

WHOLE GENOME SEQUENCING
 high-throughput screening
 gamma secretase modulator
3-D disease model
 amyloid beta
microbiome
APOE
 innate immunity
 nanobodies
PROGRESS
 protein kinase C
NEURON
 epigenetics microglia
ALZHEIMER'S
DISEASE
 INFLAMMATION



MISSION

To fund research with the highest probability of preventing, slowing or reversing Alzheimer's disease.

curealz.org

2016 RESEARCH ANNUAL

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Message from the Co-Chairmen of Cure Alzheimer's Fund

Jeff Morby and Henry McCance



We think these [5] leadership roles make us unique among Alzheimer's disease research groups, help explain our momentum, and are important to review with you.

Dear Friends,

We are pleased to report that in 2016, our cumulative committed research funding exceeded \$50 million, and as of this writing, that number already has grown to \$60 million. In 2016, we granted a record \$13.5 million for 56 projects, an increase of 34 percent from 2015. The 2016 funding was provided by \$15.6 million in donations from 15,000 donors, our 12th record fundraising year and an increase of 33 percent from 2015.

OUR LEADERSHIP ROLES

In last year's chairman's letter, Jeff Morby summarized five leadership roles that Cure Alzheimer's Fund (CureAlz) has carried out within the field. We think these leadership roles make us unique among Alzheimer's disease research groups, help explain our momentum, and are important to review with you.

First: Leadership in the Genetics of Alzheimer's Disease. From the founding of CureAlz, we have emphasized the importance of genetics to Alzheimer's research. Our goal has been to identify all risk genes, use those genes to identify disease mechanisms, and pursue potential therapies based on the knowledge gained from AD genes.

In 2005, there were only four known Alzheimer's genes, representing about 30 percent of the genetic profile of the disease. Rudy Tanzi had co-discovered three of those four genes. Later, as head of our Alzheimer's Genome Project™ (AGP), he discovered five new AD genes; the discovery was named one of the "Top 10 Medical Breakthroughs" in 2008 by TIME magazine. Since then, one of those five, CD33, has become the second-biggest drug target for Alzheimer's in the biopharma industry (the first being the amyloid precursor protein discovered by Rudy in 1987). In 2013, we were the first research organization to use "whole genome sequencing" to sequence the entire DNA of more than 1,500 individuals from AD families, and this has led to the discovery of more than 100 new Alzheimer's candidate genes.

We now have the most comprehensive Alzheimer's genomics database in the world.

We now have the most comprehensive Alzheimer's genomics database in the world. It is being used to find the variants in the candidate genes that underlie biological risk for the disease, the results of which provide the underlying data for the Genes to Therapies™ (G2T) program. In that program we study how the newly identified Alzheimer's gene variants cause AD and how we can design therapeutic leads to prevent them from doing so.

More than two-thirds of the 5.4 million Americans living with Alzheimer's are women. In 2016, CureAlz and the Rotary International Foundation co-funded a research project to analyze databases of Alzheimer's family genomes and, for the first time, identify gene variants that impact risk differently for women than for men. We already have found several new genes involved with inflammation in the brain that influence risk differently in females versus males, and expect to find additional differences in 2017.



Second: Leadership in the Creation of Advanced Research Tools. In order to study the new AD risk gene variants identified in the AGP, a large number of new mouse models of newly discovered AD genes are being created as part of G2T. Our scientists actively are using these mice “tools” to better understand different disease processes and test new therapies. In addition to the mouse models, the new Alzheimer's genes and variants also are being studied using a unique new analytical tool developed by Rudy and Doo Yeon Kim in 2015, dubbed “Alzheimer's in a Dish” (ADiD). The New York Times referred to it as “a giant step forward” for the field.

ADiD uses human stem cells to grow Alzheimer's nerve cells and see them, which develop within an artificial mini-brain in a Petri dish gel that resembles the consistency of the brain. For the first time, this system is allowing both amyloid plaque and tau tangle pathology to be created and analyzed in a Petri dish. The system has been shared openly with the Alzheimer's field and is potentially revolutionizing Alzheimer's drug discovery; it now allows research studies to be carried

out significantly faster and cheaper than with mice. Using this system and high-throughput screening of literally thousands of drugs, G2T has led to the discovery of nearly 50 known drugs and natural products that potentially reduce Alzheimer's pathology. These drugs now are being analyzed for repurposing as Alzheimer's drugs, with the expectation that some will become candidates for human trials.

Our researchers continue to enhance the diagnostic capabilities of ADiD by adding inflammatory and blood-brain barrier genes to the ADiD mini-brain. Both inflammation and blood-brain deficiencies are major contributors to Alzheimer's disease, and ADiD already is providing us with new insights. Furthermore, ADiD has proven to be a research tool of analysis for a number of different neurological diseases, such as Parkinson's and ALS. Because of its analytical potential for the scientific community, in April 2017 CureAlz sponsored a highly successful symposium at the Alzheimer's Disease/Parkinson's Diseases Congress (ADPD) in Vienna, which updated more than 1,000 scientists on the latest advances in the ADiD model, and its use and results in drug screening.

Third: Leadership in the Development of Effective Forms of Scientific Collaboration.

Our Research Consortium and Scientific Advisory Board members are some of the world's most brilliant Alzheimer's researchers. In 2016, we added three exceptional scientists to our Research Consortium: Ben Barres of Stanford University, Bart De Strooper of the UK Dementia Research Institute, and Nancy Ip of the Hong Kong University of Science and Technology. Apart from their capabilities, what truly distinguishes all of the scientists involved with our organization is their willingness to collaborate. All of them have developed in conjunction with CureAlz an atmosphere of trust and cooperation. All are willing to share their unpublished data and insights, and to participate in quarterly brainstorming sessions focused on attacking the disease in new and creative ways. Additionally, they are guided in their research decisions by the CureAlz Roadmap, a jointly shared overall research strategy, which is updated as new scientific insights are attained.

In 2016, Rob Moir and Rudy Tanzi published a new paper on the “Anti-Microbial Hypothesis,” which *The New York Times* described as “**provocative new research.**”

Fourth: Conceptual Leadership in the Field of Alzheimer’s Research. Our emphasis on genomics, translational drug discovery, the development of new tools and databases, and our collaborative approach to scientific exploration, has allowed CureAlz and its researchers to conceptualize a comprehensive model of Alzheimer’s disease that has led to a generally accepted scientific consensus about how Alzheimer’s disease originates and creates a vicious cycle of abeta peptide accumulation, tangle formation, nerve cell death and inflammation, leading to ever-accelerating nerve cell death. This model leads to three basic strategies for medical intervention (intervention points), around which we have organized our research: 1) early-phase intervention, inhibiting abeta peptide deposition and/or clearing it from the brain; 2) mid-phase intervention, preventing the formation and spreading of tau tangles; and 3) late-phase intervention, fighting inflammation and slowing down or stopping the disease process.

In 2016, Rob Moir and Rudy Tanzi published a new paper on the “Anti-Microbial Hypothesis,” which *The New York Times* described as “provocative new research.” This revolutionary hypothesis about the underlying cause of Alzheimer’s disease shows that abeta, the peptide that forms plaque in the brain, is also a component of the brain’s innate immune system, the system that protects the brain from pathogens. Abeta traps and kills pathogens when they enter the brain, essential for the protection of the brain. However, too much of this activity, resulting from genetic or other defects, may cause the brain to overproduce, or fail to clear, abeta, thereby starting the vicious cycle of the disease. This radical new view has major implications for drug discovery.

As new discoveries are made we adjust our research strategy (the Roadmap), which defines our priorities. Recently it has been found that the human gut microbiome has the capacity to influence how our genes code for a variety of Alzheimer’s-related phenomena, including brain inflammation. Accordingly, in 2016 we funded Sam Sisodia of the University of Chicago to investigate the link between the gut microbiome and Alzheimer’s disease; that investigation is continuing. Last year, Sam published an important paper showing that the gut microbiome controls plaque formation in AD mouse models.

Fifth: Leadership in Foundation Management (Venture Philanthropy). Many of the CureAlz founders are former venture capitalists. The founders and directors have, over time, personally contributed more than \$28 million to the foundation. The founders and directors pay all operating expenses of the foundation, so that 100 percent of any contribution from our generous donors goes directly into research. We want a cure or preventive measures identified as soon as possible, and we manage CureAlz with this goal. The management processes we have set up, which we call “venture philanthropy,” distinguish us as risk takers, fast decision makers and strategists. We proactively recruit the best researchers, fund them with no bureaucracy, challenge them for high-risk/high-reward projects and insist they collaborate. This venture philanthropy approach is one of the reasons for our significant momentum and successes, which have resulted in our being awarded—for the last five consecutive years—a four-star rating by Charity Navigator, including this last year under its new rating system a prestigious Perfect 100 Percent score, given to only 50 of 8,000 charities in the United States.

GOING FORWARD

While we are proud of these leadership roles we have achieved, we always are seeking to improve, and to make our research grants more impactful. To that end, during 2016 we conducted a detailed strategic review process, performed by a third-party strategy consultant. One of the recommendations adopted by the board from that process was to fund larger initiatives, involving multi-institutional consortia, to more deeply engage our leading researchers to holistically address strategic research themes. In that vein, we have formed our first consortium, CIRCUITS, a collaboration of nine research leaders from eight institutions, to use cutting-edge functional genomics, including epigenetic analysis and computational biostatistical tools, to model cellular pathways and gene interactions too complex to have been captured by past genetic analysis of protein production alone.

The dataset of AD-relevant genes, variants, cell types and regulators produced will be made available to the entire field, providing a powerful new data-driven model to accelerate identification of potential points for therapeutic intervention. A second consortium focused on the APOE gene is in the process of being formed, as is a third, which is focused on the role of the brain's immune response in Alzheimer's disease. At the same time, we will continue to fund smaller "high-risk" and "proof-of-concept" grants, which have been such productive sources of our progress to date.

As we enter 2017, we acknowledge that Alzheimer's disease increasingly has been in the news. On the positive side, awareness of AD in the public continues to increase, and research funding was a presidential campaign issue. On the negative side, several recent pharmaceutical-driven AD trials failed, and the initial 2017 U.S. budget proposal calls for cuts in National Institutes of Health research funding. All of this makes our focus on investigating the fundamental causes of Alzheimer's disease even more important. We must have a comprehensive understanding of this disease to develop the successful therapies of the future. In the nearer term, we have two potential therapies entering clinical trials in 2017. One is a gamma secretase modulator, which has the potential of being a "statin" for AD. The second, Amylyx, is a medicine developed to protect neurons from cell damage, and is potentially useful for both ALS and AD.

All of the above would not have been possible without your generous support. We are forever grateful to each and every one of our donors, to our wonderful researchers—who are more excited with their progress than ever—and to our dedicated staff. We will not rest until a set of effective therapies rids this planet of this dreaded disease; with your help, we will succeed.

Very Best Wishes,

Jeffrey L. Morby
Henry F. McCance
Co-Chairmen and Co-Founders



We have formed our first consortium, CIRCUITS, a collaboration of nine research leaders from eight institutions, to use cutting-edge functional genomics, including epigenetic analysis and computational biostatistical tools, to model cellular pathways and gene interactions too complex to have been captured by past genetic analysis of protein production alone.

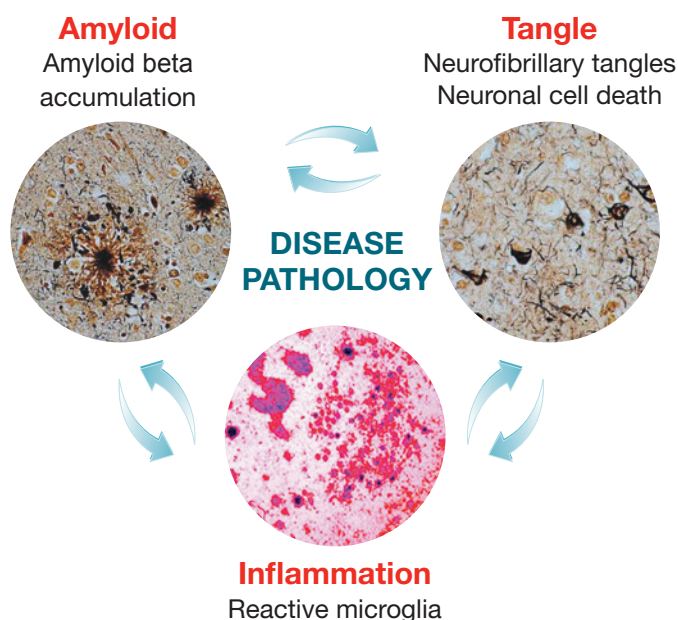
OUR RESEARCH IS MAKING A DIFFERENCE

Cure Alzheimer's Fund has a focused plan to end Alzheimer's disease, and we make significant strides toward that goal each year. Our research has contributed to a more thorough understanding of how Alzheimer's pathology progresses from the earliest to latest stages of the disease, including the identification of key genes and the functions of these genes.

