which is sent to the bladder for storage and then expelled via urination (“peeing”). However, solid deposits can develop when there is a high concentration of certain minerals and salts in the urine. Commonly referred to as kidney stones, clinicians and scientists also use the term “urinary stone disease” for this phenomenon because although most often initiated in the kidney, these deposits can form and/or cause problems almost anywhere in the urinary tract. Kidney stones vary in size, shape, and type. Quite often people have them and pass them out of the body without knowing it or without an intervention. However, kidney stones can also get stuck in the kidneys or in the tubes, called ureters, that carry urine to the bladder. This can cause severe pain and/or bleeding, frequently sending people to the emergency

room for care. The prevalence of kidney stones has nearly doubled in the past 15 years, and current estimates are that 1 in 11 people in the United States is affected. Kidney stones are more common in men than in women, but they are increasingly occurring in younger women, and they can also occur in adolescents and youth. Importantly, persons who have had a kidney stone in the past are more likely to have another.

Advances in treatment, such as using targeted high energy shock waves to blast larger stones into small pieces that can be passed more easily, means that some stones can be treated noninvasively. There are also some medicines available to help prevent recurrence of specific types of stones. However, not all treatment is noninvasive or fully effective. In fact, the majority of symptomatic kidney stones are treated using a surgical procedure in which a thin, flexible telescope is inserted into a ureter to help the doctor either snare a stone directly or use a tiny laser to fragment it for subsequent removal or natural passage, often accompanied by temporary placement of a small tube (stent) in the ureter to help with drainage. Moreover, estimates indicate that urinary stone disease is one of the costliest urologic conditions. Thus, a double-pronged approach focusing on both prevention and therapeutics is necessary.

### THE URINARY STONE DISEASE RESEARCH NETWORK

To address the burden that kidney stones place on people and the health care system, in 2016 the NIDDK funded a coordinating center and four clinical centers across the country to form the collaborative Urinary Stone Disease Research Network (USDRN). USDRN scientists are designing and conducting research on kidney stones in adults and children to learn more about who is at higher risk for kidney stones, symptoms

# PERSONAL PERSPECTIVE

associated with the placement of stents in a ureter, and how to prevent stones from forming again. By developing a robust evidence base, it is hoped that USDRN studies will provide a foundation for novel approaches and management strategies for kidney stones and their prevention.

There are several factors associated with the development and recurrence of kidney stones, but one of the main risk factors is inadequate hydration. This leads to low daily urine volumes and thus higher salt and mineral concentrations. Evidence suggests that significantly increasing fluid consumption to produce 2.0 to 2.5 liters (about 8.5 to 10.5 cups) of urine daily can prevent or delay recurrence of kidney stones, and would be a low risk, inexpensive, and effective approach. However, although people who have had kidney stones are commonly counseled to increase their fluid intake, this can be challenging for many reasons. These range from simple forgetfulness to limited bathroom access to having other urinary symptoms that cause hesitancy about drinking more fluids. Another reason is that it is easy to lose sight of how helpful a behavioral change can be once an immediate health crisis—such as severe pain from passing a stone—has ended. As a result, not many people who have had a kidney stone end up increasing their fluid consumption significantly.

In light of these challenges, the USDRN developed and launched a clinical trial to test a strategy that could help people who have had at least one kidney stone effectively increase and maintain their fluid intake and prevent stone recurrence. This trial is called the Prevention of Urinary Stones with Hydration, or PUSH.

## THE PUSH CLINICAL TRIAL: A NOVEL APPROACH TO INCREASING HYDRATION

PUSH is a randomized clinical trial testing the effectiveness of a multi-component behavioral strategy to increase water intake and consequent urine output enough to prevent kidney stone recurrence in adolescents and adults. PUSH has enrolled volunteers ages 12 and older with a history of stones, daily urine

output lower than the study target, and who own a smartphone or tablet they are willing to use for the study. The trial set an enrollment goal of 1,642 people, which it expects to meet in early 2022.

Once enrolled, a PUSH participant is randomly assigned into one of two groups for a 2-year follow-up period: the usual care “control” group or the intervention group. In both groups, participants are given a commercially available “smart” water bottle that tracks water consumption through a smartphone or tablet app. Participants in the usual care group receive standard recommendations to increase their overall fluid intake in order to achieve daily (24 hour) urine output of at least 2.5 liters or, for adolescents weighing less than about 165 pounds, an increase in urine output scaled to their weight. They are allowed but not required to use the smart water bottle to help monitor their water intake. They also receive monetary compensation for completion of study activities.