Alzheimer's Disease Model

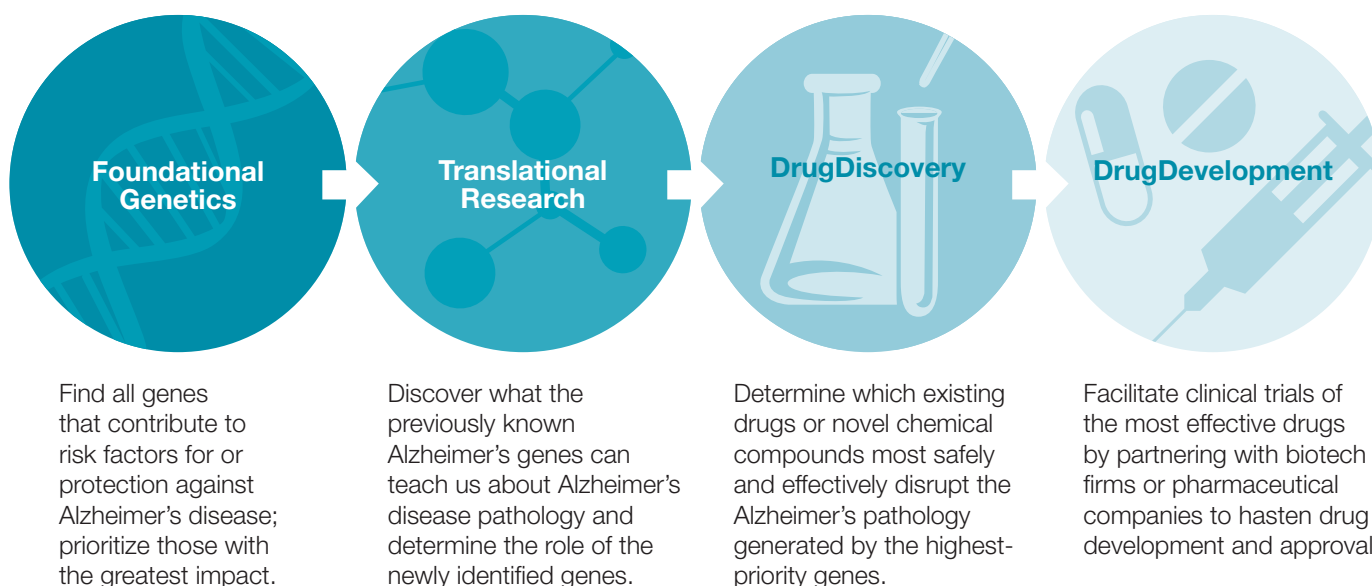
This model of Alzheimer's disease allows us to identify three basic strategies for intervention in the process:

1. An early-stage intervention inhibiting the production of the amyloid beta protein (the primary component of plaques characteristic of Alzheimer's disease), and/or clearing it from the brain after it forms.
2. An early- to mid-stage intervention that would inhibit the formation of tau tangles (proteins abundant in the central nervous system that have become defective and twisted into microscopic strands) and protect neurons from undue stress.
3. A late-stage intervention that would fight inflammation, and thus slow down or even stop the disease process.



Research Roadmap

By addressing Alzheimer's at its origins and finding the major causes of the disease, we are accelerating developments that may lead to effective therapies. The projects we fund are based on this research roadmap, which we think will be the quickest way to a cure.



Nine Areas of Research Focus


The four roadmap categories focus our funding and have helped us make significant progress in our understanding of the disease. This growing understanding has allowed us to further delineate our roadmap into the following areas of focus:




Genes to Therapies™/ Drug Screening
Finding mechanisms of action by which Alzheimer's genes affect risk and identifying targets for potential drug intervention




Identification and Early Detection
Improving our methods of detecting Alzheimer's in its early stages, opening the doors for treatments that stop the disease before symptoms begin or progress




Innate Immunity
Understanding the role of the brain's immune response—including inflammation—in the development and progression of Alzheimer's disease




Microbiome
Understanding the ways in which microbes living in the body may affect brain health




Pathological Pathways and Systems
Researching the underlying causes and mechanisms of Alzheimer's, such as amyloid beta accumulation, tau tangle propagation and inflammation



Stem Cell Models
Using stem cells to create highly accurate models of the Alzheimer's brain in the lab, in order to observe disease pathology and test possible therapies



Therapeutic Strategies
Identifying, testing and perfecting small molecules that could function as therapies for Alzheimer's disease



Whole Genome Sequencing and Epigenetics
Assessing the entire human genome, including intergenic DNA, for variants that influence risk for Alzheimer's



Individual Projects (Other)
Studying Alzheimer's disease using compelling methods or perspectives not covered by our other Areas of Focus

By funding the most promising research in each of these categories, we will build an in-depth and multifaceted picture of Alzheimer's disease. Approaching the disease from these perspectives will allow convergence on the ultimate goal—a **cure**.

RESEARCH PROJECTS

Cure Alzheimer's Fund distributed **\$13.5 million to support 56 research projects** across our focus areas, an all-time high that allowed us to fund even more of the most innovative research in 2016.

Visit curealz.org/research/funded-research to read about all of our current research projects.

Project/Researcher	Distribution Amount
Genes to Therapies™/Drug Screening	
A 3-D Human Neural Cell Culture System for Studying Neuron-Microglia Interaction in Alzheimer's Disease Hansang Cho, Ph.D., University of North Carolina at Charlotte	\$150,000
Role of Blood-Brain Barrier Function in Alzheimer's Disease Pathogenesis Investigated Using a 3-D Microfluidic Platform Se Hoon Choi, Ph.D., Massachusetts General Hospital, and Roger D. Kamm, Ph.D., Massachusetts Institute of Technology	\$293,844
The Biological Impact of TREM Locus Mutations in Alzheimer's Disease Marco Colonna, M.D., and David Holtzman, M.D., Washington University School of Medicine	\$250,000
Investigating the Mechanism of Entorhinal Cortex Hypermetabolism in APOE4 Targeted Replacement Mice Karen Duff, Ph.D., and Tal Nuriel, Ph.D., Columbia University	\$150,000
Discovery of CK1 Activators for Inducing the Autophagic Degradation of Amyloid Precursor Protein (APP) Beta-CTF Paul Greengard, Ph.D., The Rockefeller University	\$450,000
Microglial Core/CD33 and Alzheimer's Disease: From Biology to Therapy Ana Griciuc, Ph.D., and Rudolph Tanzi, Ph.D., Massachusetts General Hospital	\$400,000
Therapeutic Modulation of TREM2 Activity Christian Haass, Ph.D., DZNE Munich	\$150,000
Characterization of Certain Human APOE Targeted Gene Replacement Mice David Holtzman, M.D., and Jason Ulrich, Ph.D., Washington University School of Medicine	\$49,600
3-D Neural Core/High-Throughput Drug Screening for Alzheimer's Disease Using 3-D Human Neural Culture Systems Doo Yeon Kim, Ph.D., Massachusetts General Hospital	\$400,000
Extracellular Vesicle-Based Targeting of CD33-Mediated Pathology for Alzheimer's Disease Therapy Casey Maguire, Ph.D., Massachusetts General Hospital	\$150,000
Alzheimer's Disease-Associated Mutations in Protein Kinase C Alexandra Newton, Ph.D., University of California, San Diego	\$250,000
In Vitro and In Vivo Analysis of Amyloid Precursor Protein (APP) Variants Sangram S. Sisodia, Ph.D., University of Chicago	\$250,000
Development and Breeding of Transgenic Mice for Genes to Therapies™ Projects Taconic Biosciences	\$1,026,131
Alzheimer's Genome Project™ Rudolph Tanzi, Ph.D., Massachusetts General Hospital	\$1,500,000
BIN1 in Alzheimer's Disease Gopal Thinakaran, Ph.D., University of Chicago	\$150,000
Studying the Functional Consequences of Alzheimer's Disease Risk Variants in the CLU and ABCA7 Genes Using Both Human and Mouse Models Li-Huei Tsai, Ph.D., Massachusetts Institute of Technology	\$250,000
Genes to Therapies™ (G2T) Centralized Research Core Oversight Wilma Wasco, Ph.D., Massachusetts General Hospital	\$173,750
Use of High-Content Drug Screening and Systems Biology Modeling on a Novel 3-D Cell Model to Repurpose Known Drugs for Alzheimer's Disease Stephen Wong, Ph.D., Houston Methodist Research Institute	\$150,000
Alzheimer's Disease Drug Discovery in 3-D Weiming Xia, Ph.D., Boston University	\$150,000
SORLA Attenuates Amyloid Beta Toxicity Through Interactions with EphA4 Huaxi Xu, Ph.D., Sanford Burnham Prebys Medical Discovery Institute	\$150,000
PICALM Gene Therapy and Drug Screening for Amyloid Beta Clearance Berislav Zlokovic, M.D., Ph.D., University of Southern California	\$225,000
Innate Immunity	
Impact of Inflammasome Deactivation on Alzheimer's Disease Vishwa Deep Dixit, D.V.M., Ph.D., Yale School of Medicine	\$75,000
Amyloid Beta Expression Protects the Brain from Herpes Simplex Virus Robert D. Moir, Ph.D., Massachusetts General Hospital	\$175,000
Targeting Beneficial Innate Immunity in Alzheimer's by Interleukin-1 Receptor-Associated Kinase M (IRAK-M) Deletion Terrence Town, Ph.D., University of Southern California	\$150,000
Microbiome	
Mechanisms by Which the Gut Microbiome Influences Amyloid Deposition and Neuroinflammation in Mouse Models of Alzheimer's Disease Sangram S. Sisodia, Ph.D., University of Chicago	\$250,000
Pathological Pathways and Systems	
Understanding Reactive Astrocytes and Their Roles in Alzheimer's Disease Ben Barres, M.D., Ph.D., Stanford University	\$150,000
Intersection of Microglial Transcriptomes to Identify Key Alzheimer's Pathways of Brain Phagocytes Sam Gandy, M.D., Ph.D., Icahn School of Medicine at Mount Sinai Hospital	\$150,000

Pathological Pathways and Systems (continued)	
Uncovering Determinants of Neuronal Vulnerability in Alzheimer's Disease Paul Greengard, Ph.D., The Rockefeller University	\$250,000
Modeling DNA Methylation Changes in Alzheimer's Disease Using Induced Human Pluripotent Stem (iPS) Cells Rudolf Jaenisch, M.D., Whitehead Institute	\$150,000
The Role of Meningeal Lymphatics in Cleansing the Brain: Implications for Alzheimer's Disease Jonathan Kipnis, Ph.D., University of Virginia	\$150,000
Cell Cycle Re-entry in 3-D Human Neuron Cultures John S. Lazo, Ph.D., and George S. Bloom, Ph.D., University of Virginia	\$100,000
Regulation of Microglial Lysosome Acidification Frederick R. Maxfield, Ph.D., Weill Cornell Medical College	\$120,006
Will Restoration of Normal Glymphatic Function Slow Progression of Cognitive Decline and Amyloid Plaques in a Murine Alzheimer Model? Maiken Nedergaard, M.D., D.M.Sc., University of Rochester	\$150,000
Early Role of Microglia in Synapse Loss in Alzheimer's Disease Beth Stevens, Ph.D., Boston Children's Hospital	\$150,000
Systemic Inflammatory Networks in Alzheimer's Disease Filip Swirski, Ph.D., and Matthias Nahrendorf, M.D., Ph.D., Massachusetts General Hospital	\$200,000
Role of Neurexins in Alzheimer's Disease Pathophysiology Rudolph Tanzi, Ph.D., Massachusetts General Hospital	\$150,000
Rejuvenation of Microglia in Brain Aging and Neurodegeneration Tony Wyss-Coray, Ph.D., Stanford University	\$150,000
Therapeutic Strategies	
Development of Novel Amyloid Precursor Protein Dimerization Inhibitors That Lower Amyloid Beta Levels Carmela R. Abraham, Ph.D., Boston University	\$129,373
Nanobodies to Cross the Blood-Brain Barrier Bart De Strooper, M.D., Ph.D., and Maarten Dewilde, Ph.D., VIB-KU Leuven	\$150,000
Identification of a Protective Human Immune Response for Alzheimer's Disease Charles Glabe, Ph.D., University of California, Irvine	\$125,000
Activation of the 26S Proteasome for the Treatment of Alzheimer's Disease Alfred L. Goldberg, Ph.D., Harvard Medical School	\$150,000
The APOE Mimetic Therapeutic Peptide CN-105 Attenuates Alzheimer's Disease Pathology and Improves Functional Outcomes in a Murine Model of Alzheimer's Disease Daniel Laskowitz, M.D., M.H.S., Duke University	\$112,662
A Combination of Anti-Amyloid Beta and Growth Factor Therapy for Alzheimer's Disease Mark H. Tuszynski, M.D., Ph.D., University of California, San Diego	\$125,000
Acceleration of U.S. Food and Drug Administration-Required Good Laboratory Practice Gene Toxicity Studies With Gamma Secretase Modulator BPN-15606 Steven Wagner, Ph.D., University of California, San Diego	\$144,450
Binding Site Characterization of a Novel Pyridazine-Derived Class of Gamma Secretase Modulators Steven Wagner, Ph.D., University of California, San Diego	\$194,950
Whole Genome Sequencing and Epigenetics	
CIRCUITS: Epigenetic Determinants of Human Cognitive Aging Lars Bertram, M.D., University of Lübeck	\$199,600
CIRCUITS: Utilizing Functional Maps to Prioritize Therapeutic Targets in Alzheimer's Disease Winston Hide, Ph.D., The University of Sheffield	\$225,308
CIRCUITS: Induced Pluripotent Stem (iPS) Cells and the Human Brain Bradley T. Hyman, M.D., Ph.D., Massachusetts General Hospital	\$145,004
CIRCUITS: Whole Genome Characterization of DNA Methylation Changes in the Aged and Alzheimer's Disease Human Brain Rudolf Jaenisch, M.D., Whitehead Institute, and Joseph Ecker, Ph.D., Salk Institute	\$250,000
CIRCUITS: Production Center for Reference and Variation Gene-Regulatory Maps Manolis Kellis, Ph.D., Broad Institute, and Li-Huei Tsai, Ph.D., Massachusetts Institute of Technology	\$750,000
Analytical and Statistical Tools for Sequence Analysis for Alzheimer's Disease Christoph Lange, Ph.D., Harvard School of Public Health	\$250,000
CIRCUITS: Interpreting Alzheimer's Disease-Associated Genetic Variation at Enhancer Regions Andreas R. Pfennig, Ph.D., Carnegie Mellon University	\$189,610
Search for Female-Specific Genetic Factors Contributing to Risk for Alzheimer's Disease Rudolph Tanzi, Ph.D., Massachusetts General Hospital	\$375,000
CIRCUITS: Functional Analysis of Alzheimer's Disease Risk Genes Using Human-Induced Pluripotent Stem (iPS) Cells Li-Huei Tsai, Ph.D., Massachusetts Institute of Technology, and Manolis Kellis, Ph.D., Broad Institute	\$400,000
Individual Projects (Other)	
Pathway Crosstalks Associated With Sex and Risk for Alzheimer's Disease Murali Doraiswamy, M.D., Duke University	\$151,655
Modeling Neuronal Aging in Specific Subtypes of Human Neurons by MicroRNA-Mediated Neuronal Reprogramming Andrew S. Yoo, Ph.D., Washington University School of Medicine	\$150,000
Total Funded	\$13,455,943

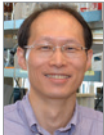
Genes to Therapies™

Finding mechanisms of action by which Alzheimer's genes affect risk and identifying targets for potential drug intervention



A 3-D Human Neural Cell Culture System for Studying Neuron-Microglia Interaction in Alzheimer's Disease

\$150,000



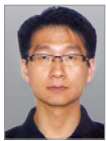
Hansang Cho, Ph.D.

Assistant Professor, Department of Mechanical Engineering and Engineering Science, and Department of Biological Sciences, University of North Carolina at Charlotte

In this proposal, we aim to dramatically improve the current microglial chemotactic model into a hybrid brain model that recapitulates pathological cascades of Alzheimer's disease, including amyloid beta deposits, microglial recruitment/clearance of amyloid beta, neurofibrillary tangles and possibly neuronal death. To do this, we have been collaborating closely with Drs. Doo Yeon Kim and Rudolph E. Tanzi (Massachusetts General Hospital) to combine their novel "three-dimensional Alzheimer's in a Dish" model with our microglial chemotactic model. Drs. Kim and Tanzi's 3-D Alzheimer's disease human neurospheroid successfully recapitulates amyloid beta and tau pathology in their system. By combining our microglia model and their 3-D Alzheimer's disease neuron model, we can analyze the impact of human microglial cells on amyloid beta and tau pathology. Furthermore, we can assess a hypothesis that human microglial cells are recruited to amyloid beta deposits in a 3-D human neural culture model, and therefore contribute to Alzheimer's disease pathogenesis.

Role of Blood-Brain Barrier Function in Alzheimer's Disease Pathogenesis Investigated Using a 3-D Microfluidic Platform

\$293,844



Se Hoon Choi, Ph.D.

Assistant Professor of Neurology, Massachusetts General Hospital

Roger D. Kamm, Ph.D.

Cecil and Ida Green Distinguished Professor of Biological and Mechanical Engineering, Massachusetts Institute of Technology

Alzheimer's disease is the most common form of dementia among older people. The blood-brain barrier (BBB) is a highly selective permeable barrier that separates the brain from circulating blood. It is formed by brain endothelial cells and prevents harmful materials from the blood from entering the brain. Evidence identifying BBB dysfunction in Alzheimer's disease or patients at risk (i.e., those with mild cognitive impairment) continues to accumulate. In cerebral amyloid angiopathy (CAA), which is a unique form of Alzheimer's disease, a toxic molecule generated in Alzheimer's disease brain, called amyloid beta, deposits within the blood vessels of the brain. The deposition of amyloid beta leads to BBB impairment, including microhemorrhages, which contribute to Alzheimer's disease pathogenesis in CAA. However, little is known about the role of the BBB's function in Alzheimer's disease pathogenesis. We propose to elucidate the function of BBB in Alzheimer's disease progression and investigate whether toxic molecules generated in the Alzheimer's disease brain cause BBB impairment.

The Biological Impact of TREM Locus Mutations in Alzheimer's Disease

\$250,000



Marco Colonna, M.D.

Professor, Pathology and Immunology, Professor of Medicine, Washington University School of Medicine

David Holtzman, M.D.

Andrew B. and Gretchen P. Jones Professor and Chairman of Neurology; Charlotte and Paul Hagemann Professor of Neurology, Molecular Biology and Pharmacology; Associate Director of the Alzheimer's Disease Research Center, Washington University School of Medicine; Scientific Director of the Hope Center for Neurological Disorders

Whole genome sequencing has identified certain polymorphisms affecting genes encoding triggering receptors expressed on myeloid cells (TREMs) with increased risk of nonfamilial (sporadic) Alzheimer's disease. TREM signaling is known to be important in the innate immune response, particularly in the inflammatory response. However, the relationship between the function of TREM receptors and Alzheimer's disease pathology largely is unresolved. This project will investigate the relevant polymorphisms to determine how they facilitate Alzheimer's disease and how they might be targeted to improve brain immune function.



Investigating the Mechanism of Entorhinal Cortex Hypermetabolism in APOE4 Targeted Replacement Mice

\$150,000



Karen Duff, Ph.D.

Professor of Pathology and Cell Biology, Taub Institute, Columbia University



Tal Nuriel, Ph.D.

Postdoctoral Research Fellow, Columbia University Medical Center

Carriers of the APOE4 gene are at significantly increased risk for developing Alzheimer's disease. We have discovered that aging mice that express the APOE4 gene show increased activity in a region of the brain that is implicated in the development of Alzheimer's disease, the entorhinal cortex, and we think this increased activity may be an important link between APOE4 and Alzheimer's disease pathology. In order to understand the cause of this increased brain activity, we will utilize sophisticated techniques to measure important genes and proteins known to increase brain activity. We anticipate the results of this study will yield significant insights into the biology of APOE4 and Alzheimer's disease, as well as how Alzheimer's disease may be treated or prevented in these individuals.

Discovery of CK1 Activators for Inducing the Autophagic Degradation of Amyloid Precursor Protein (APP) Beta-CTF

\$450,000



Paul Greengard, Ph.D.

Head of the Laboratory of Molecular and Cellular Neuroscience, Vincent Astor Professor, The Rockefeller University

Nobel Prize in Physiology or Medicine, 2000

Alzheimer's disease is a neurodegenerative disorder that affects more than 5 million people in the United States. One of the hallmarks of Alzheimer's disease is the accumulation of amyloid plaques in the brain of patients. The amyloid plaque is composed of the amyloid beta peptide, which originates from an amyloid precursor protein (APP). Multiple lines of evidence suggest that a defective clearance mechanism is involved in the pathogenesis of Alzheimer's disease. Our laboratory has discovered a novel molecular pathway regulating protein clearance, which represents an attractive therapeutic target for developing drugs for Alzheimer's disease. This project represents a direct effort to search for small molecule compounds that can restore protein clearance.

Microglial Core/CD33 and Alzheimer's Disease: From Biology to Therapy

\$400,000



Ana Griciuc, Ph.D.

Instructor in Neurology, Harvard Medical School; Assistant in Neuroscience, Massachusetts General Hospital

Rudolph Tanzi, Ph.D.

Joseph P. and Rose F. Kennedy Professor of Neurology, Harvard Medical School; Director of the Genetics and Aging Research Unit and Vice Chair of Neurology, Massachusetts General Hospital

Our current inability to prevent or delay Alzheimer's disease and the expected increase in the prevalence of Alzheimer's disease are predicted to give rise to a global Alzheimer's disease pandemic. We recently have identified a novel pathway for amyloid beta clearance in the aging brain that is highly relevant to Alzheimer's disease pathogenesis. In a very large, family-based, genome-wide association study, (GWAS) we identified the CD33 gene as a novel late-onset Alzheimer's disease risk factor. CD33 encodes a transmembrane sialic acid-binding immunoglobulin-like lectin that regulates innate immunity.

We found that CD33 is specifically expressed in microglial cells and exhibits an increased expression in Alzheimer's disease. Using microglial cell cultures, we showed that CD33 inhibits uptake and clearance of amyloid beta 42, a process that requires the sialic acid-binding domain of CD33. CD33 knockout led to a marked reduction in insoluble amyloid beta 42 levels and amyloid plaque burden in mouse models of Alzheimer's disease. We also found that CD33 knockout in Alzheimer's disease mice results in skewing of adult microglia from the M1 (pro-inflammatory, neurotoxic) to the M2 (pro-phagocytic, neuroprotective) activation phenotype. Thus, CD33 activity in microglial cells promotes amyloid beta pathology, and CD33 has emerged as a novel target for drug development in Alzheimer's disease.

Here, we propose to inhibit CD33 activity 1) to induce uptake and clearance of amyloid beta 42, and 2) to enhance skewing of microglia from the pro-inflammatory M1 toward the pro-phagocytic M2 activation state. We will identify and validate effective CD33 inhibitors by performing an unbiased high-throughput screen of 1,280 small molecules in microglial cells. These compounds are from Prestwick Chemical and have been approved by the U.S. Food and Drug Administration and other agencies. We also will develop CD33 inhibitors by screening CD33-specific antibodies for their ability to inhibit CD33 function in microglial cell-based assays. Successful compounds and CD33-specific antibodies that inhibit CD33 activity in amyloid beta clearance and M1/M2 cytokine release assays will be further tested in mice. These studies might result in a novel and powerful therapeutic approach for Alzheimer's disease.

Therapeutic Modulation of TREM2 Activity

\$150,000



Christian Haass, Ph.D.

Professor, Department of Metabolic Biochemistry, Ludwig Maximilians University of Munich; Speaker, German Center for Neurodegenerative Diseases (DZNE)

There is strong evidence that inflammation occurs in different stages of Alzheimer's disease. Understanding this process can help us to design new therapeutic approaches. TREM2 is a protein directly related to the inflammation process that occurs in the brains of patients with Alzheimer's disease. Mutations in this protein increase the risk of developing Alzheimer's disease up to threefold. A fragment of this protein, namely soluble TREM2 (sTREM2), increases at very early stages of Alzheimer's disease, and this increase occurs in parallel to an increase of biomarkers for neuronal cell death. We have evidence that increased sTREM2 reflects a protective response; however, this could not be maintained in later stages of Alzheimer's disease.

Characterization of Certain Human APOE Targeted Gene Replacement Mice

\$49,600



David Holtzman, M.D.

Andrew B. and Gretchen P. Jones Professor and Chairman of Neurology; Charlotte and Paul Hagemann Professor of Neurology, Molecular Biology and Pharmacology; Associate Director of the Alzheimer's Disease Research Center, Washington University School of Medicine; Scientific Director of the Hope Center for Neurological Disorders

Jason D. Ulrich, Ph.D.

Assistant Professor of Neurology, Washington University School of Medicine

APOE4 is the strongest identified genetic risk factor for late-onset Alzheimer's disease. Strong evidence from amyloid beta-deposition mouse models and humans indicate that APOE4 influences the metabolism of amyloid beta within the brain, promoting amyloid beta plaque pathology. The precise mechanism(s) by which APOE isoforms influence amyloid beta are not completely clear, although numerous *in vivo* and *in vitro* studies suggest that APOE4 slows the clearance of amyloid beta from the brain and facilitates the aggregation of monomeric amyloid beta. There are a variety of other effects that APOE may have in both the normal brain as well as in the setting of Alzheimer's disease and other neurological diseases. Studies on the effects of APOE4 on amyloid pathology in mouse models have relied on targeted replacement mice, where exons 2-4 of murine APOE were replaced with exons 2-4 of human APOE isoforms and a neomycin selection cassette. Recently, Cure Alzheimer's Fund supported the generation of new human APOE targeted replacement mice containing floxed alleles to support the study of cell-type specific roles for human APOE or temporal control of APOE expression. These next-generation human APOE replacement mice will accelerate research into the mechanism by which APOE4 influences the onset and progression of Alzheimer's disease, and by which APOE2 is protective.

3-D Neural Core/High-Throughput Drug Screening for Alzheimer's Disease Using 3-D Human Neural Culture Systems

\$400,000



Doo Yeon Kim, Ph.D.

Assistant Professor of Neurology, Harvard Medical School;
Genetics and Aging Research Unit, Massachusetts General Hospital

The “amyloid beta cascade hypothesis” of Alzheimer's disease has provided a major framework for Alzheimer's disease drug discovery and has led to many current clinical trials. However, to date, no single *in vitro* or *in vivo* Alzheimer's disease model has been able to recapitulate the presumed patient pathophysiology: amyloid beta deposition directly leading to tangles and neurodegeneration. Recently, we created a novel three-dimensional (3-D) human neural cell culture model of Alzheimer's disease using genetically engineered human neural stem cells. Using this unique model, we showed for the first time that expression of amyloid precursor protein (APP) and presenilin 1 (PSEN1) with familial Alzheimer's disease mutations is sufficient to induce extracellular amyloid beta deposits and robust tauopathy, including hyperphosphorylated tau and detergent-resistant, silver-positive neurofibrillary tangles. (Choi et al., 2014). This human 3-D culture model has great potential to innovate and accelerate the current Alzheimer's disease drug screening process. We now propose to use high-throughput drug screening in combination with our 3-D human cellular Alzheimer's disease model to identify and characterize novel Alzheimer's disease drugs and drug targets that can reduce both amyloid beta and tau pathologies, which is not feasible using current Alzheimer's disease mouse models.

In Aim 1, we will develop a high-throughput screening (HTS) system based on 3-D human cellular Alzheimer's disease models (3-D AD-HTS). In Aim 2, we will carry out 3-D AD-HTS using U.S. Food and Drug Administration (FDA)-approved drug libraries and validate the primary hits that reduce amyloid beta and/or tau pathologies and finally, in Aim 3, we will explore whether the validated candidate drugs rescue the neuronal injuries and functional deficits in the 3-D Alzheimer's disease culture model. The overarching goals of this study are to 1) establish a 3-D HTS Alzheimer's disease drug screening system based on human 3-D neural cell culture models; 2) find potential Alzheimer's disease drug candidates among the FDA-approved drugs (drug repurposing); and 3) identify novel cellular pathways that can regulate both amyloid beta and tau pathologies. Since no current Alzheimer's disease mouse model of amyloid beta deposition leads to tangles and neurodegeneration, which are both critical aspects of the disease, the human neural cell culture model could serve as a novel, crucial drug discovery platform for Alzheimer's disease.