Participants randomly assigned to the PUSH intervention group receive usual care information and have the same urine output targets, but they are also engaged in a three-pronged behavioral change program intended to help achieve and maintain the targets, consisting of the following:

- They are provided with individualized, calculated “fluid prescriptions” indicating the additional amount of water they need to consume daily via the smart water bottle to achieve the PUSH target urine output; this water is in addition to any other fluids they normally consume.
- They are provided small financial incentives for meeting their daily water goal, set up as a banked total amount each month in which the incentive available on a particular day is lost if a goal is not met; participants are notified about whether they’ve kept or lost incentives via encouraging text messages. The incentive is available every day at the beginning of the trial, but is then tapered down in phases, such that during the last 6 months of the trial there is no longer a financial incentive.

# PERSONAL PERSPECTIVE

- In parallel, during year 1, they are provided access to health coaches and structured problem-solving sessions and “boosters” to help overcome individual barriers to meeting fluid intake goals. Participants who still have challenges with adherence to their fluid goals after year 1 have access in year 2 to alternative, “low touch” interventions that they select themselves, such as text or telephone reminders or engaging a support partner.

At the end of the trial, PUSH researchers will compare data from the two groups regarding the number and timing of symptomatic stone events as defined by the study. This way, they can determine whether or not the structured, phased, behavioral change intervention provides a significant benefit over usual care by preventing or delaying recurrence of stones that either induce symptoms and/or require medical intervention.

According to the original study design, during the 2 years a person is enrolled, all participants are also asked to perform periodic 24-hour urine collections at home, have kidney imaging performed at the beginning and end of their participation, and fill out online questionnaires about general health status and their stone disease every 3 months, and, for adult participants, questionnaires about urinary symptoms every 6 months. These measures can provide insights into changes in urine output as a result of the intervention, changes in preexisting stones present in the kidney, and how the intervention affects urinary symptoms.

## COVID-19 AND PUSH: KEEPING IT GOING

The PUSH clinical trial began in September 2017. In March 2020, the COVID-19 pandemic led to an abrupt shutdown of laboratories and clinical trials, as new safety measures were put in place to protect investigators, study volunteers, and others involved in or supportive to the biomedical research enterprise. At that time, just over half of the anticipated volunteers had been assigned to an arm of the PUSH clinical trial.

In response to COVID-19, the USDRN rapidly developed a plan to restart PUSH in a way that would enable continued recruitment of volunteers while keeping both them and study personnel safe. This included considering how to lower barriers to participation and retention, especially as people might be hesitant to join a clinical study in person or undergo in-person procedures, such as imaging. In consultation with the trial’s Data Safety Monitoring Board and working with the Institutional Review Boards at each clinical site—all of which have a role in ensuring clinical trial participant safety and welfare—the PUSH clinical trial received approval for remote recruitment. This includes obtaining informed consent by phone or video, enrolling participants, and randomly assigning them to one of the two trial groups.

As a result, the PUSH trial not only implemented a change to make consenting processes safer and easier for potential participants and staff, but it leveraged this change to reach out and recruit participants from across the Nation. This lack of geographic barriers is enabling PUSH to enroll a more diverse population. It has also formed partnerships with health care systems—in particular, pediatric kidney stone care systems, which has accelerated identification of adolescents interested in joining PUSH. The USDRN also determined that PUSH had sufficient data from already enrolled patients to enable them to waive other in-person requirements, such as on-site kidney imaging procedures at enrollment and/or close out, and instead would use as possible images and other data obtained during regular clinical care going forward. What the USDRN learned from necessity, and the flexibility it added due to COVID-19, may serve as a model for design of other clinical trials.

## COORDINATING FOR SUCCESS

During the PUSH study—and, indeed, during most clinical trials—volunteers interact with a variety of trial personnel, but a common face is that of the study coordinator. Study coordinators, or clinical research coordinators, are linchpins in a clinical trial. Their wide array of responsibilities includes helping recruit and

# PERSONAL PERSPECTIVE

enroll study volunteers, serving as liaisons with study investigators and the research institution, monitoring progress and maintaining all required forms, ensuring compliance with all regulations, preparing reports, and being a key point of contact for study volunteers throughout the duration of a clinical trial—a role that is often crucial to helping participants see a study through to the end in the midst of busy lives, responsibilities, and other challenges.

In a behavioral change study such as PUSH, in which participants know whether they are in the intervention or the control group, study coordinators are also critical to helping keep the study “blinded” to most of the staff and to participants’ health care providers. That is, it is crucial that study investigators, clinicians, treatment providers, and the statisticians who analyze the study results have no idea about who has been assigned to which study group, or about certain individual characteristics—that way, there is less risk of biased actions occurring during the trial, analyses of data, and in the interpretation of results.

Thus, PUSH study coordinators are key go-betweens. Study coordinators also get to see firsthand the challenges that volunteers may be having in adhering to trial requirements, and also help them in navigating these challenges so that they are more likely to complete the trial successfully. In a study such as PUSH, the challenges that arise toward achieving the goals of the intervention can also be different for youth and adults.