Extracellular Vesicle-Based Targeting of CD33-Mediated Pathology for Alzheimer's Disease Therapy

\$150,000

**Casey Maguire, Ph.D.**

Assistant Professor of Neurology, Harvard Medical School
Principal Investigator, Massachusetts General Hospital

Alzheimer's disease is a devastating disease for patient and family alike. Unfortunately, there is no effective treatment and conventional, drug-based therapies have failed. Our lab has developed a therapeutic virus vector that the body's immune defenses will tolerate and that efficiently delivers nucleic acid-based material into cells. In this case, the material is a gene therapy specifically tailored to manipulate the expression of CD33, a gene seen in mouse models to slow amyloid beta clearance and thus contribute to Alzheimer's disease pathology.

Alzheimer's Disease-Associated Mutations in Protein Kinase C

\$250,000

**Alexandra Newton, Ph.D.**

Professor of Pharmacology, University of California, San Diego

This proposal addresses whether a key protein that is turned off in cancer, a disease characterized by uncontrolled cellular growth and survival, is excessively active in Alzheimer's disease, a degenerative disease. This protein, called protein kinase C, is an information processor, or "signal transducer," that regulates cellular activities. Its activity needs to be precisely balanced to maintain normal cellular function. Reduced function promotes cell survival, a hallmark of cancer. Analysis of genetic mutations identified in the Genes to Therapies™ program by Rudy Tanzi reveals that mutations found in some patients with Alzheimer's disease enhance the function of protein kinase C. This project examines whether enhanced signaling by protein kinase C generally is associated with the pathology of Alzheimer's disease, identifying protein kinase C as a promising therapeutic target.

In Vitro and In Vivo Analysis of Amyloid Precursor Protein (APP) Variants

\$250,000



Sangram S. Sisodia, Ph.D.

Thomas Reynolds Sr. Family Professor of Neurosciences, Director of the Center for Molecular Neurobiology, University of Chicago

It is widely accepted that amyloid beta peptides, the principal component of senile plaques, play a causative role in the pathogenesis of Alzheimer's disease. Amyloid beta is derived from larger amyloid beta precursor proteins (APP). Early-onset, familial AD (FAD) is caused by inheritance of mutations in genes encoding APP or presenilin (PS1 or PS2) variants. Mutations in APP lead to the production of elevated levels of amyloid beta or affect the ratio of amyloid beta 42/40 peptides. Importantly, all of the known APP mutations reside proximal to or within the amyloid beta peptide sequence. Very recently, Rudy Tanzi and colleagues (unpublished) have identified additional mutations in APP that co-segregate with two early-onset FAD pedigrees, but surprisingly, these mutations are quite a distance from the amyloid beta sequence. The mechanism(s) by which these novel variants influence amyloid beta production and/or deposition is not known, but our current proposal seeks to clarify these important issues in cell culture and transgenic mouse models.

Development and Breeding of Transgenic Mice for Genes to Therapies™ Projects

\$1,026,131



Taconic Biosciences

Taconic Biosciences, a global provider of genetically modified mouse models and associated services, is providing customized mouse models (transgenic, conventional/conditional knock out, conventional/conditional knock in) for each specific gene and type of mutation that will be studied in the Genes to Therapies™ project. Taconic also is providing breeding services for these projects.

Alzheimer's Genome Project™

\$1,500,000



Rudolph Tanzi, Ph.D.

Joseph P. and Rose F. Kennedy Professor of Neurology, Harvard Medical School;
Director of the Genetics and Aging Research Unit and Vice Chair of Neurology,
Massachusetts General Hospital

Over this past funding cycle, we reported the results of our imputed genome-wide association studies (GWAS) on six GWAS datasets (two large family-based samples—NCRAD and the NIA—and four case-control samples—TGEN2, ADNI, GenADA and NIA-LOAD, together with four additional quantitative trait-imputed GWAS datasets (cerebrospinal fluid (CSF) biomarkers from Sweden; Bonn, Germany, CSF biomarkers data; Massachusetts General Hospital Alzheimer's disease brain pathological measures; and brain amyloid imaging data using Pittsburgh compound B (PiB) scans from Australia); overall, more than 10,000 samples. In this study, we performed a systematic family-based genome-wide association and meta-analysis on close to 15 million imputed variants from three large collections of Alzheimer's disease families (approximately 3,500 subjects from 1,070 families).

In the coming funding cycle, we will utilize our comprehensive 1.5 petabyte database of whole genome (WGS) and whole exome (WES) sequences from our own and all available Alzheimer's disease family-based and case-control samples to search for Alzheimer's disease-linked functional genomic variants in the five Alzheimer's disease gene candidates: OSBPL6, PTPRG, PDCL3, CDKAL1 and SLCA25A24, which we identified over the past funding cycle as part of the Alzheimer's Genome Project™, as well as in Alzheimer's disease gene candidates identified in a genome-wide agnostic WGS screen. The primary WGS and WES samples that we are analyzing include the National Institute of Mental Health (NIMH) Alzheimer's disease family sample WGS (N=1,440 siblings from 450 families, which we specifically sequenced by WGS).

In the upcoming funding period, we will begin employing a novel analytical method of genetic association analysis with extended families, which we have developed based on adjusting for heterogeneous ascertainment bias. In family-based association analysis, each family is typically ascertained from a single proband, which renders the effects of ascertainment bias heterogeneous among family members. This is contrary to the situation in case-control studies, and may introduce sample or ascertainment bias. Statistical efficiency is affected by ascertainment bias, and careful adjustment can lead to substantial improvements in statistical power. However, genetic association analysis often has been conducted using family-based designs, without addressing the fact that each proband in a family has had a great influence on the probability for each family member to be affected.

Over the past funding period, we developed a powerful and efficient new statistic for genetic association analysis that takes into consideration the heterogeneity of ascertainment bias among family members, under the assumption that both prevalence and heritability of disease are available. With extensive simulation studies, we have shown that the proposed method performed better than the existing methods, particularly for diseases, like Alzheimer's disease, with large heritability. We now will use this method to carry out an entirely novel GWAS on the NIMH sample using the both the Affymetrix 6.0 and Illumina 2.5M single-nucleotide polymorphism (SNP) datasets. Preliminary studies of the Affymetrix 6.0 data have implicated four genomic regions harboring novel Alzheimer's disease candidate loci (Won et al, *submitted*). We will report more specific data on these findings in the next progress report.

BIN1 in Alzheimer's Disease

\$150,000

**Gopal Thinakaran, Ph.D.**

Professor, Neurology and Neurobiology, University of Chicago

The goal of this proposal is to investigate how one of the recently identified late-onset Alzheimer's disease risk genes, namely BIN1, contributes to neuropathology. BIN1 is an adaptor protein that regulates membrane dynamics in a variety of cellular contexts. Only limited information is available on BIN1 expression and function in the brain. As such, there is much to be learned about the precise biological and mechanistic connection between BIN1 and Alzheimer's disease. We propose to use an integrated approach employing cultured cells and BIN1 transgenic mice to test specific hypotheses regarding BIN1 function and dysfunction in Alzheimer's disease.

Studying the Functional Consequences of Alzheimer's Disease Risk Variants in the CLU and ABCA7 Genes Using Both Human and Mouse Models

\$250,000

**Li-Huei Tsai, Ph.D.**

Director of the Picower Institute for Learning and Memory; Picower Professor of Neuroscience, Massachusetts Institute of Technology; Senior Associate Member, Broad Institute of MIT and Harvard

The vast majority of people with Alzheimer's disease suffer from the sporadic (late-onset) form, the causes of which remain completely unknown. From studies involving thousands of people, researchers have identified a number of genetic variants that may increase one's risk for sporadic Alzheimer's disease (AD). However, little is understood regarding how carrying these variants impacts one's sporadic Alzheimer's disease risk.

To better understand the roles of genetic variants in sporadic Alzheimer's disease, we are using the cutting-edge genome editing technology of CRISPR/Cas9 and induced pluripotent stem cells (iPSCs) to engineer human stem cells to have mutations for two of the genes conveying increased risk of sporadic Alzheimer's disease: ABCA7 and CLU. We have used these iPSCs to generate brain-specific cell types, including neural progenitor cells, neurons, astrocytes and microglia.

We have discovered that neurons carrying a mutation of ABCA7 produce such AD-like cellular hallmarks as elevated levels of soluble amyloid beta and such abnormal organelles as enlarged early endosomes. We further show that when this ABCA7 mutant is incorporated into an engineered tissue-organ model, the model displays AD pathologies seen in the brains of Alzheimer's patients, such as the accumulation of amyloid deposits.

These exciting discoveries, when integrated with our planned comprehensive analysis from the other cell types, including microglia and astrocytes, of the mutational effects of ABCA7 and CLU on AD pathologies and contribution of ABCA7 mutation to familial AD using the 5XFAD mouse model, will provide valuable insights into the molecular pathways by which these genetic risk variants contribute to sporadic AD.



Genes to Therapies™ (G2T) Centralized Research Core Oversight

\$173,750



Wilma Wasco, Ph.D.

Associate Professor of Neurology, Harvard Medical School; Associate Geneticist, Massachusetts General Hospital

Wilma Wasco, Ph.D., is responsible for the day-to-day organization of the Genes to Therapies™ (G2T) Centralized Research Core. She meets routinely with Dr. Rudy Tanzi as well as the members of the Steering Committee and Meg Smith (Cure Alzheimer's Fund) to outline and discuss progress with timelines and investigations, as well as reagent generation and budgets. She will be responsible for determining what reagents are available from investigators or commercial sources while investigators are being recruited. In addition, she is the point person for Taconic and all other commercial or academic sources that will be used to generate reagents, as well as for the investigators who have been and will be recruited to work on each gene. It is envisioned that this will require, at a minimum, weekly email and phone interactions with each commercial or academic source and investigator. Dr. Wasco will travel to appropriate scientific meetings and to meet with G2T investigators. If necessary, Dr. Wasco will be involved in any experimental work that is carried out within the Genetics and Aging Research Unit at Massachusetts General Hospital, which may include cell culture and reagent testing and confirmation. Dr. Wasco has longstanding expertise in Alzheimer's disease genetic studies—she played a significant role in the original discovery of the presenilin genes and is familiar with the techniques that will be used for the gene investigations—and has a history of administrative project management.

Use of High-Content Drug Screening and Systems Biology Modeling on a Novel 3-D Cell Model to Repurpose Known Drugs for Alzheimer's Disease

\$150,000



Stephen Wong, Ph.D.

John S. Dunn Sr. Presidential Distinguished Chair in Biomedical Engineering, Professor of Systems Medicine and Bioengineering, Houston Methodist Research Institute

Taking advantage of recent research progress, we propose to carry out a compound screen to find potential drugs to treat Alzheimer's disease. The screen will use a revolutionary 3-D stem cell model, recently developed by Drs. Rudy Tanzi and Doo Yeon Kim at Massachusetts General Hospital, which for the first time faithfully recapitulated major pathological hallmarks of Alzheimer's disease in a dish. The screen also will use the previously developed neuronal image processing software packages, which are able to automatically and accurately assess the cell phenotype to evaluate the compound effect. Finally, we specifically will screen compounds that are known to be bioactive, including the compounds that already are used as drugs in clinics. Such a “drug repositioning” strategy will greatly reduce the cost of drug development, enable faster time to market and quickly translate the scientific discovery to patient bedsides.

Alzheimer's Disease Drug Discovery in 3-D

\$150,000

**Weiming Xia, Ph.D.**

Acting Associate Director of Research, Bedford VA Medical Center;
Boston University Alzheimer's Disease Center

We propose to examine biological fluids from cells treated with individual drugs that have been approved previously by the U.S. Food and Drug Administration for the treatment of numerous diseases and disorders. We will determine whether these drugs reduce the levels of the toxic proteins known to cause Alzheimer's disease. Newly identified drugs or similar modified compounds will be developed as Alzheimer's therapeutics.

SORLA Attenuates Amyloid Beta Toxicity Through Interactions with EphA4

\$150,000

**Huaxi Xu, Ph.D.**

Jeanne and Gary Herberger Leadership Chair in Neuroscience; Professor and Director, Neuroscience Initiative; Professor, Degenerative Diseases Program; Sanford Burnham Prebys Medical Discovery Institute

SORLA protein is a genetic risk factor in Alzheimer's disease, but it is unclear how changes in the SORLA abundance can trigger the onset of Alzheimer's disease in the elderly. So far, studies have shown that SORLA can limit the amount of neurotoxic amyloid beta generated in the brain. However, since high levels of amyloid beta also are seen in aged individuals without symptoms of dementia or cognitive decline, neuroprotective mechanisms are likely in place to protect neurons from amyloid beta damage. We describe here that SORLA can limit the activation of a cell surface component, EphA4, that is activated in the presence of amyloid beta, and which can damage synaptic function in the brain. Our preliminary results indicate that SORLA overexpression can limit amyloid beta-dependent activation of EphA4 in response to normal EphA4 activators such as Ephrin ligand, and amyloid beta in cultured neurons. Our pilot experiments indicate that mouse models overexpressing SORLA are less vulnerable to amyloid beta injected into the mouse hippocampus. Our study here will confirm whether SORLA can limit toxic signals from EphA4 responding to amyloid beta, and whether molecular strategies to enhance SORLA/EphA4 interactions can further protect neurons from synaptic damage from amyloid beta. Together, this study may provide insight into a new pathway to protect neurons from amyloid beta damage, which may lead to strategies to improve cognition in Alzheimer's disease patients.



PICALM Gene Therapy and Drug Screening for Amyloid Beta Clearance

\$225,000



Berislav Zlokovic, M.D., Ph.D.

Director of the Zilkha Neurogenetic Institute; Mary Hayley and Selim Zilkha Chair in Alzheimer's Disease Research; Professor and Chair, Department of Physiology and Biophysics, Keck School of Medicine, University of Southern California

Our novel theory regarding PICALM, published last year in *Nature Neuroscience*, is that its genome-wide association study (GWAS)-linked impact lowering Alzheimer's disease risk is due to its role internalizing amyloid beta into brain endothelial cells and then out into the bloodstream, effectively increasing amyloid clearance across the blood-brain barrier. We hope to increase PICALM expression to improve amyloid clearance by using an adeno-associated virus (AAV) as a vehicle to deliver gene therapy to increase endothelial expression of PICALM or, alternatively, by using a drug already safety-approved by the U.S. Food and Drug Administration (FDA) retested and shown to have this effect. We will test both the gene therapy and the top drug hits *in vivo* for both amyloid load and rescue of behavioral deficits. Therapeutic strategies that upregulate PICALM expression in the overall brain vasculature could lead to rapid advancements in Alzheimer's disease treatment.

Innate Immunity

Understanding the role of the brain's immune response—including inflammation—in the development and progression of Alzheimer's disease



Impact of Inflammasome Deactivation on Alzheimer's Disease

\$75,000



Vishwa Deep Dixit, D.V.M., Ph.D.

Professor of Comparative Medicine and Immunobiology, Yale School of Medicine

This research proposal from the Dixit laboratory, which will be pursued in interdisciplinary collaboration with the lab of Dr. Rudy Tanzi at Massachusetts General Hospital, emanates from our original findings that NLRP3 inflammasome activation in microglia controls age-related inflammation in the central nervous system (CNS), and that CD33-dependent inhibition of amyloid beta uptake by microglia reduces interleukin 1 beta (IL-1beta) to protect against Alzheimer's disease. Inflammasome is a high molecular weight protein complex that assembles in the cytosol of microglia and myeloid-lineage cells upon encounter with "damage-associated molecular patterns" such as amyloids, lipotoxic fatty acids or extracellular adenosine triphosphate derived from necrotic cells. Upon assembly, this causes caspase-1 dependent release of pro-inflammatory cytokines IL-1beta, IL-18 and a special form of cell death called pyroptosis 1.

Studies from our lab have shown that the NLRP3 inflammasome controls development of inflammation-associated degenerative diseases during aging. Consistent with our data, independent studies also have demonstrated the increased activation of NLRP3 inflammasome in Alzheimer's disease in humans, and that genetic loss of NLRP3 protects against dementia in the amyloid precursor protein/presenilin 1 (APP/PS1) mouse model. Although it is established that inflammation plays a pivotal role in development of Alzheimer's disease, therapeutic approaches that impact specific innate immune mechanisms in the microglia remain to be identified. Interestingly, our preliminary results show that a subset of the elderly is protected from age-related inflammation and microglial activation despite the presence of amyloid beta plaques. This implies there must be endogenous protective mechanisms that maintain homeostasis in aging by preventing the sensing of aberrant amyloid beta deposits in microglia, thereby resulting in reduced inflammatory damage. Thus, the long-term goal of this proposal is to identify and harness anti-inflammasome regulators as therapeutic targets to prevent or treat.



Impact of Inflammasome Deactivation on Alzheimer's Disease (continued)

This proposal is based on our discovery that ketone metabolite BHB and NLRP3 inflammasome constitute an immunometabolic checkpoint of binary opposition — endogenous polar signals that work in concert to regulate the innate immune response. Ketone bodies, BHB and AcAc support mammalian survival during periods of starvation by serving as a source of ATP for the tricarboxylic acid (TCA) cycle for brain function. Intriguingly, we have found that macrophages and microglia highly express the key ketogenic enzyme 3-Hydroxy-3-MethylGlutaryl-CoA Lyase (HMGCL). This suggests that microglia can produce ketone bodies and that local levels of BHB in the brain may function as a regulatory metabolite that restrains runaway inflammasome activation. In addition, this suggests that medium-chain triglycerides (MCTs), which can cross the blood-brain barrier, serve as substrates for production of BHB in microglia. Given MCTs are under investigation to lower Alzheimer's disease severity, the mechanism of microglial-derived BHB as regulatory anti-inflammasome metabolite has high clinical impact. Thus, based on our findings, the central hypothesis of this project is that a ketogenic substrate switch underlies the regulatory microglial responses that protect against Alzheimer's disease by inhibition of CD33 and deactivation of the NLRP3 inflammasome. The corollary is that elevating CNS ketogenesis and BHB signaling may serve as an anti-inflammatory intervention against Alzheimer's disease.

Amyloid Beta Expression Protects the Brain from Herpes Simplex Virus

\$175,000



Robert D. Moir, Ph.D.

Assistant Professor of Neurology, Harvard Medical School/Massachusetts General Hospital

Alzheimer's disease is characterized by two hallmark pathologies. The first is accumulation of amyloid beta peptide outside neurons as insoluble amyloid plaques. The second is aggregation inside of neurons of tau protein as filamentous structures called neurofibrillary tangles. High amyloid loads in brain induce tauopathy, and amyloid/tauopathy together appear to cause the neurodegeneration associated with Alzheimer's disease.

The aggregation of amyloid beta and tau was thought to be intrinsically abnormal, with no physiological function and leading only to neuron-damaging pathology. However, we recently demonstrated that amyloid beta is a natural antibiotic that protects against infection. Moreover, the normal protective actions of amyloid beta involve the peptides' aggregation and eventual entrapment of invading pathogens within amyloid plaques. This raises the possibility of an infection etiology for amyloid plaque accumulation.

The infection hypothesis for Alzheimer's disease has a long history (Alois Alzheimer, who first described Alzheimer's disease in 1907, proposed an infection etiology for the disease). However, only recently has the hypothesis drawn widespread interest among Alzheimer's disease researchers. The increased focus on an infection etiology for Alzheimer's disease



Amyloid Beta Expression Protects the Brain from Herpes Simplex Virus (continued)

has been driven in part by the recent identification of multiple links between Alzheimer's disease Amyloid Beta Expression Protects the Brain from Herpes Simplex Virus (continued) and immunity genes, and our own findings on the antibiotic role for amyloid beta and amyloid plaques. During the last 12 months we have been investigating the possible role herpes simplex 1 (HSV-1) virus may play in inducing amyloid plaques using animal models. HSV-1 is the pathogen most strongly linked to Alzheimer's disease pathology. Our findings confirm amyloid beta protects against HSV-1 infection and that the invading virus particles are trapped and inactivated by amyloid plaques.

Here we request an additional six months of funding for our study to investigate a novel model of Alzheimer's disease pathology that has emerged from our findings. In this model we will test if HSV-1 infection can lead to amyloid plaque generation that, in turn, drives tau aggregation and generates neurofibrillary tangles. This would, for the first time, serially link viral infection, amyloid plaque deposition and neurofibrillary tangle generation in a single model. We believe confirmation of our model would be a significant advance in understanding the possible role of infection in Alzheimer's disease and likely would impact current and future treatment strategies.

Targeting Beneficial Innate Immunity in Alzheimer's by Interleukin-1 Receptor-Associated Kinase M (IRAK-M) Deletion

\$150,000



Terrence Town, Ph.D.

Professor of Physiology and Biophysics, Keck School of Medicine,
University of Southern California

A defining feature of Alzheimer's disease is brain accumulation of toxic plaques that induce memory loss. In the healthy brain, innate immune cells are protective; however, in an Alzheimer's patient's brain, these cells fail to prevent plaque formation. Innate immune cells express a molecule named Interleukin-1 receptor-associated kinase M (IRAK-M) that ensures immune responses to invading bacteria and viruses are kept under tight control. Yet, this type of immune response is dysfunctional in the Alzheimer patient brain. Our hypothesis is that "rebalancing" the brain's immune response by blocking IRAK-M will enable plaque clearance. With Cure Alzheimer's Fund support over this past year, we now report that removal of the anti-inflammatory IRAK-M gene from mice that develop amyloid plaques with age restores deficits in learning and memory. Thus far, this project has provided crucial data on the role of IRAK-M and the innate immune system in Alzheimer's disease, and this additional year of funding will expand on this work to deeply understand the nature of this beneficial brain immune response. This important work represents a major step toward developing a novel immunological therapy for this devastating disorder of the mind.

Microbiome

Understanding the ways in which microbes living in the body may affect brain health



Mechanisms by Which the Gut Microbiome Influences Amyloid Deposition and Neuroinflammation in Mouse Models of Alzheimer's Disease

\$250,000



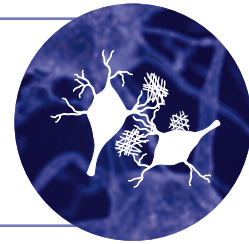
Sangram S. Sisodia, Ph.D.

Thomas Reynolds Sr. Family Professor of Neurosciences, Director of the Center for Molecular Neurobiology, University of Chicago

In our earlier proposal, we tested the hypothesis that the composition of the intestinal microbiome might play a key role in modulating neuroinflammation that, in turn, influences amyloid beta deposition. We have demonstrated that long-term treatment with an antibiotic cocktail (ABX) does not alter total bacterial abundance in either the cecum or feces, but rather induces a distinct perturbation in gut microbial diversity. The alterations in bacterial diversity are paralleled by selective changes in the levels of several circulating cytokines/chemokines in the blood sera. More importantly, we demonstrated that amyloid plaque deposition and plaque size are significantly reduced in the brains of male ABX-treated animals co-incident with an elevation in soluble amyloid beta peptides. Finally, ABX-induced perturbations in gut microbial diversity also influenced neuroinflammatory responses by conferring reduced plaque-localized gliosis and altered microglial morphology. Collectively, our findings indicate that the gut microbial composition regulates neuroinflammatory responses in the brain that ultimately can attenuate amyloid beta deposition. These studies serve as the foundation for our current proposal, in which we seek to define the mechanism(s) by which alterations in microbial diversity affect amyloid deposition and neuroinflammation. We now propose to investigate the role of the microbiome in modulating neuroinflammation, microglial phenotypes and amyloid beta pathology in an independent APPSWE/PS1L166P transgenic mouse model in which the transgenes are driven by a neuronal-specific Thy1 promoter. In addition, we will determine whether ABX-induced alterations in gut microbial community structure translate to alterations in specific metabolites (e.g., short chain fatty acids (SCFAs)) in APPSWE/PS1L166P and APPSWE/PS1ΔE9 mice. Finally, we will assess amyloidosis and neuroinflammation in germ-free APPSWE/PS1ΔE9 and APPSWE/PS1L166P mice and ascertain if reconstitution of the gut with microbial populations or microbial metabolites can alter phenotypes.

Pathological Pathways and Systems

Researching the underlying causes and mechanisms of Alzheimer's, such as amyloid beta accumulation, tau tangle propagation and inflammation



Understanding Reactive Astrocytes and Their Roles in Alzheimer's Disease

\$150,000



Ben Barres, M.D., Ph.D.

Professor of Neurobiology, Developmental Biology and Neurology,
Stanford University School of Medicine

We are investigating the mechanisms that cause neurodegeneration in Alzheimer's disease. Our recent studies have led us to realize that a toxic protein is unexpectedly secreted by a class of brain cells called astrocytes in the setting of Alzheimer's disease. Our goal in this proposal is to identify this protein so that in future studies we can test whether drugs that block the production or action of this protein will be useful as new therapies for Alzheimer's disease.

Together our findings from the first year of this project provide strong evidence that the formation of A1 neurotoxic reactive astrocytes is a fundamental neuropathological response of the mammalian brain to acute injury and neurodegenerative disease that has important implications for the development of new treatments for these diseases. We hope to develop new drugs that help patients with acute central nervous system injuries, and Alzheimer's disease and other neurodegenerative diseases, by targeting A1s in order to prevent their formation, or revert them back to normal astrocytes or to good/reparative A2 reactive astrocytes, or to block this neurotoxin or its receptor. Many questions remain about why the brain would ever generate a neurotoxic type of reactive astrocyte, however. One possibility is that these astrocytes evolved to fight off bacterial and viral infections, possibilities we have begun to investigate. Another interesting question is whether this neurotoxin is formed by epithelial cells in non-central nervous system tissues and participates in other diseases of cell death—for instance, Type 1 diabetes.



Intersection of Microglial Transcriptomes to Identify Key Alzheimer's Pathways of Brain Phagocytes

\$150,000



Sam Gandy, M.D., Ph.D.

Professor of Neurology and Psychiatry, Icahn School of Medicine at Mount Sinai Hospital;
Director, Center for Cognitive Health and NFL Neurological Care at Mount Sinai Medical Center

Computational analysis of gene expression profiles of moderate to severely affected Alzheimer's disease patients (Zhang et al., *Cell*, 2013) indicates the DAP12/TYROBP gene is an important "hub" or "driver" for the pathogenesis of typical, late-onset, sporadic Alzheimer's disease. This is the first genetic risk factor for this common form of the illness to be predicted by this new "big data" approach. Conveniently, this prediction dovetails well with a potent risk factor, the TREM2 gene, recently identified as mutated in some patients with late-onset Alzheimer's disease. In those patients, some TREM2 mutations appear to be equipotent with APOE4 in risk causation. This is notable because some had predicted that APOE4 is the most potent risk factor that would ever be identified in Alzheimer's disease. The computations network approach forms the core of the recently created multi-institutional National Institute on Aging Accelerating Medicines Partnership-Alzheimer's Disease (NIA AMP-AD) program. Herein, we propose comprehensive studies of genetically manipulated mouse brain neurons and microglia in order to advance our understanding of how the DAP12/TYROBP/TREM2 system exerts effects on Alzheimer's disease risk. The specific aim of this proposal is to characterize brain and microglial gene expression in mice that express a severe amyloid-forming phenotype and also are deficient in either DAP12 or TREM2. These mice were chosen because their pathological phenotype would be predicted to be the most similar to the pathology of the brains studied in the Zhang et al. *Cell* 2013 paper.