The additional challenges imposed by COVID-19 have also meant increased training for PUSH staff and new strategies and flexibility for them and for study volunteers. Yet, together, they are coordinating for ultimate success of the PUSH trial and its goal of determining whether a behavioral change strategy focused on increased hydration can indeed help people avoid recurrent kidney stones and the associated pain, life disruption, and burden of care.

To learn more about the PUSH trial, see: <https://usdrn.org/participate/push>.

## BRITTNEY’S EXPERIENCE AS A PUSH STUDY COORDINATOR



Brittney

Brittney, 29, grew up in Georgia and describes herself as both a quiet lover of books and an avid sports fan, especially football. She laughs as she says she has found herself “screaming at the television” when rooting for her favorite teams, the Philadelphia Eagles and the University of North Carolina Tar Heels. Adding that she is “an introvert, but enjoys working on great teams,” she shared her experience as a research study coordinator for the University of Pennsylvania/Children’s Hospital of Philadelphia, or CHOP. Brittney joined the Prevention of Urinary Stones with Hydration, or PUSH, clinical trial team at CHOP in March 2017, shortly after completing her master’s degree in public health. CHOP soon

# PERSONAL PERSPECTIVE

enrolled its first PUSH trial participant from among kids 12 and older, in October 2017. Brittney remembers how the first enrollments took “an hour and a half, and the patients were all so nice!” as everyone got through the process together, from reviewing how to use the smart water bottle to going for imaging in other parts of the medical complex. Later, she generally worked with three to four participants per day who were at various stages in the trial. To facilitate the study and make participation easier, she constantly sought out ways “to prevent having any barriers or burdens ... on the parents as much as possible,” such as coordinating kids’ research and clinical appointments at CHOP well in advance.

*Brittney, a study coordinator for the PUSH clinical trial, constantly sought out ways “to prevent having any barriers or burdens ... on the parents as much as possible” to facilitate the study and make kids’ participation easier.*

Brittney keenly remembers the rapport she and participants built during their initial visits, which helped kids and their families throughout their time in the trial. They felt comfortable texting her with questions and asking for help related to study activities. She did a lot of listening and problem-solving, saying that probably the biggest frustration for kids and families was when they felt they were doing everything right, but for some reason the bottle “just would not work.” Another challenge for some was wanting to keep life simple and use just one bottle for all their drinks, whereas for kids in the PUSH intervention group the smart water bottle was meant to provide a prescribed amount of supplemental water per day. “We really tried our hardest to make sure we could compromise where we could, and let them know where we couldn’t,” says Brittney. It was also super important to make

sure that kids as well as parents were on board with the study, she says, noting that, “one of our big points [when enrolling someone] was that ‘this was one of the things you have to want to do for yourself, because it is supposed to be a behavior change.’” Along these lines, she adds that although kids might not always have been excited about getting automated daily messages to help with behavior change, it was “funny ... how quickly they would respond” if they got a message saying that they had missed out on the financial incentive when they knew they had met their hydration goal—and, of course, she would fix it for them.

When the COVID-19 pandemic temporarily halted PUSH recruitment in early 2020, Brittney took on double duty, becoming part of the COVID-19 clinical volunteer team at CHOP while continuing to send materials and make remote calls to support kids already enrolled in PUSH. As PUSH reopened and there was approval for remote recruitment, Brittney began contacting potential new participants by phone. She continued strategizing to keep participation easy, appealing, and safe in light of COVID-19-related uncertainties and fears—including letting families know when procedures or samples they needed for kids’ clinical care could be done and covered as part of their PUSH participation, and reassuring worried parents that their kids could now opt out of end of study kidney imaging. Brittney left CHOP in July 2020 to enroll in medical school back in Georgia, noting that she still “loves urology” and is hoping to pursue a career in surgery. She remembers with deep appreciation how some of the families told her at the end of their participation how the time and care she took with them “really changed their lives.” From her time at CHOP and the PUSH study, particularly as they faced the pandemic, she says she remains inspired by the kids and families with whom she met and worked. How to capture her feelings in one word? “Resilience,” says Brittney.