Uncovering Determinants of Neuronal Vulnerability in Alzheimer's Disease

\$250,000



Paul Greengard, Ph.D.

Head of the Laboratory of Molecular and Cellular Neuroscience,
Vincent Astor Professor, The Rockefeller University
Nobel Prize in Physiology or Medicine, 2000

Neurofibrillary tangles (NFTs) and neurodegeneration occur only in very specific regions at early stages of Alzheimer's disease (AD), while many regions remain virtually unaffected. Using the bacTRAP technology the lab developed to isolate mRNAs from specific neuron types, we molecularly profiled these very vulnerable neurons and other neurons that are much more resistant to pathological lesions of AD. We looked for genes enriched in vulnerable neurons compared with resistant neurons, and were able to pinpoint a list of these candidate vulnerability genes.



Modeling DNA Methylation Changes in Alzheimer's Disease Using Induced Human Pluripotent Stem (iPS) Cells

\$150,000

**Rudolf Jaenisch, M.D.**

Professor of Biology; Member, Whitehead Institute; Member, Institute of Medicine, Massachusetts Institute of Technology

Alzheimer's disease is associated with changes to DNA methylation, a modification of DNA that can alter the expression of genes in the brain. How DNA methylation changes contribute to Alzheimer's disease, however, has been hard to determine. Our proposed work aims to study the causes of DNA methylation changes in Alzheimer's disease and to better understand how these changes might affect Alzheimer's disease patients. To do this, we are using a "disease-in-a-dish" approach of generating human neurons with the hallmarks of Alzheimer's disease from human-induced pluripotent stem (iPS) cells.

The Role of Meningeal Lymphatics in Cleansing the Brain: Implications for Alzheimer's Disease

\$150,000

**Jonathan Kipnis, Ph.D.**

Harrison Distinguished Teaching Professor and Chair, Department of Neuroscience; Director, Center for Brain Immunology and Glia, University of Virginia School of Medicine

Blood vessels supply our organs with oxygen and nutrients. Another set of vessels, called the lymphatic vessels, perform other very important roles in the maintenance of tissues removing all the waste and toxic compounds the organ produces, and also serving as a path for immune cells from organs back to the lymph nodes. The mammalian central nervous system (CNS) previously was considered one of the only organs devoid of lymphatic vessels, and it was not fully understood how toxic compounds were removed from the brain. Our group has identified and described the basic biology of a novel meningeal lymphatic vascular system that serves a tissue clearance function for the brain. Absence of these meningeal lymphatics results in attenuated clearance of macromolecules from within the CNS. We will address the role of this main drainage pathway in regulation of amyloid beta removal, its dysfunction as a contributor to Alzheimer's disease and, finally, using a pharmacological approach, we will enhance its function and assess the effect on Alzheimer's pathology.



Cell Cycle Re-entry in 3-D Human Neuron Cultures

\$100,000



John S. Lazo, Ph.D.

Professor of Pharmacology and Chemistry, University of Virginia School of Medicine

George S. Bloom, Ph.D.

Professor of Biology and Cell Biology, University of Virginia School of Medicine

The well-known behavioral symptoms of Alzheimer's disease are caused by the loss of connections, or synapses, among neurons that control memory and cognition, and by the death of those neurons. A major goal of our labs is to unravel the seminal molecular pathways that convert normal healthy neurons into neurons that will die long before the Alzheimer's disease patients themselves. To that end, we recently made major strides toward understanding what may be the most common pathway for neuron death in Alzheimer's disease: cell cycle re-entry (CCR), which represents the aberrant reactivation of innate processes for neuronal cell division. Whereas normal, fully differentiated neurons never attempt to divide, up to 5 to 10 percent of the neurons in brain regions affected by Alzheimer's disease show signs of CCR over the course of many years. These neurons, which typically have duplicated much of their DNA, evidently never divide, but instead eventually die, and may account for as much as 90 percent of the massive neuron loss that occurs in Alzheimer's disease. During the past few years we have defined many features of a complex biochemical signaling web that causes CCR. This process is initiated by soluble amyloid beta oligomers (A β Os), which are the building blocks of the insoluble amyloid plaques that accumulate in Alzheimer's disease brain, and requires soluble forms of tau, the protein that aggregates inside Alzheimer's disease neurons to form insoluble neurofibrillary tangles.

Our principal strategy so far has been to model CCR in two-dimensional (2-D) cultures of mouse brain cells, and to test the *in vivo* relevance of our findings through parallel studies of transgenic Alzheimer's disease model mice and human brain tissue samples. Now we would like to test the hypothesis that A β O-induced, tau-dependent CCR can be observed in human neurons grown in three-dimensional (3-D) culture. Such cultures recently were shown to accumulate plaques and amyloid beta-dependent tangles, and thereby recapitulate human Alzheimer's disease features that have not been achieved by any other cultured cell model. If successful, this effort will establish 3-D cultures of human neurons as a viable platform for screening potential drugs that block CCR and for revealing new diagnostic markers for this seminal process in Alzheimer's disease pathogenesis.



Regulation of Microglial Lysosome Acidification

\$120,006



Frederick R. Maxfield, Ph.D.

Vladimir Horowitz and Wanda Toscanini Horowitz Distinguished Professor in Neuroscience and Chairman, Department of Biochemistry, Weill Cornell Medical College

Microglia are the main immune cells of the central nervous system. They normally carry out diverse functions, including removal of dead cells and other debris from the brain. Under some circumstances they have been shown to degrade Alzheimer's disease amyloid plaques in acidic organelles called lysosomes. We have shown that the acidity of microglial lysosomes is controlled by signaling processes, and that resting microglia in cell culture are ineffective at degrading amyloid because of poor lysosome acidification. In this project we will use modern optical imaging methods to measure the acidity of microglial lysosomes in living mice. We will test the hypothesis that mechanisms to regulate acidity that we have observed in cell culture also operate *in vivo*. Our goal is to manipulate these signaling processes to regulate degradation of amyloid plaques and inflammatory activation of microglia.

Will Restoration of Normal Glymphatic Function Slow Progression of Cognitive Decline and Amyloid Plaques in a Murine Alzheimer Model?

\$150,000



Maiken Nedergaard, M.D., D.M.Sc.

Frank P. Smith Professor of Neurosurgery; Co-Director, Center for Translational Neuromedicine, University of Rochester; Professor Departments of Neurology and Neuroscience, University of Copenhagen

This proposal will map glymphatic function as a function of aging in a mouse model of Alzheimer's disease. We also will test the hypothesis that exercise and improved sleep can slow the progression of cognitive decline by improving glymphatic clearance.



Early Role of Microglia in Synapse Loss in Alzheimer's Disease

\$150,000



Beth Stevens, Ph.D.

Associate Professor of Neurology, Harvard Medical School, Boston Children's Hospital;
Member, Broad Institute of MIT and Harvard

In Alzheimer's disease, our brain's major communication hubs—called synapses—are lost, leading to dementia. This loss of synapses occurs in specific areas of our brain, in particular an area called the hippocampus, where memory is formed and stored. A big question scientists are exploring is what makes synapses in the hippocampus vulnerable to damage and loss in Alzheimer's disease. Interestingly, we know more about how synapse loss occurs in the healthy developing brain, as synapse loss is a normal developmental process that is required for proper brain wiring. The brain makes many synapses during development that then need to be culled. A few years ago, our lab identified one of the key ways this normal synapse loss occurs in the developing brain. It involves a group of immune molecules called "complement," where complement molecules (C1q and downstream C3) mark specific synapses to be pruned. Then, brain immune cells called microglia eliminate the complement-marked synapses. Without the complement marking of synapses, we found that the brain was left with excess synapses.

In the mature healthy adult brain, when the remodeling phase largely is over, this pruning pathway is mostly turned off; that is, we see few complement proteins and also less phagocytic microglia in these brain regions. Our recent results suggest that complement and microglia can act as improper mediators of synapse loss in early stages of Alzheimer's disease. We show that blocking the complement pathway can lead to rescue of synapse loss in multiple models, using both genetic and antibody-mediated approaches. Thus, our results show complement and microglia are crucial components of synapse loss in the Alzheimer's disease brain. Furthermore, our data suggest that this pathway potentially can be targeted therapeutically, although more research is under way to study this in detail.



Systemic Inflammatory Networks in Alzheimer's Disease

\$200,000



Filip Swirski, Ph.D.

Associate Professor of Radiology, Massachusetts General Hospital/
Harvard Medical School

Matthias Nahrendorf, M.D., Ph.D.

Associate Professor of Radiology, Harvard Medical School, Director
of the Mouse Imaging Program at the Center for Systems Biology,
Massachusetts General Hospital

A common early symptom of Alzheimer's disease is short-term memory loss. As the disease worsens, symptoms can include problems with language, disorientation, mood and behavior changes, confusion about events, difficulty speaking, swallowing and walking. Alzheimer's disease is the most common form of dementia and worsens over time, accounting for approximately 70 percent of dementia cases. It is a neurodegenerative disease characterized by loss of normal brain function as a result of damage and destruction of nerve cells. The damage occurs when structures called plaques, which are protein deposits called amyloid beta, and tangles, which are fibers called tau, build up in the brain and interfere with nerve function. Brains of Alzheimer's disease patients also contain inflammatory cells, but the relevance of inflammation to disease development and exacerbation is unknown. Inflammation is a biological response to damage, stress and infection. It is a natural defense process that manifests itself as heat, pain, redness and swelling, playing an essential role in disease.

Depending on the extent, type and duration of inflammation, the process either can be helpful or harmful, because it can remove the offending pathogen, but it also can cause damage to healthy tissue. This grant will investigate how inflammation in the body and inflammation in the brain influence Alzheimer's disease. The main hypothesis of this project is that inflammation exacerbates Alzheimer's disease and is thus a major component of Alzheimer's disease pathology and a potential therapeutic target.



Role of Neurexins in Alzheimer's Disease Pathophysiology

\$150,000



Rudolph Tanzi, Ph.D.

Joseph P. and Rose F. Kennedy Professor of Neurology, Harvard Medical School; Director of the Genetics and Aging Research Unit and Vice Chair of Neurology, Massachusetts General Hospital

Pathogenesis of Alzheimer's disease is directly linked to levels of the toxic amyloid beta peptide in the brain. Amyloid beta levels and amyloid deposition increase in aging and in Alzheimer's disease, yet age-dependent factors that increase amyloid beta levels at the synapse remain largely unknown. In large family-based genome-wide association studies (GWAS), we recently have discovered a strong association between a neurexin gene and late-onset Alzheimer's disease, and identified a number of neurexin variants that are associated with increased risk for Alzheimer's disease. Neurexin, a transmembrane protein, expresses mainly at the presynaptic side of the neuron and plays an essential role in synapse formation, learning and memory. Our preliminary studies also identified neurexin as a novel amyloid precursor protein (APP)-interacting protein and have further shown that expression of neurexin in neuronal cells reduces amyloid beta generation. Association of neurexin with risk for late-onset Alzheimer's disease and reduced amyloid beta generation with neurexin-APP binding strongly advocate for an important role of the neurexins in the pathology of Alzheimer's disease. The goal of this project is to study the functional significance of neurexin and its genetic variants in regulating amyloid pathology in aging and Alzheimer's disease. Our studies will greatly facilitate the understanding of Alzheimer's disease pathology and may contribute to the development of novel strategies for the prevention and treatment of this debilitating disease.

Rejuvenation of Microglia in Brain Aging and Neurodegeneration

\$150,000



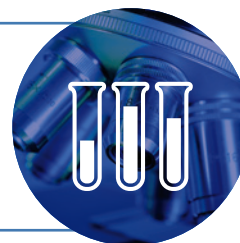
Tony Wyss-Coray, Ph.D.

Professor of Neurology and Neurological Sciences, Stanford University School of Medicine, Palo Alto Veterans Institute for Research

Aging impacts nearly every tissue and function in an organism, and the associated deterioration is the primary risk factor for major human diseases, including cancer, cardiac disease and such neurodegenerative diseases as Alzheimer's disease. The underlying cause of aging is likely a multifaceted yet interconnected tangle of processes, but there is growing evidence that in the brain, microglia—which are the only resident immune cell—have a major role. We discovered that these cells show profound changes with aging, and that soluble factors in the blood of young mice can rejuvenate these cells. We propose here to study how these cells age and what the mechanism of rejuvenation is. Our studies will help characterize the role of microglia in brain aging and Alzheimer's disease models, and may uncover new ways to rejuvenate these cells and slow down brain aging and degeneration.

Therapeutic Strategies

Identifying, testing and perfecting small molecules that could function as therapies for Alzheimer's disease



Development of Novel Amyloid Precursor Protein Dimerization Inhibitors That Lower Amyloid Beta Levels

\$129,373



Carmela R. Abraham, Ph.D.

Professor of Biochemistry and Medicine, Boston University School of Medicine

Diseases that affect learning and memory are of fundamental biological importance and are among the most challenging biomedical problems of our time. We recently demonstrated that compounds that inhibit amyloid precursor protein (APP) dimerization and enhance APP phosphorylation reduce the levels of amyloid beta, the peptide responsible for the neurotoxicity seen in Alzheimer's disease. In this project, we are further studying the mechanism of action and therapeutic potential of inhibitors of the proteins involved in the phosphorylation of APP and reduction of amyloid beta, and expect that results obtained from this study will produce candidate molecules for future studies aimed at reducing amyloid beta *in vivo* and in clinical trials of Alzheimer's patients.

Nanobodies to Cross the Blood-Brain Barrier

\$150,000



Bart De Strooper, M.D., Ph.D.

Director, VIB-KU Leuven Center for Brain and Disease Research; Professor of Molecular Medicine; Scientific Director, Department of Molecular and Developmental Genetics, KU Leuven; Visiting Professor; Director, UK Dementia Research Institute, University College London

Maarten Dewilde, Ph.D.

Staff Scientist, Laboratory for the Research of Neurodegenerative Diseases, KU Leuven

The blood-brain barrier (BBB) is a vital barrier between the bloodstream and the brain. This barrier tightly controls which molecules can enter the brain. As a consequence of this barrier, the majority of currently available drugs are unable to enter the brain. Importantly, to treat Alzheimer's disease, drugs need to reach the brain. The aim of this project is to generate a universal tool that can transport drug molecules into the brain.



Identification of a Protective Human Immune Response for Alzheimer's Disease

\$125,000



Charles Glabe, Ph.D.

Professor of Molecular Biology and Biochemistry, University of California, Irvine

Immunotherapy is a leading strategy for preventing cognitive decline in Alzheimer's disease. In 2015, Biogen-Idec reported the first disease-modifying treatment that prevented cognitive decline in human trials. It showed that high doses (10 mg/kg) of a monoclonal antibody that targets amyloid beta aggregates, aducanumab, prevented cognitive decline in mild cognitively impaired patients over a one-year period. While this is an exciting breakthrough, there are a number of problems that need to be overcome before this therapy is widely available. A number of side effects were noted that may be associated with the high amount of antibody administered, and the costs associated with the amount of antibody required will prevent its widespread utilization. Other monoclonal antibodies have shown a more limited effectiveness in clinical trials and some antibodies have shown no clinical benefit, raising the question of why some antibodies are better than others and whether we can find an antibody or combination of antibodies that is therapeutically more effective than aducanumab. Identifying a more effective antibody or antibody mixture is the goal of this proposal. We have devised a novel and innovative screening approach that has the potential to identify individual human antibodies that are associated with protection against Alzheimer's disease from the serum of Alzheimer's disease and unaffected humans. The goal of this project is to develop this peptide microarray and validate its ability to identify individual anti-amyloid antibodies.

Ultimately, we hope to use this screen to test whether a particular antibody can be identified that is associated with protecting people from Alzheimer's disease. This information also has the potential to identify a nonhuman peptide sequence that can be used to elicit a protective immune response, potentially a cost-effective and safe vaccine for Alzheimer's disease.

Activation of the 26S Proteasome for the Treatment of Alzheimer's Disease

\$150,000



Alfred L. Goldberg, Ph.D.

Professor of Cell Biology, Harvard Medical School

One fundamental feature of Alzheimer's disease (and several related neurodegenerative diseases) is the buildup in neurons of abnormal protein aggregates composed of the protein tau. One of the primary mechanisms that cells employ to prevent the accumulation of such misfolded, potentially toxic proteins is rapid degradation by the 26S proteasome, a degradative particle present in thousands of copies in all our cells. Our lab long has been investigating proteasome functions and molecular mechanisms. We recently made the unexpected discovery that the capacity of cells, including brain neurons, to destroy misfolded proteins, such as tau, can be increased by drugs that raise the levels of the



Activation of the 26S Proteasome for the Treatment of Alzheimer's Disease (continued)

cell-signaling molecule cAMP. A rise in cAMP causes a chemical modification and activation of the proteasome. These findings are exciting because they strongly suggest a new, rational approach for drug development against Alzheimer's and other neurodegenerative diseases.

In order to determine which treatment might be most effective in promoting tau degradation, we plan to compare the capacity of different phosphodiesterase inhibitors, a class of drugs that raise cAMP, to cause a similar modification and activation of brain 26S proteasomes. In addition, we shall investigate whether other signaling molecules also may activate the proteasome and enhance the degradation of misfolded disease-associated proteins. Finally, our recent collaborative studies showed that aggregated tau can be damaging to cells by impairing the function of proteasomes and thus disrupting cell regulation. We shall investigate further how these toxic proteins actually impair protein degradation by the proteasome and how neurons may compensate for such damage by increasing the production of new proteasomes or by inducing autophagy (a distinct cellular system for degrading aggregated proteins).

The APOE Mimetic Therapeutic Peptide CN-105 Attenuates Alzheimer's Disease Pathology and Improves Functional Outcomes in a Murine Model of Alzheimer's Disease

\$112,662



Daniel Laskowitz, M.D., M.H.S.

Vice Chair and Professor of Neurology, Neurobiology and Anesthesiology, Duke University Medical Center; Director of Neuroscience Medicine, Duke Clinical Research Institute; Director, Duke Neurovascular Laboratories

Increasing evidence suggests that brain inflammation plays an important role in mediating progression of Alzheimer's disease. In particular, it has been established that the APOE gene plays a critical role in mediating neuroinflammation and disease pathology. We have developed specific APOE-based peptides that are rationally derived from the receptor-binding region of this protein, and we have demonstrated that these compounds are well tolerated, cross the blood-brain barrier, and reduce brain inflammation in preclinical models of Alzheimer's disease and acute brain injury. We now test the hypothesis that chronic infusion of the neuroprotective pentapeptide CN-105 is well tolerated, and reduces progression of pathology in a clinically relevant animal model of Alzheimer's disease.



A Combination of Anti-Amyloid Beta and Growth Factor Therapy for Alzheimer's Disease

\$125,000



Mark H. Tuszynski, M.D., Ph.D.

Professor of Neurosciences, Director of Center for Neural Repair,
University of California, San Diego

We propose to test a combination of two potentially potent therapies for Alzheimer's disease: brain-derived neurotrophic factor (BDNF) and an anti-amyloid treatment (gamma secretase modulator (GSM)). In numerous animal models, modulation of amyloid beta levels (by immunotherapy or secretase blockade/modulation) has exhibited an ability to reduce Alzheimer's disease-related neuropathology and improve functional outcomes. However, amyloid beta-modifying therapies have yielded disappointing results in clinical trials in which treatment is initiated after disease onset; for this reason, current clinical approaches are focusing on treating pre-symptomatic patients. Separately, we and others have shown that the nervous system growth factor BDNF can reduce neuronal loss, stimulate synaptic markers, improve transcriptional activity and ameliorate behavioral deficits in animal models of Alzheimer's disease, ranging from amyloid precursor protein (APP) transgenic (APP tg) mice to aged nonhuman primates. We propose to test whether a combination of these therapies will exhibit additive or multiplicative benefits in animal models of Alzheimer's disease on molecular, cellular, biochemical and functional outcomes. The combination will be tested in APP tg mice (line 41) and on human induced pluripotent stem cell (iPSC)-derived neurons from Alzheimer's disease patients.

Thought leaders in the Alzheimer's disease field, together with the National Institutes of Health and the U.S. Food and Drug Administration, are encouraging the exploration of combinatorial therapies for Alzheimer's disease. We propose to pursue this promising approach using two potent candidate therapies. We have performed first-in-human clinical trials of gene therapy in Alzheimer's disease, and positive results of this work similarly could lead to human translation.

Acceleration of U.S. Food and Drug Administration-Required Good Laboratory Practice Gene Toxicity Studies With Gamma Secretase Modulator BPN-15606

\$144,450



Steven Wagner, Ph.D.

Associate Professor, University of California, San Diego School of Medicine

As a result of an Ames positive result in a single salmonella strain (T98), the U.S. Food and Drug Administration (FDA) is requiring that, prior to our pre-investigational new drug (IND) meeting with them, we perform a good laboratory practice (GLP) Ames assay with a form of BPN-15606 (besylate salt form) that is equivalent to what will be used in our IND-enabling



Acceleration of U.S. Food and Drug Administration-Required Good Laboratory Practice Gene Toxicity Studies With Gamma Secretase Modulator BPN-15606 (continued)

toxicity studies and Phase 1 clinical trials. In addition, the FDA recommended that we perform two additional *in vivo* gene toxicity assays in rodents: the Micronucleus assay and the Comet assay. This requires first converting the seven grams of the freebase form of BPN-15606 that we currently have available to the besylate salt form and characterizing this material with extensive analytical chemistry in order to show equivalence. The conversion chemistry (including analytical chemistry) will be carried out at Albany Molecular Research Inc. (AMRI). This will be a two-week process. The next step is to perform a GLP Ames assay with that material (besylate salt form of BPN-15606). The second step will be performing the *in vivo* Micronucleus and Comet genetic toxicology assays in rodents, which will be carried out at BioReliance. These three genetic toxicology tests will take approximately eight weeks. The final step will be the bioanalytical analysis of the plasma samples collected from the rodents that underwent the *in vivo* Micronucleus and Comet assays. This is in order to confirm that the rodents were exposed to the compound. This bioanalytical analysis requires a validated GLP bioanalytical assay. These latter studies will be carried out at the Stanford Research Institute (SRI).

Binding Site Characterization of a Novel Pyridazine-Derived Class of Gamma Secretase Modulators

\$194,950



Steven Wagner, Ph.D.

Associate Professor, University of California, San Diego School of Medicine

This research will identify the critical sites of interaction between novel pyridazine- derived soluble gamma secretase modulators (SGSMs) and their molecular target, as well as provide valuable information toward fostering an improved understanding of the mechanism by which these therapeutically relevant small molecules affect the production of specific amyloid beta peptide variants without inhibiting the enzyme's activity.

Despite the development of numerous potent SGSMs, the precise molecular target and the mechanism of action of this clinically relevant pyridazine series remain unknown. Our lab at the University of California, San Diego, and the Tanzi lab at Massachusetts General Hospital have developed an extremely promising series of soluble pyridazine-derived GSMs that inhibit the formation of the aggregation-prone amyloid beta 42 peptide in favor of shorter, less pathogenic amyloid beta isoforms.

We have synthesized an active pyridazine SGSM-photoprobe based on our SGSM currently under clinical consideration for cross-linking studies to demonstrate the binding site of these ligands within the gamma secretase enzyme. Additional experiments will be conducted using related GSM compounds, structurally unrelated yet mechanistically related GSMs, and gamma secretase inhibitors (GSIs) in competitive labeling studies in order to further elucidate the binding site within the gamma secretase enzyme. These experiments are critical for validating the mechanism of action of GSMs and differentiating this from that of gamma secretase inhibitors (GSIs), including the putative Notch-sparing GSIs.

Whole Genome Sequencing and Epigenetics

Assessing the entire human genome, including intergenic DNA, for variants that influence risk for Alzheimer's



CIRCUITS: Epigenetic Determinants of Human Cognitive Aging

\$199,600



Lars Bertram, M.D.

Professor of Genome Analytics, Institute for Neurogenetics, University of Lübeck, Germany

Much like many other human traits, cognitive decline and the development of Alzheimer's disease are determined by the concerted action of genetic, epigenetic and nongenetic factors. Over the last decade, genetics research in Alzheimer's disease has progressed at an unprecedented pace owing to the application of high-throughput genotyping technologies in the context of genome-wide association studies (GWAS). However, it is becoming increasingly evident that variants of the DNA sequence themselves do not fully explain the Alzheimer's disease phenotypic picture and that other mechanisms, such as those related to epigenetics, must make substantial contributions to disease development and progression. For this project, we propose to perform one of the largest epigenome-wide association studies (EWAS) to date on Alzheimer's disease-relevant neuropsychiatric phenotypes in an extremely well and deeply characterized cohort of healthy at-risk individuals from Berlin, Germany. In an auxiliary aim, we will correlate patterns of epigenetic variation in human brain samples with those derived from buccal swabs to facilitate the interpretation of our primary EWAS results. Together, the experimental data derived from this project will elucidate novel molecular mechanisms underlying cognitive decline and the onset of dementia.

CIRCUITS: Utilizing Functional Maps to Prioritize Therapeutic Targets in Alzheimer's Disease

\$225,308



Winston Hide, Ph.D.

Professor of Computational Biology, The University of Sheffield

Discovery of the causes of and treatments for Alzheimer's is confounded by the complexity of the disease, the interplay among environment and genetic bases of the disease and the disparate approaches taken by different groups looking at specific aspects of the disease. Progress has been slow, and there is an urgent need to deliver treatments that are effective and have few side effects. Current studies seek specific genes as treatment targets.



CIRCUITS: Utilizing Functional Maps to Prioritize Therapeutic Targets in Alzheimer's Disease (continued)

Usually there is a strong bias by each group as to which genes and processes they think are responsible for the disease. Failure rates are high.

This consortium will generate many different types of high-dimensional “omics” data and will integrate them together in an unbiased manner to systematically and objectively deliver the key processes and genes that appear to be responsible for the onset and progression of the disease. Using an existing industry-sponsored model pioneered at the Sheffield Centre for Genome Translation, this project will coordinate the data being generated by the consortium, and will ingest the disparate data—and the key genes and pathways resulting from them—to objectively rank the genes and pathways by their likely impact on the disease. In turn, ranked pathways then will be matched for their suitability for targeting by existing drugs. The resulting drug/pathway/gene models will provide invaluable reagents for industry and academia to assess in terms of their direct clinical outcomes on Alzheimer's treatment and progression.

CIRCUITS: Induced Pluripotent Stem (iPS) Cells and the Human Brain

\$145,004



Bradley T. Hyman, M.D., Ph.D.

Director, Massachusetts Alzheimer's Disease Research Center;
John B. Penney Jr. Professor of Neurology, Harvard Medical School; Alzheimer's Unit
Director, MassGeneral Institute for Neurodegenerative Disease

There is no doubt that induced pluripotent stem (iPS) cells derived from peripheral cells have enormous promise for personalized medicine, biomarker development, individualized treatment strategies and fundamental understanding of neurodegenerative disease. The ability to differentiate fibroblasts, for example, into relevant central nervous system cells, including cells that appear to be neurons and glia, is fascinating new science. Yet the connection between the cells that are in the dish, and the actual neurons and glia in the brain of the same individual, is truly unknown. A critical assumption of the field is that iPS cell-derived neurons and glia reflect the actual biology of the mature cells that have evolved in the brain, and then matured over the course of the person's lifetime. Importantly, this assumption has never been tested.