## HOLLY'S EXPERIENCE AS A PUSH STUDY COORDINATOR

Seattle area native and mother of two, Holly, 41, has been a clinical research study coordinator for over 14 years—4 of them with the Prevention of Urinary Stones with Hydration (PUSH) clinical trial site at the University of Washington (UW), which is enrolling adult participants. When not spending time with her sons at Mariners Stadium and other activities as the COVID-19 pandemic allows, she is mainly focused on her study coordinator role, a career that she says she “fell into” and trained for while working at a private practice, and found she really enjoys. Along with data collection and administrative aspects, “I enjoy the volunteers’ participation and ... face-to-face contact [with them],” she says with a smile. On a daily basis, Holly spends much of her time determining whether potential PUSH participants meet eligibility criteria for the study, going over the study with eligible candidates so they are fully informed prior to joining, and randomly assigning new participants to one of the two PUSH study groups. She does this either in person or now also remotely, including people from across the country. So far, “we’ve had success enrolling people from Oklahoma, Tennessee, and New York,” as well as Arizona and



Holly

California, she says. Because the PUSH trial didn’t require in-person visits for most of a participant’s involvement even before the pandemic, Holly adds that “we were already comfortable helping people along the way for 2 years remotely,” including with

issues such as smart water bottles breaking or the associated tracking app not working.

Holly emphasizes that a big goal for her and the rest of the PUSH team is to make sure participants do not feel burdened by the study, but rather feel it is something they can integrate into their daily lives. “Being a prevention study, we want it to be a positive experience for them,” she says. As part of the study, she often reaches out to participants to help with their 3-month follow ups as needed, but also hears about their frustrations at other times, such as when they don’t meet hydration goals. “Sometimes people just had to take a break,” she says, adding that when she randomizes participants she makes sure to let them know that, “we understand life gets busy and, yes, you can’t meet your goal every day, but we are here to help you.”

Holly has found that, despite some of the frustrations they encounter, the PUSH participants she has worked with are really positive about their experience. She says that most of the people who volunteer for PUSH “are very excited to start the study,” adding that “one of the best things is that ... at the end of their 24 months, they are really proud of themselves” regarding improvements in 24-hour urine collections and water consumption, and “that’s always good to see.” She observes that they know that not only are “they helping themselves, but also really contributing to kidney stone disease” research. In terms of their daily experience, she says that participants in the intervention group “really [do] love the smart water bottle” and being able “to see where they [are] at during the day for their goal” and whether they’ve met it, via the app. Participants are allowed to keep their bottles at the end of their study time, and Holly says a few have said at that point that

# PERSONAL PERSPECTIVE

they are “sad that they weren’t able to continue on” in PUSH, although of course they can continue to use the bottle to monitor water consumption.

*Holly, a study coordinator for the PUSH clinical trial, emphasizes that a big goal for her is to make sure participants do not feel burdened by the study: “Being a prevention study, we want it to be a positive experience for them.”*

For Holly, as the PUSH clinical trial continues its work through the evolving challenges of the COVID-19

pandemic, teamwork and support are critical—both personally and for the participants. It’s “not only us trying to be positive and supportive,” but also, “here at the clinical site we have support from the principal investigators, ... the monitors, and the coordinators [and assistants] supporting each other during this difficult time,” she says. She was especially thankful for this support when dealing with wildfires near her home in 2020. Talking again about the study volunteers, Holly recalls fondly how one participant even gave the UW PUSH team the moniker “Team Hydrate.” This seems to capture the spirit of PUSH—go Team Hydrate!

## ACKNOWLEDGMENTS

January 2022

### Research

The NIDDK gratefully acknowledges the researchers whose studies are described in this report, and the volunteers who participate in clinical research studies.

### Writing and Production

Overall Production

Patrick Donohue, Ph.D., Office of Scientific Program and Policy Analysis

Sandeep Dayal, Ph.D., Office of Scientific Program and Policy Analysis

Highlights of Research Advances and Opportunities

#### *Staff of the NIDDK Office of Scientific Program and Policy Analysis*

Heather Rieff, Ph.D., Director

Eleanor Hoff, Ph.D.

Lisa Gansheroff, Ph.D., Deputy Director

Dung Pham

Rebecca Cerio, Ph.D.

B. Tibor Roberts, Ph.D.

Sandeep Dayal, Ph.D.

Megan Singh, Ph.D.

Patrick Donohue, Ph.D.

Jaime J. Smith, Ph.D.

Mary Hanlon-Tilghman, Ph.D.

Robert Tilghman, Ph.D.

Erica R. Hennes, B.A.

Julie A. Wallace, Ph.D.

*Input from staff within the NIDDK's scientific program and research divisions: the Division of Diabetes, Endocrinology, and Metabolic Diseases; the Division of Digestive Diseases and Nutrition; the Division of Kidney, Urologic, and Hematologic Diseases; the Division of Extramural Activities; and the Division of Intramural Research.*

*Contributions from the NIDDK Office of Communications and Public Liaison*

Alyssa M. Voss, M.P.H.

For more information on the NIDDK and its mission, and links to other sites, visit: [www.niddk.nih.gov](http://www.niddk.nih.gov).





National Institute of  
Diabetes and Digestive  
and Kidney Diseases

NIH Publication Number: 22-7962