We propose to develop a resource to support the Collaboration to Infer Regulatory Circuits and to Uncover Innovative Therapeutic Strategies (CIRCUITS) consortium to overcome this critical and fundamental problem. Through our well-established neurodegenerative disease brain bank and tissue repository, we now have institutional review board approval to obtain fibroblasts at autopsy and grow them and convert to iPS lines for “omics” discovery, while at the same time using the same individual's brain tissue for 1) definitive diagnosis; and 2) newly developed homogenization/cell separation technologies to provide enriched populations of microglia, astrocytes, endothelial cells and neurons. We will be able to provide the matched fibroblast and isolated brain cell samples to investigators in the CIRCUITS consortium for “omics” analyses to understand in what ways iPS cells resemble, and in what ways iPS cells differ, from the cell populations they are intended to model; provide underlying deeply phenotyped data, both neuropathologically and clinically,



CIRCUITS: Induced Pluripotent Stem (iPS) Cells and the Human Brain (continued)

to CIRCUITS collaborators to provide a context for “omics” analyses; and explore new methods to culture the isolated cell types from post-mortem brain in culture to have an *in vitro* paradigm to test interventions directly in adult, mature brain cells.

CIRCUITS: Whole Genome Characterization of DNA Methylation Changes in the Aged and Alzheimer’s Disease Human Brain

\$250,000



Rudolf Jaenisch, M.D.

Professor of Biology; Member, Whitehead Institute; Member, Institute of Medicine, Massachusetts Institute of Technology

Joseph Ecker, Ph.D.

Professor, Plant Molecular and Cellular Biology Lab; Director, Genomic Analysis Laboratory, The Salk Institute for Biological Studies; Howard Hughes Medical Institute and Gordon and Betty Moore Foundation Investigator; Salk International Council Chair in Genetics

Alzheimer’s disease is the most common age-related neurodegenerative disorder. Both normal aging and Alzheimer’s disease have been correlated with changes to the patterns of DNA methylation in the brain. DNA methylation is an epigenetic mark with the capacity to stably alter gene expression. The importance of changes to DNA methylation in Alzheimer’s disease has been difficult to assess. This proposed work would characterize the alterations of genome-wide DNA methylation patterns in post-mortem human neurons in the context of normal aging and Alzheimer’s disease. We also will characterize DNA methylation patterns in human neurons generated *in vitro*. These foundational experiments are a critical first step in understanding the epigenetic mechanisms that contribute to Alzheimer’s disease.

CIRCUITS: Production Center for Reference and Variation Gene-Regulatory Maps

\$750,000



Manolis Kellis, Ph.D.

Professor of Computer Science and Head, Computational Biology Group, Massachusetts Institute of Technology; Member, Broad Institute of MIT and Harvard

Li-Huei Tsai, Ph.D.

Director of the Picower Institute for Learning and Memory; Picower Professor of Neuroscience, Massachusetts Institute of Technology; Senior Associate Member, Broad Institute of MIT and Harvard

Alzheimer’s disease is a devastating neurodegenerative disorder, afflicting 1 in 3 dying seniors and costing \$236 billion annually in the United States alone. Its prevalence is increasing rapidly in an aging population, and currently there is no cure. Recent genetic studies provide new hope for therapeutic avenues, but translating genetic results into therapeutics has been remarkably difficult, due primarily to the fact that most genetic mutations do not alter protein function directly, but instead affect the expression of nearby genes in subtle ways. Here, we seek to overcome this limitation by directly profiling the gene-regulatory differences in Alzheimer’s patients in order to understand the cell types, regulatory regions, target genes



CIRCUITS: Production Center for Reference and Variation Gene-Regulatory Maps (continued)

and upstream regulators whose function is affected in disease. We will profile epigenomic differences in Alzheimer's disease across 600 individuals, we will dissect the cell-type-specific action of these differences in neurons, astrocytes and microglial cells, and we will map the detailed circuitry of brain regulatory regions across Alzheimer's patients and controls. The resulting datasets will be released broadly to the scientific community, and also will form the foundation for computational and experimental work by the Cure Alzheimer's Fund Collaboration to Infer Regulatory Circuits and to Uncover Innovative Therapeutic Strategies (CIRCUITS) consortium, in order to translate the resulting datasets into mechanistic insights and new therapeutic avenues for Alzheimer's.

Analytical and Statistical Tools for Sequence Analysis for Alzheimer's Disease

\$250,000



Christoph Lange, Ph.D.

Professor of Biostatistics, Harvard T.H. Chan School of Public Health;
Assistant Professor of Medicine, Harvard Medical School

The availability of next-generation sequencing data in large-scale association studies for Alzheimer's disease provides a unique research opportunity. The data contains the information that is required to identify causal disease susceptibility loci (DSL) for Alzheimer's disease and many other mental health phenotypes and psychiatric diseases. In order to translate the wealth of information into DSL discovery for Alzheimer's disease, powerful statistical methodology is required. So far, a large number of rare variant association tests have been proposed. However, they do not incorporate all the important information about the variants. None of the existing approaches takes the physical location of the variant into account. Under the assumption that deleterious DSLs and protective DSLs cluster in different genomic regions, we will develop a general association analysis framework for Alzheimer's disease that is built on spatial clustering approaches. The framework will be able to handle complex phenotypes, e.g., binary, quantitative, etc., and be applicable to different study designs, e.g., family-based studies and designs of unrelated subjects. If the DSLs cluster, the increase of statistical power of the approach will be of practical relevance, enabling the discovery of DSLs. In the absence of DSL clustering, our approach will achieve similar power levels as existing methodology.

Furthermore, in order to test larger genomic regions for association, we will develop network-based association methodology. The network-based approach will have sufficient power for larger genomic regions than do existing approaches and, at the same time, will provide an intuitive understanding of the complex relationships among the variants that drive the association, fostering new biological insights. The approach can incorporate complex phenotypes and different design types. All the proposed methodology will be implemented in user-friendly software packages with existing user communities, e.g., PBAT, NPBAT and R. We will test, validate and compare the proposed approaches with the existing methodology, using large-scale simulation studies and by applications to the whole-genome sequencing family study for Alzheimer's disease from the Tanzi lab.



CIRCUITS: Interpreting Alzheimer's Disease-Associated Genetic Variation at Enhancer Regions

\$189,610



Andreas R. Pfenning, Ph.D.

Assistant Professor in Computational Biology, Carnegie Mellon University

Treating Alzheimer's disease is one of the greatest challenges we face in the coming years; the disease has the potential to have an enormous impact on human health. Despite its importance, there still are no highly effective treatments for Alzheimer's disease, due in large part to a limited understanding of the underlying disease mechanisms. Our laboratory, as a member of the Consortium to Infer Regulatory Circuits and to Uncover Innovative Therapeutic Strategies (CIRCUITS), aims to make progress toward a cure using genomic approaches. Starting from recent insights into the genetic basis of Alzheimer's disease, we will use a combination of machine learning and experimental techniques to systematically work toward the underlying biological processes, cell types, pathways and potential drug targets.

Search for Female-Specific Genetic Factors Contributing to Risk for Alzheimer's Disease

\$375,000



Rudolph Tanzi, Ph.D.

Joseph P. and Rose F. Kennedy Professor of Neurology, Harvard Medical School; Director of the Genetics and Aging Research Unit and Vice Chair of Neurology, Massachusetts General Hospital

This multidimensional investigation will seek to elucidate sex-linked factors that determine Alzheimer's disease risk, age of onset and rate of progression, powerful information that will contribute to the pursuit of a cure for both sexes. Women make up more than two-thirds of the Alzheimer's patient population, yet very little is known or understood about why this is the case or what it means about the disease's mechanisms of action, risk factors and progression. Epidemiological evidence suggests that a woman at age 65 faces almost twice the risk of developing Alzheimer's disease in her lifetime and nearly three times at age 75 than does a man of the same age, differences that are not explained by age of expected mortality alone. We propose to carry out a comprehensive, family-based association meta-analysis of all three Alzheimer's disease family sample Whole Genome Sequencing (WGS) datasets separately for each sex, an important and novel investigation since virtually no Alzheimer's disease genome-wide association study carried out to date has considered sex-based differences. More specifically, we also will calculate the overall genetic component explained by each of the more than 47 million single nucleotide variants (SNV) identified in the National Institute of Mental Health WGS project and proceed to estimate the genetic correlation/overlap between males and females. More specific appreciation of the factors that increase or decrease risk of disease, age of onset and rate of progression would benefit both sexes, yet the collection, analysis and publication of scientific data have not yet caught up to this recognized need. Common gender-specific lifestyles, X-linked genetic variants, female hormonal profiles over the lifecycle and other differences offer compelling subjects for investigation as contributing factors.



CIRCUITS: Functional Analysis of Alzheimer's Disease Risk Genes Using Human-Induced Pluripotent Stem (iPS) Cells

\$400,000



Li-Huei Tsai, Ph.D.

Director of the Picower Institute for Learning and Memory; Picower Professor of Neuroscience, Massachusetts Institute of Technology; Senior Associate Member, Broad Institute of MIT and Harvard

Manolis Kellis, Ph.D.

Professor of Computer Science and Head, Computational Biology Group, Massachusetts Institute of Technology; Member, Broad Institute of MIT and Harvard

The vast majority of people with Alzheimer's disease suffer from the sporadic (late-onset) form, the causes of which remain completely unknown. From studies involving thousands of people, researchers have identified a number of genetic variants that may increase one's risk for sporadic Alzheimer's disease. However, little is understood regarding why these small changes impact one's risk of developing Alzheimer's disease. In this work, we will use the cutting-edge genome editing technique CRISPR/Cas9 to introduce Alzheimer's disease-associated genetic variants identified through genome-wide analysis into reprogrammed human stem cells. We will differentiate human stem cells harboring these variants into various cell types populating the brain—including neurons, astrocytes and microglia—and study the effects of these variants in these different cell types. The proposed study will provide mechanistic insights into why some genetic variants found in the population may predispose some individuals to an increased risk for Alzheimer's disease.

Individual Projects (Other)

Studying Alzheimer's disease using compelling methods or perspectives not covered by our other Areas of Focus



Pathway Crosstalks Associated With Sex and Risk for Alzheimer's Disease

\$151,655



Murali Doraiswamy, M.D.

Professor of Psychiatry and Behavioral Sciences; Professor in Medicine; Director, Neurocognitive Disorders Program, Duke University School of Medicine

Some recent findings suggest that women with mild cognitive impairment may progress to Alzheimer's disease at faster rates than men. However, the biological basis of the sex differences in Alzheimer's disease still is debated and warrants a more detailed examination. We propose to use novel "big data" computational methodologies to discover how cross-talk among biological pathways may be linked to sex and cognitive decline in people at risk for Alzheimer's.

Modeling Neuronal Aging in Specific Subtypes of Human Neurons by MicroRNA-Mediated Neuronal Reprogramming

\$150,000



Andrew S. Yoo, Ph.D.

Assistant Professor, Department of Developmental Biology, Washington University School of Medicine

The ability to derive and grow human neurons in tissue culture from elderly individuals will offer invaluable tools to study how advancing aging, the strongest risk factor for Alzheimer's disease, affects neuronal properties later in life. My research team developed an experimental approach to convert (reprogram) skin fibroblast cells from human individuals directly into neurons without the usual requirement of reverting the cells back to stem cell stages. Our method utilizes small molecules called microRNAs, which can be combined with additional genetic factors to generate specific types of neurons. Here, we propose to devise a microRNA-based reprogramming technique to generate neuronal subtypes affected in early stages of Alzheimer's disease with high efficiency and specificity. Using this approach, we will generate human neurons from donors from varying age groups, and analyze age-related signatures in converted neurons across the age spectrum. If this project succeeds, we will be able to generate human neurons reflecting all ages, and to discover the biological changes that occur at different stages of life. With these powerful tools in hand, we then will be able to elucidate how neurons age and function differently across the age spectrum. Our work eventually will offer insights into the cellular properties intrinsic to aging neurons that make them susceptible to neurodegenerative diseases later in life. By devising biomarkers that indicate the aging status of neurons, our work ultimately will lead to an experimental platform to screen for drugs that one day may promote healthy brain function throughout life.

RESEARCH LEADERSHIP

Cure Alzheimer's Fund is guided scientifically by a Research Consortium, governed by a Board of Directors and administered by a small, full-time staff. Go to curealz.org/research/researchers to read the full bios of all of our researchers and curealz.org/about/people to read the full bios of all of our board members and staff.

Research Consortium

The volunteer members of Cure Alzheimer's Fund's Research Consortium develop and update our research areas of focus to identify the most promising opportunities for slowing, stopping and/or reversing Alzheimer's disease.

Members pursue their own research projects consistent with these priorities and others whose work will hasten development of effective therapies for and prevention of Alzheimer's disease.

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Scientific Advisory Board

A group independent of our Research Consortium, members of the Scientific Advisory Board advise and counsel Cure Alzheimer's Fund regarding the research priorities' overall scientific soundness. They also review individual grant proposals for consistency with the roadmap and with CAF's objectives.

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Cure Alzheimer's Fund is honored to have received
the prestigious designation of

PERFECT **100**

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the award is based on excellence in financial management
and transparency.

This award is in addition to the rating of 4 stars
received for the sixth consecutive time.



CHARITY NAVIGATOR

Four Star Charity





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Cure Alzheimer's Fund is a "doing business as" name for the Alzheimer's Disease Research Foundation,
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